

Spin Transition Molecular Materials: Intelligent Contrast Agents for Magnetic Resonance Imaging

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Abstract: Some preliminary results from a model of temperature-sensitive contrast agents are reported. This paramagnetic system switches from a spin $S = 0$ diamagnetic state to a $S = 2$ paramagnetic one at a temperature that can be tuned according to chemical composition. The magnetic susceptibility jump and the subsequent R_2^* effect as a function of temperature have been followed by means of spectroscopy, relaxometry, and imaging, demonstrating sharp and reversible transitions. Potential applications of this kind of system could be found in therapy by hyperthermia or in material science.

Introduction

After the successful phase of nonspecific paramagnetic chelates (Gd-DTPA, Gd-DTPA-BMA, Gd-DOTA or Gd-HP-DO3A), a second generation of contrast agents with an improved targeting ability has arisen (Gd-EOB-DTPA, Gd-BOPTA, etc.). Today, efforts are directed toward the development of markers for molecular imaging as well as of "intelligent" systems. The relaxivity of the latter ones should reflect local conditions such as pH, pO_2 , temperature, or the concentration of a particular metabolite.¹⁻⁷ A major problem of the compounds reported so far, however, is that the measured response, i.e., the variation of the image intensity, can be ambiguous, since it depends on both the relaxivity of the agent and its local concentration. In this work, we present preliminary results obtained on temperature reporters that, independently of their local concentration, switch from a magnetically inactive state to an active one at a given temperature. Such materials could be useful, for example, in the context of radiotherapy.

The most spectacular example of molecular bistability is the spin transition phenomenon, especially in the chemistry of iron.⁸⁻¹⁰ Some derivatives with 6-fold coordinated iron(II) show a transition between the diamagnetic state (spin = 0) existing under a critical temperature, T_c , and a paramagnetic state (spin = 2) above this one. When ferrous ions are linked in the solid,

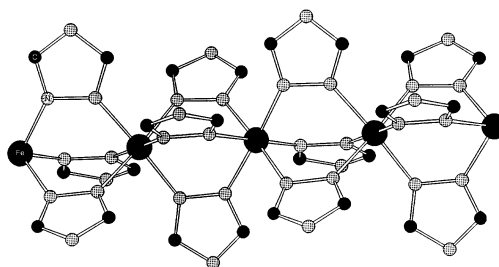


Figure 1. Polymeric structure of the particles.¹²

the transition $S = 0$ to $S = 2$ can become cooperative. The transition between the electronic states can then occur and shows a thermal hysteresis characterized by two distinctive temperatures $T_{c\uparrow}$ and $T_{c\downarrow}$, corresponding respectively to the warming up and to the cooling down pathways.^{11,12} Particles of Fe(II)/triazole/aminotriazole (Figure 1) exhibit such a molecular bistability and the temperatures of their magnetic hysteresis can be adjusted by an appropriate choice of the particle composition (triazole/aminotriazole stoichiometry).¹³ In the paramagnetic state, the particulate system is expected to induce strong magnetic field inhomogeneities which will alter, through T_2^* effects, the NMR signal in spectroscopy as well as in imaging. Since this polymeric structure of the compound is lost in water, coating of the particles will be required for use in aqueous media.

Experimental Section

Chemicals. 1,2,4-Triazole, aminotriazole, and ascorbic acid were purchased from Aldrich (Bornem, Belgium). Other organic solvents were purchased from Fluka (Bornem, Belgium). Particles of Fe(II)/triazole/4-aminotriazole were synthesized according to the method of Kröber et al.¹³

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Preparation of the $[\text{Fe}(\text{Htrz})_3](\text{ClO}_4)_2$.¹³ 1,2,4-Triazole (6 mmol, 414.4 mg) in 100 mL of methanol are added at room temperature to 100 mL of a methanolic solution of 2 mmol of hexahydrated iron(II) perchlorate (725.6 mg) and 10 mg of ascorbic acid. The mixture becomes limpid and is shaken for 1 h. The methanol is evaporated to dryness and the product is isolated as a white powder that becomes violet on cooling. The ascorbic acid prevents oxidation of the Fe(II) in Fe(III). Mixed samples are obtained according to the same experimental protocol by using appropriate quantities of 1,2,4-triazole and 4-amino-1,2,4-triazole.

Coated Particles. A 0.5 mL portion of collodion (nitrate of cellulose)¹⁴ is diluted in a mixture of 2 mL of dry ethanol and 20 mL of dry ether under vigorous stirring. Particles (1 g) are added to the solution and the suspension is shaken during 45 min at room temperature and then evaporated. Different samples were synthesized from particles with different spin transition.

Magnetic Resonance Spectroscopy and Imaging. Proton magnetic resonance spectroscopy was run on a Bruker AMX-300 spectrometer ($B_0 = 7.05$ T) (Bruker, Karlsruhe, Germany). The sample was prepared by adding 0.15–0.17 mL of liquid (cyclohexane or water, see below) to ca. 175 mg of particles so that the filling height of the soaked and packed crystals in the 5 mm o.d. NMR tube was approximately 2 cm.

The transition temperatures were defined as those corresponding to half of the line width variations. Considering the sensitivity of the raw particles to water, the preliminary study was run in cyclohexane, whereas water was used for the sample containing the coated particles. Images were obtained at 1 T on a Siemens Magnetom Impact system (Siemens, Erlangen, Germany) and at 4.7 T on an Avance-200 system (Bruker, Karlsruhe, Germany). Imaging experiments at 1 T were carried out on slurries of uncoated crystals contained in a phantom, the structure of which is shown in Figure 3. During imaging at 4.7 T, temperature was controlled by a BVT 3000 unit (Bruker, Karlsruhe, Germany) (calibration was performed with ethylene glycol ^1H spectrum).

Results and Discussion

In structures such as $[\text{Fe}(\text{Htrz})_3](\text{ClO}_4)_2$ (Htrz = 1,2,4-triazole), the metal ions are complexed by nitrogens in the 1,2 or, more rarely, 2,4 positions, giving polymeric structures that exhibit spin transition (Figure 1).¹² $[\text{Fe}(\text{Htrz})_3](\text{ClO}_4)_2$ is characterized by a $T_{c\uparrow}$ of 313 K and a $T_{c\downarrow}$ of 296 K.¹² These transition temperatures can be modified by partly substituting Htrz by 4-NH₂trz (4-amino-1,2,4-triazole) to get a doped compound of molecular composition $[\text{Fe}(\text{Htrz})_{3-3x}(4\text{-NH}_2\text{trz})_{3x}](\text{ClO}_4)_2 \cdot n\text{H}_2\text{O}$. Kröber et al. reported that $T_{c\uparrow}$ and $T_{c\downarrow}$ decrease when x increases from 0 to 0.1, according to equations: $T_{c\uparrow} = 313 - 180x$ and $T_{c\downarrow} = 296 - 160x$.¹³

Uncoated Particles. Spectroscopy. The stoichiometry $(\text{Fe}(\text{triazole})_{2.94}(\text{aminotriazole})_{0.06})$ was chosen in this study to fix the magnetic transitions around the physiological temperature ($T_{c\uparrow} = 309.4$ K and $T_{c\downarrow} = 292.8$ K). The line width at half-height of the cyclohexane protons of the sample containing two batches of particles displays a 6-fold increase when switching from the low spin state to the high one (Figure 2). The relatively large value characterizing the diamagnetic state is due to the inherent susceptibility of the “diamagnetic” material. The hysteresis phenomenon is very clear for both samples, but $T_{c\uparrow}$ and $T_{c\downarrow}$ are larger than expected ($T_{c\uparrow} \cong 312$ K and $T_{c\downarrow} \cong 308$ K). Between 295 and 316 K, the line width increases from approximately 5000 to 27 000 Hz. When the temperature decreases ($T = 292$ K), the line width returns to its initial value (Figure 2).

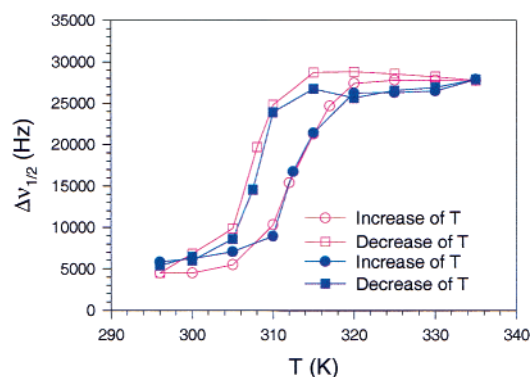


Figure 2. Line widths at half-height of the proton resonance of cyclohexane at 7.05 T as a function of temperature during heating and subsequent cooling (empty and filled symbols correspond to two different samples containing respectively 183 and 173 mg of crystals in 0.22 mL of cyclohexane).

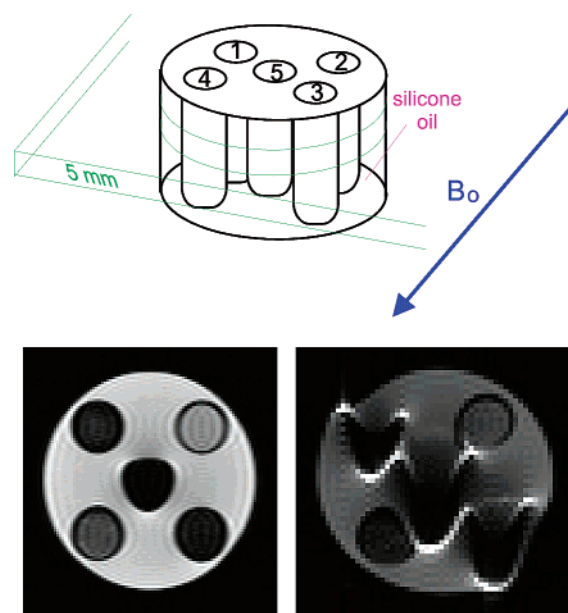


Figure 3. Imaging phantom [(1) 25% particles–75% $\text{KCl}/\text{C}_6\text{H}_{12}$, (2) C_6H_{12} , (3) 50% particles–50% $\text{KCl}/\text{C}_6\text{H}_{12}$, (4) $\text{KCl}/\text{C}_6\text{H}_{12}$, (5) 100% particles/ C_6H_{12}] and the corresponding spin-echo images obtained at 1 T below $T_{c\downarrow}$ (left-hand side) and above $T_{c\uparrow}$ (right-hand side) (TR = 1000 ms, TE = 12 ms). As expected, “dilution” of the material with diamagnetic KCl crystals induces a reduction of the extent of the artifact ($5 > 3 > 1$).

MR Imaging at 1T. Images of the phantoms recorded at low (298 K) and high (328 K) temperatures display a significant susceptibility effect of the material above the magnetic transition temperature (Figure 3). As expected from the geometry and the orientation of the sample, a typical arrowhead artifact is observed.¹⁵ The image on the left has been produced below $T_{c\downarrow}$ (and $T_{c\uparrow}$), while the one on the right has been obtained above $T_{c\uparrow}$ (and $T_{c\downarrow}$). The lower intensity of the signal arising from the silicon oil at high temperature is due to the increase of its proton longitudinal relaxation time.

Coated Particles. Spectroscopy. The coating of the particles with collodion described above does not alter the molecular bistability, as shown by the color change upon heating and subsequent cooling of the sample, but induces a decrease of $T_{c\uparrow}$ as measured on the dry sample ($T_{c\uparrow}$ decreased from 312 K for the noncoated sample to 306 K for the coated sample). The

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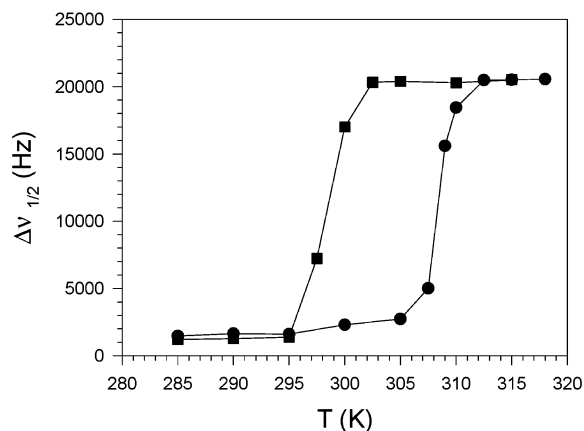


Figure 4. Line widths at half-height of the proton resonance of water at 7.05 T as a function of temperature during heating and subsequent cooling for $\text{Fe}(\text{triazole})_3$ particles coated with collodion.

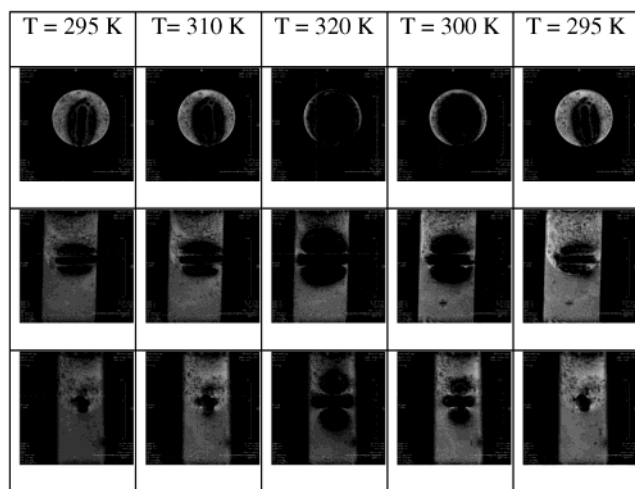


Figure 5. Axial (top), sagittal (middle), and coronal (bottom) images obtained at 4.7 T for five different temperatures with coated particles (heating from 295 to 320 K and cooling to 295 K).

collodion coating was therefore performed on particles of $[\text{Fe}(\text{Htrz})_3](\text{ClO}_4)_2$ to obtain a higher $T_{c\uparrow}$. The $T_{c\uparrow}$ and $T_{c\downarrow}$ estimated from the proton NMR line widths of water are approximately 308 and 298 K (Figure 4). Coated particles of collodion are stable in water. After 3 weeks, spin transitions are still visually

observed through the color shift. This spin transition can be performed several times without alteration of the phenomenon.

Imaging. Coated particles were incorporated in an agarose gel, but reproducible dispersion in the gel failed; therefore, the phantom was designed as a tube of 10 mm external diameter containing a layer of 4% agarose gel on which a closed polyethylene (Intramedic) tube containing particles was disposed. This tube was covered with a second layer of agarose gel. The images (Figure 5) were obtained with a gradient echo GEFI sequence (TR, 300 ms; TE, 11.7 ms; flip angle, 25°; FOV, 1.5 cm; slice thickness, 1 mm; three slices; matrix, 256 × 256; NA, 4). The temperature was first increased from 295 to 320 K and then decreased back to 295 K. Reversible susceptibility effects are clearly observed on the different axial, sagittal, and coronal slices.

Conclusions

The spin transition phenomenon appears to be a promising way to develop new intelligent contrast agents, the response of which could be tuned for specific conditions. The type of compounds tested in the present study could be used in the context of temperature monitoring in hyperthermia treatment of tumors.¹⁶ They would give a fast and unambiguous indication with respect to temperature limits, switching to a “homogeneity spoiling” state as a warning sign. It has to be stressed that this type of transition from a low to a high susceptibility when going from low to high temperature is quite unique. Hydrophobic coating with collodion has been shown to be appropriate for the use of such materials in aqueous medium, although some further investigation is needed to produce materials of suitable properties.

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