Synthesis and Physico-chemical Characterization of Nanomagnets Grafted by Biovectorizing Molecules

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INTRODUCTION:

One of the main challenges in medicine is to develop efficient and reliable tools for early and specific diagnosis of pathologies, among which cancers. One way is the use of nanomagnets grafted with molecules specifically targeting one kind of pathological cells. Superparamagnetic particles are used as contrast agents allowing for the detection of very small tumors by Magnetic Resonance Imaging (MRI) through their property to modify locally the signal intensity of the images. This contrast enhancement is due to stimulation of the water proton nuclear relaxation rates. The grafting of vectorizing molecules on the coating of these particles could affect their magnetic properties, hence their efficiency. The aim of this work is to describe i) the synthetic pathway for the production of nanomagnets grafted with vectorizing molecules and ii) the methodology of their physico-chemical characterization.

MATERIAL AND METHODS:

Colloidal nanomagnets coated with dextran are obtained by coprecipitation of a solution of ferric and ferrous ions with ammonia in the presence of dextran. The reproducibility of the synthesis is achieved by the use of a mini-reactor allowing an accurate control of the mixing conditions. The target molecules are linked covalently in two or three step-reaction sequence: epichlorhydrin is first coupled to the hydroxyl groups of the dextran coating of the Fe_3O_4 crystals to give a derivative with a terminal halogen derivative which can be used to link amine containing molecules (peptide, protein or ammonia). The folate group is subsequently bound to the aminated nanoparticle surface through the reaction between NHS-ester activated and the amino groups (figure 1). The NMRD profiles, which show the evolution of relaxivity (the increase of the relaxation rate of an aqueous suspension of particles containing 1 millimole of iron per liter) with the magnetic field were recorded at 37°C on a Fast Field Cycling Relaxometer (Stelar, Mede, Italy). The relaxivities (r_1) were measured over a field range extending from 0.24 mT to 1.2 T. Additional measurements at 20, 60 and 300 MHz were respectively obtained on Minispec PC-20 and Mq Series systems and on an AMX-300 spectrometer (Bruker, Karlsruhe, Germany). The values of the average sizes and specific magnetizations were obtained from the fitting of the magnetometric curves (magnetometer VSM-NUVO, MOLSPIN, Newcastle Upon Tyne, UK).

RESULTS AND DISCUSSION:

Relaxometric and magnetic characterizations of the nanomagnets are performed before and after the linking of reactive moieties. Magnetometric curves of the diversely grafted nanoparticles are not significantly different. The values of the diameter and of the specific magnetization given by the fitting of those curves by a Langevin function are similar for all particles. The vectorization has thus no marked influence on the magnetic and surface properties of the superparamagnetic crystal. Comparison between the relaxivity of vectorized and unvectorized particles reveals small, but significant differences of the longitudinal relaxivity i.e. an increase of the maximum of relaxivity in the NMRD curve of vectorized particles (figure 2). The theoretical fittings seems to indicate that the modification of the NMRD profiles results from a decrease of the water diffusion coefficient induced by the chemical modification. The vectorizing moiety seems thus to affect the properties of the coating. In addition, a twofold increase of the transverse relaxivity is observed (figure 3). This last feature is likely to be beneficial for the use of those systems as MRI contrast agents. One biological application of these vectorized systems is described in the poster presented by C. Burtea et al.

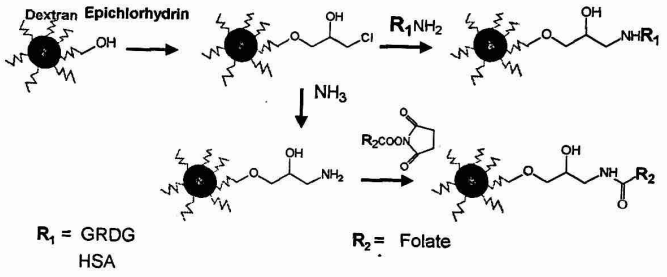

Figure 1: Synthesis of the grafted particles

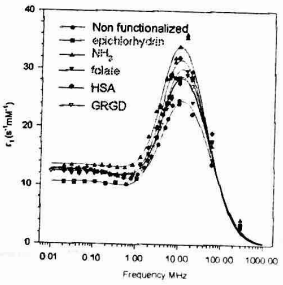

Figure 2: NMRD curves of the original and grafted superparamagnetic particles

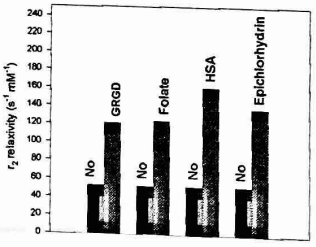

Figure 3: Transverse relaxivity of the particles

CONCLUSIONS:

Functionalization of the dextran coating of nanomagnets does not affect significantly the magnetometric properties but causes an increase of the maximum amplitude of the NMRD curve. The increase of the relaxometric efficiency of the functionalized particles at the magnetic fields used in MRI should thus be beneficial for their use *in vivo*.

ACKNOWLEGDMENTS:

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