Applicability of radioactive $^{99m}\text{Tc-O}_4^{-}$ magnetic fluid to nuclear medicine

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Abstract Magnetite nanoparticles were synthesized with solution of ferrous and ferric chlorides and ammonia water by sonochemical method. The hydrophilically radioactive magnetic fluids were prepared by labeling technetium pertechnetate ($^{99m}\text{Tc-O}_4^{-}$) and then adsorbing alginic acid on the magnetite particles. In order to measure some properties of the dispersed particles, the magnetic fluids were freeze-dried down to -70 °C, and were dried in vacuum. The total size of the particles was about 15 nm with the core diameter of 12 nm and their superparamagnetic saturation magnetization was 63 emu/g for the core-shell of Fe$_3$O$_4$/Algin and 52 emu/g for that of Fe$_3$O$_4/^{99m}\text{Tc-O}_4^{-}$/Algin. The labeling of radioactive $^{99m}\text{Tc-O}_4^{-}$ to the magnetite particles was efficient to about 70 %. The fluid of magnetic particles on which the radioisotopic substance is labeled with such an efficiency level may be applied as a tracer for diagnosis in nuclear medicine.

1. Introduction

Functional and biocompatible particles can be prepared by conjugating organic matters to an inorganic core. In general, such a core-shell combination is important in nano-bio technology: e.g., superparamagnetic magnetite nanoparticles [1] are moved in an external magnetic or electromagnetic field and adsorbing chemically drugs onto a colloid carrier [2] influences favorably their properties by decreasing the toxicity because a side effect due to direct administration can be avoided [3,4]. It is well-known that in order to transport the drugs to organs and tissues via blood flow, the intravenous injection is very effective. From this point of view, the permeability of colloid particles through the blood vessel is important in delivering the drugs to the target area for diagnosis and treatment. Since the capillary endothelial lining cells are finely connected in nervous systems of back bone, the very small particles can pass through the vessel wall and diffuse toward the outside. Therefore, a limit size of the particles is only about 20 nm for the kupffer cell in liver and the reticuloendothelial system of macrophage in spleen, medulla and lymph. The particles above the size of 50 nm are practically difficult to diffuse freely to the internal tissue because the tissues of muscle, skin and lung possess a continuous capillary vessel [5]. In order to be delivered to application parts by passing the endoderm of capillary and to be discharged out of the living body, it is desired that the magnetic particles are spherical with 10–15 nm diameter. Accordingly, the magnetic fluids need to be superparamagnetic with high saturation magnetization and also to satisfy simultaneously the
controlled particle size, the biocompatibility and the colloidal dispersibility for medical applications [6,7].

The substitution of Fe elements in the magnetite with a nuclide of copper is very effective for tumor therapy due to its β-emission and also γ-radiation close to the energy of Technetium-99m($^{99mTc}$) [8]. The $^{99mTc}$ is widely used as a radioisotope-tracer for diagnosis in nuclear medicine because it radiates only a single gamma ray and has a short physical half life of 6.02 h. In this work, superparamagnetic magnetite nanoparticles were synthesized by sonochemical method, whose colloid was obtained by adsorbing algicnic acid ($C_6H_8O_6$), a biocompatible surfactant, on the magnetic particles. Technetium pertechnetate ($^{99mTc}$-$O_4^{-}$) was labeled as a diagnosis agent on the magnetic particles. The applicability of the radioactive magnetic fluids was investigated by estimating the medical effect with calculation of the labeling efficiency.

2. Experimental

FeCl$_2$•4H$_2$O(1.72 g) and FeCl$_3$•6H$_2$O(4.70 g) with a stoichiometric ratio of 8.65 mmol Fe$^{2+}$/17.30 mmol Fe$^{3+}$ = 0.5 were solved in 80 ml of distilled water. The black precipitate of magnetic oxide was obtained by adding up to pH 10 ammonia water (28~30 % pure) under ultrasonication of 20 kHz at 140 W for 1 h to the mixed iron solution heated previously to 80 °C under Ar atmosphere [9]. The black gel was washed ten times by magnetic decantation until the pH value reached around 8. 50 ml of distilled water was added to the washed gel, which was heated to 80 °C under Ar atmosphere again. After 0.4~0.5 ml of technetium pertechnetate corresponding to triple of the practical dosage was diluted with 5 ml of saline, the radioactive solution was poured at a time into the gel solution in the ultrasonication of 30 min. The suspension was washed five times by magnetic decantation to remove the $^{99mTc}$-$O_4^{-}$ substance unlabeled on the particles. 60 ml of saline was added to the labeled magnetic particles, which was reheated to 80 °C under Ar atmosphere. The stable colloid was prepared by adding 25 ml of alginic acid 0.2 g to the solution of labeled particles in the ultrasonication of 1 h. The colloid volume was about 80 ml when cooled to room temperature. A process for preparing the $^{99mTc}$- $O_4^{-}$ labeled magnetic fluids by ultrasonication is shown in figure 1.

![Figure 1. Preparation process of technetium pertechnetate-labeled magnetic fluids by sonochemical method.](image-url)
Microstructure of the fluid particles were observed by transmission electron microscopy (TEM) where the crystallinity was analyzed by selected-area electron diffraction (SAED) and crystal phase of the particles were analyzed by X-ray diffraction (XRD). Their magnetic properties were measured in the field range of ±10 kOe using vibrating sample magnetometer.

3. Results and Discussion

In general, when a cation surfactant are adsorbed on the particles of iron oxide, the polar radicals of internal chemical adsorption layer are attached directly on the surface of core particles inwards and those of external physical adsorption layer face towards the outside. Since the weakly bonded external layer can be removed by washing with acid solution, the chemically bonded internal layer finally remains as the shell. The addition of anion surfactants to the colloid with this mono-molecular layered particles leads to the formation of the dual adsorption layer on the core particles by which the hydrophilic group is located outwards. Alginic acid, a kind of polysaccharide, possesses excellent chemical affinity due to the typical hydrophilic group. The water-based magnetic fluids can be prepared even with the monolayer of alginic acid on the magnetic particles which is formed by the adsorption process of only one step.

In order to obtain the layered particles from the magnetic fluids, the fluids were freeze-dried down to -70 \(^\circ\)C and then dried in vacuum using freeze-drier. In result, the ratio of weight of layered particles to fluid volume, i.e. colloid concentration was 27 mg/ml. Since the ratio of weight of core particles to fluid volume is 25 mg/ml from the formation amount of magnetite compound of 0.00865 mol × 231.54 g/mol = 2.0 g, the amount of shell is 2 mg/ml and totally becomes 160 mg. This shell weight corresponds mostly to that of the surfactant layer. Therefore, the addition of alginic acid 0.2 g seems to be sufficient to prepare the water-based magnetic fluid in this experimental condition.

Figure 2 shows that both of the magnetic particles with and without \(^{99m}\text{Tc-O}_4^-\) are spherical with the mean core diameter of 12 nm and the layer thickness of 2 nm to 4 nm, in which the SAED results exhibit high crystallinity.

![Figure 2. TEM images of magnetic particle fluids algin-adsorbed (a) with and (b) without \(^{99m}\text{Tc-O}_4^-\) labeling. The inserted SAED patterns represent high crystallinity.](image)

In figure 3, the XRD analysis revealed that the prepared fluid particles were highly crystallized as in the SAED with a Fe\(_3\)O\(_4\) (magnetite) phase. Although the Fe\(_3\)O\(_4\) may coexist with a \(\gamma\)-Fe\(_2\)O\(_3\) (maghemite) phase because their diffraction pattern is much the same [10], the iron oxide seems to consist mainly of Fe\(_3\)O\(_4\) in the process environment of Ar atmosphere. Also, the color of the synthesized powder was very close to the black of magnetite rather than the brown of maghemite. Lattice constants of the
magnetite particles algin-adsorbed with $^{99m}$Tc-O$_4^-$ label and with no label were measured as 0.83736 nm and 0.83727 nm, respectively. Such almost the same values in the lattice parameter suggest that labeling of a very small amount of technetium constituent has no effect on the crystalline state of magnetite particles.

Figure 3. XRD patterns of magnetic nanaoparticles algin-adsorbed with and without $^{99m}$Tc-O$_4^-$ labeling. The peak indices indicate crystal planes of magnetite.

Figure 4 shows that the synthesized particles are superparamagnetic with no remanent magnetization. Also, saturation magnetization for the core-shell systems of Fe$_3$O$_4$/Algin and Fe$_3$O$_4$/^{99m}Tc-O$_4^-$/Algin was 63 emu/g and 52 emu/g, respectively, while that of the only core particles of Fe$_3$O$_4$ was 66 emu/g. This decrease in the magnetization value is caused practically by the increase of weight ratio of nonmagnetic alginic acid and technetium pertechnetate layers.

Figure 4. Magnetization curves of magnetite nanoparticles algin-adsorbed with and without $^{99m}$Tc-O$_4^-$ labeling.
In general, the decay activity $A_d$ of radioisotope is calculated as in equation (1).

$$A_d = A_i \left(\frac{1}{2}\right)^{T/T'}$$ (1)

where $A_i$ is the initial activity measured from a radioactive substance in radiation counter before labeling, $T$ is the time elapsed until the last measurement after labeling and $T'$ is the physical half life of radioisotope. The percentage of labeling efficiency $L_{eff}$ is evaluated with $A_d$ as in equation (2).

$$L_{eff} = \frac{A_t}{A_d} \times 100$$ (2)

where $A_t$ is the terminal activity measured from a radioisotope-labeled material in radiation counter. The addition of technetium pertechnetate to the magnetite fluids was carried out three times repeatedly for reliability. Table 1 represents the numerical values of main factors observed in the labeling experiment.

| Experi. No. | Initial activity $A_i$ (mCi) | Elapsed time $T$ (min) | Decay activity $A_d$ (mCi) | Terminal activity $A_t$ (mCi) | Labeling efficiency $L_{eff}$ (%) |
|-------------|-----------------------------|------------------------|--------------------------|-------------------------------|-----------------------------|
| 1           | 35.6                        | 120                    | 28.3                     | 20.4                          | 72.1                        |
| 2           | 33.0                        | 160                    | 24.3                     | 16.6                          | 68.3                        |
| 3           | 22.7                        | 250                    | 14.1                     | 9.6                           | 68.1                        |

The labeling efficiency of technetium pertechnetate was satisfactory at the level of about 70 % even though the improved adsorption of the radioisotope would be more effective in observing the internal organs. It is suggested that the $^{99m}$Tc-O$_4^-$ is chemically well bonded on the surface of the magnetite particles. It has been also reported that the adsorption rate of 5-aminolevulinic acid for photodynamic therapy onto the magnetite nanoparticles was only 12.5 % [11]. A distribution experiment on animals of such radioactive fluids will be hereafter carried out to apply to nuclear medicine.

4. Conclusions
Magnetite nanoparticles were chemically synthesized with the addition of ammonia water to a solution of ferrous and ferric chlorides in ultrasonication. Radioactive magnetic fluids were prepared by labeling technetium pertechnetate ($^{99m}$Tc-O$_4^-$) and then adsorbing alginic acid on the particles. The saturation magnetization of superparamagnetic magnetite particles was 63 emu/g for the core-shell system of Fe$_3$O$_4$/Algin and 52 emu/g for that of Fe$_3$O$_4$/^{99m}Tc-O$_4^-$/Algin. The size of the fluid particles was 12 nm for the diameter of magnetite core and 2~4 nm for the thickness of $^{99m}$Tc-O$_4^-$ and algin. The labeling efficiency of $^{99m}$Tc-O$_4^-$ on the magnetite core particles was a high level of 70 %. Therefore, the $^{99m}$Tc-O$_4^-$ labeled magnetite fluids could be potentially used as a tracer for diagnosis in nuclear medicine.

Acknowledgments
This work was supported from Research Center for Advanced Magnetic Materials of Chungnam National University and by Basic Research Program of Agency for Defense Development.

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