Development of magnetic force-assisted gene transfer system using biopolymer-coated ferromagnetic nanoparticles

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Received 5 January 2006; received in revised form 10 February 2006; accepted 11 February 2006
Available online 19 May 2006

Abstract

The concept of drug-accumulation in the blood vessel utilizing magnetic force was a novel concept in the cancer treatment. In the present study, calculation of the applied magnetic force for the ferromagnetic particles inside the blood vessel was performed in order to clarify the effect of magnetic field gradient on the accumulation possibility of the magnetic particles and the accuracy of the targeted site in the blood vessels. The development of a two-dimensional (2D) navigation system of ferromagnetic particles in a flow system was performed. In order to improve the practice of using externally-applied magnetic fields for targeting the magnetic particles to a circumscribed body region, we tested the feasibility of a novel navigation system, made by applying a strong external (magnetic) field through a GdBaCuO bulk superconductor. A theoretical model is proposed and used in order to evaluate the efficiency of the navigation/retention of magnetic particles in the flow system. Furthermore, an experimental model system was made and the efficiency of a prototype system was examined. Comparisons of experimental and the corresponding calculation results were made to design the magnet for magnetically targeted drug delivery system in practical use.

Keywords: Drug delivery system; Gene therapy; Magnetic targeting; Bulk superconductor; Ferromagnetic particle

1. Introduction

Progress in gene therapy requires a novel drug delivery system. Magnetically targeted drug delivery system (MT-DDS) is a candidate of the new regional drug therapies presently investigated for cancer treatment [1]. The striking advantage of MT-DDS is the reduction of side effects of chemotherapeutic agents for the patient due to applying the agents mainly to the desired target volume at the tumor. As the first stage of development in MT-DDS, we prepared the magnetic drug for gene therapy, which is based on vectors bound to ferromagnetic magnetic nanoparticles such as magnetite (Fe₃O₄) and maghemite (γ-Fe₂O₃) (diameter:15–40 nm) coated by biocompatible polymer [1]. For the second stage, a magnet for MT-DDS should be designed, built and tested, because the accumulation of the magnetic vectors in the tumors vasculature is the key step for determining the efficiency of MT-DDS. In general, once the magnetic vectors were injected to artery, a magnet positioned close to the tumor drags the magnetic vectors from the artery to the vasculature of the tumor and finally concentrates them in the tissue to be treated. The magnetic field has to be applied for a certain time, until the bond between the vectors and the magnetic particle decays and the DNA in vectors has transferred into the tumor-cells. This step basically depends on the interplay of the magnetic and hydrodynamic forces acting on the magnetic vectors and on their adhesion to the walls of the blood vessel [2]. The magnetic force in turn depends on the magnetic moment of the magnetic vectors and the gradient of the applied magnetic field. However, when MT-DDS is adopted as a therapeutic approach, the current approach of using only an external magnet faces several challenges [3]. One problem is associated with the high blood velocities, typically varying between 100 and 500 mm/s in large arteries and veins, imposing adverse hydrodynamic conditions on the magnetic collection of magnetic drug carrier particles at the target site. Also, because the magnetic field intensity decreases abruptly
Therefore, magnetic force $F$ on the bioflow velocity $U$ is given by:

$$F = k V, \quad \text{where} \quad k = \mu_0 M_0 \text{H}$$

$m$ is the mass of the ferromagnetic particle, $\mu_0$ is the permeability of free space, $M_0$ is the magnetization of the ferromagnetic material, and $\text{H}$ is the magnetic field strength.

In order to experimentally clarify the effect of strong magnetic field gradient on the accumulation possibility of the magnetic particles, the model system where the magnet was externally placed at the almost same distance from the flow tube was used and the accumulation test was achieved.

## 2. Calculation method

### 2.1. Calculation of the trajectory of ferromagnetic particle in cylindrical tube

A 2D schematic of particle control system utilized for modeling the targeting of magnetic particle by the magnetic force arising from the magnet placed outside the blood vessel is represented in Fig. 1. Forces that act on the magnetic particle are shown in this figure. The particle feels viscous drag force when it begins to move. On the other hand, magnetic force acts on floating ferromagnetic particles when the particles in the cylindrical tube are placed under the magnetic field.

When the magnetic particle is assumed to be spherical, energy $U$ arising from magnetic force is as follows:

$$U = -\frac{1}{2}mH = -\frac{2}{3} \pi b \mu_0 H^2 (\chi_p - \chi_f)$$

where $\chi_p$ and $\chi_f$ are magnetic susceptibilities of the particle and fluid, respectively. And $b$ is a radius of the magnetic particle. Therefore, magnetic force $F_M$ acts the magnetic particle as follows:

$$F_M = -\Delta U = \frac{4}{3} \pi b^3 \mu (\chi_p - \chi_f) \nabla H$$

Magnetic force is product of magnetic field strength and magnetic gradient.

When velocity $v_p$ of the magnetic particle is different from the blood flow velocity $v_f$, the particle is subjected to the drag force $F_D$ from the fluid, and thus Stokes’s expression can be used on the magnetic particle:

$$F_D = 6 \pi \eta b (v_f - v_p)$$  

where $\eta$ is a viscous coefficient of the blood.

The inner walls of the tube are assumed to be two parallel planes also placed perpendicular to the plane of the figure, as shown. The blood accesses the inner wall from the initial position located in the left side in this figure with a velocity defined by a parabolic profile of initial velocity $v_0$ and it transports the magnetic particle to be captured by the wall. Finally, the magnetic particle is subjected to a homogeneous magnetic field $H_0$ positioned perpendicular to the blood flow. The model accounts for magnetic force and hydrodynamic force while neglects the effect of gravity force as well as any effect due to the inner walls. The fluid dynamics in the cylindrical tube is described by the Eqs. (2) and (3) mentioned above for an incompressible Newtonian fluid.

The trajectory of the magnetic particle is calculated by using Eqs. (2) and (3). When we initially set the magnetic particle in the cylindrical tube with a velocity defined as an initial velocity $v_0 = v(S_0)$ at the location, $S_0[X_0, Y_0]$ shown in Fig. 1, a particle acceleration, $A_0 = F(S_0)$ at the $S_0[X_0, Y_0]$ is calculated using the following equation:

$$F = \frac{F_D + F_M}{m_p}$$

where $m_p$ is a mass of the magnetic particle. The second position, velocity, and acceleration are calculated using the following equations:

$$S_2 = S_1 + v_1 dt + \frac{1}{2}A_1 dt^2$$

$$v_2 = v_1 + A_1 dt$$

$$A_2 = F(S_2)$$

Therefore, the $n$th position, velocity, and acceleration after $n$th infinitesimal time are represented by:

$$S_n = S_{n-1} + v_{n+1} dt + \frac{1}{2}A_{n-1} dt^2$$

![Fig. 1. Schematic of the controlling particle system utilized for modeling the targeting of magnetic particle inside the blood vessel using by the magnetic force. 2Rf is a diameter of the blood vessel. The final position $[X_n, Y_n]$ is calculated based on the equations of drug force ($F_D$) and magnetic force ($F_M$) using a recursive formulae.](image)
The calculation was made using recursive formulae for the particle with an averaged diameter of 2 mm of magnetite under the various magnetic gradients from 1 to 500 T/m in the blood flow (averaged flow velocity: 100 mm/s).

2.2. Simulation of the trajectory of ferromagnetic particles in Y-shape tube

The motion of ferromagnetic particles in the blood stream is modeled using by a two-dimensional system, which consists of the Y-shape tube of flow system and superconducting magnet generating 100 T/m of magnetic gradient. The schematic of the controlling particle system for the magnetically targeting inside the tube was shown in Fig. 2. The particle feels a viscous drag force when it begins to move. Simultaneously, the magnetic force acts floating ferromagnetic particles. The parameters used for calculating the particle trajectory are summarized in Table 1. The black and gray color in the flow system represents a measure of the flow velocity. The flow velocity is high at the center of the tube (black color), and low near the inner surfaces of the flow system (gray color). The velocity distribution of the flow system was calculated using ANSYS 9.0. In Fig. 3, calculated magnetic flux density using ANSYS 9.0 induced by the magnet placed outside the flow system was also shown. The black and gray color in the figure represents the strength of the magnetic flux density (gray color represents strong and black color weak).

The trajectory of the magnetic particles is calculated according to the following model. When the magnetic particle is assumed to be spherical, the total energy $-\Delta U$ arising from magnetic force is represented by the Eqs. (1) and (2). When velocity $v_p$ of the magnetic particle is different from the flow velocity $v_f$, the particle is dragged by the fluid and the drag force $F_D$ can be expressed using Stokes’s law as the Eq. (3).

Therefore, the equation to represent the particle motion is

$$ F = ma = F_M + F_D $$

Substituting (2) and (3) into (11),

$$ \frac{dv_p}{dt} = \frac{6\pi \eta b}{m} \left( v_p - v_f + \frac{F_M}{6\pi \eta b} \right) $$

and solving, we obtain

$$ v_p = v_f + \left( \frac{F_M}{6\pi \eta b} + c_1 e^{(-6\pi \eta b/m)} \right) $$

where $c_1$ is a constant of integration. Thus, the displacement, $S_p$ can be written as

Table 1

| Calculation parameters                        | Values          |
|----------------------------------------------|-----------------|
| Inner radius of simulated blood vessel       | 2 mm            |
| Velocity in X-direction ($v_f$ max)          | 200 mm/s        |
| Diameter of the ferromagnetic particle       | 2 µm            |
| Magnetic susceptibility of ferromagnetic particles | 0.1            |
| Saturated magnetization of ferromagnetic particles | 0.2 T          |
| Viscosity $\eta$                             | 0.0028 m Pa s   |
| Mass density of the ferromagnetic particle   | 5000 kg/m³      |
where \( c_2 \) is a constant of integration.

To obtain \( c_1 \) and \( c_2 \), we calculate \( S_p \) and \( v_p \) at two different times \( (t_N, t_{N+1}) \) and use \( v_f \) and \( F_M \) calculated through ANSYS 9.0.

Substituting Eqs. (15) and (16) into Eqs. (13) and (14), we obtained \( v_N \) and \( S_N \) at \( t=t_N \). When we calculate the integrated constant, \( v_f \) and \( F_M \) at \( t=t_N \) are taken from the analyzed data using ANSYS 9.0 for magnetic field and flow system, respectively.

3. Calculation result

3.1. Calculations of magnetic gradient for accumulation of the magnetic particles

The calculated particle trajectory was shown in Fig. 4. This figure can allow us to realize how strong magnetic gradient is needed to accumulate the magnetic particle on the wall of the blood vessel. For example, when we apply the magnetic gradient of 500 T/m, a magnetic particle can be easily accumulated on the wall. Simultaneously, this figure can give us the information how long distance will be required to accumulate the particle on the wall of the blood vessel even in blood flow. Fig. 5 clearly shows an effect of the magnetic gradient on the particle displacement under the blood flow. The displacement does not change so much in the range of magnetic gradient over 100 T/m.

3.2. Simulations results of the trajectory of the magnetic particles in Y-shape tube

The calculated particle trajectory is shown in Fig. 6. In Fig. 6(b), the accumulated particles are seen at the right-bottom wall of the flow system. This result indicates the feasibility of the magnetically targeted drug delivery system. The simulation results also show the efficiency of the accumulation of the magnetic particles at the target site (final position). In the simulation without the magnet (Fig. 6(a)), 50% of the particles go into upper tube after branching point and the rest of the particle (50%) go into the lower tube. This result shows that the accumulation does not occur. On the other hand, in the case of simulation with the magnet (Fig. 6(b)), the number of the accumulated particles in the lower tube after the branching...
point was 2.7 times higher than that or those going to upper tube. But, 27% of the number of the particles was captured before branching point due to the shape of the induced magnetic field. The obtained results clearly show that the magnetic accumulation in the flow system was possible using the superconducting magnet even in the case where magnet was placed outside the flow system.

4. Experimental

4.1. Magnet

A photograph of the bulk type of superconducting magnet system used in this experiment was shown in Fig. 7. The magnet was a GdBaCuO bulk superconductor (Nippon Steel, Japan) of cylindrical shape (45 mm large and 15 mm thick). Magnetization was performed by the pulse magnetization method (PFM) that can be done with copper wire [4]. The bulk superconductor can generate a strong magnetic field in an open space. Therefore, the magnetic field generated by bulk superconducting magnet is wide range and feasible to MT-DDS. Fig. 8 shows the magnetic flux density of a vertical element of the magnet against the distance from the surface of the cover. The calculated value of magnetic flux density with the distance shows good agreement with the measured values (Fig. 8). This suggests that the result of the magnetic field analysis by the finite element method is correct and can allow us to design the magnetic field and the magnet itself.

4.2. Experimental model system

The photograph shown in Fig. 7 also involves the experimental model system. The Y-shape tube with a branching point was placed at the same distance in the case of a rat where the vena cava was assumed to be located at about 35–50 mm from the body surface of a rat. The variation of special position of the vena cava from 35 to 50 mm is dependent on the size of the rat body.

The size of the model system was 90 mm × 90 mm × 95 mm (H), made of acrylic box. The diameter of the tube was 2 mm
and the length was 100 mm. The center of the tube in the flow direction has a branching point with an angle of 60°. Ferromagnetic particles flow from the left side (corresponding to the initial position in Fig. 2) and feel the drag force and the magnetic force. These ferromagnetic particles have an averaged diameter of 2 μm and a magnetic susceptibility of 68 emu/g prepared by a precipitation process in aqueous solution. The XRD pattern of the prepared particles gives only the magnetite peaks. The reason why we used the 2 μm ferromagnetic particles is that observation and video recording is so easier than using smaller particles as to prove the concept of the magnetic targeting using superconducting magnet.

5. Result and discussion

The obtained result of the accumulating test using a glass tube clearly shows that the ferromagnetic particles are accumulated in a local part of the glass tube by the magnetic force in the both cases where superconducting magnet was placed at the distance of 35 and 50 mm. This experimental result shows that particles can be accumulated, as predicted (see Fig. 6(b)) even at a flow velocity of 200 mm/s. The rough estimation of the accumulation efficiency was made by UV absorbance. About 70% of particles were found to be accumulated by magnet, which is almost the same value as obtained by simulation.

Ally et al. [5], Grief et al. [6], and Asmatulu et al. [7] have been individually investigating the magnetic targeting in model systems where the magnetic gradient is ranging of 0.03–10 T/m. Most of the papers on the magnetic targeting have been dealing with accumulation under weaker magnetic gradient such as 1 T/m, because their idea is basically based on the usage of the traditional electromagnet or permanent magnet. Therefore, their experimental work and/or calculations can demonstrate the possibility to accumulate the ferromagnetic particles when the magnet was placed at the very close to the flow tube. On the contrary, in the present work, we assumed to utilize superconducting magnet for magnetic targeting. Therefore, calculations of the magnetic gradient were emphasized on the range from 1 to 500 T/m. As a result, it was found that accumulation of the ferromagnetic particles with the size of 2 μm under the blood stream requires the gradient of 100 T/m at least for the practical application. In this sense, this paper will open the way for usage of the superconducting magnet for magnetic targeting.

6. Conclusion

This theoretical and experimental study shows the feasibility of a novel magnetically targeted drug delivery system, where a strong magnet can accumulate magnetic micro/nanoparticles in the flow system. In the present paper, an in vitro flow model system was established to examine the theoretical model and evaluate the efficiency of bulk superconducting magnet to accumulate of ferromagnetic particle, 2 μm diameter, inside flow system using an externally placed magnet at the distance of 35–50 mm from the flow system. In vitro flow experiments were used to check the accuracy of the theoretical model of a strong magnet for accumulating magnetic particle under flow system. Comparisons of experimental and corresponding modeling data verified theoretical predictions. A further study using a bulk superconducting magnet confirmed the efficiency of accumulating particles even at flow velocities of 200 mm/s. The obtained results show that the magnetic accumulation in the flow system was possible even in the case where magnet was placed at the distance of 35–50 mm from the flow system.

Two important findings were obtained from this study. The first is that our 2D computational approach is sufficient to examine the relatively simple magnetic system, as we see good correlation between model and experimental results. Secondly, the capture efficiencies are sufficiently enhanced with the placement of the bulk superconducting magnet to warrant further investigation into the clinical efficacy of this magnetic drug targeting system.

Acknowledgements

The authors would like to thank Prof. Hiroyuki Fujishiro (Iwate University, Faculty of Engineering) for allowing us to use the bulk superconducting magnet, many useful discussions, and support of this work. This work was partly supported by grants-in-aid for Scientific Research (no.16560669) from the Ministry of Education, Culture, Sports, Science and Technology.
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