J.Pharm.Dyn., **3**, 264—267 (1980)

BIOMEDICAL APPLICATIONS OF MAGNETIC FLUIDS. I. MAGNETIC GUIDANCE OF FERRO-COLLOID-ENTRAPPED ALBUMIN MICROSPHERE FOR SITE SPECIFIC DRUG DELIVERY *IN VIVO*

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(Received February 4, 1980)

Magnetic guidance of magnetic albumin microsphere for site specific drug delivery was investigated *in vivo*. After intravenous injection in mice, magnetic microspheres localized in the site (lung) at which two permanent magnets were placed. Injection into the renal artery in rats also indicated that the microspheres were concentrated at the kidney by a magnetic field. When magnet was not applied, however, the microspheres were concentrated mainly in the liver. Such preferential localization by magnetic means suggested that magnetic albumin microspheres could become effective drug carriers with site specificity for the delivery of chemotherapeutic agents in cancer therapy.

Keywords—magnetic albumin microsphere; magnetic fluid; drug carrier; site specificity; drug delivery; magnetic guidance

Localization of chemotherapeutic agents to specific sites would reduce the systemic dose of a given drug while still achieving effective local concentration of the drug. One chemical approach to targeting chemotherapeutic agents is inclusion of the agents within carriers. Recent examples include entrappment of daunomycin into DNA[1] and other antitumor agents into liposomes.[2,3] We recently reported that 5-fluorouracil entrapped in albumin microspheres was present in high levels in the liver of mice after intravenous injection,[4,5] and suggested sustained release and prolonged action of entrapped drug occurred in Ehrlich ascites[6] and solid carcinoma.[7] However, intravenous injection of such liposomes and microspheres results in their uptake predominantly by the reticuloendothelial system,[4,5,8] especially the Kupffer cells in the liver.[9] More recently, Widder *et al.*[10] suggested that since magnetic microspheres injected into the ventral caudal artery could be localized to some extent to a predetermined tail segment by an externally applied magnetic field, magnetically guided albumin microspheres entrapped drugs would be useful as a drug delivery system with site specificity.

The experimental developments in this approach have been prevented by two difficulties, *i.e.* the control of a magnetic field at a target topical site and the preparation of an active drug with a magnetically responsive character. If site specific drug delivery of antitumor agents could be achieved with magnetic means, this delivery system may eliminate adverse side effects that are often the sequelae of generated systemic drug distribution. This paper describes the utility of magnetic albumin microsphere as a drug carrier with target specificity by measuring microsphere levels in the lung and kidney after intravenous and intra-renal-arterial administration, respectively.

Magnetic albumin microspheres were prepared with bovine serum albumin, ^{125}I-human serum albumin (50 μCi/ml, Japan Radioisotope Association) and magnetic fluid* (Type W-35, Taiho Industries Co., Ltd.) by a modification of the method of Widder *et al.*[11] The final

* Average diameter of the colloidal particles is 100—200 A.

microspheres contain about 50% magnetite (Fe_3O_4). Two types of microspheres, namely small microsphere (1 μm in average diameter) and large microsphere (3 μm), were used in the experiments.

In the first experiments, the mouse lung was selected as a model for *in vivo* testing of microspheres for two reasons. (i) The lung tumor occupies high ratios in many malignancies. (ii) The microspheres after intravenous injection pass through the lung until sequestration in the liver. One mg of small or large microsphere was injected into the tail vein in ICR mice, weighing about 30 g, as 0.2 ml of a suspension. Two magnets (Super Disc Magnet, No. 30730, Edmund Scientific Co.) with a magnetic introduction of about 3000 Gauss were placed on the breast and back of mice throughout the experiment so as to concentrate the microsphere into the lung. Mice were killed 10 min after administration, and ^{125}I-labeled microspheres in various tissues were determined by an Auto-gamma scintillation spectrometer (Type 5110, Packard).

The rat kidney was selected for a model in the second experiments. It is difficult to collect the microspheres in other tissues except the lung or heart after intravenous injection, then the route of the administration was changed to intra-artery. Administration of the microspheres through the renal artery was carried out by cannulating a polyethylene tubing and forming a new vessel bypassing the original artery, because the polyethylene tubing is easy for injection. One mg of small microsphere was injected into the left renal artery in male Wistar rats weighing about 220 g. Two magnets were directly placed throughout the experiments at the both sides of left kidney which was exposed by a midline abdominal incision. Distribution of microsphere 10 and 60 min after administration was examined by the method described previously. The distribution of ^{125}I-albumin microspheres to various organs is represented as % of dose per gram tissue or that in whole tissue.

The experimental results in mice were shown in Fig. 1. After intravenous injection of small microspheres in mice without magnet (control), about 3.9% of the administered dose (15.8%/g tissue at the concentration) was found in the lung. When the particle size of the microspheres was enlarged from 1 to 3 μm in diameter, uptake of microsphere in the lung increased from 3.9% of dose (15.8%/g tissue) to 10.7% (51.7%/g). This results may indicate, considering that blood capillary is narrow, that the large microspheres lead to the occlusion of the capillary. When two magnets were applied to the lung, however, the microsphere level in the lung increased about four-fold for small microsphere and twice for large microsphere compared with each control. After injection of large microsphere, a peak lung

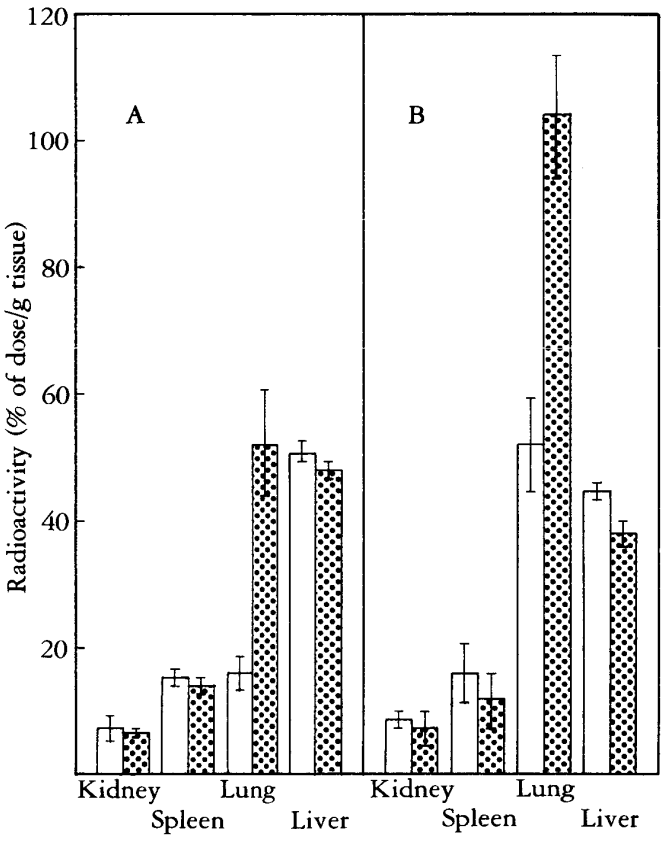

FIG. 1. *Tissue Distribution of Radioactivity at 10 min after Intravenous Injection of Magnetic Microsphere (1 and 3 μm in diameter)*

A; 1 μm in average diameter, B; 3 μm in average diameter. ☐ ; control (no magnet), ▩ ; treatment with two magnets throughout the experiments. Each column represents the mean value of 3—5 experiments. Vertical bars indicate S.E.M.

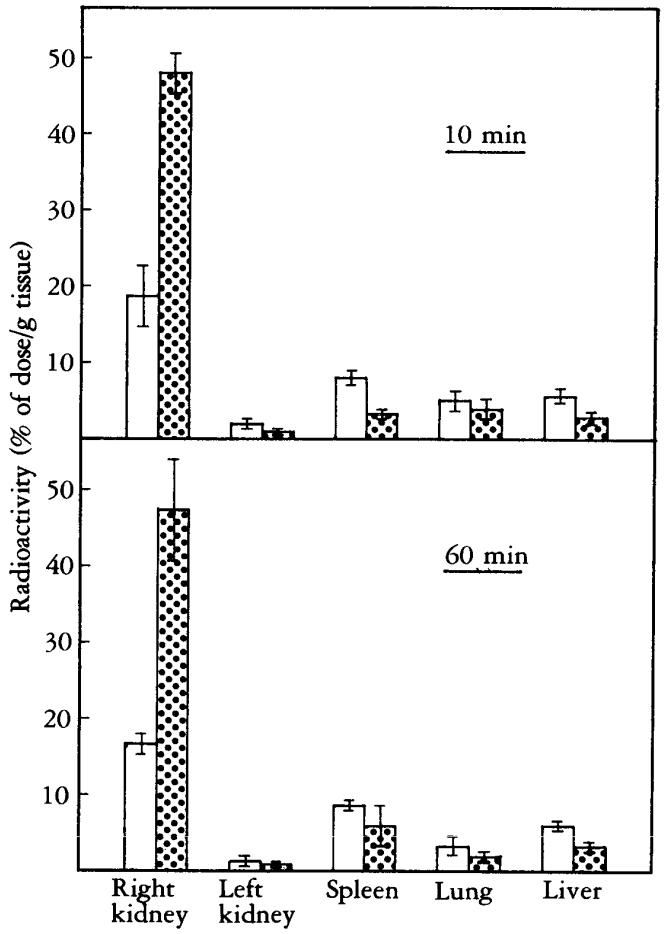

FIG. 2. *Tissue Distribution of Radioactivity at 10 and 60 min after Administration into the Left Renal Artery of Magnetic Microsphere (1 μm in diameter)* ▭ *; control (no magnet),* ▩ *; treatment with two magnets throughout the experiments. Each column represents the mean value of 3 experiments. Vertical bars indicate S.E.M.*

level (104.0%/g tissue) was measured, but the amount of the microspheres in the lung was only 21.6% of the dose. When the strength of magnetic field is increased, this value (21.6%) might be improved considering that Widder *et al.*[10] used high magnetic field strength (8000 oersteds) to retain 37—65% microspheres into the tail after infusion through the ventral caudal artery.

After intravenous injection, the microspheres did not concentrate in the kidney and localized mainly in the liver as mentioned above.* Though the data are not illustrated, intravenous injection in rats applied two magnets at the kidney did not increase in the microsphere level in the magnet site.** Microsphere level in the kidney after intra-renal-arterial administration (Fig. 2, control) was higher than that in the experiments of intravenous injection with and without magnets. Administration into the rat kidney with magnets, on the other hand, concentrated about 56% of the dose (48%/g tissue) in the kidney at 10 min as shown in Fig. 2 (treatment with two magnets), and the value was about 2.5-fold higher than that in the control. The microsphere level in the kidney at 60 min was not different from that at 10 min, which suggested that the microspheres in the kidney were retained by the magnets.

A carrier system for the site specificity of chemotherapic agents by magnetic means is a unique idea and magnetic albumin microspheres may provide a new means of treatment in cancer chemotherapy. Presently, we plan to investigate the applicability of such drug carriers to several experimental tumor systems.

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* After intravenous injection of the microsphere without magnets, the level in the kidney was 1.5% of the dose (1.2%/g tissue).

** After intravenous injection with magnets, the level in the kidney was 1.7% of the dose (1.4%/g tissue).

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