Communications to the Editor

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Drug-carrier Property of Albumin Microspheres in Chemotherapy. I. Tissue Distribution of Microsphere-entrapped 5-Fluorouracil in Mice[1)]

After intravenous injection in mice, 5-fluorouracil-6-^{3}H entrapped in albumin microspheres localized mainly in the liver, and the disappearance rate of radioactivity in microspheres from the tissue was very slow in comparison with that of free drug. Such preferential localization and sustained release of entrapped drugs suggested that albumin microspheres are useful as drug-carrier in chemotherapy.

Keywords——albumin microsphere; antitumor agent; drug-carrier; reticuloendothelial system; drug distribution

It is desirable that the drug selectively reaches its target tissues in drug therapy. After the drug is absorbed or injected into the blood stream, it may be distributed into the interstitial, cellular, and transcellular fluids. The rate, extent, and pattern of the distribution are determined by the physicochemical characteristics of the drug. And the undesired effects of the drug sometimes arise because of the accumulation in other tissues except for target tissues. In particular, since most antitumor agents lack tumor specificity thereby resulting in dose-limiting systemic toxicity, these drugs should be delivered to target tissues correctly.

Past approaches to find method of directing drug-carrier to paticular tissues have been largely related to drug-containing liposome (phospholipid vesicles). Gregoriadis and his coworkers[2)] reported that liposome-entrapped drugs intravenously injected into rats were concentrated at the liver and the spleen by the phagocytosis of the reticuloendothelial system. Tanaka, *et al.*[3)] also reported similar results that liposome-entrapped ^{14}C-inulin after intravenous injection were recovered mainly in the liver and the spleen. Trouet, *et al.*[4)] reported reduced toxicity and increased effectiveness in the treatment of leukemia when DNA complexes of the antitumor agent daunomycin are pinocytized from solution. Radiologists utilized the phagocytic activity of the liver and the spleen to study and diagnose the function of the reticuloendothelial system by using radiolabeled albumin aggregates, albumin microspheres and sulfur colloids.[5)] Kramer[6)] suggested the possibility that albumin microspheres could be utilized as the prominent drug-carrier with tissue specificity. Drug-carrier property of albumin microspheres in chemotherapy also appeared to be of interest to us. In this paper we wish to report the tissue distribution of 5-fluorouracil (5-FU) entrapped in albumin microspheres after intravenous injection in mice. Albumin microspheres are physically and chemically stable, are selectively removed from blood stream by the reticuloendothelial system, and are nonantigenic and metabolizable within the body. Wagner, *et al.*[7)] reported

1) Part of this work was presented at 97th Annual Meeting of Pharmaceutical Society of Japan, Tokyo, April, 1977.
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3) T. Tanaka, K. Kobayashi, K. Okumura, S. Muranishi, and H. Sezaki, *Chem. Pharm. Bull.* (Tokyo), **23**, 3069 (1975).
4) A. Trouet, D.D. Campeneere, and C. De Duve, *Nature* (London), **239**, 110 (1972).
5) *a*) I. Zolle, B.A. Rhodes, and H.N. Wagner, Jr., *Int. J. Appl. Radiat. Isotop.*, **21**, 155 (1970); *b*) U. Scheffel, B.A. Rhodes, T.K. Natarajan, and H.N. Wagner, Jr., *J. Nucl. Med.*, **13**, 498 (1972).
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7) H.N. Wagner, Jr., D.C. Sabiston, Jr., J.G. McAffee, D. Tow, and S. Stern, *New Engl. J. Med.*, **271**, 377 (1964).

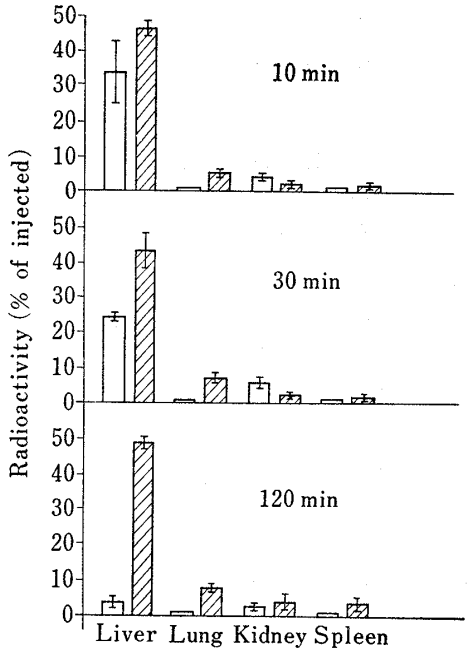

Fig. 1. Time Course of Uptake and Persistence of 5-Fluorouracil-6-^{3}H in Tissues

▨: microsphere-entrapped 5-fluorouracil.
□: non-entrapped 5-fluorouracil.
Each column represents the mean value of 3—5 experiments.
Vertical bars indicate S.E.M.

that they administered aggregated albumin in more than 1200 studies in over 300 subjects and were unable to detect any evidence of antigenicity. Therefore the results satisfy the criteria which a substance should possess if it is to be satisfactorily employed as drug-carriers.

Bovine serum albumin microspheres containing the antitumor agent 5-fluorouracil were prepared by a modification of the method of Scheffel, *et al.*[5b] Size distribution of microspheres was determined with a coulter counter and the main part was in the range 0.4—1.0 microns.

In animal experiment, 5-fluorouracil-6-^{3}H was injected into the mouse through the tail vein (ICR, 22—32 g), as 0.5 ml of a solution (0.2% polysorbate 80 in 0.9% NaCl solution) or a suspension of microspheres. And mice were killed at certain time intervals. Radioactivity in the lung, liver, spleen, and the kidney was determined by using a sample oxidizer and a liquid scintillation counter.

Soon after injection of non-entrapped 5-fluorouracil-6-^{3}H, most radioactivity was removed from the circulation and some of it transientry was in the liver and the kidney. But radioactivity in the liver decreased from 34.30% (10 min) to 3.14% (120 min). In contrast, entrapped 5-fluorouracil-6-^{3}H was mainly accumulated in the liver, and a considerable amount of the drug accumulated was still retained by the tissue 120 min later as shown in Fig. 1. This is similar to a phenomenon that albumin macroaggregates as scanning agent in clinical use today are phagocytized by the reticuloendothelial system of the liver. The albumin microspheres also are delivered into the reticuloendothelial system because of its high phagocytic activity. These results suggested that entrapment of the drug in albumin microspheres led to a dramatic change in its retaining in the liver.

After the microspheres were administered into the mice, disappearance of the radioactivity in the liver was very slow. Other work from our own laboratory has shown that *in vitro* drug release from microspheres entrapped 5-fluorouracil continued over a week, although the release rate was slow. From these results, the microspheres in the body may be disintegrated gradually and 5-fluorouracil entrapped in microspheres may be released. And it is expected that the albumin microspheres are useful for maintenance of the clinical effectiveness or therapeutic concentration in the tissue. Such good localization and prolonged action of entrapped drugs in the liver suggested that albumin microspheres are potential as drug carrier in chemotherapy. Work is now in progress on the effect of albumin microspheres on the tumor bearing mice.

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