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Effect of Liposome Size and Drug Release Properties on Pharmacokinetics of Encapsulated Drug in Rats[1]

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ABSTRACT

The organ distribution and half-life in blood of liposome-entrapped [^{14}C]sucrose was examined in rats for two compositions of liposomes having widely differing drug release properties as determined by an *in vitro* assay in the presence of serum at 37°C. The liposomes were composed of 1) egg phosphatidylcholine-cholesterol, 2:1 molar ratio and of 2) bovine brain sphingomyelin-phosphatidylcholine, 4:1 molar ratio. The two types of liposome were determined to have half-lives for release of contents *in vitro* in the presence of serum of 2.6 and 35 hr, respectively. Organ distribution and blood values were followed with time for small unilamellar, multilamellar and reverse-phase evaporation liposomes of both compositions. Free [^{14}C]sucrose was almost totally eliminated from rats within 0.5 hr of injection. Liposome-entrapment increased the circulation time of sucrose *in vivo* in the following order: small unilamellar > reverse-phase evaporation > multilamellar liposomes. For all sizes of liposomes, the composition with slower leakage as determined by *in vitro* assay (sphingomyelin-phosphatidylcholine, 4:1) resulted in significantly longer half-lives in blood and in whole body. Liposomes containing sphingomyelin were taken up by liver to a lesser extent and by spleen to a greater extent than phosphatidylcholine-cholesterol liposomes of all sizes.

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Considerable attention has been focused in recent years on the use of "drug carriers" or "drug delivery systems" to localize drugs at target sites in specific tissues *in vivo* (Juliano, 1980; Gregoriadis and Allison, 1980). Liposomes (phospholipid vesicles) have many properties which make them attractive as drug carriers including their low intrinsic toxicity, biodegradability and relative lack of immunogenicity. In addition, it is a simple matter to vary liposome size, charge and lipid constituents. A number of authors have commented on the substantial alterations in pharmacokinetics of drugs when they are liposome-associated (Juliano and Stamp, 1978; Mauk and Gamble, 1979; Ellens *et al.*, 1981). If liposomes are to be successful as drug carriers or for targeting drugs *in vivo*, then it is important, in most instances, that the liposome-drug package have a circulatory half-time in blood of the order of a few hours.

Two factors which have a major impact on the circulation half-life of the liposome-drug package are 1) the tendency of liposomes, by virtue of their particulate nature, to rapidly localize in the reticuloendothelial system (Jonah *et al.*, 1975; Kimelberg *et al.*, 1976) and 2) the tendency of liposomes of many compositions to rapidly release their drug contents under the influence of serum components (Kimbelberg *et al.*, 1975; Scherphof *et al.*, 1978; Allen and Cleland, 1980). Various techniques have been applied in an attempt to overcome these limitations. Abra *et al.* (1980) have shown that a blockade of hepatic uptake sites by predosing mice with empty liposomes resulted in a prolonged circulation time of liposome-entrapped [^{14}C]inulin. Juliano and Stamp (1975) have reported that SUV of neutral or positive charge have a longer circulatory half-life than large MLV. Several authors have recently shown that manipulation of liposome phospholipid and cholesterol content produces liposomes which are more refractory to serum-induced leakage of their contents (Gregoriadis and Senior, 1980; Ellens *et al.*, 1981; Allen, 1981).

In the present study, we have examined in rats the effect of liposome size and composition on the tissue distribution and circulation half-life of liposome-entrapped *vs.* free [^{14}C]sucrose. Two liposome compositions were chosen on the basis of their widely differing susceptibility to lysis by serum components *in vitro*; namely, PC-CHOL, 2:1 molar ratio, and SM-PC, 4:1 molar ratio. Liposomes of these compositions have been determined to have half-lives for the release of their contents *in vitro*, in the presence of 99.5% serum, of 2.5 and 35 hr, respectively (Allen, 1981). We were interested in seeing whether increased resistance to serum components in *in vitro* assays would translate into increased circulation half-lives *in vivo*. SUV, REV and MLV were employed in these studies.

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ABBREVIATIONS: SUV, small unilamellar liposomes; MLV, multilamellar liposomes; PC-CHOL, egg phosphatidylcholine-cholesterol; SM-PC, bovine brain sphingomyelin-egg phosphatidylcholine; REV, large unilamellar liposomes; PBS, phosphate-buffered saline.

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Materials and Methods

SM was purchased from Avanti Biochemicals (Birmingham, AL), PC from Makor Chemicals (Jerusalem, Israel) and CHOL from Sigma Chemical Co. (St. Louis, MO). [^{14}C]Sucrose (3.6 mCi/mmol) was purchased from New England Nuclear (Boston, MA). All other chemicals used were reagent grade.

Preparation of liposomes. Chromatographically pure lipids in organic solvent were taken to dryness under vacuum in a rotary evaporator. MLV were prepared in the following fashion: 100 μmol of dried phospholipid was suspended in 2 ml of sterile PBS containing 0.125 mCi of [^{14}C]sucrose. The sample was then vortexed in a N_2 atmosphere until all the phospholipid was suspended in buffer (10–20 min). The vortexing step was aided by brief pulses (10 sec) of sonic energy from a bath-type sonicator (Laboratory Supplies Inc., Hicksville, NY). From this procedure we obtained a hetergenous population of MLV containing 1 to 2 liters of entrapped [^{14}C]sucrose solution per mole of phospholipid. Olson *et al.* (1979) have determined that MLV containing similar trapped volumes have a mean diameter of 1.3 μ. Free [^{14}C]sucrose was removed from liposome-entrapped sucrose by passage over Sephadex G-50 in sterile PBS. SUV were made by a similar procedure except 0.2 to 0.215 mCi of [^{14}C]sucrose were added to 150 μmol of lipid in 2.25 ml of buffer. After the vortex step, liposomes were sonicated to a clear, opalescent solution (30–180 min) in a bath-type sonicator and free [^{14}C]sucrose was removed by Sephadex G-50 chromatography. Sephrose CL-4B chromatography of SUV prepared in this fashion, followed by phosphate analysis, shows that less than 10% of the total lipid is present as MLV. These liposomes consist of a homogenous population averaging 30 to 50 nM in diameter and containing 0.1 to 0.2 liters entrapped solute per mole of phospholipid. Reverse-phase evaporation liposomes were prepared according to the technique of Szoka and Papahadjopoulos (1978). We determined that these liposomes contain a trapped volume of 4 to 6 liters of solute per mole of phospholipid from which an average liposome diameter of 100 to 200 nM can be calculated. In all liposome preparations we adjusted the total phospholipid and amount of [^{14}C]sucrose added to the lipid so as to end up with approximately 8 μmol of phospholipid entrapping 0.5 to 1 × 10^6 cpm of [^{14}C]sucrose in a volume of 0.25 ml of PBS.

Animal experiments. Female Sprague-Dawley rats weighing an average of 200 g were anesthetized with methoxyflurane and injected in the femoral vein with 0.25 to 0.3 ml of sterile PBS containing an average of 8 μmol of phospholipid and a minimum of 0.5 × 10^6 cpm of [^{14}C]sucrose. Liposomes were either MLV, SUV or REV (reverse-phase). Control rats received 10^6 cpm of [^{14}C]sucrose in 0.3 ml of PBS. Rats were sacrificed 0.5, 2, 6 or 18 hr postinjection and 100-mg samples of blood, brain, thymus, lung, heart, liver, spleen and kidney were collected. All experiments were done in triplicate and all results are expressed as mean ± S.D. Tissue samples were dissolved in 1 ml of NCS tissue solubilizer (Amersham Corp., Arlington Heights, IL). After dissolution, the samples were decolorized with 100-μl aliquots of 30% hydrogen peroxide and acidified to pH 6 with concentrated HCl. Aquasol (15 ml; New England Nuclear) was added to each acidified sample, the samples were dark-adapted for 3 days (to minimize chemiluminescence) and counted on a Beckman LS 7500 scintillation counter with automatic quench compensation.

Results

Significant differences occurred with liposome size when we examined the percentage of administered [^{14}C]sucrose remaining in the blood over 18 hr (fig. 1, A and B). [^{14}C]Sucrose trapped in SUV remained in circulation longer than that trapped in REV, which in turn had a longer circulation time than that in MLV. Free [^{14}C]sucrose was rapidly eliminated from blood (fig. 1, A and B). From a comparison of figures 1 A and B, it is apparent that [^{14}C]sucrose trapped in liposomes composed of SM-PC, 4:1, had a significantly longer circulation time for all types of liposomes than did [^{14}C]sucrose trapped in liposomes composed of PC-CHOL, 2:1. Although we do not have enough data points to determine the exact kinetics of elimination, there appear to be at least two components to the elimination curve, an initial rapid phase, followed by a slower phase of elimination of trapped [^{14}C]sucrose from the blood. The slower phase of elimination is approximately the same for all three liposome types of the same phospholipid composition. The differences in rate of removal from blood between liposomes of different sizes is, therefore, a reflection of the differences in rate of the initial, rapid component of elimination.

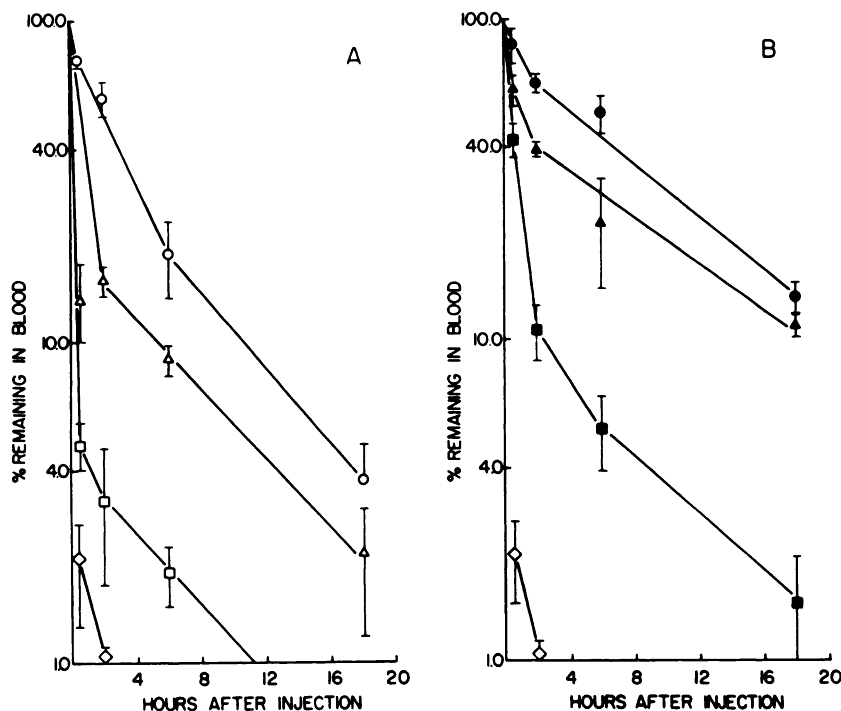

Fig. 1. Percentage of administered liposome-entrapped [^{14}C]sucrose present in rat blood at various time postinjection. The blood volume of the rat was estimated to be 7% of its body weight. A, liposomes composed of PC-CHOL, SUV (○), REV (△) and MLV (□). B, liposomes composed of SM-PC, SUV (●), REV (▲), MLV (■), and free [^{14}C]sucrose (◇).

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TABLE 1
Percentage of injected [^{14}C]sucrose in liver plus spleen 0.5 hr postinjection

	PC-CHOL, 2:1	SM-PC, 4:1
MLV	65.9 ± 4.0	50.5 ± 11.2
REV	49.2 ± 0.9	29.6 ± 3.2
SUV	12.2 ± 0.6	8.6 ± 3.2

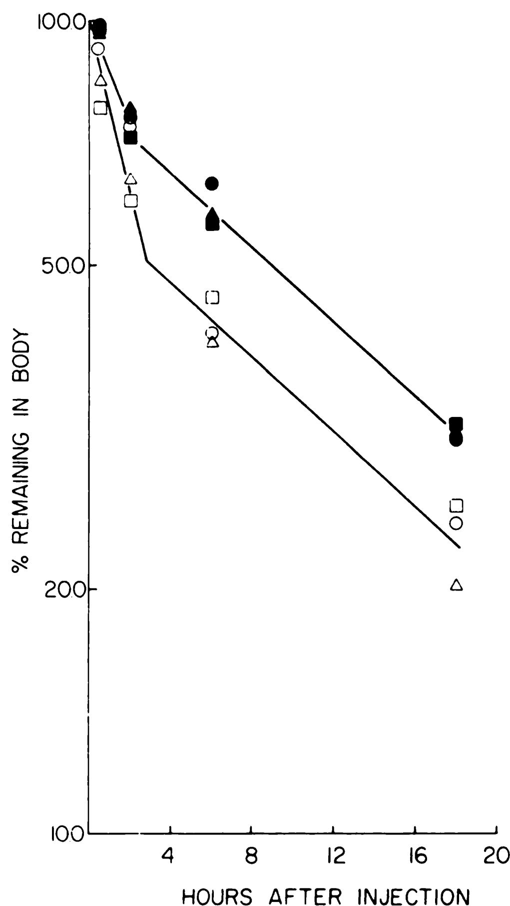

Fig. 2. Percentage of administered liposome-entrapped [^{14}C]sucrose remaining in the rat body at various times postinjection. The amount of [^{14}C]sucrose remaining in the body was calculated as the sum of [^{14}C] sucrose in blood, liver, spleen, heart, lung, brain, thymus and kidney. Liposomes were composed of PC-CHOL, 2:1 molar ratio, and SM-PC, 4:1 molar ratio. SUV of SM-PC (●), REV of SM-PC (▲), MLV of SM-PC (■), SUV of PC-CHOL (○), REV of PC-CHOL (△) and MLV of PC-CHOL (□).

The initial, rapid phase of elimination of liposomes from blood is due in large part to uptake of liposomes by the reticuloendothelial system. Table 1 shows that the percentage of the administered [^{14}C]sucrose in reticuloendothelial tissue (liver and spleen) 0.5 hr postinjection varies with both liposome size and liposome composition. Large liposomes are taken up by liver and spleen much more rapidly than are small liposomes. Liposomes composed of PC-CHOL, 2:1, localize in the liver and spleen to a greater extent than do liposomes composed of SM-PC, 4:1 (table 1).

The percentage of injected [^{14}C]sucrose remaining in the rat bodies at various times after injection for the two liposome compositions is seen in figure 2. It is apparent that [^{14}C]sucrose trapped in liposomes composed of SM-PC, 4:1 (fig. 2, closed symbols), is retained in the body for a significantly longer time for all three liposome types than is [^{14}C]sucrose trapped in PC-CHOL, 2:1. Both liposome compositions resulted in much longer residence times for [^{14}C]sucrose in the body than for free [^{14}C]sucrose. Less than 5% of free [^{14}C]sucrose remained in the body 0.5 hr after injection. We therefore assume for the purposes of this paper that all [^{14}C]sucrose found in the body is liposome-associated and that the amount of free [^{14}C]sucrose is negligible. The liposome size (SUV *vs.* REV *vs.* MLV) did not appear to significantly affect the rate of elimination from the body for both liposome compositions. Entrapped [^{14}C] sucrose was initially eliminated from the body at a rapid rate and subseqeuntly at a slower rate. The second, slow phase of elimination occurred at the same rate for both liposome compositions, whereas the initial phase was more rapid for liposomes composed of PC-CHOL, 2:1.

More than 95% of the [^{14}C]sucrose remaining in the body at any time could be accounted for by the [^{14}C]sucrose content in four tissues, liver, spleen, lung and blood. The percentage of administered dose present in these tissues for all three liposome types and both liposome compositions is presented in histogram form in figures 3, A to D. REV and MLV composed of PC-CHOL have a pronounced and immediate tendency to localize in liver as compared with liposomes composed of SM-PC. On the other hand, MLV and REV composed of SM-PC reach higher levels in spleen than PC-CHOL liposomes with spleen concentrations not peaking until 2 hr or more postinjection. SUV of both liposome compositions result in high blood levels which are not substantially different for the first 2 hr postinjection, but SUV of SM-PC result in significantly higher blood levels after longer time periods. MLV of PC-CHOL had the greatest tendency to localize in lung tissue. REV of both liposome compositions were intermediate between SUV and MLV in their tendency to localize in various tissues, reflecting their intermediate size distribution.

In tables 2 and 3 are found the relative tissue content of [^{14}C] sucrose for the three liposome types and two liposome compositions, corrected for tissue blood content. The results are expressed as counts per minute per milligram of tissue and can easily be converted into moles of sucrose per milligram of tissue by multiplying by the appropriate correction factor depending on the initial concentration of sucrose which was trapped in liposomes. Each rat in this series of experiments received approximately the same total amount of sucrose and to compensate for small differences in numbers of counts of [^{14}C] sucrose injected we standardized the radioactive counts to 10^6 cpm of sucrose injected per rat at each injection time.

SUV of PC-CHOL resulted in peak concentrations in liver 6 hr postinjection and in spleen 2 hr postinjection. When the liposomes were composed of SM-PC, peak concentrations were further delayed, with highest levels in liver not achieved until 18 hr after injecting SUV, 6 hr after administration of REV and 2 hr after injecting MLV. For liposomes composed of SM-PC, peak spleen levels were achieved 6 hr after injection of SUV and 2 hr after administration of reverse-phase and MLV. Overall, much lower levels of drug were found in liver and much higher levels were found in spleen when the liposomes were composed of SM-PC. Unexpectedly, SUV and MLV of both compositions resulted in higher lung concentrations of drug than did reverse-phase liposomes and the peak concentration of drug in lung did not occur until 2 hr after injecting REV.

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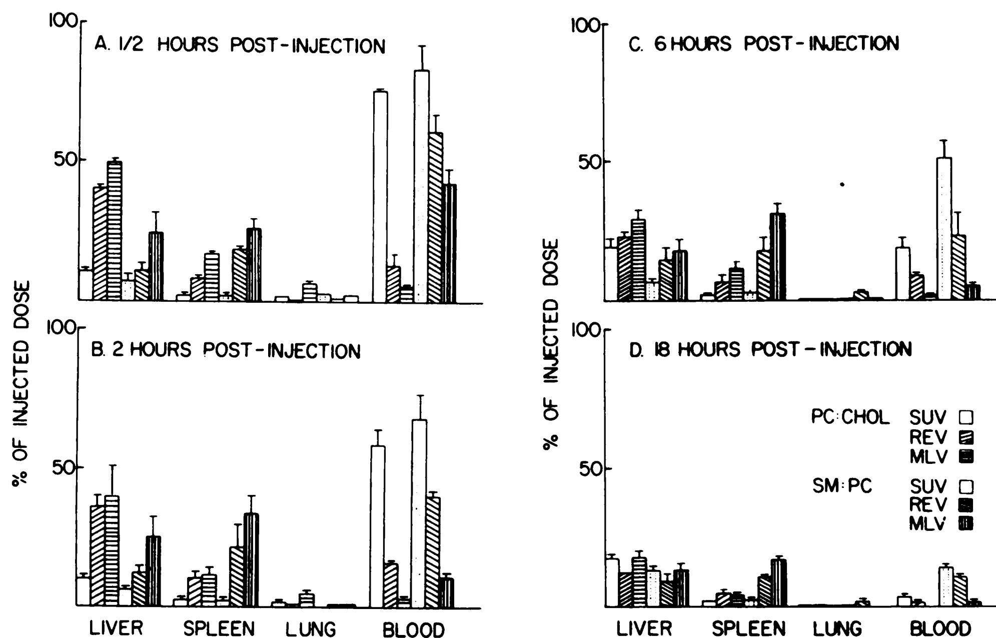

Fig. 3. Percentage of administered liposome-entrapped [^{14}C]sucrose present in liver, spleen, lung and blood at various times postinjection.

Discussion

The amount of drug remaining in the blood (fig. 1, A and B) at any time is dependent on two main factors: rate of liposome lysis by serum components and rate of uptake of the liposome-entrapped drug by tissues. In our experiments, the [^{14}C]sucrose which was released by the action of serum components was rapidly excreted and therefore uptake of free [^{14}C]sucrose by the tissues is not likely to be a major consideration. The initial, rapid drop in the level of circulating drug is due, in large part, to rapid initial uptake of liposomes by the liver (and spleen to some extent) and is dependent on liposome size, with larger liposomes removed more rapidly than smaller liposomes from the circulation (table 1). The subsequent slower decrease in blood levels of [^{14}C]sucrose may be due in part to slow tissue uptake of circulating liposomes or may be a reflection of gradual lysis of liposomes by serum components with rapid excretion of the free drug. Liposomes composed of SM-PC, 4:1, were taken up less rapidly by the reticuloendothelial system for all liposome sizes as is evidenced by the differences in initial phase of elimination in figure 1B *vs.* fig. 1A and in the data in table 1. They also were less susceptible to lysis by serum components which is probably a contributing factor to the somewhat slower final phase of removal of SM-PC liposomes *vs.* PC-CHOL liposomes. These two factors combined resulted in a significant increase in circulation half-life for liposomes composed of SM-PC, 4:1, for all three liposome sizes as compared with liposomes composed of PC-CHOL.

The amount of drug ([^{14}C]sucrose) remaining in the body (fig. 2) at various times postinjection was estimated from the sum of the [^{14}C]sucrose in blood, kidney, liver, spleen, lung, brain, heart and thymus. Drug trapped in liposomes of PC-CHOL, 2:1, was initially removed from the body at a more rapid rate than drug entrapped in liposomes of SM-PC, 4:1. The size of the liposomes did not appear to be an important factor in elimination of sucrose from the body in contrast to the situation in blood. The initial difference in rate between the two liposome compositions most likely represents the more rapid breakdown of PC-CHOL liposomes by serum components with rapid elimination of free [^{14}C]sucrose from the body (fig. 2). Subsequently, both liposome compositions resulted in similar rates of elimination of sucrose from the body and this may provide an indication that tissue degradation of both types of liposomes may occur at the same rate.

The wide variation in tissue levels of drug observed for liver and spleen could be interpreted in the following manner. Liposomes, when injected i.v., initially pass through the heart and lungs and then circulate through the liver and other organs. Because of the large blood volume circulating through the liver (approximately 30% of blood volume in each pass), most of the injected liposomes traverse the liver within the first three or four passes. The length of time to achieve saturation of liver uptake sites depends on liposome size and composition, with larger liposomes taken up more rapidly than smaller ones and liposomes of PC-CHOL taken up more rapidly than liposomes of SM-PC, for unknown reasons. Similar results have been reported by Ellens *et al.* (1981) and by Hwang *et al.* (1980). The spleen, on the other hand, being a much smaller organ, receives only a small proportion of the circulating blood at each pass and it therefore take longer to become saturated. If substantial numbers of liposomes escape the liver, then high spleen levels may be achieved, but only after sufficient blood (carrying liposomes) has circulated through the spleen (which may take 2–6 hr). This effect has also been reported by Abra *et al.* (1980). The spleen appears to have a greater capacity for liposome uptake than does the liver, as witnessed by the high drug concentrations achieved in spleen, and peak spleen levels of drug are achieved before peak liver levels if the liposomes have sufficient access to the spleen. This can be seen in the case of SUV of both compositions and reverse-phase liposomes of SM-PC (tables 2 and 3).

After correction for tissue blood content, we observed little tendency for liposomes to be taken up by thymus and heart at the early time periods. Low levels of uptake were noted at later time periods for these tissues. Low levels of [^{14}C]sucrose were observed in brain at all time periods, but the possibility of free [^{14}C]sucrose contributing to brain (and kidney) levels at the early time points cannot be discounted.

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TABLE 2

Content of [^{14}C]sucrose in counts per minute per milligram of tissue[a] at various times postinjection of SM-PC liposomes

Rats were injected in the femoral vein with 0.25 to 0.3 ml of PBS containing 8 μmol of SM-PC, 4:1, liposomes with approximately 10^6 cpm of trapped [^{14}C]sucrose. Rats were sacrificed at the indicated time intervals, tissues were sampled and counts per minute of sucrose per milligram of tissue was calculated. Tissue sucrose levels were corrected for blood content using the correction factors of Hwang *et al.* (1980).

	SUV				REV				MLV			
	0.5 hr	2 hr	6 hr	18 hr	0.5 hr	2 hr	6 hr	18 hr	0.5 hr	2 hr	6 hr	18 hr
Brain	0.8 ± 0.2	0.9 ± 0.2	0.4 ± 0.1	0.2 ± 0.1	4.5 ± 6.0	3.2 ± 4.3	0.6 ± 0.2	0.2 ± 0.1	0.7 ± 0.1	0.5 ± 0.2	0.6 ± 0.2	0.7 ± 0.1
Thymus	0	0	0	0	0	1.7 ± 2.4	1.2 ± 2.1	0	0	0	0.1 ± 0.1	0.7 ± 0.1
Lung	16.2 ± 3.4	11.3 ± 5.3	2.9 ± 1.3	4.1 ± 1.0	1.7 ± 1.5	5.1 ± 0.5	4.1 ± 0.3	3.8 ± 0.9	17.7 ± 8.0	6.3 ± 0.6	3.5 ± 0.5	2.8 ± 0.4
Heart	0	0	0	1.0 ± 0.2	0	0.4 ± 0.2	1.0 ± 0.5	0.7 ± 0.1	1.9 ± 2.1	0.5 ± 0.1	0.6 ± 0.2	0.6 ± 0.1
Liver	5.5 ± 2.8	5.5 ± 1.3	5.9 ± 1.0	12.3 ± 2.3	11.2 ± 3.1	12.8 ± 1.9	13.9 ± 5.1	9.3 ± 3.4	35.1 ± 8.5	38.5 ± 14.3	23.3 ± 6.1	19.7 ± 6.1
Spleen	27.3 ± 3.3	36.7 ± 7.8	48.2 ± 3.3	43.9 ± 3.7	325.2 ± 3.6	386.8 ± 149.7	265.5 ± 67.6	222.4 ± 65.6	503.4 ± 22.7	647.4 ± 99.1	606.5 ± 197.0	311.0 ± 9.1
Kidney	10.2 ± 0.9	5.9 ± 1.0	4.7 ± 0.4	2.7 ± 0.2	9.2 ± 5.4	3.6 ± 0.6	4.1 ± 0.3	4.7 ± 0.3	4.7 ± 0.3	3.1 ± 0.5	3.0 ± 0.6	2.1 ± 0.3
Blood	50.3 ± 6.2	38.5 ± 10.3	30.6 ± 2.2	8.5 ± 0.5	41.8 ± 1.1	25.2 ± 2.2	14.3 ± 6.7	7.4 ± 0.3	27.2 ± 8.3	7.4 ± 1.0	3.7 ± 1.2	1.1 ± 0.4

[a] Standardized to 10^6 counts injected.

TABLE 3

Content of [^{14}C]sucrose in counts per minute per milligram of tissue[a] at various times postinjection of liposomes composed of PC-CHOL

Rats were injected in the femoral vein with 0.25 to 0.3 ml of PBS containing 8 μmol of PC-CHOL, 2:1, liposomes with approximately 10^6 cpm of trapped [^{14}C]sucrose. Rats were sacrificed at the indicated time intervals, tissues were sampled and counts per minute of sucrose per milligram of tissue was calculated. Tissue sucrose levels were corrected for blood content using the correction factors of Hwang *et al.* (1980).

	SUV				REV				MLV			
	0.5 hr	2 hr	6 hr	18 hr	0.5 hr	2 hr	6 hr	18 hr	0.5 hr	2 hr	6 hr	18 hr
Brain	1.2 ± 0.2	0.5 ± 0.2	0.6 ± 0.1	0.3 ± 0.0	1.9 ± 1.5	2.1 ± 0.8	1.2 ± 0.2	0.2 ± 0.1	1.6 ± 1.2	1.7 ± 1.2	1.0 ± 0.4	0.5 ± 0.2
Thymus	0	0	0	0.3 ± 0.2	0	1.2 ± 2.0	0.4 ± 0.3	0.5 ± 0.2	0.9 ± 0.6	0.8 ± 0.2	1.1 ± 0.2	1.0 ± 0.3
Lung	12.7 ± 2.5	9.6 ± 7.3	2.0 ± 0.3	1.5 ± 0.5	2.1 ± 2.0	6.3 ± 1.4	4.7 ± 1.2	2.4 ± 0.6	50.4 ± 2.1	37.6 ± 11.4	24.2 ± 5.6	16.7 ± 2.2
Heart	0	0.2 ± 0.3	0.3 ± 0.4	0.9 ± 0.1	0.1 ± 0.1	3.5 ± 4.5	0.6 ± 0.2	0.5 ± 0.1	1.4 ± 0.4	2.4 ± 0.4	1.4 ± 0.2	0.8 ± 0.1
Liver	8.9 ± 1.0	11.4 ± 0.4	24.5 ± 5.9	19.9 ± 7.9	42.2 ± 1.9	41.6 ± 2.1	25.1 ± 0.8	11.1 ± 1.4	47.1 ± 1.0	39.4 ± 4.1	34.4 ± 6.4	16.2 ± 3.8
Spleen	29.6 ± 0.8	44.2 ± 11.3	41.8 ± 9.6	36.2 ± 7.7	158.0 ± 38.3	194.7 ± 40.7	124.8 ± 58.0	79.9 ± 29.2	250.1 ± 29.2	205.8 ± 60.2	220.3 ± 61.7	70.8 ± 2.6
Kidney	5.0 ± 0.5	4.4 ± 1.3	2.6 ± 0.4	2.6 ± 0.9	4.5 ± 0.6	3.2 ± 0.8	3.5 ± 1.7	1.5 ± 0.3	4.4 ± 0.6	4.0 ± 1.2	2.9 ± 0.7	1.8 ± 0.2
Blood	45.2 ± 2.4	38.2 ± 6.3	11.8 ± 3.6	2.2 ± 0.9	7.9 ± 2.4	10.0 ± 0.8	5.4 ± 1.1	1.2 ± 0.5	2.7 ± 0.6	1.9 ± 1.0	1.2 ± 0.2	0.3 ± 0.1

[a] Standardized to 10^6 counts injected.

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The explanation for why high lung concentrations are achieved with SUV and not reverse-phase liposomes is not clear. Fidler *et al.* (1980) have reported that negatively charged reverse-phase liposomes are more similar to MLV than SUV in their ability to be taken up by lung. They did not report data for neutral reverse-phase liposomes, such as those used in this study. It is possible that reverse-phase liposomes may be too small to be physically trapped by lung capillaries but too large to enter into interstitial spaces, as may happen with SUV.

In conclusion, both liposome size and liposome composition can be varied in order to achieve a wide variation in concentration of drug in blood, liver, spleen and lung. These manipulations result in much less effect on levels of drug achieved in other tissues of the body. Drugs trapped in SUV and reverse-phase liposomes composed principally of sphingomyelin resulted in circulation half-lives of up to 6 hr for [^{14}C]sucrose as compared with 2 hr or less for liposomes composed principally of phosphatidylcholine and only minutes for free [^{14}C]sucrose. This points out the potential value of liposomes as slow release systems for drugs, particularly those drugs which are rapidly broken down *in vivo*. Manipulation of liposome properties will allow the rate of release to be controlled. The long circulation half-life of sphingomyelin-containing liposomes is of potential benefit in targeting liposomes and their associated drugs to specific tissues in the body.

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