GAMMA SCINTIGRAPHIC STUDY OF ACCUMULATION OF 99M TECHNETIUM RADIO LABELLED LIPOSOMES WITHIN INFLAMMATORY TISSUE

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Scintigraphic imaging of actively inflamed rheumatoid arthritic (RA) joints has been reported following the intravenous (i.v.) administration of 99mTechnetium radiolabelled 100nm cholesterol-poor liposomes, (Williams 1987; O'Sullivan 1988). However HPLC analysis of the stability of these cholesterol-poor liposomes has shown them to be relatively unstable both in vitro and in vivo, Love (1990).
Due to their instability the accumulation of intact cholesterol-poor liposomes at inflamed tissue sites cannot be assumed, making the interpretation of scintigraphic images difficult. By incorporating cholesterol into the liposome structure, a vesicle was formed which was shown by HPLC to be resistant to degradation by serum components, Love (1990). These serum stable vesicles have been termed "cholesterol-rich" liposomes, and subsequent gamma scintigraphic studies have utilised these vesicles. Attempts have been made to repeat the scintigraphic observations obtained with the RA patients in the adjuvant-induced arthritis rat model and to demonstrate that 99mTc-radiolabelled cholesterol-rich liposomes accumulate within the inflamed paw tissue.
Cholesterol-rich liposomes, 120nm, were injected i.v. into normal and adjuvant-arthritic rats (n=6). Scintigraphs were taken of the hind paws using a GEC Maxicamera 400A with a pin-hole collimator at time intervals of 50, 135 and 1300 minutes.
Analysis of the scintigraphic data obtained is shown in Table 1. At each time point there was significantly greater activity, (normalised with respect to time and unit area), located within the arthritic paws compared to the normal paws, (ANOVA, $p<0.001$). There was no significant difference, (ANOVA, $p>0.25$), between the amount of activity associated with the inflamed paws as a function of time. However there was a significant difference between the amount of radioactivity associated with normal paws at the three time points, (ANOVA, $p<0.001$).

Table 1. Liposome activity associated with normal and arthritic rat hind paws with time, (expressed as counts/s/100 elements).

	50min	135min	1300min
Normal	1.5±0.7	0.9±0.2	0
Arthritic	5.5±1.3	6.1±1.3	4.2±2.4

The scintigraphs obtained demonstrated the ability of the 99mTc-cholesterol-rich liposomes to accumulate within the inflamed paws enabling the imaging of the inflammatory tissue at 1300 minutes, by which time >99.3% of the injected liposome dose had been removed from the circulation. At the same time point no liposomal activity was associated with the normal rat paw tissue.
These data support the view that i.v. administered cholesterol-rich 120nm liposomes are able to extravasate intact from the circulation to areas of inflammation. We therefore envisage the potential application of liposomes for drug targeting to or imaging of inflammatory sites.

Williams, B.D. et al (1987) Ann.Rheum.Dis. 46: 314-318
O'Sullivan, M.M. et al (1988) Ann.Rheum.Dis. 47: 485-491
Love, W.G. et al (1990) J.Microencap. 7: 105-112