ANTIBACTERIAL ACTIVITY OF LIPOSOME ENTRAPPED CHLORAMPHENICOL

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An interesting application of liposomes is the delivery of antibiotics to bacteria located within the host cell where, under normal circumstances, they may be protected from drug action (Bonventre & Gregoriadis 1978). This preliminary report describes the in vitro interaction of liposome entrapped chloramphenicol (CAP) with Escherichia coli.

Dried lipid films, comprising egg or dipalmitoyl lecithin (Sigma), cholesterol and ± dicetyl phosphate, molar composition 7:2:1 were hydrated under N_2 with 6.7mM pH 7.4 phosphate buffered saline (PBS) or a solution of 2mg ml^{-1} CAP in PBS. Hydrated dispersions were probe sonicated (10 min charged, 20 min neutral vesicles) under N_2 and free and liposomal CAP separated by gel filtration (Sepharose 6B, PBS eluant). CAP content, determined by HPLC, of typical pooled liposome suspensions was 5ug ml^{-1}, charged and 3ug ml^{-1}, neutral.

A range of concentrations of free or liposomal CAP, sterilised by filtration, in nutrient broth were inoculated with an overnight broth culture of E. coli (NCTC 1001) to a final cell density of $1\text{-}3x10^5$ ml^{-1} and after 24 hr at 37° the number of colony forming units (CFU) determined directly (surface plates) on nutrient agar after a 10^5 x dilution of each concentration. The number of CFU was calculated as a percentage of the appropriate control.

It was found (Fig. 1) that concentrations of CAP known to significantly reduce the number of CFU had no such effect when used in liposomal form although active CAP could be recovered from such vesicle suspensions by dissolution of the liposomes in 50% v/v propanol. Empty liposome suspensions had no effect on E. coli growth rate and addition of empty liposomes to free CAP solution did not reduce its inhibitory activity. Thus in spite of the modifying effects of cholesterol, dicetylphosphate and CAP on the transition temperature (-15° for egg, 41° for dipalmitoyl) and fluidity of the constitutent phospholipids, the vesicle bilayer was not sufficiently permeable to CAP to allow diffusion of a bacterio-

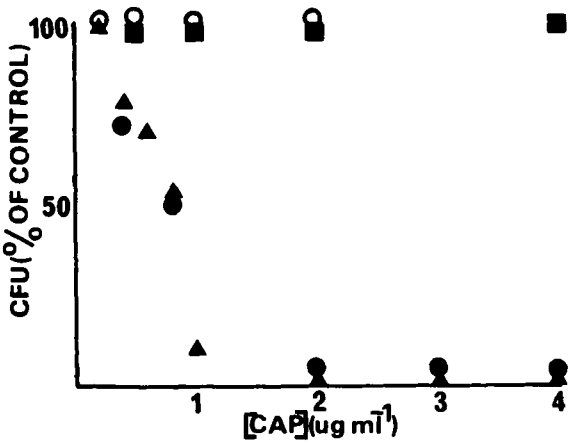

Fig. 1. Reduction in numbers of colony forming units (CFU) of E.coli after exposure to free and liposomal chloramphenicol at 37°. Free drug, ▲; drug in egg or dipalmitoyl lecithin liposomes, anionic ■, neutral, ○; drug extracted from liposomes, ●.

static quantity into the culture medium. In vivo therefore disruption either of essentially fluid or solid liposomes would be a prerequisite for an antibacterial effect.

Benventre, P., Gregoriadis, G.(1978) Antimicrob. Agents Chemother 13: 1049-1051

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