The kinetics of deposition of ciclosporin (CSA) into the dermis of hairless mouse skin after topical application of liposomally encapsulated ciclosporin, was determined using both in vitro and in vivo experiments. The overall mass balance was much lower for in vitro studies than for in vivo studies suggesting that excess formulation may have been lost despite attempts to control the process by anesthetizing the animals periodically to minimize their movements. The recoveries were generally about 70% in vitro compared to about 90% in vivo. The amounts of CSA found in the tape stripings were roughly similar for in vivo and in vitro topical applications at all time points studied. The drug content in the deeper skin strata, however, was very different. CSA deposition in the deeper skin strata 4 hr after topical application was significantly greater with the in vivo studies. Further, CSA levels decreased rapidly as a function of time in vivo whereas these levels increased steadily with time in vitro. The in vivo drug profile may be due to the clearance of drug into the systemic circulation by way of the dermal vasculature. For the in vitro diffusion experiments, the diffusional resistance of the hydrated dermis to a very hydrophobic drug such as CSA appeared to transport into the receiver and results in a build-up of CSA levels in the dermis. The results of this study also suggest that the state of hydration of the stratum corneum plays an important role in determining the kinetics of drug transfer from liposomes into skin.

Topical delivery of peptide drugs has gained increasing attention and is considered to be one of the beneficial routes for the delivery of these drugs in view of its by-pass of gastrointestinal degradation and better patient compliance. In the case of skin disorders, systemic delivery of peptides may be ineffective because of rapid clearance from the circulation and an inability to deliver the molecule to the appropriate cells. Peptides usually are large molecular weight drugs of hydrophilic nature which are not readily topically absorbed. The objectives of the study were to study the topical absorption of gamma-interferon and transforming growth factor-alpha (TGF-α) in vitro by using a liposomal formulation. This study also describes the evaluation of the effect of lipid composition and the concentration of lipids on the deposition of the growth factors into the various strata of hairless mouse and pig skin in vitro, in an attempt to find an optimum formulation for optimal drug deposition. The deposition of drug and lipid into the deeper strata of skin upon application of dehydrated/rehydration liposomes was examined using radiolabeled drug and lipid markers. The studies have shown that by using a liposomal formulation, it is possible to deliver peptide drugs to the stratum corneum, as well as to the deeper skin strata where the basal cell layers reside. The extent of deposition of hydrophilic drugs from liposomal preparations following topical application is dependent on a variety of factors, particularly the liposomal lipid composition, the drug to lipid ratio and the tissue/follicular density of the application site. The drug deposition into the deeper skin strata (epidermis and dermis) was optimum for formulations where the drug/lipid ratio was less than 1:10. For all formulations tested, deposition into the deeper skin strata was much higher for the animal species having a more significant follicular route. By using liposomal formulations it is thus possible to deliver these large peptide molecules into the skin where they can exert their pharmacological effect.

**P231 AMIKACIN-LOADED LIPOSOMES • PRODUCTION, CHARACTERIZATION AND TREATMENT EFFICIENCY IN MYCOBACTERIUM AVIUM INTRACELLULARE COMPLEX (MAC) INFECTED MICE**

W.E. Buckle, H.C.R. Hilsenrath, S. Elbers, J.E. Diederichs, R.H. Müller, H. Hohn

1. Institut für Pharmazie, Pharmazeutische Technologie, 30166 Berlin, Germany
2. Institut für Pharmazeutische Technologie, 30129 Berlin, Germany

Amikacin, an aminoglycoside, is active in vivo against MAC infections [1]. The encapsulation of amikacin in liposomes increases the treatment efficiency [2]. Many published results using liposomes as vehicle for this drug lack of precise characterization and little or no mention was given to an adequate pharmaceutical quality [3]. However it is a prerequisite for registration by the regulatory authorities. In this study, general aspects of the influence of physico-chemical data of liposome formulation on treatment efficiency were investigated.

Production of liposomes was performed using an ethanol injection method. A phospholipid/stabilizer/ethanol mixture was injected into an aqueous drug solution at high speed stirring. This led to liposomes of 180nm to 250nm average diameter, accessible for vascular filtration. We adjusted the surface charge and the phospholipid bilayer rigidity (micropolarity) by additives, e.g., tocopherol acetate or retinol palmitate. Particle size and polydispersity index were measured by photon correlation spectroscopy, the zetapotential by laser doppler anemometry. Micropolarity was determined by fluorescence polarization measurements using diphenylhexatriene as indicator. These additives increased the phospholipid bilayer rigidity from 390°/Psec to a maximum of 1800°/Psec. The higher rigidity improves physical stability and can reduce the diffusion of drug into the outer phase. Furthermore the pharmaceutical quality of drug-loaded liposomes was compared with a commercial fat emulsion for parenteral nutrition as far as particle size is concerned. The maximum droplet diameter and total number of large particles per volume unit were analyzed by Coulter Counter technique. The control large particles, which can potentially block blood capillaries, was similarly low in liposome dispersion and commercial, registered emulsions.

Infection of mice by Mycobacterium avium strain TMC 724 (1x10⁷ CFU/mouse) was treated with a regimen of liposomes-encapsulated amikacin. The suspension was administered i. v. three times weekly for two weeks. Bacterial counts and histological staining were performed on spleen, liver and lung obtained 7 days after cessation of treatment. Two different stages of infection – one and five weeks after inoculation – were studied. Microbiological and histopathological differences could be detected depending on the drug formulation (liposomes-encapsulated vs. aqueous), its dosage and the grade of disease when treatment was started. Liposomal amikacin was superior in every aspect studied. Kinetic studies were initiated to quantify the prolonged release effect of liposomes and to optimize administration intervals.