ELSEVIER

International Journal of Pharmaceutics 109 (1994) 147–154

international journal of pharmaceutics

The effect of mixed micellar systems, bile salt/fatty acids, on the solubility and intestinal absorption of clofazimine (B663) in the anaesthetised rat

Joan R. O'Reilly, Owen I. Corrigan, Caitriona M. O'Driscoll *

University of Dublin, Department of Pharmaceutics, School of Pharmacy, Trinity College, 18 Shrewsbury Road, Dublin 4, Ireland

(Received 26 January 1994; Accepted 4 March 1994)

Abstract

Clofazimine (B663) is a highly lipophilic drug used in the treatment of leprosy. The solubility and gastrointestinal membrane permeability (P_{app}) of B663 in mixed micellar systems were examined. Membrane permeability was determined using a rat gut perfusion model and, in addition, these studies incorporated the hydrophilic marker PEG 4000. The mixed micellar systems studied contained the bile salt, sodium cholate (NaC), in association with different fatty acids including caprylic acid, oleic acid and linoleic acid. At a set concentration of NaC (40 mM) the solubility of B663 increased with increasing concentration of each fatty acid. Relative to NaC, the maximum enhancement in solubility (16-fold) was obtained with the NaC/linoleic acid (40:40 mM) system. An optimum bile salt/fatty acid ratio of 1:1 existed for maximum solubility enhancement. All mixed micellar systems enhanced the absorption of B663 relative to the simple micelle. The P_{app} tended to increase with increasing fatty acid concentration, maximum enhancement being obtained with the NaC/linoleic acid 40:40 mM system. With each mixed micellar system a higher P_{app} was obtained with lower drug loading. The effects of the mixed micellar systems on the absorption of PEG 4000 varied with fatty acid loading. These results have shown that mixed micelles can enhance the absorption of B663 to a greater extent relative to non-micellar and simple micellar systems. Maximum enhancement (> 800-fold) in the rate of B663 absorption was obtained with the NaC/linoleic acid 40:40 mM system. These results offer a possible explanation for the reported enhancement in gastrointestinal absorption of B663 when co-administered with fatty materials.

Key words: Clofazimine; Polyethylene glycol 4000; Sodium cholate; Fatty acid; Mixed micelle; Solubility; Membrane permeability

1. Introduction

Clofazimine ($C_{27}H_{22}Cl_2N_4$) is a rimno-phenazine derivative used in the treatment of dapsone resistant leprosy. It is a lipophilic compound the gastrointestinal absorption of which is enhanced by lipids (Yawalker and Vischer, 1979). We have previously shown (O'Reilly et al., 1994) that the solubility of B663 is significantly enhanced in the presence of a range of bile salts including sodium cholate, a trihydroxy unconjugated bile salt. Stud-

* Corresponding author.

SSDI 0378-5173(94)00074-F

ies on rat gut membrane permeability of B663 from a range of different concentrations of sodium cholate indicated that significant enhancement occurred only at the higher concentration of 80 mM.

In the gastrointestinal tract during lipid digestion bile salts are found associated with phospholipids, fatty acids and monoglycerides. In combination with bile salts these lipoidal compounds form mixed micelles. Many studies have reported enhanced bioavailability of drugs when administered in combination with mixed micellar solutions (Palin, 1985; Tomita et al., 1988; Park et al., 1992). The aim of this study was to extend the previous simple micelle work and to investigate the effect of bile salt/fatty acid mixed micellar systems on the solubility, and gastrointestinal absorption of clofazimine in the rat.

2. Theory

The apparent permeability coefficient (P_{app}) of the drug is an indication of the overall permeability of the gastrointestinal barrier to the drug and is defined in Eq. 1 (Ho et al., 1977, 1983):

$$P_{app} = 1/\left[(1/P_{aq}) + (1/P_m)\right] \quad (1)$$

where P_{aq} is the permeability coefficient of the aqueous boundary layer for drug molecules (cm s^{-1}) and P_m represents the membrane permeability coefficient (cm s^{-1}).

Values of P_{aq} and P_m may be calculated as follows:

$$P_{aq} = D_{aq}/h \quad (2)$$

where D_{aq} is the aqueous diffusion coefficient of all drug species (cm s^{-1}) and h denotes the effective thickness of the aqueous boundary layer.

$$P_m = P_o + P_p \quad (3)$$

where P_o is the permeability coefficient of the lipoidal pathway of the membrane for the non-dissociated drug species and P_p represents that of the aqueous pore pathway.

The experimental apparent permeability coefficient was calculated for each system using the following expression:

$$P_{app} = -(Q/2\pi rl) \cdot \ln(C(l)/C(0)) \quad (4)$$

where Q is the flow rate (ml s^{-1}) and $C(l)/C(0)$ denotes the fraction of drug remaining in the intestinal lumen of length l and effective lumenal radius r.

The rate of drug absorption (R), mg s^{-1}, was also calculated for each system according to the following:

$$R = (C(0) - C(l)) \cdot Q \quad (5)$$

where $(C(0) - C(l))$ is the difference in concentration of drug entering and leaving the lumen.

3. Experimental

3.1. Materials

All bile salts and fatty acids of least 98% purity were obtained from Sigma Chemical Co. Clofazimine (B663) was obtained from the Health Research Board Laboratories, Trinity College, Dublin. [^{14}C]PEG 4000 (specific activity 15 mCi/g) was obtained from Amersham. All solvents used were of HPLC grade. Sorenson's phosphate buffer was prepared using Analar grade phosphate salts.

3.2. Preparation of mixed micelles

Mixed micellar solutions were prepared by adding the fatty acid incrementally to concentrated solutions of the relevant bile salt in buffer (pH 7.2). All systems were stirred continuously at 37°C. Saturated solutions were prepared by adding excess drug to the system and sonicating for 60 min. The solutions were then filtered twice through a 0.2 μm HPLC grade filter.

3.3. Determination of solubility

The saturated solubility of clofazimine in each of the systems under investigation was determined using the method of Hamlin and Higuchi (1966). Samples were assayed by UV at 283 nm.

3.4. Rat gut perfusion studies

Absorption studies were conducted using a rat intestinal perfusion technique (Komiya et al.,

1980), and samples were assayed for B663 by HPLC, as previously described (O'Reilly et al., 1994). To assess membrane integrity all systems incorporated a 'non-absorbable' marker, [^{14}C] PEG 4000, which was assayed by liquid scintillation counting (O'Reilly et al., 1994). All perfusate samples collected were weighed, the results obtained indicating no significant change in flow output rate during perfusion. The P_{app} values were calculated using the actual fluid flow rate based on the weight of perfusate sample.

3.5. Measurement of diffusion coefficients

Diffusion coefficients were measured by two separate techniques: (1) quasi-elastic light-scattering analysis was used to determine the diffusion coefficients of micellar aggregates. Solution viscosities were determined using a U-tube viscometer; and (2) diffusion cell method as described by Goldberg and Higuchi (1968). A silver membrane filter of diameter 25 mm and pore size 0.45 μm (Millipore AG4502500) was used. The cell was calibrated using benzoic acid as a standard substance of known diffusion coefficient.

4. Results and discussion

The effects of mixed micellar solutions, formed by incorporating various fatty acids into the bile salt sodium cholate (NaC), on the solubility and gastrointestinal absorption of B663 were investigated.

4.1. Solubility of clofazimine in mixed micelles

Previous studies (O'Reilly et al., 1994) have shown that a range of simple bile salt micellar systems enhance the solubility of clofazimine; at a concentration of 40 mM this enhancement varied from 17- to 30-fold, with sodium cholate and sodium deoxycholate, respectively. In this study the solubility of B663 in mixed bile salt/fatty acid micelles was measured. Initially four fatty acids were examined: caprylic acid, a medium-chain saturated fatty acid ($C_{8:0}$); stearic acid, a long-chain saturated fatty acid ($C_{18:0}$); oleic acid, a long-chain unsaturated fatty acid ($C_{18:1}$); and linoleic acid, a long-chain unsaturated fatty acid ($C_{18:2}$). Attempts to make mixed micelles using stearic acid were unsuccessful, within the concentration range studied.

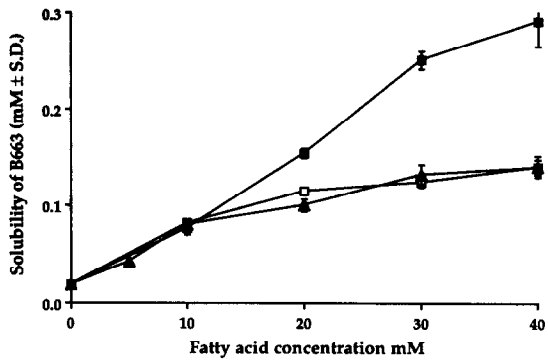

Fig. 1. Solubility of clofazimine (B663) (mean ± S.D.) in mixed micellar systems containing sodium cholate (40 mM) and varying concentrations of fatty acids: (■) linoleic acid; (▲) oleic acid; (□) caprylic acid.

The solubility of clofazimine in the presence of sodium cholate (40 mM)/fatty acid mixed micelles, at pH 7.2, with increasing concentration of fatty acid is shown in Fig. 1. The solubility of B663 increased with increasing concentration of each fatty acid, the plots tending to show a negative curvature at higher concentrations. It appears that as the ratio of bile salt/fatty acid approaches 1:1 the increase in solubility begins to plateau. The solubility enhancements observed in the presence of caprylic acid and oleic acid were very similar. The relative enhancement in the presence of linoleic acid (16-fold) was approximately double that of the other fatty acids, in the bile salt/fatty acid 1:1 systems.

The solubility of B663 in the presence of sodium cholate/caprylic acid mixed micelles at various concentrations of both the bile salt and the fatty acid was measured. These results show that as the concentration of fatty acid increased the solubility of B663 reached a maximum and then began to decrease (Fig. 2). This decrease appeared to occur at a bile salt/fatty acid ratio of < 1. These systems with sodium cholate/caprylic acid were optically transparent when prepared.

Bile salts possess the ability to solubilise fatty acids into mixed micelles. The fatty acid is likely to be solubilised within the hydrocarbon portion of the micelle. The bile salt micelle swells appreciably on the addition of even a small amount of fatty acid, e.g., the micellar weight of the sodium deoxycholate micelle in 0.1 M sodium chloride increases from 4950 to 7810 for solutions containing 1 mol of oleate to 5 mol of deoxycholate (Small, 1971). In this study, the observed increase in solubility in the presence of the mixed micelle is attributed to an increased affinity of the drug for the lipophilic mixed micelle interior. The decrease in solubility, at bile salt/fatty acids ratios of < 1, may be the result of a phase change within the mixed micellar systems. Staggers et al. (1990) prepared equilibrium phase diagrams corresponding to the aqueous lipid composition of the upper small intestinal contents during lipid digestion. Analysis of the hydrodynamic radius of the micellar species present showed that, with increases in the ratio of mixed intestinal lipids (MIL) to mixed bile salts (MBS), the composition of the micellar phase changed. At a ratio of 0.8–1.2 the radius increased abruptly to values typical of those of vesicles and two phases co-existed, namely, cholesterol and MIL saturated micelles along with unilamellar vesicles. Alternatively, the decrease in solubility may be indicative of competition between the drug and the fatty acid for the bile salt micelle (Crooks et al., 1973).

The differences in solubility enhancement observed with different fatty acids may be dependent on structural factors. The solubility enhancement increased with an increase in the degree of unsaturation of the fatty acids. In addition, the lower melting point fatty acid resulted in greater drug solubilization.

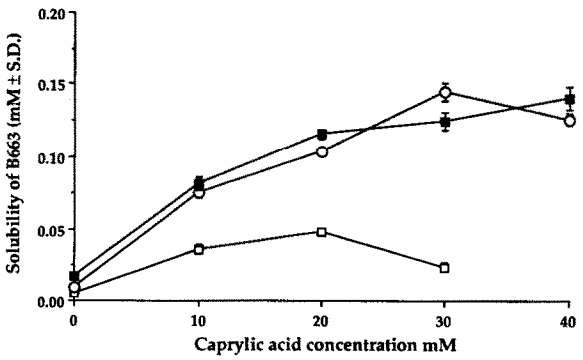

Fig. 2. Solubility of clofazimine (B663) (mean ± S.D.) in mixed micellar systems containing sodium cholate (NaC) and caprylic acid: (■) 40 mM NaC; (○) 30 mM NaC; (□) 20 mM NaC.

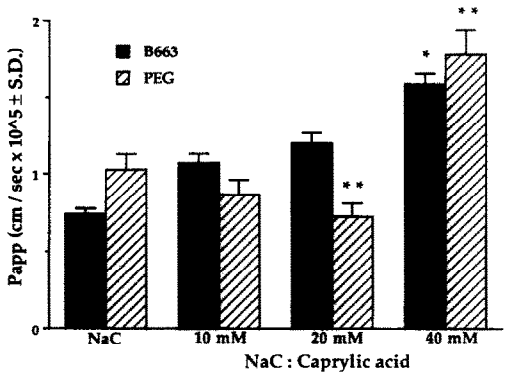

Fig. 3. Apparent permeability coefficients (mean ± S.D.) of clofazimine and PEG 4000 from mixed micellar systems containing sodium cholate (NaC) (40 mM) and varying caprylic acid loading (statistically significant from the P_{app} in 40 mM NaC at the * 99% and the ** 95% level).

4.2. *Absorption from mixed micellar systems*

The effects of changing a range of parameters, including (1) the type of fatty acid used, (2) the loading of the fatty acid into the micelle, and (3) the drug loading in the micelle, on the gastrointestinal absorption of B663 and PEG 4000 from mixed micelles were estimated.

4.2.1. *Type of fatty acid*

In saturated solutions of B663 all mixed micellar systems studied enhanced drug absorption relative to the simple micelle (Fig. 3 and 4). Maximum absorption was observed with linoleic acid, the long-chain unsaturated fatty acid. The effect of the mixed micellar systems on the absorption of PEG 4000 varied with the fatty acid loading (Fig. 3 and 4).

4.2.2. *Fatty acid loading*

4.2.2.1. Clofazimine. The effect of fatty acid loading on the absorption of B663, in saturated solu-

tions, was also examined. The P_{app} tended to increase with increasing fatty acid concentration. In all mixed micellar systems containing 40 mM fatty acid a significant increase in P_{app} of B663 relative to the simple NaC micelle was obtained (Fig. 3 and 4). Maximum enhancement was obtained with the NaC/linoleic acid systems, at 40:40 mM a 4-fold increase in the P_{app} of B663 relative to the simple micelle was observed. Decreasing the linoleic acid concentration from 40 to 20 mM did not significantly affect the P_{app} of B663 (Fig. 4).

All mixed micellar systems investigated increased the rates of absorption of B663 relative to non-micellar and simple micellar systems. Maximum enhancement of the order of ~800-fold was obtained with the NaC/linoleic acid 40:40 mM system (Table 1).

The enhanced P_{app} of B663 in the presence of bile salt fatty acid mixed micelles may be attributed to a combination of the mixed micelle facilitating diffusion of the drug through the ABL, and, an effect on the permeability of the membrane thus facilitating absorption of the micellised drug. The effect of the mixed micelle on the ABL may be to reduce the thickness of the mucus layer in a fashion similar that of the simple bile salt micelle (O'Reilly et al., 1994). Several reports in the literature have suggested that fusogenic fatty acids may affect the permeability of the

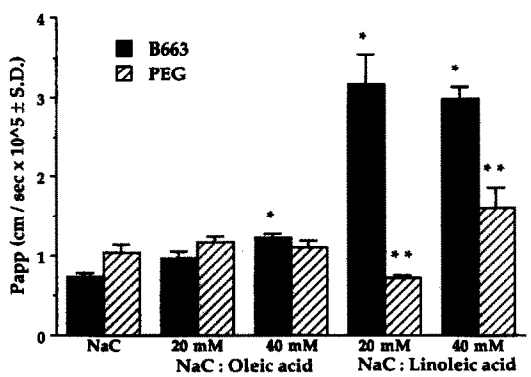

Fig. 4. Apparent permeability coefficients (mean ± S.D.) of clofazimine and PEG 4000 from mixed micellar systems containing sodium cholate (NaC) (40 mM) and varying oleic acid and linoleic acid loadings (statistically significant from the P_{app} in 40 mM NaC at the * 99% and the ** 95% level).

Table 1
Rate of absorption of clofazimine from mixed bile salt/fatty acid (40:40 mM) micellar systems

System	Rate ($mg\ s^{-1}$) ($\times 10^5$) (± S.D.)	Relative enhancement
Buffer (pH 7.2)	0.013 ± 0.002	–
Sodium cholate (NaC) (40 mM)	0.216 ± 0.009	16.64
NaC/oleic acid (40:40 mM)	2.505 ± 0.107	193
NaC/caprylic acid (40:40 mM)	3.240 ± 0.155	249
NaC/linoleic acid (40:40 mM)		
Saturated with B663	10.470 ± 0.381	805
Unsaturated with B663	2.630 ± 0.120	202

intestinal membrane to a drug (Maggio and Lucy, 1976; Muranishi, 1985). Muranishi et al. (1981) investigated the effects of various fatty acids on membrane fluidity and reported that the lower the melting point of the fatty acid (long-chain unsaturated fatty acids have lower melting points than saturated acids) the more the membrane was disordered. It was suggested that the enhanced membrane permeability caused by fusogenic lipids was assocciated with disorder in the hydrophobic region of the lipid bilayer and the interaction of the incorporated lipid with the polar head group of the membrane phopholipid.

Kajii et al. (1988) have demonstrated that caprylic acid exerted a membrane perturbing action on rat small intestine brush border membrane vesicles and liposomes in vitro, and that this effect was related to interaction with a membrane protein. The enhanced P_{app} of B663 in the presence of mixed micelles containing the medium-chain saturated caprylic acid may therefore be explained in terms of a slightly different mechanism to that of the fusogenic oleic and linoleic acids.

4.2.2.2. PEG 4000. The effects of the mixed micellar systems on the P_{app} of the drug and the PEG 4000 were not directly correlated. At low concentrations of caprylic and linoleic acids (10–20 mM) the P_{app} of PEG 4000 decreased compared to that obtained with the simple bile salt micelle. However, as the concentration of fatty acid was increased this trend was reversed, a

> 2-fold increase in the P_{app} for PEG 4000 being obtained when caprylic acid was increased from 20 to 40 mM (Fig. 3). This effect was not obvious with oleic acid. However, when the membrane permeability of PEG 4000 (P_p) was calculated, as previously described (O'Reilly et al., 1994), the P_p increased from 1.357×10^{-5} cm s^{-1} with the simple NaC micelle (40 mM) to 2.783, 1.600, and 2.159×10^{-5} cm s^{-1} with mixed micellar systems (40 : 40 mM) containing caprylic, oleic and linoleic acids, respectively.

We previously reported that the P_{app} of PEG 4000 increased with increasing concentration of the bile salt NaC (O'Reilly et al., 1994). It was postulated that this enhancement was due to a change in the paracellular permeability of the membrane caused by the effect of the bile salt on the tight junctions.

In the case of the mixed micellar systems used in the current study, the fatty acids at low concentrations appear to exercise a protecting effect on the membrane, i.e., masking the effect of the bile salt on the tight junctions. This effect may be due in part to the fatty acid reducing the CMC of the bile salt and therefore reducing the active molecular concentration of bile salt available. However, at high fatty acid concentrations (bile salt/fatty acid ratio of 1 : 1) the 'protective' effect is reduced.

The enhanced P_{app} of PEG 4000 at the high fatty acid concentration may be due to a change in the paracellular permeability. This is consistent with the work of Tomita et al. (1988), who investigated the absorption enhancement mechanisms for a range of fatty acids and bile salt/fatty acid mixed micelles using an everted sac procedure. The colonic pore radius was reported to increase from control values of 0.8–0.9 to 1.1–1.2 nm in the presence of sodium caprylate and to 1.3–1.6 nm in the presence of mixed micelles. In addition, Tomita et al. (1988) reported that the thickness of the ABL in the perfused colonic loop was reduced to approximately one quarter the control in the presence of sodium caprate and from approx. 0.112 to 0.072 cm in the presence of the mixed micellar system. These results suggesting that a decrease in the resistance of the ABL represents an additional promotional mechanism are consistent with the proposed mechanism of enhancement for B663.

Table 2
Experimental permeability coefficients (P_{app}) of clofazimine (B663) from simple bile salt (40 mM) and mixed bile salt/fatty acid (40 : 40 mM) micellar systems unsaturated and saturated with B663

Micellar system	P_{app} (cm s^{-1}) ($\times 10^5$) ($\pm$S.D.)	
	Unsaturated	Saturated
Sodium cholate (NaC)	1.164 ± 0.150	0.738 ± 0.041
NaC/caprylic acid	2.710 ± 0.183 [a]	1.590 ± 0.067
NaC/oleic acid	2.470 ± 0.237 [a]	1.217 ± 0.060
NaC/linoleic acid	3.890 ± 0.280 [a]	2.974 ± 0.152

[a] Statistically significant from P_{app} of corresponding saturated system at $\geq 99\%$.

4.3. Drug loading

To investigate the effect of drug loading in the mixed micelle, absorption was investigated from corresponding systems unsaturated with drug. The initial concentration of drug in each of the perfusates was constant at 0.0264 mg ml^{-1}. The saturation levels varied depending on the solubility of the drug in the relevant system. The degrees of saturation were 38.9, 39.9 and 19.16% for the NaC systems with caprylic, oleic and linoleic acids, respectively. With each fatty acid system a higher P_{app} was obtained with lower drug loading (Table 2). This trend is consistent with the results obtained for simple bile salt micellar systems (O'Reilly et al., 1994). The rates of absorption of B663 from the unsaturated systems were lower than from the corresponding saturated systems (Table 1).

The aqueous diffusion coefficients (D^*_{aq}) of NaC/caprylic acid systems saturated and unsaturated with B663 were measured to assess the effect of micelle loading on the permeability of the ABL to the micellised species (P^*_{aq}). The D^*_{aq} were measured by two methods, quasielastic light scattering and a diffusion cell technique (Table 3). The D^*_{aq} of the mixed micelle is of the order of 10×10^{-7} cm^2 s^{-1} in the absence of drug. The values are essentially the same for the mixed micelle containing a 2 : 1 and a 1 : 1 ratio of bile salt to fatty acid. Addition of B663 results in a

Table 3
Aqueous diffusion coefficients (D^*_{aq}) of sodium cholate/caprylic acid mixed micellar systems

Micellar system	D^*_{aq} ($cm^2\ s^{-1}$) ($\times 10^7$) ($\pm$S.D.)	
	Method A [a]	Method B [b]
40:40 mM NaC/caprylic acid	10.78	–
40:20 mM NaC/caprylic acid	10.98	–
40:40 mM NaC/caprylic acid		
Unsaturated with B663	10.34	8.650 ± 0.44
Saturated with B663	0.341	0.655 ± 0.18

[a] Quasi-elastic light-scattering data.
[b] Diffusion cell method.

decrease in D^*_{aq} with an increase in saturation. The mean D^*_{aq} of the NaC/caprylic acid 40:40 mM system unsaturated with drug is 9.21×10^{-7} $cm^2\ s^{-1}$. This is a slightly lower rate of diffusion than the micelles in the absence of drug. Increasing the concentration of drug to form a saturated system, however, led to a further reduction and a mean D^*_{aq} of 0.55×10^{-7} $cm^2\ s^{-1}$. The magnitude of the difference in D^*_{aq} was unexpected. Amidon et al. (1982), using progesterone in Tween 80 as a model micelle solubilised system, showed that the difference in D^*_{aq} for a micellar system saturated and unsaturated with progesterone was insignificant.

The increase in D^*_{aq} of the unsaturated, relative to the saturated, and the consequent increase in the P^*_{aq} was not sufficient to explain the observed enhancements in permeability coefficient. In addition to the effect on P^*_{aq}, it is postulated that the enhanced effective permeability is due in part to direct transfer of drug from the micelle to the membrane.

5. Conclusions

The bile salt/fatty acid mixed micellar systems, containing either caprylic, oleic or linoleic acids, resulted in further enhancements in the P_{app} of B663 compared to those previously reported for simple micelle systems (O'Reilly et al., 1994). The rates of absorption of B663 also increased, maximum enhancements of the order of 800-fold being obtained with the NaC/linoleic acid 40:40 mM system.

The alterations in the absorption of B663 in the presence of bile salt/fatty acid micelles arise from a combination of a number of factors including increased solubility due to micellar solubilization, reduction in the resistance imposed by the ABL, a specific fatty acid related effect on membrane permeability, and an effect on the diffusion coefficient.

The effects of the mixed micellar systems on the absorption of B663 and PEG 4000 were not directly correlated. At low concentrations of fatty acid the absorption of PEG 4000 tended to decrease relative to the simple micelle; this trend was reversed at higher fatty acid concentrations. The increase in the P_{app} of PEG 4000 at the higher fatty acid concentrations was consistent with increases in paracellular membrane permeabilities.

Acknowledgements

The authors wish to thank Dr J.F. O'Sullivan, formerly of the Health Research Board Laboratories, Trinity College Dublin, Ireland, for the supply of clofazimine. We also thank Dr N. Crowther, School of Pharmacy, University of Bradford, U.K., for the quasi-elastic light-scattering analysis.

References

Amidon, G.E., Higuchi, W.I. and Ho, N.F.H., Theoretical and experimental studies of micelle-solubilized solutes. *J. Pharm. Sci.*, 71 (1982) 77–84.

Crooks, M.J. and Brown, K.F., A note on the solubilization of preservative mixtures by cetomacrogol. *J. Pharm. Pharmacol.*, 25 (1973) 281–284.

Goldberg A.H. and Higuchi, W.I., Improved method for diffusion coefficient determination employing the silver membrane filter. *J. Pharm. Sci.*, 57 (1968) 1583–1585.

Hamlin, W.E. and Higuchi, W.I., Dissolution rate-solubility behavior of 3-(1-methyl-2-pyrrolidinyl)-indole as a function of hydrogen ion concentration. *J. Pharm. Sci.*, 55 (1966) 205–207.

Ho, N.F.H., Park, J.Y., Morozowich, W. and Higuchi, W.I., Physical model approach to the design of drugs with

improved intestinal absorption. In Roche, E.B. (Ed.), *Design of Biopharmaceutical Properties through Prodrugs and Analogues,* Am. Pharm. Assoc., Washington, 1977, pp. 136–227.

Ho, N.H.F., Park, J.Y., Ni, P.F. and Higuchi, W.I., Advancing quantitative and mechanistic approaches in interfacing gastrointestinal drug absorption studies in animals and humans. In Crouthamel, W. and Sarapu, A.C., (Eds), *Animal Models for Oral Drug Absorption in Man,* Am. Pharm. Assoc., Washington, 1983, pp. 27–106.

Kajii, H., Horie, T., Hayashi, M., Awazu, S., Fluorescence study of the membrane-perturbing action of sodium caprylate as related to promotion of drug absorption. *J. Pharm. Sci.,* 77 (1988) 390–392.

Komiya, I., Park, J.Y., Kamani, A., Ho, N.F.H. and Higuchi, W.I., Quantitative mechanistic studies in simultaneous fluid flow and intestinal absorption using steroids as model solutes. *Int. J. Pharm.,* 4 (1980) 249–262.

Maggio, B. and Lucy, J.A., Polar-group behaviour in mixed monolayers of phospholipids and fusogenic lipids. *Biochem. J.,* 155 (1976) 353–364.

Muranishi, N., Takagi, N., Muranishi, S., Sezaki, H., Effect of fatty acids and monoglycerides on permeability of lipid bilayer. *Chem. Phys. Lipids,* 28 (1981) 269–279.

Muranishi, S., Modification of intestinal absorption of drugs by lipoidal adjuvants. *Pharm. Res.,* 3 (1985) 108–118.

O'Reilly, J.R., Corrigan, O.I. and O'Driscoll, C.M., The effect of simple micellar systems on the solubility and intestinal absorption of clofazimine (B663) in the anaesthetised rat. *Int. J. Pharm.,* 105 (1994) 137–146.

Palin, K.J., Lipids and oral drug delivery. *Pharm. Int.,* 6 (1985) 272–275.

Park, G.B., Shao, Z. and Mitra, A.K., Acyclovir permeation enhancement across intestinal and nasal mucosae by bile salt-acylcarnitine mixed micelles. *Pharm. Res.,* 9 (1992) 1262–1267.

Small, D.M., The physical chemistry of the cholanic acids. In Nair, D.P. and Kritchevsky, D. (Eds), *The Bile Acids, Chemistry, Physiology and Metabolism,* Vol. 1, Plenum, New York, 1971, pp. 249–356.

Staggers, J.E., Hernell, O., Stafford, R.J. and Carey, M.C., Physical-chemical behavior of dietary and biliary lipids during intestinal digestion and absorption: I. Phase behavior and aggregation studies of model lipid systems patterned after aqueous duodenal contents of healthy adult human beings. *Biochemistry,* 29 (1990) 2028–2040.

Tomita, M., Shiga, M., Hayashi, M., Awazu, S., Enhancement of colonic drug absorption by the paracellular permeation route. *Pharm. Res.,* 5 (1988) 341–346.

Yawalker, S.J. and Vischer, W., Lamprene (clofazimine) in leprosy. *Lepr. Rev.,* 50 (1979) 135–144.