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Study of the influence of sodium taurocholate (STC) and sodium glycocholate (SGC) on the mass transfer of certain drugs. Diethylstilbestrol

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Summary

Diethylstilbestrol had a solubility of 1.05 mg/100 ml and a negligible dissolution efficiency in phosphate buffer. The presence of bile salts raised the solubility to a value as high as 219.5 and 218.25 mg/100 ml and the dissolution efficiency to 57.7 and 46.4% for STC and SGC systems, respectively. The addition of lecithin, cholesterol, lecithin plus cholesterol or fatty acids (lauric, myristic, palmitic) or monoglycerides (glyceryl monolaurate, glyceryl monomyristate or glyceryl monostearate) to STC or SGC aqueous solutions did not have any significant effect on the solubility or dissolution rate of the drug. However, the simultaneous presence of these additives in the medium enhanced the solubility but the dissolution rate remained practically unchanged.

Introduction

Diethylstilbestrol (DES) is a powerful synthetic oestrogen which is not a steroid but has actions and uses similar to those of oestradiol (Martindale, 1977). It is practically insoluble in water and reported to be excreted in bile (Caldwell and Cline, 1976).

The solubilities of the closely related non-steroidal oestrogens, dienosterol, hexestrol and DES, were found to increase in the presence of ionic and non-ionic surfactants (Nakagawa, 1954). Again, the solubilization of hexestrol by bile salt solutions was studied by Bates et al. (1966a, b, c) who investigated the influence of the addition of inorganic electrolytes and lipids on the solubilization of this drug by

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individual bile salts as well as the influence of mixed bile salts on such solubilization.

As in previous publications issued from this laboratory (Mattha et al., 1980; Kassem et al., 1980), the present publication considers the solubility and dissolution rate of diethylstilbestrol in aqueous solutions of the conjugated bile salts, STC or SGC, in the presence of some lipids prevailing in the small intestine; these were either bile constituents (lecithin and cholesterol) or products related to fat digestion (fatty acids: lauric, myristic, palmitic; monoglycerides: glyceryl monolaurate (GML), glyceryl monomyristate (GMM), glyceryl monostearate (GMS)). The measurements were carried out at a temperature of 37°C, pH of 6.4, a sodium ion concentration of 0.15 mol/l and a bile salt concentration of 0.04 mol/l, so as to assimilate physiological conditions prevailing in the human small intestine during absorption (Sjövall, 1959; Bates et al., 1966b).

Materials and methods

Materials

Sodium taurocholate from BDH Chemicals, Poole, U.K. Sodium glycocholate from I.C.N. (Pharmaceuticals, Life Sciences Group, Plainview, NY, U.S.A.) and both were used as received. Pure egg lecithin was from E. Merck, Darmstadt, Cholesterol, U.S.P.XIX was from E. Merck, Darmstadt. Lauric, myristic and palmitic acids, biochemical grade were from Akzo Chemie, Italy. Glycerol monolaurate and glyceryl monomyristate, pure were from Akzo Chemie, Italy. Glyceryl monostearate, pure was from Emery Industries, U.S.A. Diethylstilbestrol, U.S.P.XIX was from E. Merck, Darmstadt. Sodium chloride (A.R.); sodium hydroxide (A.R.); disodium hydrogen phosphate (A.R.).

Apparatus

Cecil Spectrophotometer model CE595; synchronous motor (Hanson, Research); pH meter, type PHM 22r (Radiometer-Copenhagen); membrame filters (Sartorius-

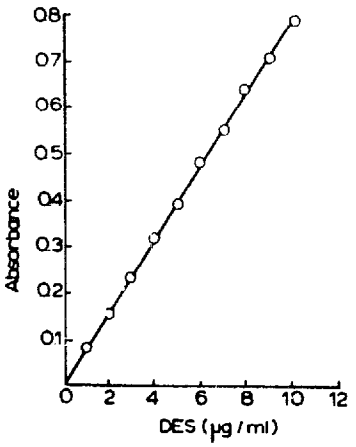

Fig. 1. Standard curve of DES.

Membrane filter GMBH-34 Göttingen, F.R.G., pore size 0.6 μm, 25 mm diameter); Warburg shaker; planimeter (type GK 800, Metrimpex, Budapest, Hungary).

Methods

The same as described in a previous publication (Mattha et al., 1980). For the determination of DES, the clear filtrate from the solubility or dissolution rate experiment was diluted with 0.1 N sodium hydroxide and the absorbance of the solution determined at a wavelength of 259 nm (Clarke, 1971) against a blank of the respective dissolution medium to correct for any absorbance of the salt or any lipid additive. The absorbance of the drug was found to follow Beer's law in the concentration range considered (Fig. 1).

Results and discussion

The influence of various lipid additives on the mass tansfer of DES is shown in Table 1 and Figs. 2, 3 and 4.

Influence of STG or SGC

Fig. 2 and Table 1 show that the addition of bile salt to phosphate buffer raises the solubility of DES in the latter solvent from 1.05 mg/100 ml to 219.5 and 216.3

TABLE 1

INFLUENCE OF LIPID ADDITIVES ON THE SOLUBILITY AND DISSOLUTION RATE (EXPRESSED IN TERMS OF DISSOLUTION EFFICIENCY, D.E.%) OF DES (37°C) IN AQUEOUS SYSTEMS CONTAINING 0.04 M STC OR SGC (SODIUM ION CONCENTRATION = 0.15 M)

Additive	STC		SGC	
	S [a]	DE%	S [a]	DE%
Phosphate buffer	1.05	-	1.05	-
Bile salt	219.50 [b]	57.50	216.25 [b]	46.39
Lecithin (0.2%)	220.75	53.89	208.75	41.94
Cholesterol (0.025%)	221.75	56.94	210.00	42.78
Lecithin + cholesterol	222.00	57.50	219.75	47.50
Lauric acid (0.4%)	220.50	56.38	221.50	47.22
Myristic acid (0.2%)	214.00	55.00	202.25	43.06
Palmitic acid (0.05%)	217.50	55.00	212.00	42.50
GML (0.4%)	218.11	52.78	223.25	47.78
GMM (0.2%)	219.00	56.67	211.00	43.06
GMS (0.025%)	219.00	56.67	219.50	46.94
All additives	230.00	58.33	228.00	48.61

[a] Solubility in mg/100 ml.

[b] Molar ratio of DES to STC $= \frac{219.50 \times 1000}{1000 \times 100 \times 268.34 \times 0.04} = 0.2$

and of DES to SGC $= \frac{216.25 \times 1000}{1000 \times 100 \times 268.34 \times 0.04} = 0.2$

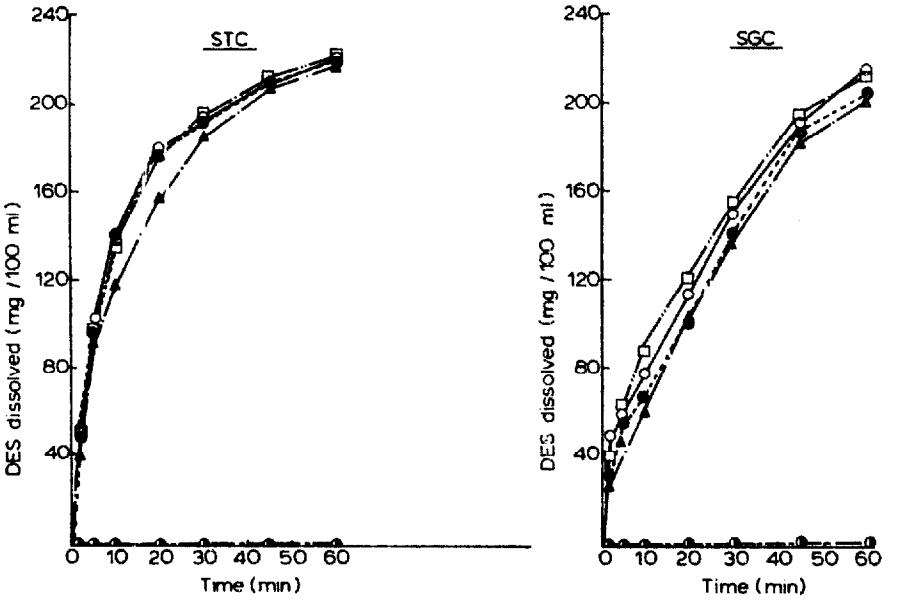

Fig. 2. Influence of lecithin and/or cholesterol on the dissolution rate of DES in buffered solutions containing trihydroxy bile salts. O------O, phosphate buffer; O——— O, bile salt; ▲——— ▲, bile salt + lecithin; ●——— ●, bile salt + cholesterol; □——— □, bile salt + lecithin + cholesterol.

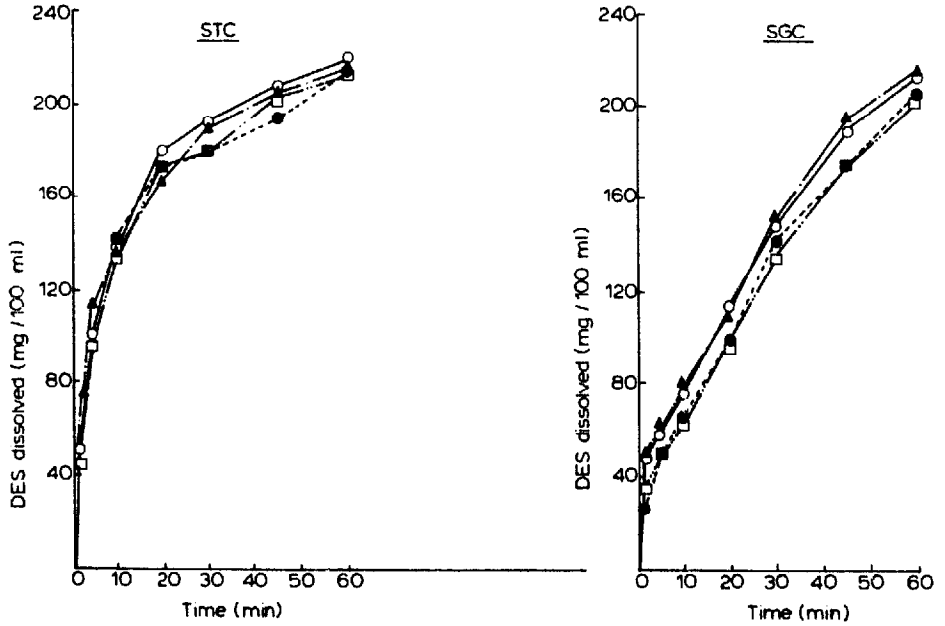

Fig. 3. Influence of fatty acids on the dissolution rate of DES in buffered solutions containing trihydroxy bile salts. O——— O, bile salt; ▲——— ▲, bile salt + lauric acid; ●——— ●, bile salt + myristic acid; □——— □, bile salt + palmitic acid.

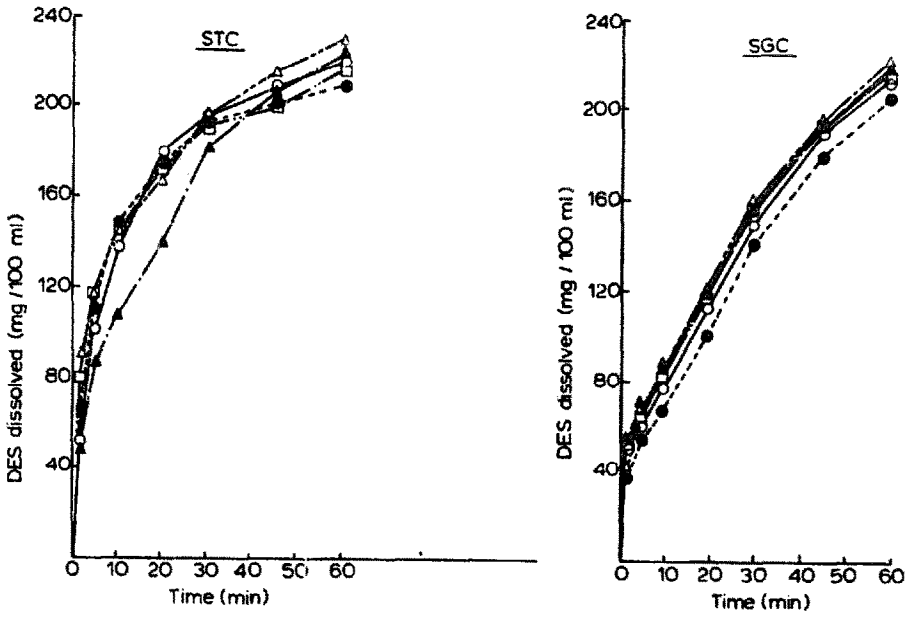

Fig. 4. Influence of monoglycerides and all additives on the dissolution rate of DES in buffered solutions containing trihydroxy bile salts. O——O, bile salt; ▲——▲, bile salt + GM laurate; ●——●, bile salt + GM myristate; □——□, bile salt + GM stearate; △——△, bile salt + all additives.

mg/100 ml and its dissolution efficiency[1] from negligible values to 57.5 and 46.4% in STC and SGC systems, respectively.

A similar dramatic increase in the solubility of the parent non-steroidal synthetic estrogen, hexestrol, was reported by Bates et al. (1966b) in simulated intestinal bile mixtures.

The molar ratio of DES at saturation to any of the two bile salts considered (known as saturation ratio (Carey and Small, 1972)), calculated on the basis of data presented in Table 1 is equal to about 0.2. On the basis of an average aggregation number[2] of 5 for the bile salt molecules, which is quite probable under the present experimental conditions (Carey and Small, 1972; Fontell, 1971), the drug would be found to obey the principle of one (or more) molecule of solubilizate per bile salt micelle (Carey and Small, 1970, 1972) which means that solubilization is most probably of micellar type.

The high solubility of DES in bile salt solutions compared to that of griseofulvin and digoxin (Mattha et al., 1980; Kassem et al., 1982) reflects its polar nature (Bates et al., 1966a, b; El-Gorab and Underwood, 1973; Fontell, 1972). The molecule of

[1] Dissolution efficiency, D.E. (%) (Khan, 1974): this is defined as the area under the dissolution curve up to a certain time, t, expressed as a percentage of the area of the rectangle described by 100% dissolution in the same time. In the present work, t = 60 min.

[2] According to Carey and Small (1972, p. 512) and Fontell (1971, p. 717), small micelles are formed by all bile salts, free or conjugated, in water at 20°C in solute concentrations greater than their critical micelle concentrations and less than 5 g/100 ml of solution. The aggregation numbers of these micelles range from 2 to 9. The aggregation numbers of trihydroxy bile salts remain less than 10 at counterion concentrations less than 1.0 M Na^+.

DES carries two phenolic groups on the hydrocarbon stilbene nucleus and would probably behave as an amphiphilic molecule.

It is most probable that DES is solubilized by bile salt micelles through a mechanism more or less similar to that described for lecithin (Small, 1967; Dervichian, 1968; Zimmerer, 1979). This is evidenced by the fact that DES, hexestrol and other estrogens with free hydroxyl groups are very soluble in some non-ionic, cationic and anionic surfactants, while those with all hydroxyl groups substituted are less soluble (Nakagawa, 1954). This suggests that the hydrophobic portion of the drug molecule interacts with the surfactant, while the strongly polar hydroxyl groups are directed to the surface of the micelle in the aqueous medium. This is in accordance with the hypothesis of Bates et al. (1966a) who suggested that hexestrol was being incorporated predominantly into the "palisade layer" of the bile salt micelles as a relatively polar solubilizate molecule would behave.

Influence of different additives

Figs. 2–4 and Table 1 show that the addition of lecithin, cholesterol, lecithin plus cholesterol, most of the fatty acids or any of the investigated monoglycerides does not have any significant effect on the solubility and dissolution rate of the drug while their presence together (all the lipids) in the medium enhances drug solubility, the dissolution rate remains practically unchanged.

However, the presence of myristic or palmitic acid in SGC solutions reduces the solubility and dissolution rate of the drug to some extent (figure 3 and table 1).

Here crops up a discrepancy between the present results and those obtained by Bates and his coworkers (Bates et al., 1966b) while studying the effect of lipid additives on the solubilization of the parent estrogen hexestrol by 0.04 M SIMULATED INTESTINAL BILE SALT MIXTURE. These authors had noted that fatty acids, monoglycerides and lecithin reduce the solubility of the drug in this mixture to extents that were much more sensible than reported in the present case (table 1).

However, this discrepancy would disappear when realizing the following facts:

a) All lipid additives used in the present study (and in Bates' study too (Bates et al., 1966b)) with the exception of lecithin and monolaurin, have been shown to act as non-polar additives in a system containing one bile salt at 37°C (Hofmann, 1963a and 1963b; Bates et al., 1966b) thereby decreasing the capacity of this system to dissolve polar solubilizates like hexestrol and DES.

b) Conjugated trihydroxy bile salts are known to be the most polar bile salts available (Carey and Small, 1972).

c) The simulated intestinal bile salt mixture used by Bates et al. included, in addition to STC and SGC (already representing a much smaller fraction of their mixture than in the present case) the two less polar bile salts sodium cholate and sodium desoxycholate.

Accordingly, it would be reasonable to assume that the tendency of the bile salt system used by Bates et al. to dissolve polar, hexestrol or DES, molecules was much lower than in the present case, accounting in this way for the results of Bates and his coworkers and for the observed discrepancy.

This is further evidenced in the present case by the lower solubility of DES in the

SGC-palmitic acid or SGC-myristic acid systems, SGC being less polar than STC (Norman, 1960a, 1960b) and myristic and stearic acids being fatty acids of very low polarity.

Conclusions

(1) The trihydroxy bile salts, STC or SGC, enhance the mass tansfer of DES.

(2) The bile salt component of the investigated systems seems to be the primary determinant in drug dissolution.

(3) Micellar solubilization is probably the mechanism involved in the enhancement of drug mass transfer in bile salt solutions in the case of DES.

It is noteworthy to mention in this respect that STC and SGC are known to be considerably less harmful to the intestinal tract than dihydroxy bile salts (Muranishi et al., 1979).

(4) Changes in the composition of the mixed bile salt–lipid micelles present in the dissolution medium seem to only slightly affect the solubilizing potential of these micelles towards DES.

(5) The combination of any of the two bile salts with all the investigated lipids of the intestinal mixture improves the dissolution of the investigated drug in bile salt solutions.

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