DRUG DEVELOPMENT AND INDUSTRIAL PHARMACY, 13(3), 547-563 (1987)

THE IN-VITRO UPTAKE OF A LOW DOSE DRUG (RIBOFLAVINE) BY SOME ADSORBENTS

Saleh A.H.Khalil, Lobna M.Mortada, M.A.Shams-Eldeen and Manal M.El-Khawas

Department of Pharmaceutics, Faculty of Pharmacy, University of Alexandria, Alexandria, Egypt.

ABSTRACT

An investigation was carried out of the in-vitro adsorption of a low-dose model drug (riboflavine) by three types of kaolin, attapulgite, magnesium trisilicate and a grade of magnesium aluminium silicate (veegum^{R}). The adsorption experiments were designed under conditions simulating in-vivo with respect to variations in pH values, volume of the adsorption medium, and the presence of electrolytes, a hydrophilic colloid and a surfactant. Under all conditions examined, riboflavine adsorption followed the sequence veegum > attapulgite > kaolin > magnesium trisilicate.

The presence of either veegum^{R}(1 g) or kaolin (4 g) in the medium reduced the level of drug in solution during dissolution rate testing of capsules. Desorption results suggest only partial release of the adsorbed drug. The results obtained emphasize the strong uptake of a potent ionic drug by the various silicates commonly used in pharmacy.

INTRODUCTION

The interaction of drugs with adsorbents represents a common type of drug interactions. Pharmaceutical materials possessing adsorptive properties are used in pharmacy and medicine either as drugs (e.g. kaolin and attapulgite) or as adjuvants (e.g. veegum^{R} and bentonite). Reports on drug-adsorbent interactions point to the strong in-vitro adsorption of anticholinergic drugs in anti-diarrheal mixtures onto kaolin and antacids (1,2). Also, aminoglycoside antibiotics (e.g. neomycin) are known to be adsorbed on negatively charged silicate adsorbents(3). The incorporation of adjuvants such as bentonite, veegum^{R} and formerly kaolin, in the dosage forms can reduce dissolution testing and/or drug availability.

The extent of such interactions would be significant with low dose drugs. The in-vivo release of riboflavine from hard gelatin capsules, formulated with kaolin as the diluent, has been studied by dissolution technique (4). Also, the uptake of thiamine hydrochloride by kaolin (added as a filler in tablets) was responsible for the

observed reduction in the vitamin extraction by 0.2 N HCl but not at higher pH values. When an acetate buffer of pH 6.0 was used, relatively higher percentages recovery were obtained (5).

The physical entrapment of organic molecules between silica-alumina layers could possibly explain the clinical failure of tolbutamide tablets in which veegum^{R} (at two concentrations) was used as a disintegrant (6). In-vitro adsorption testing may be of value as a screening tool in the studies of drug interactions. However, unless properly carried out under conditions simulating in-vivo, the results may be of little value.

The objective of the present work has been to examine the uptake and desorption of a low dose drug (represented by riboflavine) by some adsorbents under factors simulating in-vivo conditions, e.g., variations in pH, volume of the adsorption medium and presence of electrolyte, a surfactant and a hydrocolloid.

MATERIALS

Riboflavine was of B.P. grade. The adsorbents used were: attapulgite (regular), three types of kaolin namely: light kaolin (B.P.), natural kaolin (B.P.) and white fine kaolin (DAB 7), a grade of magnesium aluminium silicate (Veegum^{R}, H.V.) and magnesium trisilicate (U.S.P.). The electrolytes used were of A.R. quality. A commercial anti-diarrheal suspension known as Quintess^{R}, Eli Lilly, USA (each 30 ml contains: activated attapulgite 3 g and activated attapulgite colloidal 0.9 g) was also used.

METHODS

Adsorption Experiments:

In-vitro adsorption studies were carried out at 37±0.2°C by dissolving riboflavine in solutions having different pH values (2.1-7). Appropriate concentrations of either HCl or NaOH were used to adjust the pH values. The use of buffers was avoided in view of the effect of buffer species on the extent of adsorption. Fifty milliliters of a solution containing 1.5 to 15 mg per 100 ml of drug were placed in 100 ml stoppered conical flasks containing the adsorbent (1 g). Attapulgite, magnesium trisilicate and veegum^{R} were pretreated with varying volumes of 0.3-0.6 N HCl for one hour at 37±0.2°C, to adjust the pH of the medium to the required value. The suspensions were shaken in a constant temperature water bath till equilibrium was attained (2 h). After centrifugation, the drug concentration remaining in the supernatant was determined by measuring the absorbance at 445 nm. Two replicate runs were made and the results averaged. Preliminary experiments have shown that the filters used did not adsorb the drug to any significant extent.

Since heat sterilization at 160° is indicated to sterilize kaolin if not used extemporanously, the effect of such treatmert on the extent of adsorption was examined. The three types of kaolin were heated at 160°C for 3 h and adsorption experiments were performed on the heated and unheated samples.

Effect of Electrolytes:

The effect of some electrolytes on the extent of adsorption was examined over the concentration range 0-100 mMole

at pH 2.1. Sodium chloride, aluminium chloride, calcium chloride, sodium sulphate, sodium phosphate and citric acid were used. Percentage suppression in the extent of adsorption was calculated as follows:

$$\frac{\text{\% adsorption in the absence of electrolytes} - \text{\% adsorption in the presence of electrolytes}}{\text{\% adsorption in the absence of electrolyte}} \times 100$$

Effect of Methylcellulose and Polysorbate 80:

The effects of 0.5% w/v methylcellulose and 50 mg% polysorbate 80 on the extent of adsorption of riboflavine were investigated. The polymer and surfactant were separately equilibrated with the adsorbent for 1 h before addition of the drug solution.

Desorption Experiments:

The extent of elution of the adsorbed vitamin was determined in two media of pH values 2.0 and 6.5. The residue obtained by centrifugation of the suspension, after the adsorption run, was digested in varying volumes of the desorption medium at 37°. The amount of drug eluted over a period of 4 h was determined in an aliquot of the supernatant after centrifugation.

In-vitro Availability in Presence of Adsorbents:

Dissolution runs of capsules containing riboflavine were carried out in the presence of either kaolin, veegum^R or Quintess^R suspension. The dissolution experiments were performed in 0.01 N HCl using the USP paddle-stirrer apparatus at 100 r.p.m. The dissolution medium (900 ml) containing either 1 g veegum^R, 4 g kaolin or 30 ml Quintess^R suspension

was maintained at 37±0.2°C. At zero time, one capsule (size 0) containing 10 mg of riboflavine was allowed to sink to the bottom of the dissolution vessel before starting rotation. A small loose piece of stainless steel wire was attached to the capsule to prevent its floatation. Samples were withdrawn at various time intervals and immediately replaced by fresh volumes of the dissolution medium to maintain a constant volume. The amount of the drug in solution was determined fluorimetrically[6] according to the USP method (7).

Dialysis Experiments:

To investigate the possible binding of the drug with methylcellulose, the following dialysis experiment was carried out. Cellophane bags[7], prepared from visking tubing 24/32 inch, were filled with 10 ml solutions (pH 2.1) containing either 1.5 or 15 mg% of riboflavine and 0.5% w/v solution of methylcellulose. The bags were immersed in 10 ml solution (pH 7.0) and shaken at 37±0.2°C for 2 h. The volumes of both inside and outside solutions were accurately measured and the drug contents in both solutions were determined. Duplicate experiments were carried out and the results averaged. Preliminary experiments showed that no binding occurred of riboflavine to the membrane.

RESULTS AND DISCUSSION

Riboflavine possesses two ionizable groups of pKa_1=1.9 and pKa_2=10.2 (8). Within the pH values studied in the present work (2.1-7), the drug will carry a net positive charge at pH values below 6. The adsorption of the vitamin by the various adsorbents examined can be attributed to

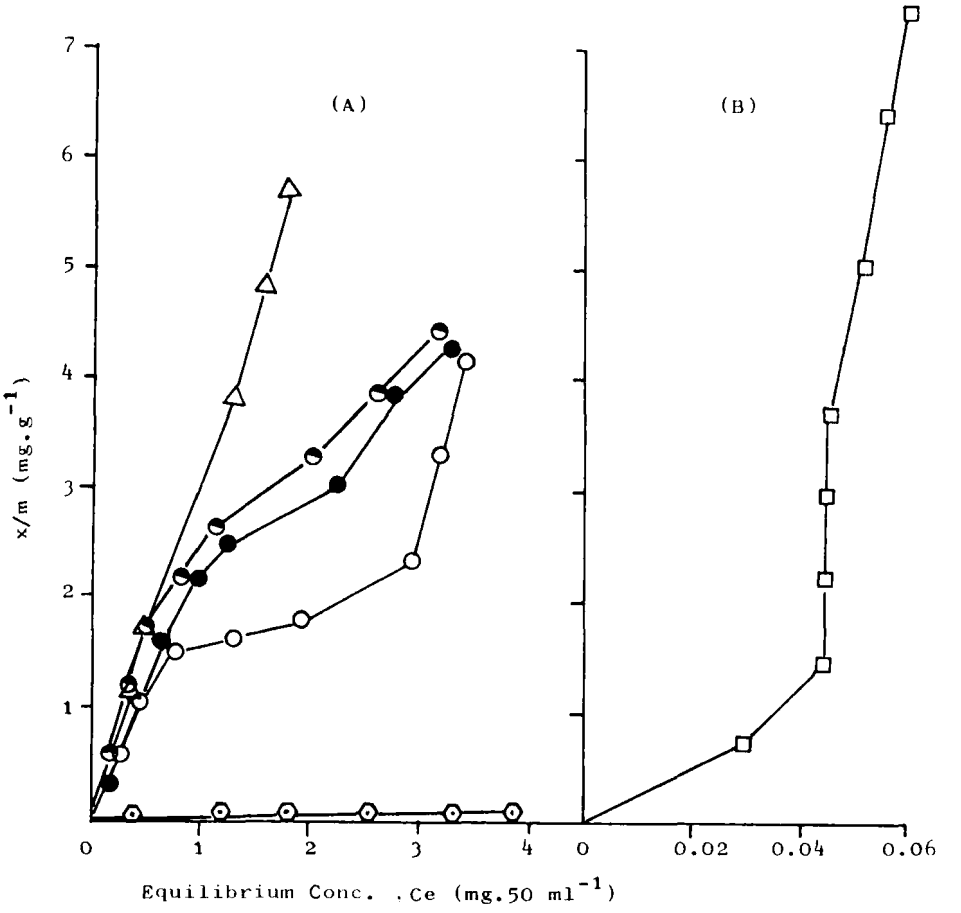

FIGURE 1

Results of adsorption of riboflavine at pH 2.1; (A) for the adsorbents: △ attapulgite; ◒ natural kaolin; ● light kaolin; ○ white kaolin and ⊙ magnesium trisilicate, and (B) for veegum □ .

charge opposition between the cationic drug and the anionic adsorbent surfaces. Also, ion exchange mechanism cannot be ruled out.

Figure 1 shows the adsorption plots of riboflavine on the various adsorbents at pH 2.1. Riboflavine was adsorbed by the different types of kaolin and attapulgite but

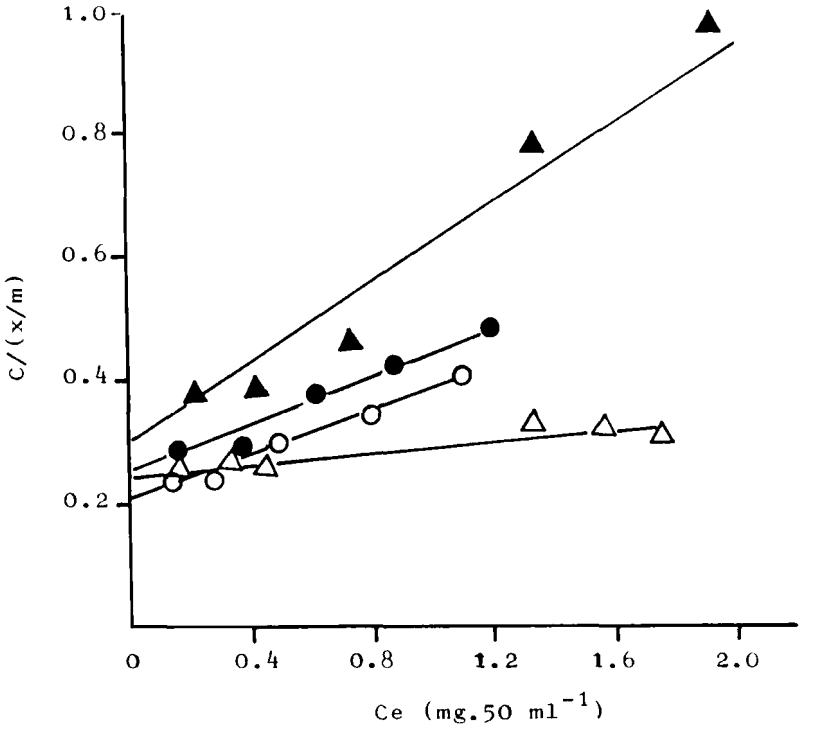

FIGURE 2

Langmuir plots showing the adsorption of riboflavine at pH 2.1; adsorbents used were: ▲ white kaolin; ● light kaolin; ○ natural kaolin and △ attapulgite.

not by the decomposition product of magnesium trisilicate (Fig.1A). At pH 2.1, veegum was relatively more efficient than either attapulgite or kaolin (Fig.1B). The data of adsorption of riboflavine onto attapulgite and kaolin fitted a Langmuir plot as shown in Fig.2. The efficiency of the adsorbents (Fig.1A) followed the sequence: attapulgite > kaolin > magnesium trisilicate. When comparing the efficacy of the three types of kaolin, both light and natural kaolins gave relatively higher adsorptive capacity than white kaolin.

For veegum, about 100% adsorption of riboflavine occurred at pH 2.1 over the concentration range 1.5 to 15 mg%.

The variation in the extent of adsorption of riboflavine by different adsorbents examined cannot be solely ascribed to the variation in the surface areas of adsorbents. Although the sample of veegum used had relatively the largest mean particle diameter (4.1u),it showed higher adsorptive capacity compared to attapulgite, kaolin and magnesium trisilicate.

The effect of pH values on the extent of adsorption of riboflavine is shown in Figures 3 and 4. Within the pH range studied (2.1 - 7), riboflavine was chemically stable when maintained at 37±0.2°C for 2 h. and kept in the dark. On kaolin, significant adsorption took place over the pH range 2.1 to 6.9. An increase in the pH value decreased the extent of adsorption (Fig.3) and produced a gradual shift in the equilibrium concentrations to higher values beyond which multilayer adsorption took place. For example, at pH values 2.1, 4.3 and 6.9 the 'critical' equilibrium concentrations were 2.9, 3.5 and 4.2 respectively. The Langmuir constants calculated at various pH values are shown in Table 1.

Magnesium trisilicate showed significant adsorption over the pH range 3.2-8 but not at pH 2.1(Fig.4). The adsorptive capacity of this antacid appears to be associated with the 'intact' magnesium trisilicate. Under a strongly acidic condition (pH 2.1) where about 90% decomposition of the antacid occurred, no riboflavine was adsorbed, hence implying that the hydrated silica gel formed had no adsor-

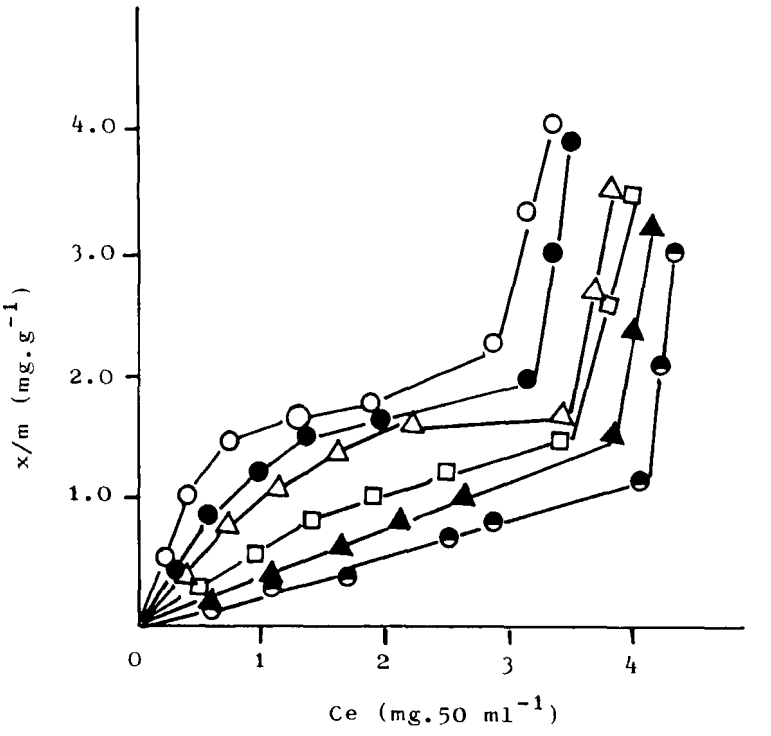

FIGURE 3

Effect of pH values on the adsorption of riboflavine; adsorbent used was white kaolin, pH values were: ○ 2.1; ● 3.2; △ 4.3; □ 5.2; ▲ 6.2 and ◓ 6.9.

ptive capacity to the vitamin. At relatively high pH values, an increase in the level of the adsorbed drug occurred since higher percentages of the antacid would be in the 'intact' form. The reverse situation is noticed in the kaolin system as the increase in the pH value decreased the extent of adsorption (Fig.3). Since charge opposition is thought to be responsible for the adsorption mechanism of riboflavine onto kaolin, it follows therefore, that a reduction in the net positive charge on riboflavine would decrease the extent of adsorption.

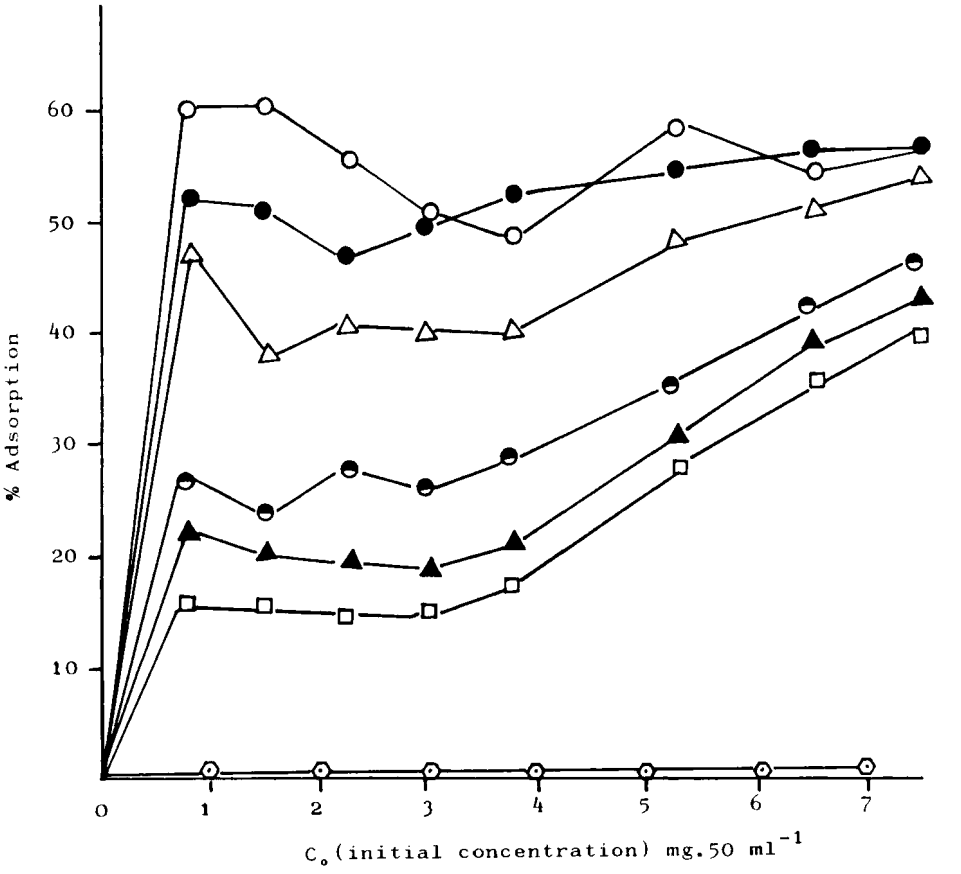

FIGURE 4

Effect of pH values on percentage adsorption of riboflavine; adsorbent used was magnesium trisilicate, pH values used were: ○ 8; ● 6.8; △ 6.2; ◓ 5.2; ▲ 4.3; □ 3.2; ⊙ 2.1.

Figure 5 shows the effect of some electrolytes on the suppression of riboflavine adsorption over the concentration range 1-100 mMole. On kaolin, sodium chloride, sodium sulphate, sodium dihydrogen phosphate, aluminium chloride and citric acid showed significant suppression effects on the extent of adsorption. On the other hand, calcium chloride had no apparent effect (Fig.5); only citrate ions at pH 7.0 reduced the extent of adsorption of riboflavine

TABLE 1

Calculated Values of the Langmuir Constants (a) and (b) of Riboflavine Adsorbed Onto White Kaolin as a Function of pH

pH	a x 10^{2*}	b($mg.g^{-1}$)**
2.1	172.4	2.43
3.2	93.9	2.78
4.2	68.7	2.63
5.2	7.4	8.48
6.1	7.1	6.90
7.0	42.9	1.76

*The adsorption coefficient of Langmuir plot.
**The reciprocal of the slope of the Langmuir plot(the limiting adsorptive capacity).

onto magnesium trisilicate. The observed suppression can be ascribed to the insulation of the ionic groups on both the drug and the adsorbent by the oppositely charged ions of the electrolytes. Significant suppression was only caused by trivalent aluminium and phosphate ions.

Table 2 shows the effect of 0.5% w/v methylcellulose on the extent of adsorption of riboflavine onto white kaolin and veegum at pH 2.1 and on magnesium trisilicate at pH 7.0. Whilst an insignificant effect was observed in the veegum system, a significant reduction in the amount of the vitamin adsorbed was found in the kaolin and magnesium trisilicate systems. Dialysis experiments were designed to test whether the suppression in the extent of adsorption was due to possible interaction between methylcellulose and

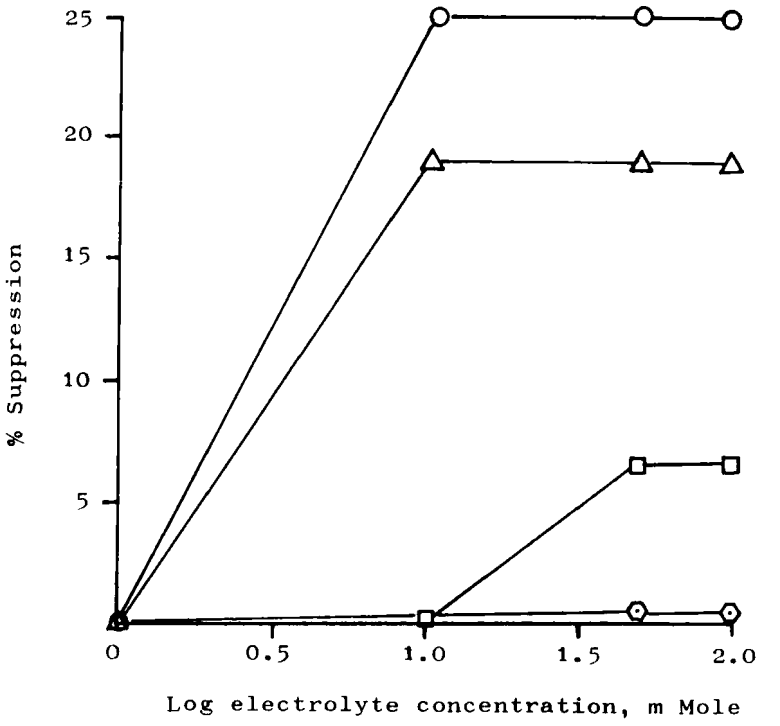

FIGURE 5

Effect of some electrolytes on the percent suppression of riboflavine adsorption at pH 2.1. Initial drug concentration 10.5 mg/100 ml; adsorbent used was white kaolin, electrolytes used were; ○ sodium chloride; △ sodium sulphate; □ citric acid; ⊙ a composite plot for aluminium chloride, sodium dihydrogen phosphate and calcium chloride.

riboflavine. The data obtained showed that no significant binding occurred between riboflavine and methylcellulose within the concentration range used and under the test conditions. Therefore, the observed suppression in adsorption can be attributed to the property of the polymer as a protective colloid. A similar finding was found for the suppressive effect of some polymers on the adsorption of benzoic acid by sulphamethazine(9).

Table 2 also shows the effect of 0.05% polysorbate 80

TABLE 2

Effects of 0.5% Methylcellulose and 0.05% Polysorbate 80 on Percent Suppression in Adsorption of Riboflavine Onto Kaolin, Veegum(at pH 2.1) and Magnesium Trisilicate(at pH 7) at Two Initial Drug Concentrations: (A) 1.5 and (B)15 mg%

	% Suppression in adsorption on					
	Kaolin		Veegum		Magnesium Trisilicate	
	(A)	(B)	(A)	(B)	(A)	(B)
0.5% Methyl-cellulose (η_{rel}=2.3)	78.0	13.9	2.7	3.0	96.3	10.5
0.05% Poly-sorbate 80 (γ=38.0 $N.m^{-1}$)	89.1	31.8	0.0	8.6	95.0	25.3

(surface tension= 38 $N.m^{-1}$) on the extent of adsorption of riboflavine. On veegum, the presence of the surfactant did not significantly affect the extent of adsorption of the drug. In kaolin and magnesium trisilicate systems, polysorbate 80 significantly decreased the extent of adsorption of riboflavine at the two initial concentrations studied. The percentages suppression in adsorption ranged from 25.3 to 95.0. Such suppression is due to the positive adsorption of the surface active agent on the adsorbent thus competing for the adsorption sites.

The data showing the in-vitro availability of riboflavine from hard gelatin capsules in the presence of kaolin (4 g), veegum (1 g) and Quintess^{R}(30 ml) are shown in Figure 6. The presence of the adsorbent in the dissolution medium had a significant suppressive effect on the level of the drug in solution. This suggests that significant uptake of the drug

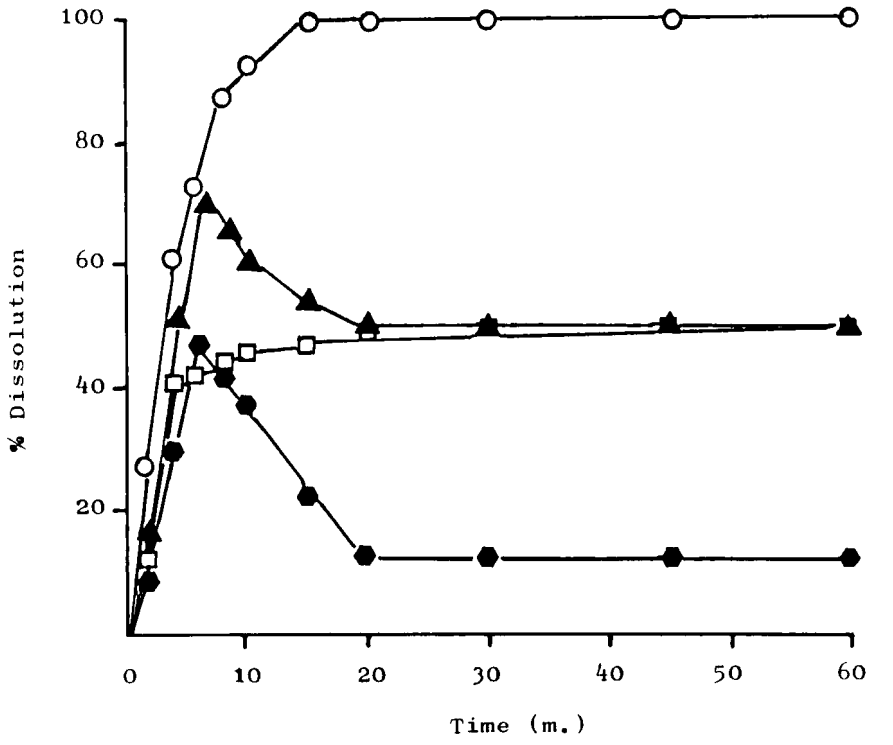

FIGURE 6

Effect of the presence of adsorbents on the in-vitro availability of riboflavine at pH 2.1; adsorbents used were: ○ no adsorbent; ▲ 4 g kaolin; ⬣ 1 g veegum; □ 30 ml Quintess.

tock place under the conditions of dissolution rate test. In this respect, the efficacy of the adsorbents followed the sequence: veegum > Quintess^{R} > kaolin. The strong entrapment of the drug molecules between silica-alumina-silica layers has been proposed to explain the strong and irreversible elution of drugs from veegum-containing systems (10).

Desorption experiments at 37±0.2°C were carried out as a function of medium composition and desorption time. The extent of desorption of the vitamin from kaolin and magnesium trisilicate systems were almost similar. A slight difference

in percent drug eluted was found in the kaolin system upon changing the pH of the medium from 2.0 to 6.5. On veegum, the percent riboflavine eluted was insignificant in the two media; percent drug eluted did not exceed 7.5.

An increase in the volume of desorption medium from 50 to 900 ml produced an insignificant effect on the extent of drug elution. Also, an increase in the desorption time from 0.5 to 4 h resulted in no significant increase in the percent drug eluted. In the systems studied, only partial desorption occurred after 4 h; maximum values of drug released were 47.9% (at pH 2.0) and 61.0% (at pH 6.5). The partial desorption of riboflavine in the presence of increasing volumes of the medium as well as the increase in the level of drug eluted by increasing the pH value suggest a chemisorption mechanism.

The results presented in this work clearly show the dependency of the extent of adsorption of a low dose ionic drug (riboflavine) on a number of factors that mimic the in-vivo conditions. The importance of considering such factors as variations in the pH values, the presence of a hydrocolloid and a surfactant when carrying out in-vitro adsorption studies, is emphasized.

The in-vivo evaluation of riboflavine-attapulgite system has been studied. The results obtained are reported by Khalil et al.(11).

REFERENCES

(1) C.W. Ridout, Pharm.Acta Helv., 43, 42 (1968).

(2) S. El-Masry and S.A.H. Khalil, J.Pharm.Pharmacol., 26, 243 (1974).

(3) M.A. Aggag and Y.H. Hussein, Egypt.J.Pharm.Sci., 18, 177 (1977).

(4) A.G. Stewart, D.J.W. Grant and J.M. Newton, J.Pharm. Pharmacol., 31, 1 (1979).

(5) M. Gupta, Ind.J.Pharm., 31, 125 (1969).

(6) A.B. Varley, J.Am.Med.Assoc., 206, 1745 (1968).

(7) "United States Pharmacopoeia", 20th rev., United States Pharmacopoeial Convection, Inc., Washington, 1980, p.926.

(8) M. Windholz, "The Merck Index", 9th ed., An Encyclopedia of Chemicals and Drugs, Merck & Co., Inc., Rahaway, N.J., U.S.A.

(9) S.A.H. Khalil and R.N. Nasipuri, J.Pharm.Pharmacol., 25, 138 (1973).

(10) K.N. Wai and G.S. Banker, J.Pharm. Sci., 55, 1215 (1966).

(11) S.A.H. Khalil, L.M. Mortada, M.A. Shams-Eldeen and M.M. El-Khawas, submitted to Drug Develop.Ind.Pharm. (1985).