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In Vitro Uptake of Oral Contraceptive Steroids by Magnesium Trisilicate

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Abstract □ Some steroids used in oral contraceptives were adsorbed significantly by magnesium trisilicate. The adsorption affinity followed the sequence: ethindrone > mestranol > norethindrone > ethinyl estradiol. Adsorption data obtained at relatively low initial concentrations fitted a Langmuir plot; the values for monolayer adsorption ranged between 0.24 and 0.32 mg/g. At higher concentrations of the steroids, multilayer adsorption occurred. The results of desorption experiments made at 37° in water and 0.05 *N* HCl suggested that desorption was incomplete and depended on the amount of steroid adsorbed. During the dissolution testing of a brand of contraceptive tablets containing norethindrone acetate, the presence of 0.5% (w/v) magnesium trisilicate in the medium resulted in almost complete reduction in the amount of the steroid remaining in solution after 1 hr.

Keyphrases □ Magnesium trisilicate—*in vitro* adsorption of various progestins, estrogens, and commercial contraceptive tablets □ Adsorption, *in vitro*—various progestins, estrogens, and commercial contraceptive tablets by magnesium trisilicate □ Progestins—ethindrone and norethindrone, *in vitro* adsorption by magnesium trisilicate □ Estrogens—ethinyl estradiol and mestranol, *in vitro* adsorption by magnesium trisilicate □ Contraceptives, oral—commercial product, *in vitro* adsorption by magnesium trisilicate □ Antacids—magnesium trisilicate, *in vitro* adsorption of various progestins, estrogens, and commercial contraceptive tablets

The adsorption of steroids at oil–water interfaces (1, 2) and at lipid surfaces (3) has been reported. In view of the adsorption on magnesium trisilicate of such drugs as prednisolone (4), digoxin (5), and digitoxin (5), the possible uptake of contraceptive steroids on this antacid cannot be ruled out.

Wagner (6) reported that the presence of a solid adsorbent interferes with drug absorption. The bioavailability of some drugs decreased when coadministered with antacids possessing adsorptive properties (7, 8). A recent report (9) confirmed the previously reported *in vitro* findings (5) that concurrent administration of digoxin with some antacids (including magnesium trisilicate) results in a significant reduction in digoxin bioavailability.

Table I—Equilibrium Aqueous Solubilities at 37 ± 0.1° of the Steroids and Monolayer Adsorption Values on 1% (w/v) Magnesium Trisilicate

Steroid	Aqueous Solubility, μmoles/liter	Monolayer Adsorption Value, mg/g
Ethindrone	2.9	0.32
Mestranol	4.8	0.25
Norethindrone	28.2	0.24
Ethinyl estradiol	33.7	—

The objective of the present work was to examine the *in vitro* adsorption on magnesium trisilicate of two progestins, norethindrone and its 10-methyl derivative ethindrone, and two estrogens, ethinyl estradiol and mestranol, of closely related chemical structure.

EXPERIMENTAL

Materials—Magnesium trisilicate powder BP[1] of 11.2-μm mean surface volume diameter was used. Ethindrone[2], ethinyl estradiol[3], mestranol[4], and norethindrone[5] were used as supplied. A batch of contraceptive tablets[6] was used in the dissolution studies. Chloroform and ethanol were analytical reagent grade[7].

Methods—*Adsorption Experiments*—Adsorption experiments were carried out at 37 ± 0.2° using 1% (w/v) magnesium trisilicate, as previously reported (5). After centrifugation, the steroid concentration remaining in the supernate was determined spectrophotometrically[8] at 240 nm (for ethindrone and norethindrone) and 280 nm (for ethinyl estradiol and mestranol) against a blank. A preextraction step with chloroform was necessary to eliminate the interference due to leachable materials from magnesium trisilicate.

Three replicate runs were made, and the results were averaged. Reproducibility was within ±3.0%.

Desorption Rates—The desorption rates of adsorbed ethindrone were determined at 37 ± 0.2° in both water and 0.05 *N* HCl over 3 hr as previously reported (5). The amount of steroid desorbed at a specified time was determined in the supernate after centrifugation as described earlier.

For ethindrone, the effect of the amount adsorbed on the extent of desorption after 3 hr was investigated.

Dissolution Testing—The dissolution rate of a brand of contraceptive tablets[6] was tested using the USP rotating-basket dissolution apparatus[9]. Two media were used: water and 0.5% (w/v) magnesium trisilicate in water. The dissolution medium (800 ml) was maintained at 37 ± 0.1°, and two tablets were used; the speed of rotation of the basket was 100 rpm. Samples were withdrawn after 0.5, 1, 2, and 3 hr. Fresh aliquots of water were added each time to maintain a constant volume.

The percentage of steroid in solution was calculated with reference to the labeled norethindrone acetate content. Determinations of the concentration of norethindrone acetate in solution were made in the aliquot withdrawn after centrifugation and extraction. Measurements were made at 240 nm.

[1] Evans Medical Ltd., England.
[2] Control No. 167017, WHO Centre for Chemical Reference Substances, Solna 3, Sweden.
[3] Batch 50187, Schering AG.
[4] Batch A 5365, Organon Laboratories Ltd., United Kingdom.
[5] Reference No. 1253, Organon Laboratories Ltd., United Kingdom.
[6] Anovlar 21, Batch 05-73-1070, Schering. Each tablet contained 4 mg of norethindrone acetate and 50 μg of ethinyl estradiol.
[7] British Drug Houses Ltd., Poole, England.
[8] Unicam SP 500 series 2 spectrophotometer.
[9] Erweka, Apparatebau GMBH, D-6056 Heusenstamm, West Germany.

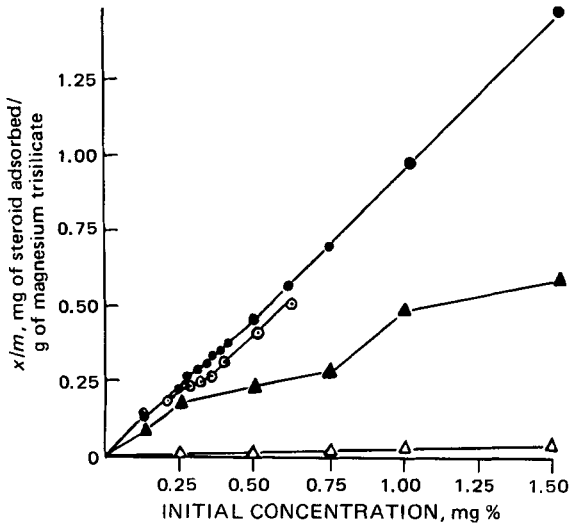

Figure 1—*Adsorption plots of some contraceptive steroids on 1% (w/v) magnesium trisilicate at 37°. Key: ●, ethindrone; ⊙, mestranol; ▲, norethindrone; and △, ethinyl estradiol.*

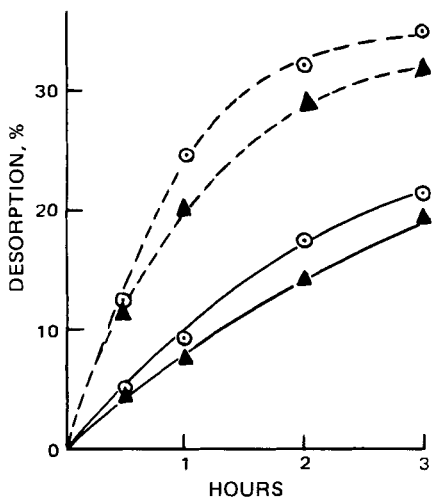

Figure 2—*Desorption rate at 37° in 0.05 N HCl (- - -) and water (—) for norethindrone (⊙) and ethindrone (▲). Amount of adsorbed steroid = 0.25 mg/g.*

Equilibrium Solubilities—The equilibrium solubilities of the steroids in water were determined by equilibrating excess steroid in glass-stoppered flasks at 37 ± 0.1° for 24 hr. Samples were filtered through a membrane filter[10] (0.2-μm mean pore diameter), which had been shown not to adsorb the steroids. The filtered samples were assayed by measurements at either 240 or 280 nm.

RESULTS AND DISCUSSION

Preliminary experiments showed that equilibrium was attained within 3–12 hr, depending on the steroid used and its initial concentrations. Figure 1 shows the adsorption plots for the four steroids. The extent of adsorption followed the sequence: ethindrone > mestranol > norethindrone > ethinyl estradiol.

Ethinyl estradiol was very slightly adsorbed (<6.5%) over the concentrations used. Unlike the norethindrone adsorption plot, the plots of both ethindrone and mestranol showed relatively less extended plateau regions over which monolayer adsorption occurred. At relatively low initial concentrations, the adsorption data fitted a Langmuir plot. Values of the monolayer adsorption, calculated from the reciprocal of slopes of the linear plots, are shown in Table I together with the equilibrium aqueous solubilities determined at 37 ± 0.1°.

The insignificant adsorption of ethinyl estradiol on the antacid was believed to be due to the partial ionization of the phenolic group of ring A at the pH 9.6 of the medium. When the pH of the medium was preadjusted to 4.5–9.0, no significant change in the extent of adsorption was observed. Only a slight increase from 6.5 to 9.0% occurred for an initial concentration of 1.0 mg % of the steroid.

[10] Sartorius, GMBH, 34 Göttingen, West Germany.

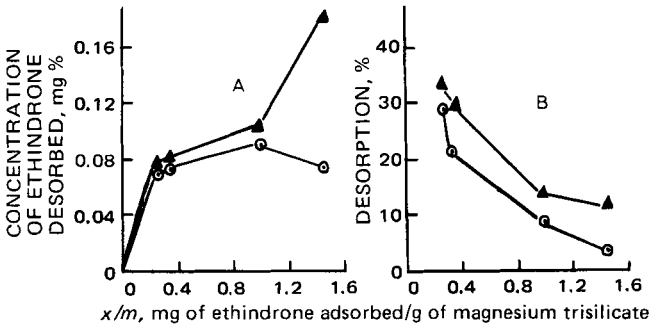

Figure 3—*Results of desorption of ethindrone after 3 hr as a function of amount of adsorbed steroid in 0.05 N HCl (▲) and water (⊙).*

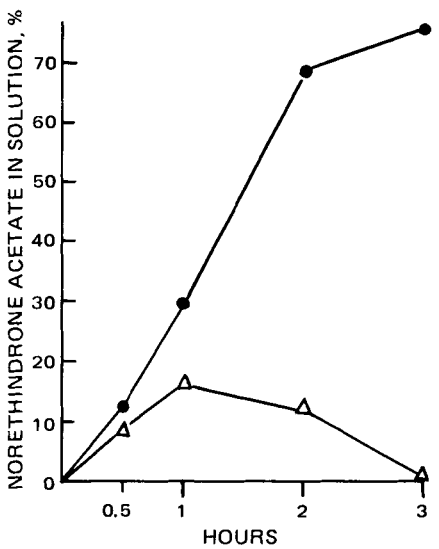

Figure 4—*Results of dissolution testing of a brand of contraceptive tablets at 37° in water (●) and in water containing 0.5% (w/v) magnesium trisilicate (△).*

The minor differences in the chemical structure of the four steroids were probably responsible for the observed variation in the extent of adsorption. While ethinyl estradiol was insignificantly adsorbed over the concentrations used, its 3-methyl ether, mestranol, was adsorbed significantly. Similarly, norethindrone was relatively less adsorbed than its 10-methyl derivative, ethindrone. That a slight change in the chemical structure of a drug affects the extent of its adsorption on magnesium trisilicate was previously shown for digoxin and digitoxin (5) and atropine and hyoscine (10).

The four steroids examined possess varying relative polarity, as reflected by the equilibrium solubilities in water (Table I). With the more polar steroids, the extent of adsorption was decreased. This result is in agreement with the findings of Patrick and Eberman (11) who concluded that, for a given solvent, the more soluble solutes are generally less strongly adsorbed than the less soluble solutes.

The results of desorption experiments using ethindrone and norethindrone are shown in Fig. 2. In the two media, norethindrone was relatively more desorbed than ethindrone. Because of the decomposition of the antacid in 0.05 *N* HCl, desorption rates in this medium were relatively higher than in water. However, in spite of the extensive decomposition of the antacid in 0.05 *N* HCl [about 82%, as was determined from the amount of magnesium ions released (5)], desorption was incomplete and did not exceed 35%. This result is probably due to readsorption on the silica gel formed from decomposition of the antacid in an acid medium.

The effect of the amount of ethindrone adsorbed (*x/m* values) on the extent of desorption after 3 hr was studied (Fig. 3). In water, variations

in the x/m values from 0.25 to 1.44 mg/g produced from 0.07 to 0.09 mg % of desorbed ethindrone (Fig. 3A). In 0.05 N HCl, the amounts of ethindrone desorbed were relatively higher. When the amounts of steroid desorbed were converted to percentages desorption (with reference to the x/m values), a gradual decrease occurred as the x/m values were increased (Fig. 3B).

The results of dissolution testing of the brand of contraceptive tablets in both water and 0.5% (w/v) magnesium trisilicate are shown in Fig. 4. The presence of the antacid in the medium drastically decreased the concentration of norethindrone acetate in solution. After 3 hr, the concentration of steroid in solution was less than 2%, compared with 75.2% in water. The reduction in concentration of steroid was a direct result of the adsorption onto the antacid particles.

It is suggested that the concurrent administration of magnesium trisilicate and oral contraceptive tablets containing the steroids tested might interfere with the steroid absorption. A recent report (9) that confirmed the decreased bioavailability in humans of digoxin in the presence of magnesium trisilicate lends supporting evidence to this suggestion.

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Improved Colorimetric Determination of Aspirin and Salicylic Acid Concentrations in Human Plasma

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Abstract □ An improvement in a previously described method for the determination of plasma salicylic acid and aspirin levels in humans is described. The procedure was simplified by employing only one plasma sample for both salicylates. More accurate estimation of salicylates, particularly aspirin, was achieved by using two different calibration curves. Salicylic acid was estimated by reaction with an aqueous solution of the Folin–Ciocalteu phenol reagent. Absorbance of the blue-colored complex, which formed on addition of sodium hydroxide, was measured at 670 nm. The influence of alkalinity in the formation of the colored complex is discussed. The average recovery of aspirin added to plasma was 94.61%; it was 214.72% by the previous method.

Keyphrases □ Aspirin—colorimetric analysis in human plasma □ Salicylic acid—colorimetric analysis in human plasma □ Colorimetry—analyses, aspirin and salicylic acid in human plasma □ Analgesics—aspirin and salicylic acid, colorimetric analyses in human plasma

Aspirin is hydrolyzed rapidly *in vivo* to salicylic acid (1–3). Determination of blood levels of both salicylates is of considerable pharmaceutical and clinical interest. Several spectrometric procedures are available (4–6) for the estimation of these salicylates after aspirin ingestion.

The colorimetric method of Smith (4), utilizing Folin–Ciocalteu phenol reagent, is used commonly to quantitate salicylates in plasma (Scheme I). The absorbance density of the colored complex formed in alkaline solution is read at 670 nm, and the salicylic acid concentration is obtained from a standard calibration curve. The aspirin concentration is estimated from the difference between free (nonhydrolyzed) and total (hydrolyzed) salicylates.

Table I—Typical Recoveries of Salicylic Acid

Added, μg[a]	Found, μg[b] Nonhydrolyzed Curve[c]	Found, μg[b] Hydrolyzed Curve[d]
5	5.25 ± 0.63	3.08 ± 0.55
10	10.50 ± 0.90	7.00 ± 0.66
20	21.67 ± 2.04	15.92 ± 1.67
25	23.90 ± 0.85	17.33 ± 1.04
40	42.33 ± 0.50	32.25 ± 0.43

[a] Amounts of salicylic acid in distilled water added per milliliter of human plasma sample. [b] n = 3. [c] Free curve, approximate pH 9.6 (Smith method, improved method). [d] Approximate pH 10.4 (improved method; used for total salicylates and aspirin determination).

When this general procedure was applied in this laboratory in bioavailability studies of various aspirin dosage forms in human volunteers, the absorbance intensity of the colored complex was extremely pH dependent. Furthermore, the Smith method (nonhydrolyzed standard curve) resulted in much higher recoveries of total salicylates and aspirin from human plasma because the final pH values of the colored solutions read at 670 nm are different for free (pH 9.6) and total (pH 10.4) samples. The average difference in pH values, about 0.8 unit higher in total (hydrolyzed) samples, is due to an additional amount of 0.5 ml of 1.5 N NaOH in the final test solution. As far as could be determined, the correlation between the alkalinity of a test solution and the absorbance of the colored complex has not been reported. Such a pH influence could be important in the estimation of salicylates, particularly aspirin.

The present report describes a modification of the Smith