Table I—Synthesis of 1,11-Diazasteroids

Number	Compound Name	Yield, %	Melting Point	Mass Spectrum, *m/e*	IR, cm^{-1}	Formula	Analysis, %	Calc.	Found
VI	2-Methoxy-1,11-diaza-8,14-seco-1,3,5,7,9-gonapentaene-12,14-dione[a]	86	143–144.5°[b]	—	1735, 1660	$C_{16}H_{16}N_2O_3$	C	67.59	67.55
							H	5.67	5.69
							N	9.85	9.71
VII	2-Methoxy-1,11-diaza-1,3,5,7,9,13-gonahexaen-12-one	35	275–276°	266	3360, 1660, 1600	$C_{16}H_{14}N_2O_2 \cdot CH_3OH$	C	68.44	68.45
							H	6.08	5.88
							N	9.37	9.88
IX	7-Methoxy-1,11-diaza-8,14-seco-1,3,5,7,9-gonapentaene-12,14-dione[a]	89	116–118°	—	1720, 1670	$C_{16}H_{16}N_2O_3$	C	67.59	67.40
							H	5.67	5.76
							N	9.85	9.95
X	7-Methoxy-1,11-diaza-1,3,5,7,9,13-gonahexaen-12-one[e]	39	247–249.5° dec.[c,d]	266	1620, 1610	$C_{16}H_{14}N_2O_2$	C	72.16	72.09
							H	5.30	5.35
							N	10.52	10.52
XII	1,11,15-Triaza-8,14-seco-D-homo-1,3,5,7,9-gonapentaene-12,14-dione	77	220–221.5°[d]	—	3320, 3160	$C_{15}H_{15}N_3O_2$	C	66.90	67.11
							H	5.61	5.45
							N	15.60	15.38
XVII	1,11-Diaza-1,3,5,7,9,13-gonahexaen-12-one 1-*N*-oxide	14	242.5–244° dec.[f]	252	3325, 1270, 850	$C_{15}H_{12}N_2O_2 \cdot H_2O$	C	66.66	66.89
							H	5.22	5.19
							N	10.36	10.42
XVIII	3,11,15-Triaza-8,14-seco-D-homo-1,3,5,7,9-gonapentaene-12,14-dione	83	186.5–188°[d]	—	1680, 1650, 1630	$C_{15}H_{15}N_3O_2$	C	66.90	66.81
							H	5.61	5.46
							N	15.60	15.56

[a] These condensations were carried out as in the preparation of XIV (see *Experimental*), except that the amine was added dropwise to a 10% excess of the keto ester over 40–60 min. [b] Crystallized from acetone–water. [c] After chromatography on Florisil. [d] Crystallized from acetone–chloroform. [e] NMR: δ 8.65 (1H, d, $J_{2,3}$ = 4 Hz, $J_{2,4}$ = 1.5 Hz, H_2), 7.99 (1H, d, $J_{3,4}$ = 8 Hz, $J_{2,4}$ = 1.5 Hz, H_4), 7.44 (1H, d, $J_{3,4}$ = 8 Hz, $J_{2,3}$ = 4 Hz, H_3), 6.71 (1H, s, H_6), 4.03 (3H, s, CH_3O), 3.00 (4H, m, $H_{15,17}$), and 2.30 (2H, m, H_{16}) ppm. [f] Crystallized from water.

exhibited *N*-oxide bands at 1240 and 850 cm^{-1}; the mass spectrum showed *m/e* 252 (also indicated the presence of water).

Anal.—Calc. for $C_{15}H_{12}N_2O_2 \cdot H_2O$: C, 66.66; H, 5.22; N, 10.36. Found: C, 66.27; H, 5.16; N, 10.26.

RESULTS AND DISCUSSION

8-Amino-2-methoxyquinoline, prepared by a minor modification of the method of Mislow and Koepeli (5), condensed with II to give an 86% yield of the secosteroid VI, which cyclized to 2-methoxy-1,11-diaza-1,3,5,7,9,13-gonahexaen-12-one in 35% yield (VII, Table I). Similarly, 8-amino-6-methoxyquinoline (VIII) and II reacted to produce IX (89%), which was cyclized to 7-methoxy-1,11-diaza-1,3,5,7,9,13-gonahexaen-12-one (X) (39%). 5-Aminoisoquinoline (XIII) and II gave XIV (90%), which was cyclized to the 3,11-diazasteroid XV (69%, see *Experimental*). Both I and XIII condensed with 3-ethoxycarbonyl-2-piperidone (XI) to give the secosteroids XII and XVIII, respectively, neither of which could be cyclized to steroids. *N*-Oxides of IV and XV were prepared by oxidation with *m*-chloroperbenzoic acid.

Diazasteroids IV, VII, X, XVI, and XVII were inactive in the National Cancer Institute screen against P-388 leukemia in mice. However, XV (NSC 265959) exhibited slight activity at 12.5–50 mg/kg; at higher dose levels, it was toxic[2].

REFERENCES

(1) I. Y. C. Tao and R. T. Blickenstaff, *Steroids*, **27**, 205 (1976).

(2) D. G. Bew and G. R. Clemo, *J. Chem. Soc.*, **1955**, 1775.

(3) R. J. Chorvat and R. Pappo, *Tetrahedron Lett.*, **1975**, 623.

(4) F. D. Popp, W. R. Schleigh, P. M. Froehlich, R. J. Dubois, and A. C. Casey, *J. Org. Chem.*, **33**, 833 (1968).

(5) K. Mislow and J. B. Koepeli, *J. Am. Chem. Soc.*, **68**, 1553 (1946).

[2] *Note added in proof:* Dr. Chinan Fan, Department of Biochemistry, Scripps Clinic and Research Foundation, La Jolla, Calif., found that XV exhibits a modest inhibitory activity ($K_i = 1.3 \times 10^{-5}$ *M*) against the L-1210 dihydrofolate reductase; 1,2,3,4,13,14-hexahydro-IV (1) and 1-methyl-1,2,3,4-tetrahydro-XII (1) were inactive against P-388 and against dihydrofolate reductase.

Structures of Silver Sulfonamides

AUKE BULT ˣ and HUUB B. KLASEN

Received June 7, 1976, from the *Laboratory for Pharmaceutical and Analytical Chemistry, State University of Groningen, Antonius Deusinglaan 2, Groningen 8004, The Netherlands.* Accepted for publication May 24, 1977.

Abstract □ The structures of silver sulfonamides were found to depend highly on the substituent at the amide nitrogen of the sulfonamide. Silver is coordinated to that nitrogen and the sulfonamide is in the amido form if no substituent is present or if the substituent is a phenyl, acetyl, or 2-pyrimidyl group. If the substituent is a 2-thiazolyl or 2-pyridinyl group, the sulfonamide is in the imido form and silver coordinates to the nitrogen of the substituent. Depending on the number of suitable donor atoms per sulfonamide, the silver compounds are charged or uncharged and the primary amino group may be involved in complexation.

Keyphrases □ Silver—coordinating properties with various sulfonamides, structures of complexes studied □ Sulfonamides, various—coordination with silver, structures of complexes studied □ Complexes—various silver sulfonamides, coordination properties and structures studied □ Anti-infectives, topical—various silver sulfonamides, coordination properties and structures studied

Interest in silver sulfadiazine as an antibacterial agent in the treatment of extensive burns has increased steadily. IR (1) and NMR (1) studies as well as X-ray analysis (2, 3) of the structure of silver sulfadiazine have been reported. The bactericidal action *in vivo* of silver sulfadiazine is superior to the related silver sulfonamides. Because the

Table I—Analytical Data of the Silver Sulfonamides

Compound	% Ag		% C		% H		% N		% S	
	Calc.	Found	Calc.	Found	Calc.	Found	Calc.	Found	Calc.	Found
I Silver benzenesulfonamide	40.86	40.3	27.27	27.2	2.29	2.3	5.31	5.2	12.14	12.0
II Silver benzenesulfonamidobenzene	31.73	31.6	42.35	42.4	2.96	2.9	4.12	4.1	9.43	9.5
III Silver 2-(benzenesulfonamido)pyrimidine	31.54	31.1	35.08	34.9	2.36	2.2	12.29	12.5	9.89	9.3
IV Silver 2-(benzenesulfonamido)thiazole	31.07	30.8	31.12	31.3	2.03	2.0	8.07	7.9	18.47	18.3
V Silver sulfanilamide	38.65	38.5	25.80	25.8	2.53	2.5	10.04	10.2	11.49	11.6
VI Silver sulfanilamidobenzene	30.13	29.1	40.56	40.9	3.12	3.2	7.89	8.1	9.03	9.0
VII Silver sulfadiazine	30.22	29.4	33.62	33.4	2.54	2.4	15.70	15.7	8.98	9.0
VIII Silver sulfathiazole	29.70	28.3	29.83	30.1	2.23	2.3	11.61	11.6	17.71	17.7
IX Silver sulfapyridine	30.29	31.5	37.07	36.8	2.83	2.7	11.80	11.8	9.00	8.9
X Silver sulfacetamide	33.60	34.0	29.90	29.8	2.82	2.7	8.73	8.9	9.99	9.9

bactericidal action is related quantitatively to the binding of silver to microbial DNA, the dissociation of the silver sulfonamide is a prerequisite. The unique property of silver sulfadiazine seems to be its moderate initial dissociation coupled with its continual release of silver (4).

This investigation systematically studied the coordinating properties of silver with sulfonamides and obtained insight into the structures of the silver compounds. A difference in structure is one factor that influences the silver release and contributes to the different biological activities of the silver sulfonamides.

EXPERIMENTAL

Equipment and Analyses—A double-beam grating spectrophotometer[1], an IR spectrophotometer[2], and a conductivity meter[3] were used. The silver was analyzed by the Volhard titration after decomposition of the compound with 65% HNO_3. Elemental analyses also were performed[4].

Materials and Reagents—All chemicals were analytical or reagent grade. The sulfanilamides and benzenesulfonamides were obtained commercially.

Synthesis—*Benzenesulfonamides*—2 - (Benzenesulfonamido)py - rimidine and 2-(benzenesulfonamido)thiazole were prepared according to literature methods (5).

Benzenesulfonamidobenzene—Benzenesulfonyl chloride, 20 g, was added dropwise, with stirring, to a cooled solution of 10 g of aniline in 80 ml of dry pyridine. After standing overnight at room temperature, the solvent was partly removed under reduced pressure to a small volume. Addition of water separated the crude product, which was isolated by filtration, washed with water, and recrystallized from ethanol–water, yielding 20 g, mp 102–103.1°.

Anal.—Calc. for $C_{12}H_{11}N_1O_2S_1$: C, 61.76; H, 4.76; N, 6.01; S, 13.75. Found: C, 61.5; H, 4.7; N, 6.0; S, 13.6.

Sulfanilamidobenzene—A solution of 15 g of aniline in 50 ml of dry pyridine was added dropwise, with stirring, to a cooled solution of 20 g of *N*-acetylsulfanilyl chloride in 50 ml of dry pyridine. The mixture was then heated for 1 hr at 80°. After the solvent was removed under reduced pressure to half volume, the mixture was poured into 4 *N* HCl; the separated product was filtered off and dissolved in 200 ml of 10% (w/v) NaOH. Then the mixture was refluxed for 1 hr and acidified. The separated product was filtered off, washed with water, and recrystallized from ethanol–water, yielding 13.2 g, mp 188.1–188.9°.

Anal.—Calc. for $C_{12}H_{12}N_2O_2S_1$: C, 58.02; H, 4.87; N, 11.29; S, 12.92. Found: C, 57.9; H, 4.8; N, 11.3; S, 12.9.

Silver Sulfonamides—The sulfonamide, 0.03 mole, was dissolved in 30 ml of 1.0 *N* NaOH and 70 ml of water. After dissolution, in some cases with gentle heating, the solution was diluted with water to 300 ml. A solution of 0.03 mole of silver nitrate in 100 ml of water was then added dropwise with stirring. The white precipitate was separated, washed with water, and dried at 120°.

The elemental analyses are given in Table I. For purification, a quantity of silver sulfadiazine (VII) was dissolved in 25% NH_3 solution and again isolated by: (*a*) the partial evaporation of the solvent in air, from which a crystalline product (A) was formed (1); and (*b*) acidifying to pH 7 with 4 *N* HNO_3, after which the precipitate (B) was filtered off, washed with water, and dried at 120°.

The elemental analyses of VII and Products A and B were nearly identical, but the IR spectra were slightly different. The spectrum of B was the most regular and well defined and closely resembled the spectrum of a commercial sample of VII[5].

RESULTS AND DISCUSSION

Silver prefers the formation of two and four coordinate complexes with linear and tetrahedral geometry, respectively (6). With the anionic ligand, L, the formation of the anionic complexes $[AgL_2]^-$ and $[AgL_4]^{3-}$ is possible (6–8). The effective formation of these complexes depends on the number of suitable donor atoms per L. One donor atom per L favors the formation of the anionic complexes; more donor atoms per L favor the formation of uncharged, polymeric complexes $(AgL)_n$.

Table II summarizes the results of the physical measurements. The IR spectra of the silver compounds II, III, and IV show that the introduction of silver in the sulfonamide is attended with the deprotonation of the amide nitrogen (^{1}N); ν(NH) is absent. The remaining stretching frequency ^{1}NH of I decreases by 70 cm^{-1}.

Comparison of $\nu^*(^4NH_2)$ [$\nu^* = (\nu_s + \nu_{as})/2$] of the silver compounds with that of the parent compounds (sulfonamides) gives information about the involvement of this group in complexation. The values of $\Delta^*\nu = \nu^*_{\text{sulfonamide}} - \nu^*_{\text{silver compound}}$ are: V = +100 cm^{-1}, VI = +155 cm^{-1}, VII = +25 cm^{-1}, VIII = −90 cm^{-1}, IX = −5 cm^{-1}, and X = +70 cm^{-1}. These results make reliable conclusions difficult because of the possible interference by hydrogen bonding. Considering the magnitude of the shift of the stretching frequency ^{1}NH in I, the involvement of $^4NH_2$ in silver complexation is possible in V, VI, and X. In VII, $^4NH_2$ is not involved in coordination (2, 3).

Of great value are the results of an IR study of sulfonamides (10). The value of ν^*(SO), the weighted average value of ν_s(SO) and ν_{as}(SO), depends on the character of the groups attached to SO_2 (Table II). If R_3 is H, the value of ν^*(SO) is about 1230–1255 cm^{-1} (the amido form) except when R_2 is 2-thiazole or 2-pyridine. In that case, ν^*(SO) is about 1200 cm^{-1} and these sulfonamides are in the imido form (5, 11). The proton R_3 = H is located on the nitrogen of R_2. These results are also supported by X-ray analysis of sulfathiazole (12).

The introduction of $^4NH_2$ in the sulfonamide (R_1 in Table II) lowers ν^*(SO) by about 15–16 cm^{-1}. When R_3 is Na, the compound is the anion of the sulfonamide. The value of ν^*(SO) is lowered to 1160–1200 cm^{-1}. The divergency of the values can be ascribed to the different possibilities of delocalization of the negative charge dependent on the nature of R_2. The lowering of ν^*(SO) correlates with the quantity of negative charge on the amide nitrogen and is the largest when R_2 is H, *i.e.*, 1156 cm^{-1} (10).

Thus, when R_3 is Ag, the value of ν^*(SO) can give information about the electronic environment of the amide nitrogen. By using the values of ν^*(SO), the silver compounds can be divided into two groups. Group A consists of I–III, V–VII, and X. The substitution of the R_3 H by Ag lowers ν^*(SO) by 50–75 cm^{-1}; ν^*(SO) of R_3 = Ag and R_3 = Na have about the same value. Group B consists of IV, VIII, and IX. The substitution of the R_3 H by Ag hardly changes ν^*(SO); the value of ν^*(SO) for R_3 = Ag is over 30 cm^{-1} higher as for R_3 = Na.

For Group B compounds, the small change of ν^*(SO) by silver substitution indicates that the electronic environment of the amide nitrogen

[1] Perkin-Elmer 124.
[2] Beckman Accu Lab 2.
[3] Radiometer type CDM^{2d} and conductivity cell type CDC104.
[4] Analytical Department of the Chemical Laboratories, University of Groningen, Groningen, The Netherlands.
[5] Philips-Duphar B. V., Weesp, The Netherlands.

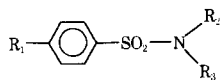

Table II—Physical Data of the Silver Sulfonamides

Compound				Λ_M, Ω^{-1} cm^2/mole[a]			ν^*(SO)[b], cm^{-1}
Number	R_1	R_2	R_3	Acetonitrile	*N,N*-Dimethylformamide	Dimethyl Sulfoxide	
—	H	H	H	—	—	—	1243
I	H	H	Ag	—	—	13.6	1168
—	H	H	Na	—	—	—	1155
—	H	Benzene	H	—	—	—	1248
II	H	Benzene	Ag	87.4	36.5	15.9	1190
—	H	2-Pyrimidine	H	—	—	—	1256
III	H	2-Pyrimidine	Ag	—	—	3.7	1204
—	H	2-Thiazole	H	—	—	—	1213
IV	H	2-Thiazole	Ag	—	5.2	8.15	1212
—	NH_2	H	H	—	—	—	1228
V	NH_2	H	Ag	—	—	7.6	1158
—	NH_2	H	Na	—	—	—	1156
—	NH_2	Benzene	H	—	—	—	1232
VI	NH_2	Benzene	Ag	—	12.4	14.9	1171
—	NH_2	Benzene	Na	—	—	—	1171
—	NH_2	2-Pyrimidine	H	—	—	—	1241
VII	NH_2	2-Pyrimidine	Ag	—	—	2.2	1178
—	NH_2	2-Pyrimidine	Na	—	—	—	1186
—	NH_2	2-Thiazole	H	—	—	—	1197
VIII	NH_2	2-Thiazole	Ag	—	5.2	7.25	1199
—	NH_2	2-Thiazole	Na	—	—	—	1166
—	NH_2	2-Pyridine	H	—	—	—	1190
IX	NH_2	2-Pyridine	Ag	—	6.6	9.75	1192
—	NH_2	2-Pyridine	Na	—	—	—	1159
—	NH_2	Acetyl	H	—	—	—	1238
X	NH_2	Acetyl	Ag	75.6	33.2	18.75	1174
—	NH_2	Acetyl	Na	—	—	—	1206

[a] Measured at 10^{-3} *M*. The Λ_M values are calculated for $[AgL_2]Ag$ (see text). Standard ranges of 1:1 electrolyte (9): acetonitrile, 120–160; *N,N*-dimethylformamide, 65–90; and dimethyl sulfoxide, 23–42. [b] Potassium bromide pellets; $\nu^*(SO) = \sqrt{[\nu_s(SO)^2 + \nu_{as}(SO)^2]/2}$.

remains almost unchanged. These sulfonamides are in the imido form and maintain that form in the silver compounds. The silver is bound to the substituent R_1 = 2-thiazole or 2-pyridine. The donor atom is the nitrogen in R_2. The same method of coordination is found with cobalt in sulfathiazole (13).

The resemblance in lowering of ν^*(SO) in Group A as a result of substitution of the R_3 H by Ag or Na indicates a similarity in the type of bonding for sodium and silver. The conclusion that silver is ionically bound like sodium cannot be correct because almost all compounds are nonconducting and the UV spectra are different. In addition, it has been shown (3) that VII has coordinative bonding. The results can be interpreted as follows. In the compounds of Group A, a coordinative bond between the amide nitrogen and silver is formed. The formation of that bond has been proved for VII (2, 3); it is very likely for I and II (the amide nitrogen is the only donor atom) and likely for III, V, VI, and X. A fraction of the lowering of ν^*(SO) may be caused by an SO···Ag interaction, but this interaction will be weak (2, 3, 6).

Conductivity—The values of Λ_M given in Table II are based on the assumption of the formation of the complex $[AgL_2]Ag$ (L is the anion of the sulfonamide). The values indicate that only II and X may have that composition. The Λ_M values of these compounds in the three solvents agree with the values found for the complex $[AgL_2]Ag$ (L is the anion of a barbiturate) (7). The Λ_M values of IV, VI, VIII, and IX in *N,N*-dimethylformamide are indicative of uncharged compounds.

All silver compounds are soluble in dimethyl sulfoxide. However, the values of Λ_M are rather variable. This variation can be attributed to the strong dissociative and solvolytic effects of dimethyl sulfoxide. This explanation is supported by the fact that Λ_M of IV, VI, VIII, and IX is larger in dimethyl sulfoxide than *N,N*-dimethylformamide, and this result is unlike the theoretical expectation (the standard ranges of *N,N*-dimethylformamide have higher values than of dimethyl sulfoxide; see footnote *a* of Table II). The values of Λ_M in dimethyl sulfoxide of III, V, and VII indicate the presence of uncharged compounds. From Λ_M, it is difficult to draw a conclusion about the electrolyte type of I, but the insolubility in acetonitrile and *N,N*-dimethylformamide indicates an uncharged compound.

UV—The UV spectra of the sulfonamides and their silver compounds in methanol are similar (III and VIII are insoluble in methanol). In general, the spectra of the sodium sulfonamides are different from the former spectra and confirm the difference in the interaction of silver and sodium with the sulfonamides.

From the results discussed so far, the following conclusions about the structures of the individual compounds can be derived: I, silver coordinates with the amide nitrogen (^{1}N); II, silver coordinates with ^{1}N, and the compound has the composition $[AgL_2]Ag$; III, silver coordinates with ^{1}N and is uncharged, and the involvement in coordination of the two nitrogen atoms of 2-pyrimidine is likely (resemblance with VII); IV, silver coordinates to the nitrogen atom of 2-thiazole and is uncharged; V, silver coordinates with ^{1}N and the primary amino group and is uncharged; VI, silver coordinates with ^{1}N and the primary amino group and is uncharged; VII, silver coordinates with ^{1}N and is uncharged, and the two nitrogen atoms of 2-pyrimidine are involved in coordination (2, 3); VIII, silver coordinates to the nitrogen atom of 2-thiazole and is uncharged; IX, silver coordinates to the nitrogen atom of 2-pyridine and is uncharged; and X, silver coordinates with ^{1}N and possibly the primary amino group, and the composition of the compound is $[AgL_2]Ag$.

The composition $[AgL_2]Ag$ of II and X indicates the presence of one suitable donor atom per L. For II, this structure agrees with expectation; for X, this structure is uncertain because of the possible involvement of the primary amino group. In the uncharged compounds (III and V–VII), at least two suitable donor atoms per L are needed. In IV, VIII, and IX, with the nitrogen of 2-thiazole or 2-pyridine as a donor atom, the second donor atom of L is uncertain. Possibly ^{1}N is the second donor atom, but the imido form must be maintained (sulfur of 2-thiazole and oxygen of SO are less probable).

The variation in structure of the silver compounds and the resulting differences in stability constants and silver release can at least partly explain the different bacterial action.

REFERENCES

(1) B. J. Sandmann, R. U. Nesbitt, Jr., and R. A. Sandmann, *J. Pharm. Sci.*, **63**, 948 (1974).

(2) A. W. Struss, *Diss. Abstr. Int. B*, **35**, 5808 (1975).

(3) D. S. Cook and M. F. Turner, *J. Chem. Soc. Perkin II*, **1975**, 1021.

(4) C. L. Fox and S. M. Modak, *Antimicrob. Agents Chemother.*, **5**, 582 (1974).

(5) T. Uno, K. Machida, K. Hanai, M. Ueda, and S. Sasaki, *Chem. Pharm. Bull.*, **11**, 704 (1963).

(6) F. A. Cotton and G. W. Wilkinson, "Advanced Inorganic Chemistry," 3rd ed., Interscience, New York, N.Y., 1972, p. 1044.

(7) A. Bult and H. B. Klasen, *Pharm. Weekbl.*, **110**, 533 (1975).
(8) J. R. Demember and F. A. Wallace, *J. Am. Chem. Soc.*, **97**, 6240 (1975).
(9) W. J. Geary, *Coord. Chem. Rev.*, **7**, 81 (1971).
(10) A. Rastelli, P. G. de Benedetti, A. Albasini, G. Vampa, and M. Melegari, *Farmaco, Ed. Sci.*, **29**, 654 (1974).
(11) P. G. de Benedetti, A. Rastelli, A. Albasini, M. Melegari, and G. Vampa, *Atti Soc. Nat. Mat. Modena*, **105**, 73 (1974).
(12) G. J. Kruger and G. Gafner, *Acta Crystallogr., B*, **27**, 326 (1971).
(13) A. Bult, *Pharm. Weekbl.*, **111**, 385 (1976).

ACKNOWLEDGMENTS

The authors are indebted to Professor Dr. D. A. Doornbos and Dr. A. S. Horn for their help.

In Vitro Uptake of Oral Contraceptive Steroids by Magnesium Trisilicate

SALEH A. H. KHALIL *x and M. IWUAGWU

Received March 12, 1977, from the *School of Pharmacy, University of Benin, Benin City, Nigeria.* Accepted for publication May 27, 1977. *Present address: Department of Pharmaceutics, Faculty of Pharmacy, Alexandria University, Alexandria, Egypt.

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.