after oral ingestion of a capsule and a sustained-release product. Their results indicated that the extent of absorption from the sustained-release product was almost twice that from a regular capsule. In addition, ~98% of the sustained-release dose was absorbed compared to an intravenous dose. The latter value is unusually high and we are not aware of any study where an oral dose of ascorbic acid is virtually completely absorbed. Another discrepancy is that the ascorbic acid half-life was determined (27) to be ~11 hr prior to saturation and 29 hr after saturation. These values are considerably greater than those reported for the half-life of the vitamin after exogenous administration (11, 17, 26, 30). The reason for the substantial differences in half-life and availability from the timed-release products reported here and by Zetler *et al.* (27) is not known.

The results of this study indicate that ascorbic acid absorption is incomplete after oral ingestion and that there is considerable intersubject variation in the extent of absorption. In addition, absorption of the vitamin is considerably less efficient from the timed-release capsule examined here compared with the other oral forms. These findings have several implications. From a practical point of view, efficient oral therapy with the vitamin can be achieved by dissolving powdered ascorbic acid in water. In addition, for the specific manufacturers' products examined in this study, tablets and chewable tablets appear comparable to a solution of the vitamin. This conclusion may not apply to tablets made by all manufacturers but that can only be determined from the evaluation of other products in a manner used in the present study. The timed-release capsule examined here appears to be a more expensive and less reliable means of providing oral vitamin therapy compared with other more conventional dosage forms. This conclusion may apply to similar dosage forms which attempt to delay or sustain the release of the vitamin; therefore, bioavailability studies for such forms are essential.

A question that remains to be answered is to what extent does variation in ascorbic acid absorption influence the results of large-scale trials designed to examine the clinical effects of the vitamin? To the authors' knowledge no consideration has been given to absorption as a parameter potentially influencing the findings of such studies. Investigators pursuing such trials should give some consideration to the possible influence of variation in ascorbic acid absorption on clinical outcome.

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Mass Spectral Fragmentation of 24,24-Diphenyl-23-ene Derivatives of Cholic Acid

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Abstract □ After electron impact in the mass spectrometer, 24,24-diphenyl-23-ene derivatives of cholic acid ejected the 17-sidechain as an ionized 1,1-diphenyl-butadiene derivative, and the 12α-acetoxy group activated this loss. This contrasts markedly with the mass spectrometric fragmentation of typical sterols having unsaturated 17-sidechains that are also devoid of functionality on the C-ring.

Keyphrases □ Mass spectra—fragmentation of 24, 24-diphenyl-23-ene derivatives of cholic acid □ Cholic acid—24, 24-diphenyl-23-ene derivatives, fragmentation by mass spectra □ Derivatives—24, 24-diphenyl-23-ene derivatives of cholic acid, mass spectral fragmentation

During the course of other investigations (1), a number of 24,24-diphenyl-23-ene derivatives of cholic acid having structural similarities to known biologically active compounds were prepared. Since the Barbier–Wieland 17-sidechain degradation is used extensively in steroid-terpenoid synthesis (2, 3) and electron impact induced loss of the 17-sidechain is of both fundamental and diagnostic importance (4–6), the observations concerning mass spectral cleavage processes of the Barbier–Wieland modified 17-sidechain of steroids and terpenoids are summarized.

RESULTS AND DISCUSSION

The preparation of the compounds in this study was unexceptional. However, isolation of pure diene (IV*a*–IV*d*) was difficult because unreacted monoene was invariably present. Also, it is likely that *E* and *Z*

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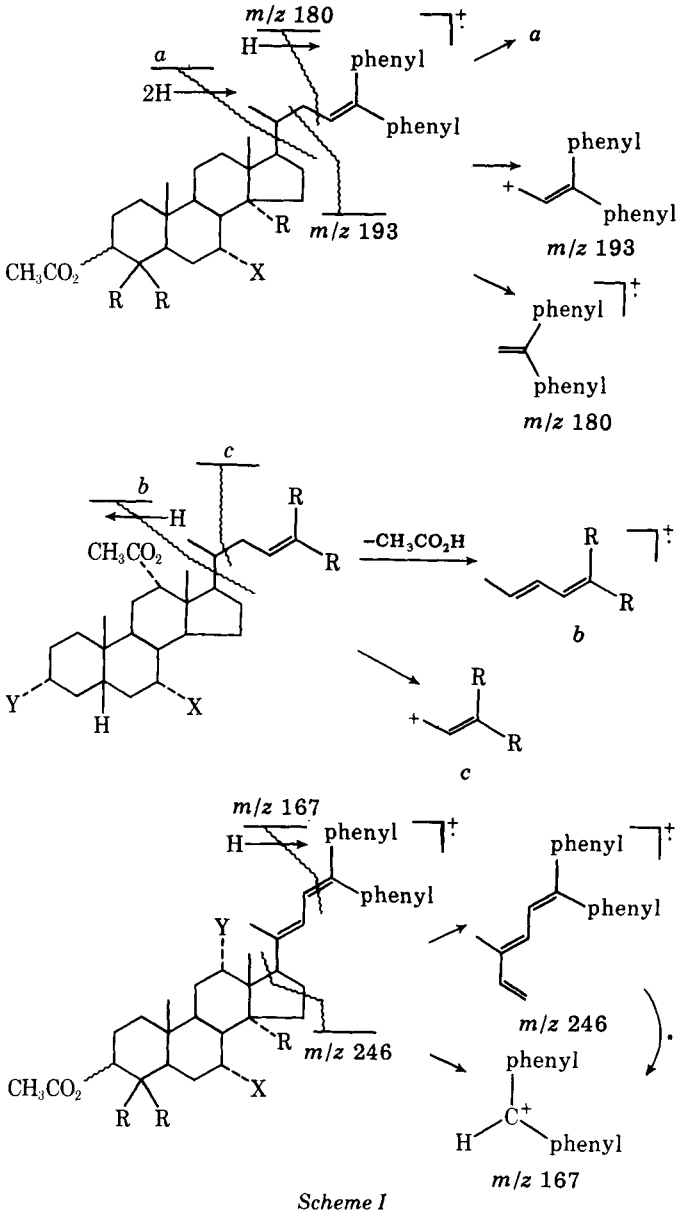
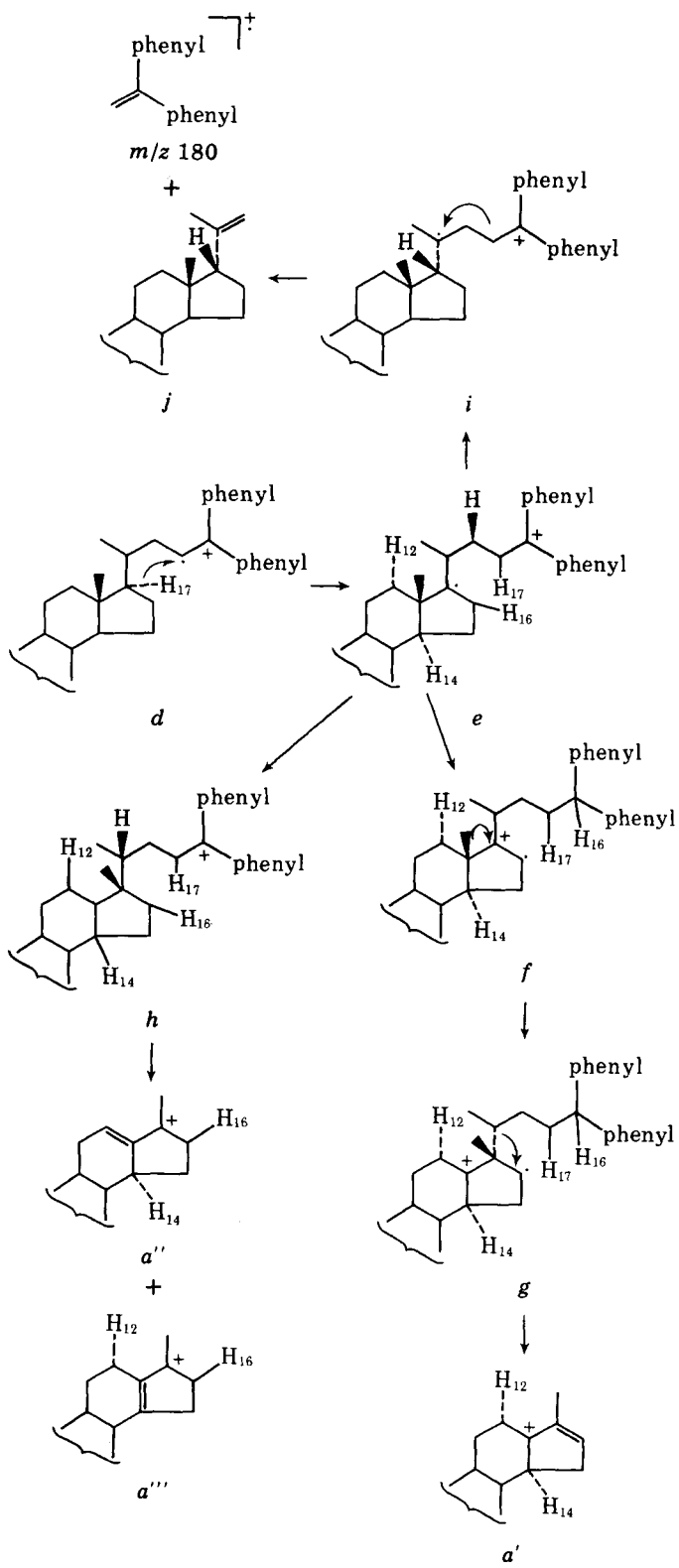


Scheme I



Scheme II

17-sidechain diene isomers may be coproducts, but this could not be verified experimentally. The propensity for acid catalyzed elimination of the 7α-acetoxy group as acetic acid for both the monoenes and dienes has been previously noted (1, 3). Table I summarizes the mass spectral data for compounds I–VIII.

Scheme I summarizes the characteristic fragmentations of 24,24-diphenyl-23-ene derivatives of cholic acid. If no functionality exists on the C-ring of the monoene, then ion *a*, the allyl carbocation (*m/z* 193), and ionized α,α-stilbene are the predominant ions observed in the mass spectra. The genesis of ion *a* was previously elucidated (4). Typical sterols having no B-ring or C-ring functionality and either a saturated or unsaturated 17-sidechain exhibit 17-sidechain and D-ring cleavage processes in which there is a drift of hydrogen atoms from the steroid skeleton to the departing neutral fragment (7). Placing functionality on the B- or C-rings (*e.g.*, an oxo group at positions 6 or 12) reverses the direction of hydrogen atom drift so that hydrogen moves from the 17-sidechain toward the steroid skeleton. The former process results in increased unsaturation in the steroid framework and corresponding charge residence in that unit upon scission, whereas the latter process produces increased unsaturation in the 17-sidechain unit and frequently results in greater charge residence in that fragment. Thus, ion *a* in Scheme I emanates from hydrogen migration to the 17-sidechain from the steroid skeleton followed by scission at the 17–20 bond; ion *b* emanates from hydrogen migration from the 17-sidechain to the steroid skeleton followed by 17–20 bond scission, where the reversal of hydrogen migration in the latter is induced by the presence of the 12α-acetoxy group. Otherwise, the process not regulated by hydrogen migration, simple allylic scission (*m/z* 193 and ion *c*), appears to be relatively unaltered in 24,24-diphenyl-23-enes either possessing or not possessing C-ring functionalization by the 12α-acetoxy group.

The specific mechanism for the genesis of ion *a* is presented in Scheme II. Charge localization on the olefinic 17-sidechain triggers site specific C-17 hydrogen transfer through a 5-membered ring to give the highly stable ion-radical *e* (4). The second itinerant hydrogen originated from positions C-12, C-14, and C-16 (4). Direct transfer of C-16 hydrogen to C-24 gives ion *f* which can undergo a 1,2-methyl shift to give ion *g*. Simple

Table I—Partial Monoisotopic Mass Spectra (70 ev) of 24,24-Diphenyl-23-ene Derivatives of Cholic Acid

Compound	$[M]^+$	$[M-2CH_3CO_2H]^+$	$[M-3CH_3CO_2H]^+$	$[M-CH_3CO_2H]^+$	$[M-CH_2CH{=}CR_2-CH_3CO_2H]^+$	$[M-CH_3CHCH_2CH_2CHR_2-CH_3CO_2H]^+$
Ia	596 (1)	536 (10)	476 (5)	—	—	—
Ib	596 (1)	536 (8)	476 (3)	—	343 (3)	313 (14)
Ic	610 (15)	550 (11)	490 (10)	—	357 (5)	—
Id	654 (3)	594 (7)	534 (4)	474 (3)	341 (7)[a]	—
IIa	724,722 (2, 3)	664,662 (4, 6)	604,602 (3, 5)	544,542 (2, 3)	341 (4)[a]	—
IIb	714 (6)	654 (4)	594 (2)	534 (2)	—	—
III	640 (5)	580 (18)	520 (39)	460 (14)	—	—
IVa	594 (8)	534 (44)	474 (5)	—	—	—
IVb	594 (6)	534 (3)	474 (2)	—	—	—
IVc	550 (44)	490 (6)	—	—	—	—
IVd	610 (13)	592 (10)[b]	532 (32)[b]	472 (6)[b]	—	—
V	536 (3)	476 (10)	—	—	—	313 (56)[c]
VI	594 (14)	534 (19)	474 (6)	—	341 (28)	311 (6)
VII	580 (3)	520 (2)	—	—	327 (29)	297 (6)
VIII	578 (9)	518 (3)	—	—	—	—

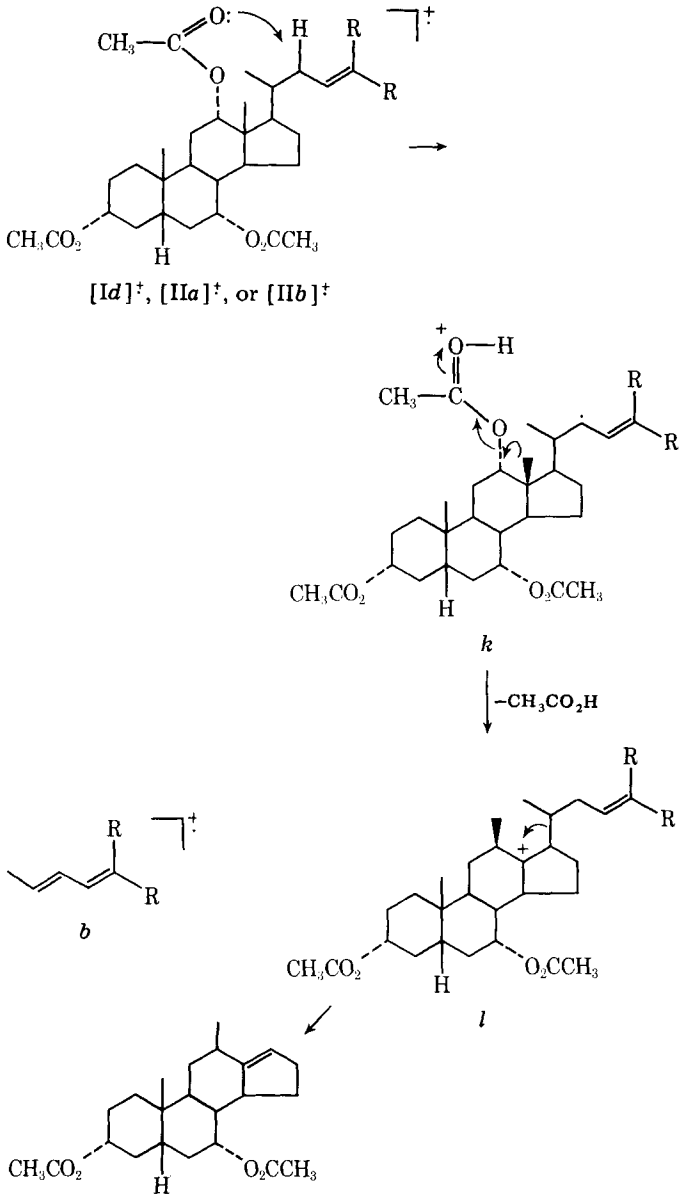
Compound	$[M-CH_2CH{=}CR_2-2CH_3CO_2H]^+$	$[M-CH_3CHCH_2CH_2CHR_2-2CH_3CO_2H]^+$	$[CH_2{=}C(CH_3){-}CH{=}CHCH{=}CR_2]^+$	$[CH_3CH{=}CH{-}CH{=}CR_2]^+$	$[CH{=}CH{-}CH{=}CR_2]^+$	$[CH_2CH{=}CR_2]^+$
Ia	283 (4)	253 (6)	246 (2)	220 (100)	205 (9)	193 (12)
Ib	283 (62)	253 (100)	246 (6)	—	—	193 (91)
Ic	297 (54)	—	246 (5)	220 (58)	205 (24)	193 (100)
Id	281 (12)[a]	—	—	220 (100)	205 (14)	193 (22)
IIa	281 (11)[a]	—	—	290,288 (64, 100)	275,273 (2, 3)	263,261 (12, 18)
IIb	—	—	—	280 (41)	265 (7)	253 (100)
III	—	—	—	206 (12)[d]	—	207 (100)[d]
IVa	—	—	246 (11)	220 (26)	205 (11)	193 (11)
IVb	—	—	246 (55)	220 (62)	205 (22)	193 (15)
IVc	—	—	246 (15)	220 (20)	205 (19)	193 (30)
IVd	—	—	246 (23)	220 (34)	205 (25)	193 (16)
V	283 (42)[c]	253 (100)[c]	246 (11)	220 (12)	205 (5)	193 (96)
VI	281 (100)	251 (18)	—	220 (24)	205 (13)	193 (68)
VII	—	—	—	—	—	193 (22)
VIII	—	—	246 (100)	220 (46)	205 (11)	—

Compound	$[CH_2{=}CR_2]^+$	$[R_2CH]^+$	Miscellaneous	Metastable Transitions
Ia	180 (4)	167 (3)	—	191 (220 → 205)
Ib	180 (72)	167 (21)	373 (1, $[M-CH_3CHCH_2CH_2CHPh_2]^+$), 314 (16)	423 (536 → 476), 233.5 (596 → 373)
Ic	180 (84)	167 (22)	—	574.5 (610 → 592), 515 (550 → 532), 496 (612 → 550) 455 (490 → 472), 436.5 (550 → 490), 332 (610 → 443)
Id	180 (7)	167 (12)	401 (1)	191 (220 → 205)
IIa	—	237,235 (6, 10)	253, 255 (30, 10, [228, 290 → 253, 255]$^+$)	488-90 (602,4 → 542,4), 259-61 (288, 90 → 273, 5) 222-4 (288, 90 → 253, 5)
IIb	240 (18)	227 (37)	145 (35)	82 (253 → 145)
III	180 (20)	167 (12)	129 (43)	466 (580 → 520), 407 (520 → 460)
IVa	—	167 (100)	—	
IVb	—	167 (100)	255 (17, [m-2HOAc—$CH_3C{=}CHCH{=}CR_2]^+$), 231 (13, [246—CH_3]$^+$)	480 (594 → 534), 217 (246 → 231), 191 (220 → 205), 113.5 (246 → 167)
IVc		167 (100)	299 (20), 259 (12), 231 (12)	515 (550 → 532), 437 (550 → 490)
IVd	180 (24)	167 (100)	550 (2), 490 (3), 365 (13), 305 (7)	575 (610 → 592), 478 (592 → 532)
V	180 (22)	167 (23)	521 (3), 461 (8), 115 (65)	446.5 (476 → 461), 506.5 (536 → 521), 68.5 (193 → 115)
VI	—	167 (13)	115 (40)	504 (534 → 519), 480 (594 → 534), 444 (474 → 459), 421 (534 → 474) 232 (341 → 281), 68.5 (193 → 115)
VII	180 (100)	—	357(17,[M-$CH_3CHCH_2CH_2C-HR_2]^+$), 343 [10]	247 (357 → 297)
VIII	—	167 (57)	231 (13, [246-CH_3]$^+$)	217 (246 → 231), 191 (220 → 205), 113.5 (246 → 167)

[a] Add one more CH_3CO_2H loss to column heading. [b] Replace one CH_3CO_2H by H_2O in column heading. [c] Delete one CH_3CO_2H in column heading. [d] Subtract or add a CH_2 group to the column heading.

scission of ion *g* leads to the allylic carbocation *a'*. Alternatively, a 1,2-methyl shift in ion *e* gives ion *h* where the tertiary radical at C-12 activates α-hydrogens at both the C-12 and C-14 positions, and their transfer to C-24 followed by simple scission at 17–20 leads to allylic ions *a''* and *a'''*, respectively. An ion analogous to the *m/z* 180 ion was not observed, and no doubt its presence in the spectra of Ib, V, and VII results from the stabilizing effects of the phenyl groups. A 1,2-hydrogen shift from C-20 to C-17 converts ion *e* into ion *i*, which can undergo simple scission to give olefin *j* and the *m/z* 180 ion.

The introduction of a 12α-acetoxy group into the C-ring of 24,24-diphenyl-23-enes results in a dramatic alteration of their mass spectral breakdown and is emphasized by comparing the mass spectra of Ia *versus*

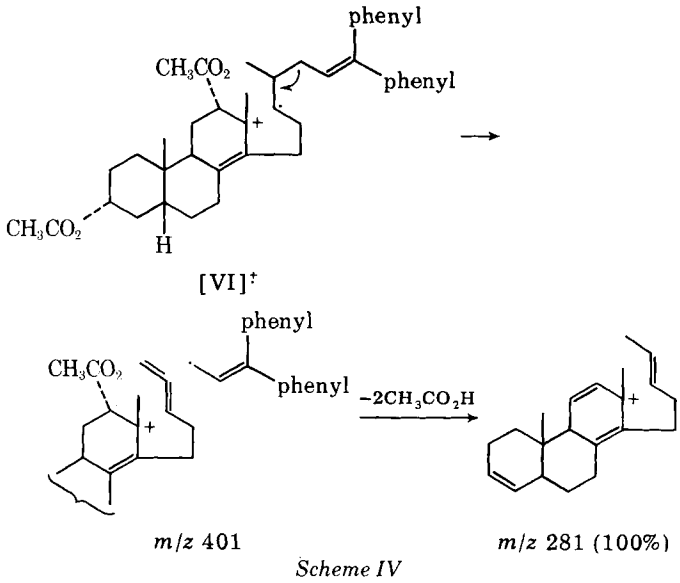


Scheme III

Ib. By selectively labeling with acetate-d_3, prior work demonstrated that in the consecutive loss of three acetic acid molecules from the molecular ion of methyl 3α,7α,12α-triacetoxy-5β-cholan-23-oate, that the first loss of acetate comes from the 12α-acetoxy group, the second from the 7α-acetoxy, and the third from the 3α-acetoxy group (8). A plausible mechanism for the formation of 1,1-biphenyl-1,3-butadiene ions (*b*) from the 17-sidechain in the mass spectra of Ia, Id, IIa, IIb, III, and VI could involve prior elimination of acetic acid from the 12α-acetoxy group to produce an 11–12 double bond which then triggers this process. However, Dreiding models suggests that it is sterically possible to transfer the activated C-22 hydrogen directly to the 12α-acetoxy carbonyl (Scheme III) through a nine-membered ring (an eight-membered ring for III) to generate the resonance stabilized ion *k*. Precedence for long-range activated hydrogen transfer to the 12α-acetoxy group in other cholic acid derivatives after electron impact has been provided (9). Extrusion of acetate from ion *k* simultaneous to 1,2-methyl shift would produce the tertiary carbocation *l* which can decompose *via* simple scission to give ion *b*.

Loss of the allyl radical ($\cdot CH_2CH{=}CR_2$) from the 17-sidechain was observed in all the monoenes and indicates that some charge localization occurs at the 12–17 bond. This is probably responsible for the 281 base peak in the mass specrum of VI, since the 8–14 double bond should activate ionization of this bond

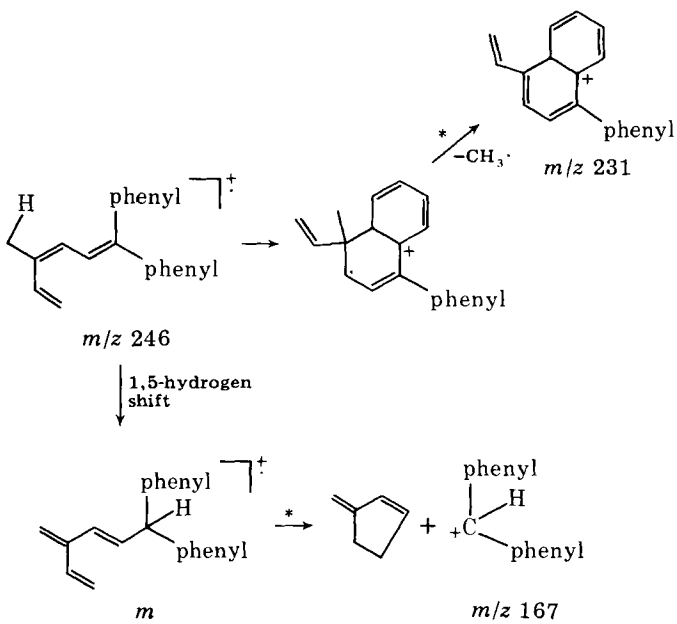
$$[(VI)^{+} \rightarrow m/z\ 401 \xrightarrow{-CH_3CO_2H} m/z\ 341 \xrightarrow{-CH_3CO_2H} m/z\ 281].$$



Scheme IV

Prior mass spectral work established that the dominant consecutive loss of one, two, and three acetic acid molecules from the 12α, 7α, and 3α-acetoxy groups of methyl cholate triacetate after electron impact led to increasingly more stable daughter ions since the relative intensities of the $[M\text{-}CH_3CO_2H]^{+}$, $[M\text{-}2CH_3CO_2H]^{+}$, and $[M\text{-}3CH_3CO_2H]^{+}$ ion peaks dramatically increased in that order (8). This observation permitted tracing of the probable movement of positive charge from the region of the D-ring to that of the A-ring. The fact that the mass spectral ions $[M\text{-}CH_3CO_2H]^{+}$, $[M\text{-}2CH_3CO_2H]^{+}$, and $[M\text{-}3CH_3CO_2H]^{+}$ are of low intensity and decrease in that order in relative intensity for Id, IIa, and IIb (Table I) provides evidence for extensive localization of charge on or near the 24,24-diphenyl-23-ene moiety. Since the critical energy for bond cleavage in a mass spectral ion is lower in the vicinity of positive charge and because there is a quasi-equilibrium fluctuation of excitation energy over the total charged molecule, it was observed that fragmentation in these compounds (I–VIII) is dominated by hydrogen migration and bond scission in the region of the 17-sidechain, which is the site of charge localization.

Simple D-ring scission of dienes IVa to IVd and VIII led to an *m/z* 246 hexatriene ion of obvious stability. Metastable peaks (Table I) corresponding to the degradation of the *m/z* 246 ion to *m/z* 167 (m* 113.5) were observed in the spectra of IVb and VIII. A plausible mechanism (Scheme V) for the formation of the dominant *m/z* 167 ion involves an orbital symmetry allowed 1,5-hydrogen shift to form ion *m* which decomposes



Scheme V

to m/z 167. The m/z 167 related ions in the spectra of the monoenes have a nominal relative abundance and no doubt have an alternate genesis.

Unidirectional migration of two hydrogens from the unsubstituted C- and D-rings of the steroid skeleton to the unsaturated 17-sidechain where charge is initially localized precedes the loss of the 17-sidechain as a neutral radical. In the presence of a 12α-acetate group, this process is superseded by the migration of a hydrogen from the ion-radical containing 17-sidechain to the C-ring acetate substituted steroid system producing an ionized butadiene derivative. Simple scission fragmentation processes occurring near the site of charge localization and not involving hydrogen migration in steroid systems should be less affected (than processes involving hydrogen migration) by the presence of local ring substituents that do not significantly alter the charge distribution in the ion.

EXPERIMENTAL

IR data, reported in inverse centimeters (cm^{-1}), were obtained as chloroform solutions; ^{1}H-NMR data[1], reported in ppm (δ) from tetramethylsilane, were obtained in chloroform-*d*. Mass spectra[2] were obtained at an ionization voltage of 12 and 70 ev (Table I), source temperature of 180°, and a trap current of 10 μamp.

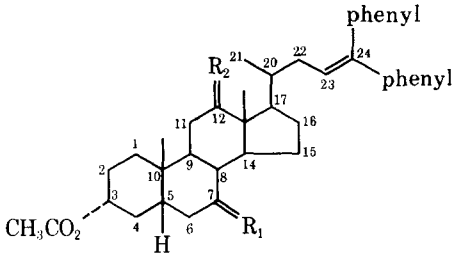
Column chromatography was performed using silica gel[3], and TLC was performed on silica gel HF_{254}[3]. The latter was usually developed with 4:1 hexane–ethyl acetate. TLC was visualized by viewing under a UV lamp and charring by brief heating after spraying the TLC plate with 2% ceric sulfate in 2 *N* sulfuric acid. All reactions were monitored by TLC. In general, the dienes had slightly smaller R_f values than the corresponding monoenes and the deoxycholate analogs had larger R_f values than the corresponding chenodeoxycholate analogs. The order of decreasing R_f value for the monoenes coincided with the order of increasing electron density in the phenyl moiety; *p*-chlorophenyl II *a* (largest R_f), diphenyl I*d*, and *p*-methoxyphenyl II*b* (smallest R_f, gives characteristic red color upon brief charring with ceric sulfate).

The synthesis of compounds I*a*, I*b*, I*d*, IV*b*, V, VI, VII, and VIII have already been described (1, 3, 10).

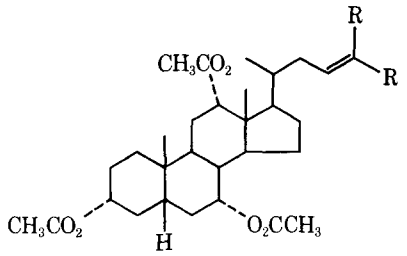
3α,7α-Diacetoxy-12-oxo-24,24-diphenyl-5β-chol-23-ene(I*c*)—Saponification of 3α,7α,12α-triacetoxy-24,24-diphenyl-5β-chol-23-ene (5 g) was achieved by heating at reflux with potassium hydroxide (10 g) and methanol (165 ml) for 2 hr. This was diluted with water and extracted with ether. The ether extract was washed with 5% HCl and concentrated on a rotary evaporator. The dry residue of triol (4 g) was selectively acetylated to the 3α,7α-diacetate by dissolving it in benzene (20 ml) and pyridine (5 ml) and reacting at room temperature with acetic anhydride (5 ml) for 12 hr. The isolated 3α,7α-diacetoxy-12α-ol (4.2 g) was dissolved in acetone (150 ml), cooled on an ice bath, and oxidized dropwise with Jones reagent. Chromatography of the isolated 3α,7α-diacetoxy-12-one I*c* yielded 83% of pure product: mp 186–188° (melts at 115° and solidifies again and then melts at 186–188°); ^{1}H-NMR: δ 7.23 (s, 10H, phenyl H), 6.15 (t, 1H, C-23), 4.97 (peak, 1H, 7β-H), 4.57 (hump, 1H, 3β-H), 2.00 (s, 6H, 3α,7α-acetates), and 1.02 (s, 6H, C-18, and C-19).

3α,7α,12α-Triacetoxy-24,24-di(*p*-chlorophenyl)-5β-chol-23-ene(II*a*)—A Grignard reagent of 4-bromochlorobenzene (45 g) and magnesium (5.8 g) was prepared in dry ether (100 ml). Methyl cholate (10 g) dissolved in dry tetrahydrofuran (100 ml) was added dropwise to the Grignard reagent. After heating at reflux for 48 hr, the contents of the reaction were poured into ice (400 g) containing 36% HCl (70 ml), the organic layer was separated, and the aqueous phase was extracted with ether. The combined ether extracts were dried over anhydrous sodium sulfate and concentrated on a rotary evaporator to yield 15.7 g of crude tetraol. This tetraol was heated at reflux in a mixture of acetic anhydride (50 ml) and acetic acid (100 ml) for 6 hr, and then the acetic acid/anhydride solvent was slowly distilled off until the volume was reduced to 30 ml. The residue was steam distilled to remove the biphenyl side-product, and the isolated product was chromatographed through silica by gradient elution with hexane–ethyl acetate to yield 11 g of II*a*:mp 180–182°; $\bar{\nu}_{max}$ 2960 (strong, C-H stret), 1730 and 1250 (strong, acetate), and 1590 (weak, olefin stret); ^{1}H-NMR δ 6.94 (4 peak multiplet, 8 H, aromatic H), 4.53 (hump, 1 H, 3β-H), 2.10 (s, 3H, 12α-acetate), 2.05 (s, 3H, 7α-acetate), 2.03 (s, 3H, 3α-acetate), 0.92 (s, 3H, C-10), and 0.73 (s, 3H, C-18).

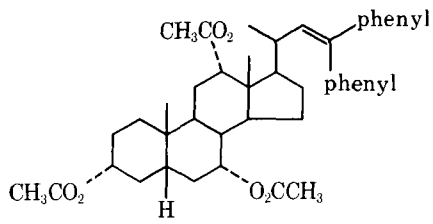
[1] Varian, models A-60 and T-60.
[2] Nuclide 12-90-G single focusing instrument.
[3] MCB Grade 62 and E. Merck.



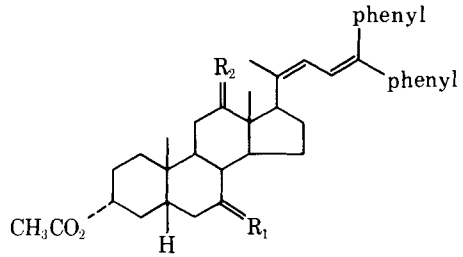
I*a*: $R_1 = H_2$, R_2 = α-OAc, β-H
I*b*: R_1 = α-OAc, β-H, R_2H_2
I*c*: R_1 = α-OAc, β-H, R_2O
I*d*: R_1 = α-OAc, β-H, R_2α-OAc, β-H



II*a*: R = *p*-ClC_6H_4
II*b*: R = *p*-$CH_3OC_6H_4$



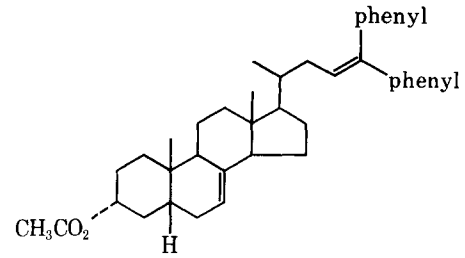
III



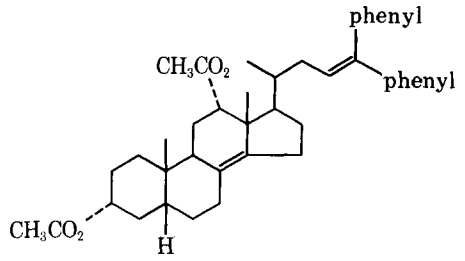
IV*a*: $R_1 = H_2$, R_2 = α-acetate, β-H
IV*b*: R_1 = α-acetate, β-H, H_2
IV*c*: $R_1 = H_2$, O
IV*d*: R_1 = α-acetate, β-H, α-OH, β-H

3α,7α,12α-Triacetoxy-24,24-di(*p*-methoxyphenyl)-5β-chol-23-ene (II*b*)—A solution of 4-methoxyphenylmagnesium bromide was prepared from *p*-bromophenol (100 g), magnesium (913 g), and dry tetrahydrofuran (300 ml). Methyl cholate (23 g) in dry benzene (150 ml) was added dropwise to the red-colored Grignard. After heating the reaction mixture at reflux for 24 hr, the reaction was halted, worked up, and purged of 4,4′-bismethoxybiphenyl by steam distillation. The 60 g of carbinol product thus obtained was heated at reflux in acetic anhydride/acetic acid (1:2, 400 ml) for 6 hr. Then the acetic anhydride/acetic acid solution was slowly distilled to a volume of 100 ml. The isolated product was column chromatographed through silica gel to yield 36 g of a glassy solid which refused to crystallize but was pure by TLC: ^{1}H-NMR δ 7.0 (m, 8H, phenyl H, 5.90 (t, 1H, C-23), 5.05 (peak, 1H, 12β-H), 4.98 (peak, 1H, 7β-H), 4.54 (hump, 1H, 3β-H), 3.81 (s, 3H, OCH_3), 3.76 (s, 3H, OCH_3), 2.11 (s, 3H, 12α- acetate), 2.07 (s, 3H, 7α-acetate), 2.02 (s, 3H, 7α-acetate), 0.92 (s, 3H, C-19), and 0.73 (s, 3H, C-18).

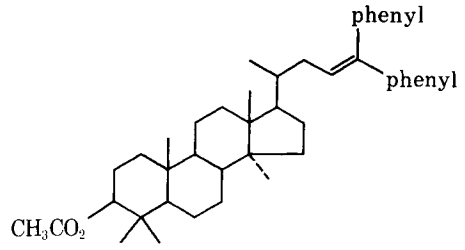
3α,7α,12α-Triacetoxy-23,23-diphenyl-24-nor-5β-chol-22-ene (III)—Methyl norcholate (2.0 g) dissolved in dry benzene (10 ml) was added dropwise to phenylmagnesium bromide (10 equivalents) in ether (25 ml), and tetrahydrofuran (10 ml). After heating at reflux for 24 hr, the reaction mixture was poured into ice (100 g) containing 36% HCl (5



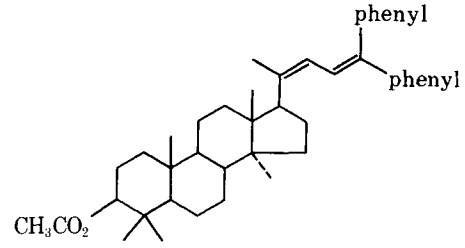
V



VI



VII



VIII

ml). This aqueous mixture was steam distilled to remove the biphenyl side product. The isolated carbinol (3.1 g) was heated at reflux with acetic anhydride (26 ml) and acetic acid (52 ml) for 3 hr, and then the volume of the acetic anhydride/acetic acid was reduced to 20 ml by slow distillation. Column chromatography of the crude triacetate through silica gel with hexane–ethyl acetate yielded 1.8 g of desired product: mp 109–111°; ^{1}H-NMR δ 7.20 (s, 10H, phenyl H), 5.80 (d, J = 10 Hz, 1H, C-22), 4.98 (peak, 1H, 12β-H), 4.88 (peak, 1H, 7β-H), 4.53 (hump, 1H, 3β-H), 2.15 (s, 3H, 12α-acetate), 2.07 (s, 3H, 7α-acetate), 2.02 (s, 3H, 3α-acetate), 0.95 (d, J = 6 Hz, 3H, C-20), 0.88 (s, 3H, C-19), and 0.52 (s, 3H, C-18).

3α,7α-Diacetoxy-12α-hydroxy-24,24-diphenyl-5β-chola-20(22),23-diene (IV*d*)—3α,7α,12α-Triacetoxy-24,24-diphenyl-5β-chola-20(22),23-diene was saponified and selectively acetylated to yield the title compound (IV*d*) after chromatography: mp 164–166°; ^{1}H-NMR δ 7.28 (s, 10H, phenyl H), 6.97 (d, J = 11 Hz, 1H, C-23), 6.03 (d, J = 11 Hz, 1H, C-22), 4.9 (peak, 1H, 7β-H), 4.53 (hump, 1H, 3β-H), 3.60 (peak, 1H, 12β-H), 2.03 (s, 3H, 7α-acetate), 2.02 (s, 3H, 3α-acetate), 1.95 (s, 3H, C-21), 0.92 (s, 3H, C-19), and 0.56 (s, 3H, C-18).

3α-Acetoxy-12-oxo-24,24-diphenyl-5β-chola-20(22),23-diene (IV*c*)—Monoene I*a* was synthesized by a published method [^{1}H-NMR δ 7.18 (s, 10H, phenyl H), 6.06 (t, 1H, C-23), 5.06 (peak 1H, 12β-H), 4.67 (hump, 1H, 3β-H), 2.07 (s, 3H, 12α-acetate), 2.01 (s, 3H, 3α-acetate), 0.91 (s, 3H, C-19), and 0.72 (s, 3H, C-18)]. The volume of a mixture of monoene I*a* (10.5 g) and carbon tetrachloride (400 ml) was reduced to 300 ml by distillation, *N*-bromosuccinimide (4.1 g) was added, and the mixture was photolyzed with a sun lamp (100 watts)while heating at reflux for 1 hr. The resulting mixture was filtered and heated at reflux for an additional 3 hr to complete the hydrogen bromide elimination. Column chromatography of the crude product through silica gel by gradient elution with hexane–ethyl acetate yielded 10 g of material that contained some unreacted monoene (<10% by NMR) but mainly comprised of diene IV*a*: ^{1}H-NMR δ 7.22 (s, 10H, phenyl H), 6.88 (d, J = 11 Hz, 1H, C-23), 5.92 (d, J = 11 Hz, 1H, C-22), 4.82 (peak, 1H, 7β-H), 4.57 (hump, 1H, 3β-H), 2.14 (s, 3H, 7α-acetate), 2.02 (s, 3H, 3α-acetate), 1.92 (s, 3H, C-23), 0.90 (s, 3H, C-19), and 0.60 (s, 3H, C-18). Saponification of IV*a* was achieved by heating at reflux with potassium hydroxide (10 g) and methanol (200 ml) for 3 hr. The reaction mixture was poured into ice (600 g) and the solid was collected by filtration. The dry solid was selectively acetylated by reacting with acetic anhydride (25 ml) and pyridine (25 ml) in benzene (125 ml) for 24 hr. Column chromatography produced 8 g of 3α-acetoxy-12α-hydroxy-24,24-diphenyl-5β-chola-20(22),23-diene: ^{1}H-NMR δ 7.30 (s, 10H, phenyl H), 6.98 (d, J = 11 Hz, 1H, C-23), 6.05 (d, J = 11 Hz, 1H, C-22), 4.75 (hump, 1H, 3β-H), 3.78 (peak, 1H, 12β-H), 2.02 (s, 3H, 3α-acetate), 1.97 (s, 3H, C-21), 0.93 (s, 3H, C-19), and 0.57 (s, 3H, C-18). Jones oxidation of this product was carried out in cold (5°) acetone (800 ml) to yield 4.0 g of IV*c*: mp 190–192°; $\bar{\nu}_{max}$ 1730 and 1240 (acetate), 1705 (12-oxo), and 1630 cm^{-1} (weak C=C stret); ^{1}H-NMR δ 7.23 (s, 10H, phenyl H), 6.90 (d, J = 11 Hz, 1H, C-23), 6.04 (d, J = 11 Hz, 1H, C-22), 4.67 (hump, 1H, 3β-H), 1.99 (s, 6H, 3α-acetate and C-20), 1.01 (s, 3H, C-18), and 0.88 (s, 3H, C-19).

Also, 0.8 g of 3,12-dioxo-24,24-diphenyl-5β-chola-20(22),23-diene [^{1}H-NMR δ 7.28 (s, 10H, phenyl H), 6.94 (d, J = 11 Hz, 1H, C-23), 6.07 (d, J = 11 Hz, 1H, C-22), 2.02 (s, 3H, C-21), 1.10 (s, 3H, C-18), and 0.92 (s, 3H, C-19)], and 1.2 g of 3α-acetoxy-12-oxo-5β-pregnan-20-one [^{1}H-NMR δ 4.68 (hump, 1H, 3β-H), 3.32 (t, 1H, 17α-H), 2.25 (s, 3H, C-21), 2.00 (s, 3H, 3α-acetate), 1.03 (s, 3H, C-18), and 0.95 (s, 3H, C-19)] were obtained.

3α,12α-Diacetoxy-24,24-diphenyl-5β-chol-23-ene (1*a*)—The synthesis of I*a* has been described (10); mass spectrum (12 ev), *m/z* (%): 596 (1, $[M]^+$), 536 (11, $[M-CH_3CO_2H]^+$), 476 (5, $[M-2CH_3CO_2H]^+$), 461 (1, $[M-2CH_3CO_2H-CH_3]^+$), 283 (6, $[M-2CH_3CO_2H-C_{15}H_{13}]^+$), 256 (9, $[M-CH_3CO_2H-C_{14}H_{12}]^+$), 246[3], 220 (100, $[C_{15}H_{13}]^+$), 205 (10, $[220-CH_3]^+$)*, 193 (10, $[C_{15}H_{13}]^+$), 180 (5, $[C_{14}H_{12}]^+$), and 167 (3, $[C_{13}H_{11}]^+$).

3α,7α-Diacetoxy-24,24-diphenyl-5β-chol-23-ene (1*b*)(1)—Mass spectrum (12 ev), *m/z* (%): 569 (1, $[M]^+$), 536 (7, $[M-CH_3CO_2H]^+$), 476 (3, $[M-2CH_3CO_2H]^+$), 343 (5, $[M-CH_3CO_2H-C_{15}H_{13}]^+$), 314[28], 313 (23, $[M-CH_3CO_2H-C_{17}H_{19}]^+$), 253 (100, $[M-2CH_3CO_2H-C_{17}H_{19}]^+$), 193 (33, $[C_{15}H_{13}]^+$), 180 (56, $[C_{14}H_{12}]^+$), and 149 [73].

3β-Acetoxy-24,24-diphenyl-5α-lanost-23-ene (VII)—Mass spectrum (12 ev), *m/z* (%): 580 (2, $[M]^+$), 565 (1, $[M-CH_3]^+$), 387 (2, $[M-C_{15}H_{13}]^+$), 357 (36, $[M-C_{17}H_{19}]^+$), 343[15], 327 (33, $[M-C_{15}H_{13}-CH_3CO_2H]^+$), 297 (6, $[M-C_{17}H_{19}-CH_3CO_2H]^+$), 283[3], 259[2], 231[3], 222[15], 217[15], 193 (22, $[C_{15}H_{13}]^+$), 180 (100, $[C_{14}H_{12}]^+$), 163[5], 149[4], 135[4], and 122[5].

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