Parke, Davis and Company

The Synthesis of Steroidal 16α,17α-Fused Isoxazolines and Isoxazolidines

T. P. Culbertson, G. W. Moersch and W. A. Neuklis

The reaction of nitrile oxides with Δ^{16}-steroids substituted at C-17 by $COCH_3$, $OCOCH_3$, CN, $NHCOCH_3$ and H afforded 16α,17α-[3,1-(2-isoxazolino)]-steroids. From the reaction of C,N-diphenylnitrone with pregna-5,16-diene-3β-ol-20-one acetate two isomeric products were isolated, 16α,17α-(2,3-diphenyl-3,1-isoxazolidino)-pregn-5-en-3β-ol-20-one acetate and 16α,17α-(2,3-diphenyl-1,3-isoxazolidino)-pregn-5-en-3β-ol-20-one acetate.

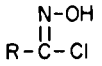
Certain steroids containing a heterocyclic ring fused at positions C-16 and C-17, such as the 16α,17α-dioxolane derivatives of 9α-fluoro-16α-hydroxyprednisolone (1) and of 16α,17α-dihydroxyprogesterone (2) have been reported by other investigators. The present investigation was undertaken in order to prepare steroids having other heterocyclic ring systems fused at these positions.

The cycloaddition of 1,3-dipolar reagents to unsaturated compounds, which has been reviewed by Huisgen (3), affords a simple method for the synthesis of five-membered nitrogen-containing heterocycles. The 1,3-cycloaddition of nitrile oxides to olefins affords Δ^2-isoxazolines (4), and it was of interest to use this reaction as a direct method for synthesizing isoxazolino-steroids. While this paper was being written Fritsch, Seidl and Ruschig (5) reported that benzonitrile oxide and acetonitrile oxide added to 3β-acetoxypregna-5,16-dien-20-one (Ia) to give isoxazolines with structure VIa (R_1 = C_6H_5, CH_3; R_2 = $COCH_3$). The structures assigned to these compounds were based on the assumption that the oxygen atom at the negatively charged end of the nitrile oxide dipole would add to the positively charged end of the α,β-unsaturated ketone dipole. However, Huisgen (3) has noted that 1,3-dipolar addition reactions are susceptible to both electronic and steric influences and that strained double bonds (such as cyclopentenes) do not necessarily require a polarizing substituent in order to react. We have carried out nitrile oxide addition reactions on Ia and other 17-substituted-Δ^{16}-steroids and have also isolated 16(17)-fused isoxazolinosteroids. The physical constants reported by the German workers for the above compounds and their 3β-hydroxy and Δ^4-3-ketone derivatives were similar to those we have found for products prepared from the same reactants. However, our compounds were shown to have the sterically less hindered structures IIIa and IIIb.

Benzonitrile oxide (IIa), acetonitrile oxide (IIb) and carboethoxyformonitrile oxide (IIc) reacted at room temperature with Ia to give 3β-acetoxy-16α,17α-[3-phenyl-3,1-(2-isoxazolino)]-pregn-5-en-20-one (IIIa), 3β-acetoxy-16α,17α-[3-methyl-3,1-(2-isoxazolino)]-pregn-5-en-20-one (IIIb), and 3β-acetoxy-16α,17α-[3-carboethoxy-3,1-(2-isoxazolino)]-pregn-5-en-20-one (IIIc), respectively (Table I). The reactions were run by generating the nitrile oxides *in situ*. Triethylamine was added to an ether or methylene chloride solution of the steroid, Ia, and the appropriate hydroxamoyl chloride, IV. The same results were obtained by reverse addition in which the hydroxamoyl chloride was added to a solution of the steroid and triethylamine. Although excess reagent was used, the elemental analyses of the products corresponded to 1:1 adducts. The fact that the Δ^{16}-double bond was specifically attacked could be seen by the disappearance of the 240 mμ band in the ultraviolet spectrum and the shift of the 20-ketone band in the infrared spectrum from 1660 cm^{-1} (conjugated) to 1714-1707 cm^{-1} (unconjugated, see Table II). Since only one isomer was obtained in each case, the cycloaddition of the nitrile oxides is considered to have been to the α-side of the steroid molecule, in conformity with other additions to the 16(17)-double bond, such as osmium tetroxide hydroxylation (6).

Addition by carbon-carbon bond formation at C-16 and carbon-oxygen bond formation at C-17 is supported by periodate titration of the aminotriols obtained by lithium aluminum hydride reduction of the isoxazolines IIIa and IIIb. Both aminotriols reacted with one mole of periodic acid and therefore must have the glycol structures Va and Vb (7). If the isoxazolines had structure VIa (reverse addition of the nitrile oxide to the double bond), lithium aluminum hydride reduction would have resulted in aminotriols with structure VII which have no vicinal hydroxyls and would not be expected to react with periodate. Additional evidence which supports the assignment of structure III is the pyrolytic ring opening of Xe (R_1 = CO_2H, R_2 = $COCH_3$), which was formed from 16-dehydroprogesterone and IIc (R_1 = $CO_2C_2H_5$), followed by hydrolysis of the ester, to give 16α-cyano-17α-hydroxypregn-4-en-3,20-dione IX (8). In this case the location of the hydroxyl group could only be accounted for if the isoxazoline ring were fused to the α-side of the steroid with the heterocyclic oxygen atom attached to the 17α-position.

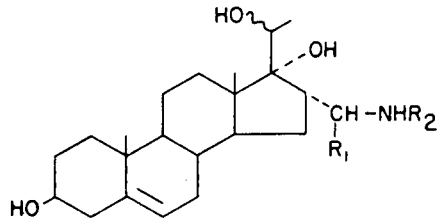
A crude isoxazoline was isolated from the reaction of androsta-5,16-dien-3β-ol (Ic) with benzonitrile oxide and purified as the 3β-acetate. In the n.m.r.



IVa, $R = C_6H_5$

IVb, $R = CH_3$

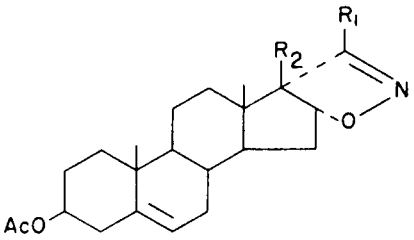
IVc, $R = CO_2C_2H_5$



V a, $R_1 = C_6H_5$, $R_2 = H$

V b, $R_1 = CH_3$, $R_2 = H$

V c, $R_1 = CH_3$, $R_2 = COCH_3$

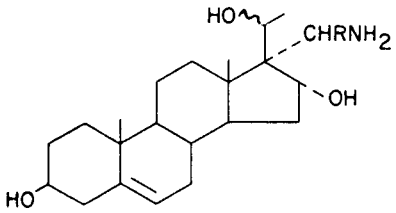


VI a, $R_1 = C_6H_5$, CH_3, $CO_2C_2H_5$; $R_2 = COCH_3$

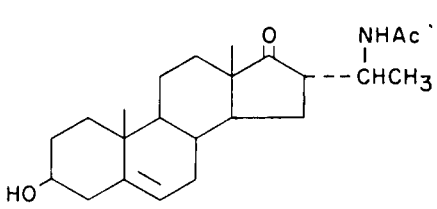
VI b, $R_1 = C_6H_5$, $R_2 = H$

VI c, $R_1 = C_6H_5$, $R_2 = OCOCH_3$

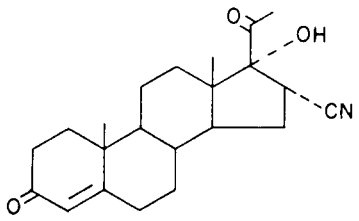
VI d, $R_1 = C_6H_5$, $R_2 = NHCOCH_3$



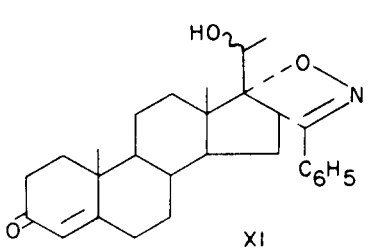
VII, $R = C_6H_5$, CH_3



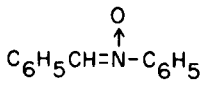
VIII



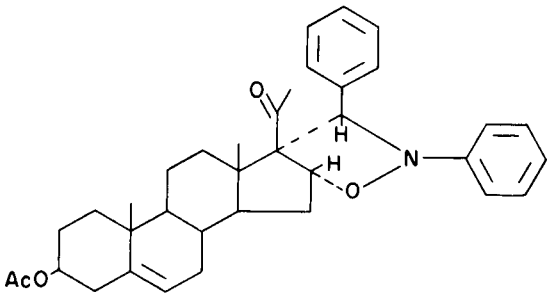
IX



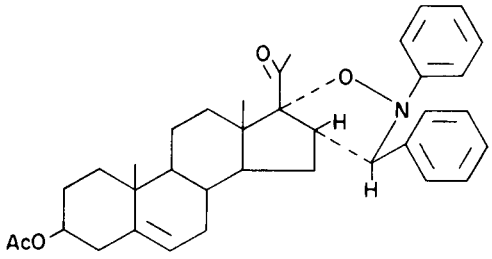
XI



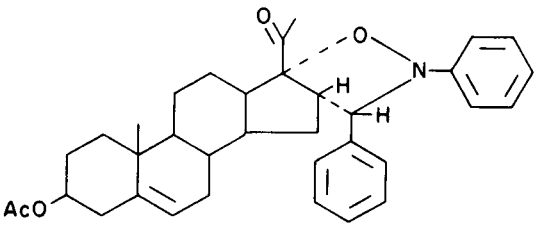
XII



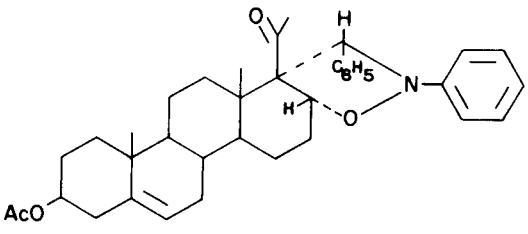
XIII



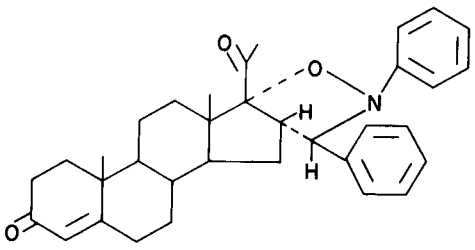
XIV



XV



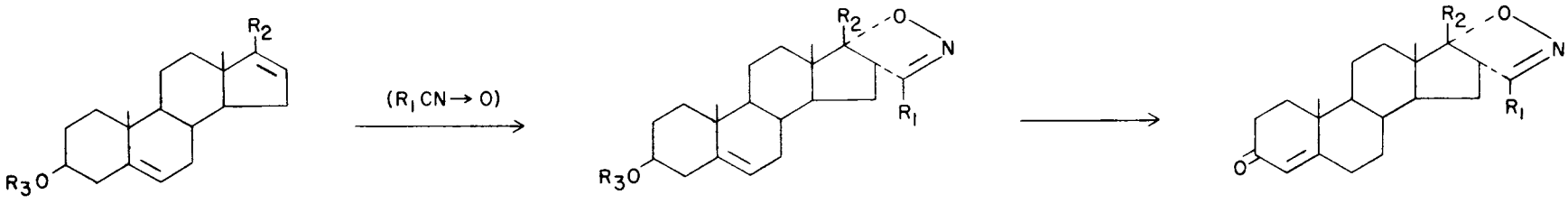
XVI



XVII

TABLE I

Isoxazolino Steroids



I II III X

Product	R_1	R_2	R_3	Starting Steroid	Yield %	M.p. °C, Solvent (a)	$[\alpha]_D^{23}$	Formula	% Carbon Calcd.	% Carbon Found	% Hydrogen Calcd.	% Hydrogen Found	% Nitrogen Calcd.	% Nitrogen Found
IIIa	C_6H_5	$COCH_3$	$COCH_3$	Ia	80	209-211, A	-70	$C_{30}H_{37}O_4N$	75.75	75.62	7.84	7.77	2.94	3.03
IIIb	CH_3	$COCH_3$	$COCH_3$	Ia	20	235-237.5, A	+43 (b)	$C_{25}H_{35}O_4N$	72.66	72.52	8.51	8.38	3.39	3.49
IIIc	$CO_2C_2H_5$	$COCH_3$	$COCH_3$	Ia	80	168-170, A	-14	$C_{27}H_{37}O_6N$	68.76	68.94	7.91	7.99	2.97	3.08
IIId	CH_3	$COCH_3$	H	IIIb	69	243-252, A		$C_{23}H_{33}O_3N$	74.36	74.42	8.95	8.87	3.77	3.79
IIIe	C_6H_5	$COCH_3$	H	IIIa		198-204, A-B		$C_{28}H_{35}O_3N$						
IIIf	C_6H_5	CN	$COCH_3$	Ib (c)	43	225-227, C	-98	$C_{29}H_{34}O_3N_2$	75.95	76.03	7.47	7.45	6.11	5.89
IIIg	C_6H_5	CN	H	IIIf	64	238-240, D-B		$C_{27}H_{32}O_2N_2$	77.86	77.81	7.75	7.67	6.73	6.56
IIIh	C_6H_5	H	$COCH_3$	Ic (d)		238-238.5, C-B (e)	-330 (c 0.5)	$C_{28}H_{35}O_3N$	77.56	77.80	8.14	8.16	3.23	3.21
IIIi	C_6H_5	$OCOCH_3$	$COCH_3$	Id (f)	94	235-237, F-E	-85 (c 0.5)	$C_{27}H_{37}O_5N$	73.29	73.03	7.59	7.45	2.85	2.71
IIIj	CH_3	$OCOCH_3$	$COCH_3$	Id	17	199-202, K-E	+11	$C_{25}H_{35}O_5N$	69.91	69.94	8.21	8.23	3.26	3.47
IIIk	$CO_2C_2H_5$	$OCOCH_3$	$COCH_3$	Id	31	245-246, A	-39	$C_{27}H_{37}O_7N$	66.51	66.57	7.64	7.71	2.87	2.92
IIIl	C_6H_5	$NHCOCH_3$	$COCH_3$	Ie (g)	88	299-301, J		$C_{30}H_{38}O_4N_2$	73.45	73.59	7.81	7.80	5.71	5.69
IIIm	C_6H_5	$OCOCH_3$	CHO	If (h)	30	190-191, I-E	-109	$C_{29}H_{35}O_5N$	72.91	72.65	7.39	7.47	2.93	2.88
Xa	C_6H_5	$COCH_3$		IIIe	53 (i)	221-222, G-H		$C_{28}H_{33}O_3N$	77.91	78.09	7.71	7.78	3.24	3.32
Xb	CH_3	$COCH_3$		IIId	56 (i)	235-239, G	+131 (j)	$C_{23}H_{31}O_3N$	74.76	74.72	8.46	8.34	3.79	3.85
Xc	C_6H_5	CN		IIIg	38 (k)	270-271, D-B		$C_{27}H_{30}O_2N_2 \cdot 1/2H_2O$	76.56	76.49	7.38	7.04	6.61	6.77
Xd	C_6H_5	$OCOCH_3$		IIIm	60 (l)	252-253.5, I	-43	$C_{28}H_{33}O_4N$	75.13	75.29	7.43	7.19	3.13	3.21

(a) Solvents of recrystallization: A, methanol; B, water; C, ethanol; D, dioxane; E, petroleum ether; F, benzene; G, acetone; H, heptane; I, ethyl acetate; J, tetrahydrofuran; K, ether. (b) Acetone solution. (c) L. Miramontes, P. Aguinaco and M. A. Romero, *J. Am. Chem. Soc.*, 82, 6153 (1960). (d) F. Sondheimer, O. Mancera, G. Rosenkranz, U. S. Patent 2,842,565, July 8, 1958. (e) The cycloaddition reaction was run on the 3β-alcohol but the product was purified as the 3β-acetate. (f) J. Fajkos and F. Sorm, *Coll. Czech. Chem. Comm.*, **24**, 766 (1959). (g) G. Rosenkranz, O. Mancera, F. Sondheimer, C. Djerassi, *J. Org. Chem.*, **21**, 520 (1956). (h) J. Fajkos, *Coll. Czech. Chem. Comm.*, **25**, 1082 (1960). (i) Oxidation procedure A. (j) Methanol solution. (k) Oxidation procedure B. (l) The 3-formate ester was oxidized directly by procedure A.

TABLE II

Ultraviolet and Infrared Spectra of Isoxazolino Steroids

Product	UV Data λ max, mμ (ε)	IR Data (cm^{-1}) 3-Acetate, or 20-ketone 17-acetate,		Misc.
IIIa	262 (12,650)	1724	1714	
IIIb		1728	1707	
IIIc	249 (4,950)	1728 (a)	1717	
IIId			1710	
IIIe	262 (13,000)		1713	3400 (OH)
IIIf	259 (13,950)	1728		2245 (CN)
IIIg				2245 (CN), 3450 (OH)
IIIh		1730		
IIIi	254 (11,800)	1728	1756	
IIIj		1729 (b)	1749	
IIIk	230 (5,910)	1733	1757	1722 (CO_2Et)
IIIl		1735		1700 ($NH-COCH_3$)
IIIm		1725 (b) (3-CHO)	1755	
Xa	245 (22,000)		1711	1670 (3-C=O)
Xb	239 (17,000)		1710	1665 (3-C=O)
Xc	246 (23,400)			1683 (3-C=O), 2240 (CN), 3460 (H_2O)
Xd	245 (26,400)		1755	1670 (3-C=O)

(a) This was coincident with the carbonyl band of the 3'-carbethoxy group. (b) Chloroform solution.

TABLE III

N.m.r. Spectra of Isoxazolino Steroids

Structure	18-Methyl	21-Methyl	N.m.r. (δ) 16-H (a)	Misc.
IIIa	0.80	2.31	4.45	7.5 (phenyl) (a)
IIIb	0.73	2.25	3.85	1.91 (3'-methyl)
IIId	0.72	2.25	3.74	1.92 (3'-methyl)
IIIf	1.04		4.36	7.5 (phenyl) (a)
IIIh	0.88		4.00	4.67 (17-H) (b), 7.5 (phenyl) (a)
IIIk	0.98		3.92	2.05 (17-OAc), 4.35 (c), and 1.37 (d) (CO_2Et)
IIIl	1.00		4.55	1.94 (N-Ac), 6.30 (N-H), and 7.5 (phenyl) (a)
Xa	0.83	2.30	4.47	5.71 (4-H), 7.5 (phenyl) (a)
Xb	0.76	2.25	3.83	1.92 (3'-methyl), 5.75 (4-H)

(a) Multiplet. (b) Doublet, J = 7 c.p.s. (c) Quartet, J = 7 c.p.s. (d) Triplet, J = 7 c.p.s.

spectrum (Table III) a doublet at δ 4.67 (J = 7 c.p.s.) appeared over a broad multiplet for the 3α-proton, indicating a hydrogen attached to a carbon bearing an oxygen atom. This could be best accounted for with structure IIIh, since the 17β-hydrogen should be split only by the 16β-hydrogen. If the nitrile oxide had added in the opposite direction to give VIb, this absorption would have come from the C-16 hydrogen, which would have been split into a more complex pattern by the C-15 and C-17 hydrogens.

Nitrile oxides IIa (R_1 = C_6H_5), IIb (R_1 = CH_3), and IIc (R_1 = $CO_2C_2H_5$) reacted readily with 3β,17-dihydroxyandrosta-5,16-diene diacetate (Id) to afford isoxazolines IIIi, IIIj, and IIIk. The 3-formate derivative IIIm was prepared by the reaction of If and IIa, and was converted directly to the Δ^4-3-ketone Xd by Oppenauer oxidation. Similarly, 17-acetamidoandrosta-5,16-dien-3β-ol acetate (Ie) reacted with IIa to give IIIl. Nitrile oxides have been reported to react with enamines (9) and with vinyl acetate (10) to form isoxazolines having two heteroatoms attached to the same carbon atom. In the infrared spectra of IIIi, IIIj, IIIk and IIIm the 17-acetoxy groups exhibited carbonyl stretching frequencies (*ca.* 1755 cm^{-1}) which were higher than those reported for most 17β-acetoxy steroids (1722-1720 cm^{-1}) (11). Freeman has observed similar shifts in carbonyl bands when carboxylic acids are esterified with electron withdrawing groups (12). For example, this band in acetal diacetate appeared at 1761 cm^{-1} even though the two acetoxy functions were separated by a carbon atom. A similar hypsochromic shift was observed for the 17-amide carbonyl band of IIIl which was found at 1700 cm^{-1} rather than at 1655 cm^{-1} as reported for 17β-acetamidoandrost-5-en-3β-ol acetate (13). Examples of the reverse addition products VIc and VId are not known, but it would seem more likely that shifts of this amount would result from structures which have the isoxazoline oxygen atom, rather than the carbon atom, attached to C-17 of the steroid.

The reaction of benzonitrile oxide with 3β-acetoxy-17-cyanoandrosta-5,16-diene (Ib) afforded the isoxazoline IIIf which was hydrolyzed to the 3β-alcohol IIIg and subsequently oxidized to the Δ^4-3-ketone Xc with chromic acid. The structure of the heterocyclic portion of these compounds was assigned by analogy with the previous examples. The Δ^4-3-keto steroids Xa and Xb were prepared from the Δ^5-3β-alcohols IIIe and IIId by Oppenauer oxidation. Treatment of IIIe with aluminum isopropylate alone in refluxing toluene resulted in formation of 20ξ-hydroxy-16α,-17α-[3-phenyl-3,1-(2-isoxazolino)]-pregn-4-en-3-one (XI). Similar aluminum isopropylate catalyzed reactions have been reported where a keto group in a steroid acts as a hydrogen acceptor for the oxidation of a hydroxyl group in the same molecule when no other ketone was added (14).

Nitrones are reported to react with olefins in a manner analogous to nitrile oxides to give isoxazolidines (3). C,N-Diphenylnitrone (XII) reacted with Ia in boiling toluene to give a crude mixture containing a considerable amount of unreacted steroid. Chromatography of the crude product and fractional crystallization afforded two isomeric isoxazolidines: A in 6% yield, m.p. 216-217° and B in 15-28% yield, m.p. 191-192°. The infrared spectra clearly showed that the reaction involved the Δ^{16}-double bond. Since diphenylnitrone has even greater steric bulk than benzonitrile oxide, it was assumed that these isomeric products also resulted from α-side addition to the steroid. This was somewhat supported by the C-18 methyl resonances of products A and B which appeared at δ 0.80 and δ 0.73, respectively, and suggested that the 17-acetyl group is β-oriented. Similar C-18 methyl peaks are found for 17α-hydroxyprogesterone (δ 0.76) (15), for 17α-methylprogesterone (δ 0.72) (16) and for the isoxazolino steroid IIIa (δ 0.80). Epimerization of a 17-acetyl group from the β- to α-side is reported to cause a downfield shift of δ 0.3 in the C-18 methyl re-

sonance (17). If β-side addition by the diphenylnitrone had occurred, the resulting oxygen atom at the 16β- or 17β-position also would be expected to deshield the C-18 methyl group (15).

The proton resonance for product A which appeared as a multiplet at δ 5.40 was assigned to the 16β-hydrogen since it was farther downfield than the 3α-proton absorption (*ca.* δ 4.6), suggesting that the electronegative portion of the isoxazolidine ring (-O-N) was attached to the C-16 carbon atom. For product B the C-16 proton signal appeared at δ 3.70, which, although shifted somewhat downfield for a methine hydrogen, was near the values found for the C-16 hydrogens on the isoxazolino steroids (δ 3.7-4.5). Additional evidence for the direction of diphenylnitrone addition across the Δ^{16}-double bond was indicated by the 20-ketone bands in the infrared spectra of A and B at 1705 cm^{-1} and 1715 cm^{-1}, respectively. Hypsochromic shifts of 20-ketones are reported for steroids when acetoxy groups are attached to the adjacent C-17 or C-21 positions (18), while 17α-alkyl substituents cause a bathochromic shift (16). Since the higher frequency carbonyl band would be expected to be associated with the more electronegative 17α-substituent (-O-N), the two products appeared to be isomers in which A had the 16α,17α-(1,3-isoxazolidino) structure and B had the reverse 16α,17α-(3,1-isoxazolidino) structure.

The C-21 methyl group of A appeared as a three proton singlet at δ 1.55 which was upfield from the usual position reported for methyl ketones (δ 2.1-2.4) (19), suggesting that it was strongly shielded through space. The benzyl proton appeared as a lone singlet at δ 4.96, shifted downfield from the expected position for a hydrogen attached to a carbon which is bound to phenyl and nitrogen (20). These data were best accommodated by structure XIII. A Dreiding model of XIII indicated that the rotations of the C-phenyl and 17β-acetyl groups should be hindered because of the close proximity between themselves, the adjacent C-18 methyl and the N-phenyl group. In the least hindered conformation the C-21 methyl hydrogens are positioned over the face of the C-phenyl ring in the positive shielding region (21). In this conformation the benzyl hydrogen lies approximately in the plane of the C-phenyl ring and should be deshielded. The aromatic protons on both phenyl rings showed a complex splitting pattern at δ 6.6-7.5. This would be expected for the N-phenyl group since resonance of the unshared pair of electrons on nitrogen would result in nonequivalent *o*, *m* and *p*-hydrogens (22). The splitting of the C-phenyl protons apparently resulted from restricted rotation of the phenyl ring such that each of its protons is not subjected to the same magnetic environment. This is somewhat analogous to the splitting of the phenyl protons in *cis*-1,2-diphenylcyclopentane (23).

In the n.m.r. spectrum of the isomeric major product, B, the C-21 methyl group was not shielded and appeared as a singlet at δ 2.40. The fact that the C-21 methyl peak appeared at a position somewhat downfield for a 20-keto steroid with a 17α-hydrogen (19) or alkyl group (16) (*ca.* δ 2.1) and compared favorably with the δ 2.31 peak found in the isoxazolino steroid IVa again suggested that the heterocyclic oxygen atom in B was attached to C-17. The protons on the N-phenyl ring again appeared as a multiplet centered at δ 7.02, but a sharp five proton singlet at δ 7.27 was evident for the C-phenyl group. This suggested that the C-phenyl ring in this case is in a position where it can rotate freely in terms of the n.m.r. time scale such that its protons would be surrounded by the same magnetic environment. These data were best accommodated by structure XIV, especially if the N-phenyl group were flipped toward the less hindered α-side. In alternative structures XV and XVI the C-21 methyl cannot be shielded by the C-phenyl ring; however, Dreiding models revealed that the rotation of the C-phenyl ring in XV should be hindered by the 15α-hydrogen and in XVI by the 12α-hydrogen. If this were the case, shielding of these protons by the aromatic ring should cause them to absorb at a higher magnetic field and perhaps be distinguishable, but no such peaks were observed.

The benzyl proton of B was found as a singlet at the expected position of δ 3.70 (20) in the center of a multiplet for the C-16 proton. The dihedral angle between the benzyl hydrogen and the C-16 hydrogen in a Dreiding model of XIV appeared to be near 90° when the isoxazolidine ring was puckered. In this conformation splitting of the benzyl proton by the C-16 proton should be at a minimum according to the Karplus equation (24). By the same argument structure XV was further ruled out because the dihedral angle between the benzyl and C-16 hydrogens would be about 30°, which should allow the benzyl proton to be split into a doublet by the C-16 proton.

The Δ^4-3-ketone XVII was prepared from compound B by hydrolysis to the 3β-alcohol and subsequent Oppenauer oxidation.

EXPERIMENTAL (25)

Hydroxamoyl Chlorides.

Benzhydroxamoyl chloride (26) (IVa) was prepared by passing one equivalent of chlorine gas into a chloroform solution of benzaldoxime cooled in an ice bath. Evaporation of the solvent at reduced pressure gave an oil which partially solidified after refrigeration overnight. The crude product was kept in the cold and could be used without purification for periods up to two weeks.

Acethydroxamoyl chloride (27) (IVb) was obtained as a crude oil by passing one equivalent of chlorine gas into an ice-cold solution of acetaldoxime in 5% hydrochloric acid. After saturating the solution with ammonium sulfate the product was extracted with ether and dried over calcium chloride. The ether solution was kept cold and used as such without further purification.

Ethyl chloroximinoacetate (IVc) was prepared according to Skinner (28) by treatment of ethyl glycinate in hydrochloric acid solution at -15° to -20° with sodium nitrite solution. The material which crystallized from the reaction mixture was filtered, washed with water, dissolved in benzene and dried. The product crystallized upon addition of petroleum ether and cooling to give fine, white needles, m.p. 79.5-80°.

Nitrile Oxide Addition Reactions.

Excess triethylamine (0.2 to 0.4 moles) in ether or methylene chloride solution was added dropwise to a stirred solution of the steroid (0.01 mole) and the hydroxamoyl chloride (0.02 to 0.03 moles) in ether or methylene chloride. After standing overnight the triethylamine hydrochloride was removed by filtration and washed with the solvent. Evaporation of the solvent gave a crude product which was recrystallized.

Hydrolysis Procedure.

The 3β-acetoxy steroids IIIa and IIIb were treated with a solution of 0.5-1% sodium hydroxide in methanol and refluxed 0.5 hr. IIIf was treated in the same manner but allowed to stand at room temperature for 2 hrs. The solutions were then poured into water and the products were filtered and recrystallized.

Oxidation Procedures.

Procedure A: A mixture of the steroid alcohol (0.01 mole) and aluminum isopropylate (0.01-0.05 mole) in 500 ml. of dry toluene with 50 ml. of cyclohexanone was refluxed for 1-1.5 hr. An excess of a saturated solution of sodium potassium tartrate (50-200 ml.) was then added and the mixture was steam distilled until the distillate was clear. The crude product was filtered, washed with water, dried, and recrystallized.

Procedure B: The steroid alcohol (0.01 mole) in 1 l. of acetone (distilled from $KMnO_4$) was treated under a nitrogen atmosphere at ice bath temperature with 0.011 mole of Jones' reagent (26.7 g. CrO_3 and 23 ml. conc. H_2SO_4 diluted to 100 ml. with water) during about 10 min. After stirring an additional 5 min., the mixture was diluted with ice water and the product was filtered, washed with water, and dried. The crude product was then refluxed for 25 min. in 200 ml. of 95% ethanol with 1.0 g. of oxalic acid dihydrate. The solvent was evaporated under reduced pressure and the residue was recrystallized.

20ξ-Hydroxy-16α,17α-[3-phenyl-3,1-(2-isoxazolino)]-pregn-4-en-3-one (XI).

Treatment of III with a 5 mole excess of aluminum isopropylate according to Oxidation Procedure A without added cyclohexanone gave XI in 16% yield, m.p. 289-291° (from acetone); λ max 245 mμ (log ε, 4.34); ν max 3450, 1676, 1622 cm^{-1}.

Anal. Calcd. for $C_{28}H_{35}O_3N$: C, 77.55; H, 8.14; N, 3.23. Found: C, 77.22; H, 8.13; N, 3.39.

3β,17α,20ξ-Trihydroxy-16α-(aminobenzyl)-pregn-5-ene (Va).

A solution of 2.00 g. of IIIa in 60 ml. of tetrahydrofuran was added dropwise at room temperature to 3.0 g. of lithium aluminum hydride in 100 ml. of tetrahydrofuran during 1 hr. The reaction was refluxed 3.5 hr. and stirred overnight at room temperature. It was then decomposed by cautious addition of 10 ml. of water and 5 ml. of 15% sodium hydroxide solution with vigorous stirring. After stirring 1 hr. the white granular precipitate was filtered and washed with tetrahydrofuran. Evaporation of the filtrate to dryness gave 2.02 g. of a white amorphous solid. When this material was shaken with dilute hydrochloric acid the hydrochloride salt was formed as a slightly soluble solid, which was filtered and dried. Two recrystallizations from methanol-water gave 0.32 g. of granular crystals of the hydrochloride salt, m.p. 265-268°.

Anal. Calcd. for $C_{28}H_{42}O_3NCl\cdot H_2O$: C, 68.06; H, 8.98; N, 2.84; Cl, 7.18. Found: C, 68.21; H, 9.05; N, 2.86; Cl, 7.10.

The hydrochloride of Va in dilute aqueous solution was treated with sodium hydroxide solution and the free base was extracted into ether. Evaporation of the ether gave a glass which was crystallized twice from benzene-hexane, m.p. 133-134°.

Anal. Calcd. for $C_{28}H_{41}O_3N$: C, 76.49; H, 9.40; N, 3.18. Found: C, 76.45; H, 9.19; N, 3.19.

The titration of Va hydrochloride with periodic acid (29) was complete within 4 hrs. at which time one mole of periodate had reacted with one mole of steroid.

3α,17α,20ξ-Trihydroxy-16α-(1-aminoethyl)-pregn-5-ene (Vb).

A lithium aluminum hydride reduction was carried out on 2.00 g. of IIIb in the same manner as for the preparation of Va to give 1.67 g. of crude product. Four recrystallizations from methanol gave an analytical sample, m.p. 204-209°.

Anal. Calcd. for $C_{23}H_{39}O_3N$: C, 73.17; H, 10.41; N, 3.71. Found: C, 72.81; H, 10.64; N, 3.68.

Periodate titration of Vb in aqueous methanol showed an uptake of 1.07 equivalents of periodic acid in 40 min. and a slow further oxidation to 1.44 equivalents after 6 hr.

The N-acetyl derivative, Vc, was prepared by reacting Vb in methanol solution with acetic anhydride for 2 days. Evaporation to dryness and recrystallization of the residue from methanol-water gave a product which softened at about 170° and melted at 190-194°; ν max 1635, 1665 3400 cm^{-1}.

Anal. Calcd. for $C_{25}H_{41}O_4N$: C, 71.56; H, 9.85; N, 3.34. Found: C, 71.22; H, 9.89; N, 3.37.

16α-(1-Acetamidoethyl)-androst-5-en-3β-ol-17-one (VIII).

A solution of 14 mg. of Vc in 6 ml. of methanol and 2 ml. of 0.1M sodium periodate solution was allowed to stand at room temperature for 39 hr. The reaction mixture was diluted with 5 ml. of water and some of the methanol was evaporated at reduced pressure. Filtration gave 7 mg. of crystalline material, m.p. 215-218°; ν max 1550, 1655, 1735 and 3350 cm^{-1}.

16α,17α-(2,3-Diphenyl-1,3-isoxazolidino)-pregn-5-en-3β-ol-20-one acetate, (XIII) and 16α,17α-(2,3-diphenyl-3,1-isoxazolidino)-pregn-5-en-3β-ol-20-one acetate (XIV).

A solution of 7.14 g. of pregna-5,16-dien-3β-ol-20-one acetate and 4.50 g. of C,N-diphenylnitrone (30) in 100 ml. of toluene was refluxed for 72 hr. Evaporation of the solvent at reduced pressure gave an orange oil. This crude material was partially purified by dissolving it in benzene and passing the solution through a column containing 100 g. of alumina (Merck, acid washed). Evaporation of the eluates gave 10.53 g. of solid material which was recrystallized from cyclohexane-petroleum ether. The first crop, 2.70 g., had a m.p. of 170-195° and contained a considerable amount of starting material as indicated by the infrared spectrum. Fractional crystallization of this material from methanol gave 0.62 g. (6%) of XIII, m.p. 216-217°, $[\alpha]_D^{23}$ -94.5° (c 0.64); ν max 1495, 1605, 1705, 1735 cm^{-1}. The n.m.r. spectrum showed singlets integrating for 3 protons each at δ 0.80 (C-18 methyl), δ 1.03 (C-19 methyl), δ 1.55 (C-21 methyl) and δ 2.02 (3β-acetate), a singlet for one proton at δ 4.96 (benzyl hydrogen), a single proton multiplet centered at δ 4.55 (3α-hydrogen), a two proton multiplet at δ 5.40 (C-6 and C-16 hydrogens) and complex multiplets integrating for 10 protons at δ 6.6-7.5 (N-phenyl and C-phenyl).

Anal. Calcd. for $C_{36}H_{43}O_4N$: C, 78.09; H, 7.83; N, 2.53. Found: C, 78.38; H, 7.81; N, 2.62.

Partial evaporation of the cyclohexane-petroleum ether filtrate gave a crude product which when recrystallized from methanol gave 1.60 g. (15%) of XIV (31), m.p. 182-184°. A sample recrystallized for analysis had m.p. 191-191.5°; λ max 250 mμ (ε, 6,370); ν max 1495, 1605, 1715, 1735 cm^{-1}. The n.m.r. spectrum showed singlets integrating for three protons each at δ 0.73 (C-18 methyl), δ 1.05 (C-19 methyl), δ 2.02 (3β-acetate), δ 2.40 (C-21 methyl). A singlet at δ 3.70 (benzyl hydrogen) along with a multiplet centered at the same position (C-16 hydrogen) integrated for two protons. Multiplets integrating for one proton each were centered at δ 4.60 (3α-hydrogen) and δ 5.30 (C-6 hydrogen). The N-phenyl protons appeared as a multiplet at δ 7.02 and the C-phenyl protons as a singlet at δ 7.27.

Anal. Calcd. for $C_{36}H_{43}O_4N$: C, 78.09; H, 7.83; N, 2.53. Found: C, 78.48; H, 8.02; N, 2.62.

16α,17α-(2,3-Diphenyl-3,1-isoxazolidino)-pregn-4-en-3-one (XVII).

A solution of 2.72 g. of XIV in 200 ml. of methanol and 2 ml. of 50% sodium hydroxide solution was refluxed 0.5 hr., cooled and poured into water. The crude product, 16α,17α-(2,3-diphenyl-3,1-isoxazolidino)-pregn-5-en-3β-ol, was filtered and dried; yield, 2.45 g., m.p. 130-140°; ν max 1712, 3450 cm^{-1}. This material was dissolved in 150 ml. of toluene and refluxed 1.5 hr. with 4.5 g. of aluminum isopropylate and 25 ml. of cyclohexanone. The mixture was steam distilled to remove volatile materials, cooled, and the crude solid was filtered and washed with water. Recrystallization from cyclohexane-petroleum ether afforded 1.80 g. of crystals, m.p. 218-226°. A sample recrystallized for analysis had m.p. 227-229°; $[\alpha]_D^{23}$ +43.9° (c 1.02); λ max 241 mμ (ε, 23,100); ν max 1618, 1675, 1712 cm^{-1}.

Anal. Calcd. for $C_{34}H_{39}O_3N$: C, 80.11; H, 7.71; N, 2.75. Found: C, 80.37; H, 7.86; N, 2.87.

Acknowledgment.

The authors wish to thank Mr. C. E. Childs and the staff of our Microanalytical Laboratory, and Dr. J. M. Vandenbelt and the staff of our Physical Chemistry Laboratory for their valuable technical assistance. We are also indebted to Professor H. S. Mosher, Stanford University, for some of the n.m.r. data and their interpretation.

REFERENCES

(1) J. Fried, A. Borman, W. B. Kessler, P. Grabowich and E. F. Sabo, *J. Am. Chem. Soc.*, 80, 2338 (1958).

(2) J. Fried, E. F. Sabo, P. Grabowich, L. J. Lerner, W. B. Kessler, D. M. Brennan and A. Borman, *Chem. and Ind.* (London), 465 (1961).

(3) R. Huisgen, *Angew. Chem. (Inter. Ed.)*, **2**, 565, 633 (1963).

(4) A. Quilico, "The Chemistry of Heterocyclic Compounds, Five-

and Six- Membered Compounds with Nitrogen and Oxygen," R. H. Wiley, Ed., Interscience Publishers, Inc., New York, N. Y., 1962, p. 95.

(5) W. Fritsch, G. Seidl and H. Ruschig, *Ann.*, 677, 139 (1964).

(6) N. L. Wendler and D. Taub, *J. Am. Chem. Soc.*, 82, 2836 (1960).

(7) The fact that a glycol structure was cleaved was confirmed by the isolation of the keto-amide VIII from the periodate oxidation of Vc.

(8) G. W. Moersch, E. L. Wittle, and W. A. Neuklis, to be published.

(9) M. E. Keuhne, S. J. Weaver and P. Franz, *J. Org. Chem.*, 29, 1582 (1964).

(10) T. Mukaiyama and T. Hoshino, *J. Am. Chem. Soc.*, 82, 5339 (1960).

(11) G. Roberts, B. S. Gallagher, and R. N. Jones, "Infrared Absorption Spectra of Steroids", Vol. II, Interscience Publishers, Inc. New York, N. Y., 1958, p. 26.

(12) J. P. Freeman, *J. Am. Chem. Soc.*, 80, 5954 (1958).

(13) P. de Ruggieri, C. Ferrari, and C. Gandolfi, *Gazz. chim. ital.*, 91, 655 (1961).

(14) C. Djerassi, *Org. Reactions*, 6, 231 (1951).

(15) R. F. Zurcher, *Helv. Chim. Acta*, 46, 2054 (1963).

(16) M. J. Weiss, R. E. Schaub, G. R. Allen, Jr., J. F. Poletto, C. Pidacks, R. B. Conrow and C. J. Coscia, *Tetrahedron*, 20, 357 (1964).

(17) M. B. Rubin and E. C. Blossey, *J. Org. Chem.*, 29, 1932 (1964).

(18) See Reference 11, p. 25.

(19) N. S. Bhacca and D. H. Williams, "Application of NMR Spectroscopy in Organic Chemistry", Holden-Day, Inc., San Francisco, 1964, p. 33.

(20) Protons on a methylene bearing phenyl and nitrogen substituents are reported to absorb at δ 3.56. L. M. Jackman, "Application of Nuclear Magnetic Resonance Spectroscopy in Organic Chemistry", Pergamon Press, London 1959, p. 60.

(21) See reference 20, p. 125.

(22) See reference 20, p. 62.

(23) D. Y. Curtin, H. Gruen, Y. G. Hendrickson and H. E. Knipmeyer, *J. Am. Chem. Soc.*, 83, 4838 (1961).

(24) M. Karplus, *J. Chem. Phys.*, 30, 11 (1959).

(25) The melting points were determined in a Thomas-Hoover melting point apparatus and are corrected. The infrared spectra were recorded in KBr disks with a Beckman IR-7 spectrophotometer. The n.m.r. spectra were recorded in deuterochloroform solution using a Varian A-60 spectrometer with resonance reported as parts per million downfield from tetramethylsilane used as an internal reference. The ultraviolet spectra were recorded in methanol solution using a Cary Model 14 spectrophotometer. Rotations were recorded in one per cent chloroform solution unless otherwise noted.

(26) A. Werner and H. Buss, *Ber.*, 27, 2193 (1894).

(27) H. Wieland, *ibid.*, 40, 1676 (1907).

(28) G. S. Skinner, *J. Am. Chem. Soc.*, 46, 731 (1924).

(29) E. L. Jackson, *Org. Reactions*, 2, 341 (1944).

(30) E. Bamberger, *Ber.*, 27, 1556 (1894).

(31) In a similar run XIV was isolated in 28% yield as the only product.

Received December 10, 1964 Ann Arbor, Michigan