

## A New Class of Natural Steroids, with Ring D Aromatic, from *Nicandra physaloides* (Solanaceae). X-Ray Analysis of Nic-10, and the Structures of Nic-1 ('Nicandrenone'), -12, and -17

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X-Ray analysis of Nic-10 from *Nicandra physaloides* shows it to be the novel  $6\alpha,7\alpha$ -epoxy-5-hydroxy-17(13  $\rightarrow$  18)*abeo*-5 $\alpha$ -pregna-2,13,15,17-tetraene-1,20-dione (1), in which ring D is aromatic and six-membered, apparently by inclusion of the C-13 methyl. The nature of the oxygenated A and B rings, however, show clearly its familial relationships with other Solanaceae steroids.

Structure work on three further aromatic (n.m.r.) steroids was carried out spectrally. Nic-1 (nicandrenone) (4) proved to have the ABCD system of Nic-10, but carried the full ergostane side-chain oxidatively elaborated into the same epoxidised cyclic hemiacetal ring E feature as previously encountered in Nic-3. Nic-12 (2) and Nic-17 (3) also have the same ABCD system as Nic-1 and Nic-10, but unlike the latter, which have C<sub>9</sub> and C<sub>3</sub> side-chains respectively, these both have a C<sub>6</sub> side-chain. The four ring-D-aromatic steroids can be arranged in a biosynthetic sequence involving plausible degradation steps. Possible mechanisms for the ring D expansion-aromatisation reaction are mentioned.

In the preceding paper<sup>1</sup> the leaf extract of *Nicandra physaloides* (Solanaceae), which has antifeeding properties towards insect larvae, was examined. The structures of three related, and highly oxygenated, 24-methyl steroids (Nic-3, -7, and -11) were assigned. We now report a second family of compounds which contain a novel steroid-like structure in which ring D is expanded by incorporation of an oxidised 13-methyl

group, and aromatised. This unique feature was first elucidated in Nic-10 by X-ray single-crystal analysis.<sup>2</sup>

The structure of Nic-10, † m.p. 207 °C, C<sub>21</sub>H<sub>22</sub>O<sub>4</sub>, was determined from diffractometer data (1 610 reflections) by direct methods and refined by least squares and difference Fourier methods to *R* 0.055. A general view of the structure is shown in Figure 1 together with the numbering used for crystallographic purposes only. Bond lengths and angles are displayed in Figures 2 and 3,

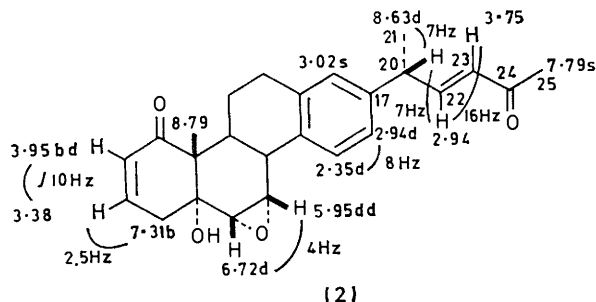
† See preceding paper for isolation and nomenclature.

<sup>1</sup> M. J. Begley, L. Crombie, P. J. Ham, and D. A. Whiting, preceding paper.

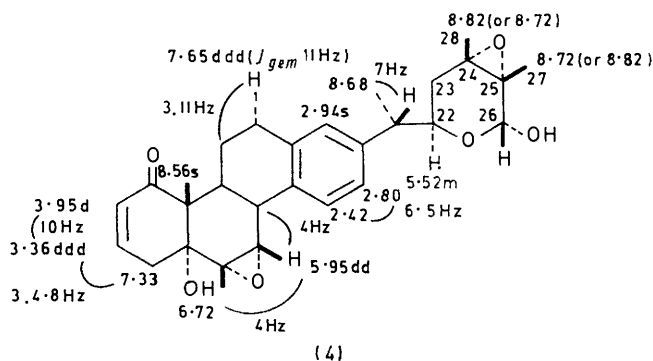
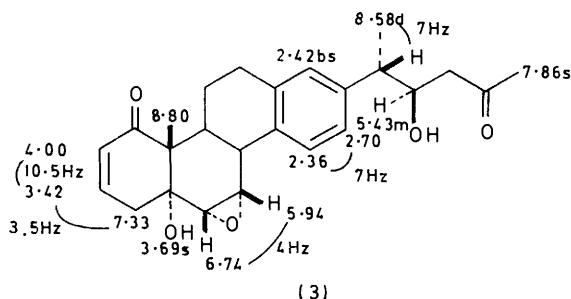
<sup>2</sup> For a preliminary report see M. J. Begley, L. Crombie, P. J. Ham, and D. A. Whiting, *J.C.S. Chem. Comm.*, 1972, 1250.



Finally, we report on Nic-1, m.p. 117 °C,\*  $C_{28}H_{34}O_6$ . Nic-1 is the extractive most like the preparation 'nicandrenone' isolated from *N. physaloides* by Fraenkel



*et al.*<sup>3</sup> and was shown to have repellent and insecticidal properties: indeed the essential identity of Nic-1 and 'nicandrenone' is assumed by Bates and Eckert.<sup>4</sup> Structure (4) was allocated to Nic-1 as a result of



detailed spectroscopic analysis, using the compound of this and our previous paper<sup>1</sup> for model data. Nic-1 has rings A—D in common with Nic-10, -12, and -17, and ring E is as found in Nic-3 and -7. The relative stereochemistry of rings A—D in compounds (2)—(4) is assumed, in the absence of any spectroscopic pointers to the contrary, to be the same as in the X-ray-derived structure of Nic-10, while the stereochemistry of the asymmetric centres in the C(20)—(26) chain is likely to be the same as in the X-ray-derived structure of Nic-3.<sup>1</sup> The absolute stereochemistry of compounds (1)—(4) is shown by their o.r.d. spectra. These are dominated by

\* One specimen, m.p. 138 °C, obtained from ethanol-light petroleum was probably a dimorph.

the strong negative Cotton effect of the (ring A) cyclohexenone R-band ( $n \rightarrow \pi^*$ ) near 330 nm, which is also shown by Nic-3, -7, and -11 and which can be correlated with the 5(R),10(R) configuration shown.<sup>1</sup> Contemporaneous with our preliminary communication,<sup>2</sup> Bates and Eckert independently suggested structure (4),<sup>4</sup> but without stereochemical assignments, as a result of detailed analysis of <sup>1</sup>H and <sup>13</sup>C n.m.r. data.

The biogenesis of these *Nicandra* extractives can be linked by the hypothesis outlined in Scheme 1. Aromatisation of ring D by inclusion of the angular methyl represents a new biosynthetic pathway which may have interest in connexion with aberrant steroid metabolism and cancer induction. Oxidation of C(18), cleavage of the C(13)—C(17) bond and recyclisation with C(18)—C(17) are required, and in the absence of any experimentally derived restrictions on the process, some speculative ways in which this might be accomplished are shown in Scheme 2. As shown in Scheme 1, nucleophilic opening of the 24,25-epoxy-function of Nic-1, shown in the  $\delta$ -hydroxyaldehyde form (5), permits retro-aldol cleavage (6), giving rise to Nic-17 and an aldehyde. Dehydration affords Nic-12, and further oxidative degradation may lead to Nic-10: one mechanistic possibility (7) is shown.

#### EXPERIMENTAL

For cultivation and extraction of *Nicandra physaloides*, and isolation of the terpenoid components, see the preceding paper.

*Nic-10* [6 $\alpha$ ,7 $\alpha$ -epoxy-5-hydroxy-17(13  $\rightarrow$  18)abeo-5 $\alpha$ -pregna-2,13,15,17-tetraene-1,2,0-dione] (1) had m.p. 207 °C from ethanol (Found: C, 74.55; H, 6.80%; *M*, 338.151; *M* - 15, 323.128.  $C_{27}H_{32}O_4$  requires C, 74.55; H, 6.55%; *M*, 338.152; *M* -  $CH_3$ , 323.128);  $\lambda_{max}$  (EtOH) 212 (33 950), 265 (18 550), and 285i nm (2 200);  $\nu_{max}$  (KBr) 3 490 (OH), 3 020, 1 675 (C=O), 1 630, 1 565, 1 362, 1 292, 1 260, 1 097, 914, and 892  $cm^{-1}$ ;  $\tau$  ( $CD_3SO_2CD_3$ ) 2.06 (1 H, dd,  $J_{15,16}$  7 Hz, 16-H), 2.31 (1 H, s, 18-H), 2.42 (1 H, d,  $J$  7 Hz, 15-H), 3.38 (1 H, ddd,  $J$  10, 5, 2 Hz, 3-H), 4.29 (1 H, bd,  $J$  10 Hz, 2-H), 5.43 (1 H, s, OH), 6.01 (1 H, dd,  $J$  4, 2 Hz, 7-H), 6.86 (1 H, d,  $J$  4 Hz, 6-H), 7.21 (3 H, s, 21-H<sub>3</sub>), and 8.87 (3 H, s, 19-H<sub>3</sub>);  $[M]^{23}_{272 nm} + 7 300^\circ$  (min.),  $[M]^{23}_{299 nm} + 9 290^\circ$  (max.), and  $[M]^{23}_{363 nm} - 3 900^\circ$  (min.).

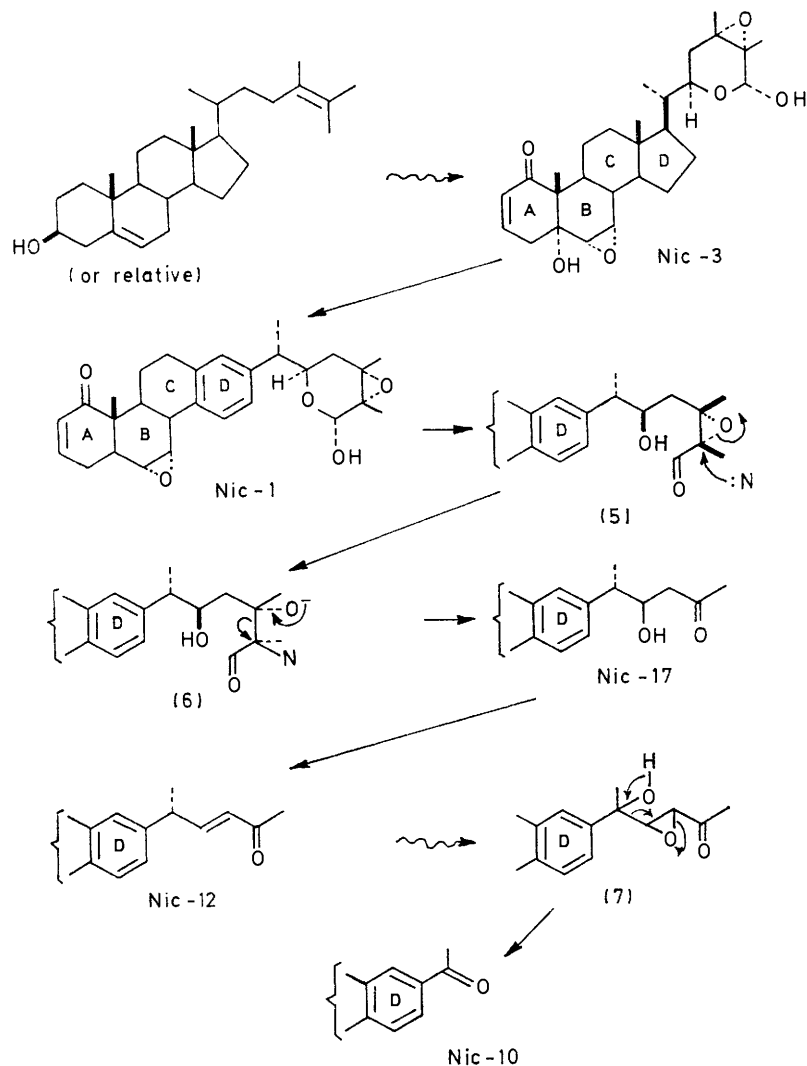
*Nic-12* [6 $\alpha$ ,7 $\alpha$ -epoxy-5-hydroxy-26,27,28-trinor-17(13  $\rightarrow$  18)abeo-5 $\alpha$ -ergosta-2,13,15,17,22-pentaene-1,24-dione] (2) had m.p. 174—175 °C from ethanol (Found: *M*, 392.196; *M* - 43, 349.182.  $C_{25}H_{28}O_4$  requires *M*, 392.199; *M* -  $CH_3CO$ , 349.180);  $\lambda_{max}$  (EtOH) 219 nm (26 500);  $\nu_{max}$  (KBr) 3 510 (OH), 1 684 (C=O), 1 665 (C=O), 1 630, 1 254, 996, 914, and 895  $cm^{-1}$ ;  $\tau$  ( $CD_3SO_2CD_3$ ) 2.57 (1 H, d,  $J$  8 Hz, 15-H), 2.98 (1 H, d,  $J$  8 Hz, 16-H), 3.01 (1 H, s, 18-H), 3.01 (1 H, m, 22-H), 3.36 (1 H, ddd,  $J$  10, 5, 2 Hz, 3-H), 4.01 (1 H, d,  $J$  16 Hz, 23-H), 4.30 (1 H, d,  $J$  10 Hz, 2-H), 5.47 (1 H, s, OH), 6.04 (1 H, dd,  $J$  4, 2 Hz, 7-H), 6.86 (1 H, d,  $J$  4 Hz, 6-H), 7.83 (3 H, s, 25-H<sub>3</sub>), 8.65 (3 H, d,  $J$  7 Hz, 21-H<sub>3</sub>), and 8.86 (3 H, s, 19-H<sub>3</sub>);  $[M]^{23}_{260 nm} + 3 300^\circ$  (min.),  $[M]^{23}_{305} + 6 150^\circ$  (max.),  $[M]^{23}_{287} + 5 650^\circ$  (min.),  $[M]^{23}_{305} + 6 250^\circ$  (max.), and  $[M]^{23}_{361} - 3 542^\circ$  (min.).

<sup>3</sup> O. Nalbandov, R. T. Yamamoto, and G. Fraenkel, *J. Agric. Food Chem.*, 1964, **12**, 55.

<sup>4</sup> R. B. Bates and D. J. Eckert, *J. Amer. Chem. Soc.*, 1972, **94**, 8258.

*Nic-17* [6 $\alpha$ ,7 $\alpha$ -epoxy-5,22-dihydroxy-2,6,27,28-trinor-17(13  $\rightarrow$  18)abeo-5 $\alpha$ -ergosta-2,13,15,17-tetraene-1,24-dione] (3) was precipitated from ethanol solution by light petroleum (b.p. 60–80 °C), m.p. 86 °C (Found:  $M$ , 410.209;  $M$  – 18, 392.199;  $M$  – 86, 324.173.  $C_{25}H_{30}O_5$  requires  $M$ , 410.209;  $M$  –  $H_2O$ , 392.199;  $M$  –  $C_4H_6O_2$ , 324.173);  $\lambda_{max}$  (EtOH) 216 (15 000), 276 (1 400), 295i (1 100), and 302 nm (1 100);  $\nu_{max}$  (KBr) 3 490 (OH), 3 450 (OH), 1 698 (C=O), 1 678 (C=O), 1 503, 1 288, 1 068, and 923  $cm^{-1}$ ;  $\tau$  ( $CD_3$ -SO- $CD_3$ )

138 °C) (Found:  $M$ , 466.236;  $M$  – 18, 448.225;  $M$  – 143, 323.165.  $C_{28}H_{34}O_6$  requires  $M$ , 466.236;  $M$  –  $H_2O$ , 448.225;  $M$  –  $C_7H_{11}O_3$ , 323.165);  $\lambda_{max}$  (EtOH) 217 (13 100), 219i (12 900), 259 (700), 267 (700), 276 (670), and 293 nm (290);  $\nu_{max}$  (KBr) 3 470 (OH), 1 685 (C=O), 1 510, 1 078, 1 028, and 923  $cm^{-1}$ ;  $\tau$  ( $CD_3$ -SO- $CD_3$ ), 2.62 (1 H, d,  $J$  6.5 Hz, 15-H), 3.02 (1 H, d,  $J$  6.5 Hz, 16-H), 3.08 (1 H, s, 18-H), 3.97 (1 H, d,  $J$  7 Hz, 26-OH), 3.23 (1 H, ddd,  $J$  10, 5, 3 Hz, 3-H), 4.30 (1 H, d,  $J$  10 Hz, 2-H), 5.14 (1 H,



SCHEME 1 Biogenetic connections among *Nicandra* extractives

2.61 (1 H, d,  $J$  7 Hz, 15-H), 2.98 (1 H, d,  $J$  7 Hz, 16-H), 3.03 (1 H, s, 18-H), 3.33 (1 H, ddd,  $J$  10.5, 5, 3 Hz), 4.30 (1 H, d,  $J$  10.5 Hz), 5.35 (1 H, m, 22-H), 5.5 (s, OH), 6.00 (1 H, d,  $J$  4 Hz, 7-H), 6.85 (1 H, d,  $J$  4 Hz, 6-H), 7.97 (3 H, s, 25- $H_3$ ), 8.80 (3 H, d,  $J$  7 Hz, 21- $H_3$ ), and 8.85 (3 H, s, 19- $H_3$ );  $[M]^{25}_{286\text{ nm}} + 5\ 670^\circ$  (min.),  $[M]^{25}_{292\text{ nm}} + 5\ 800^\circ$  (max.), and  $[M]^{25}_{361\text{ nm}} 2\ 270^\circ$  (min.).

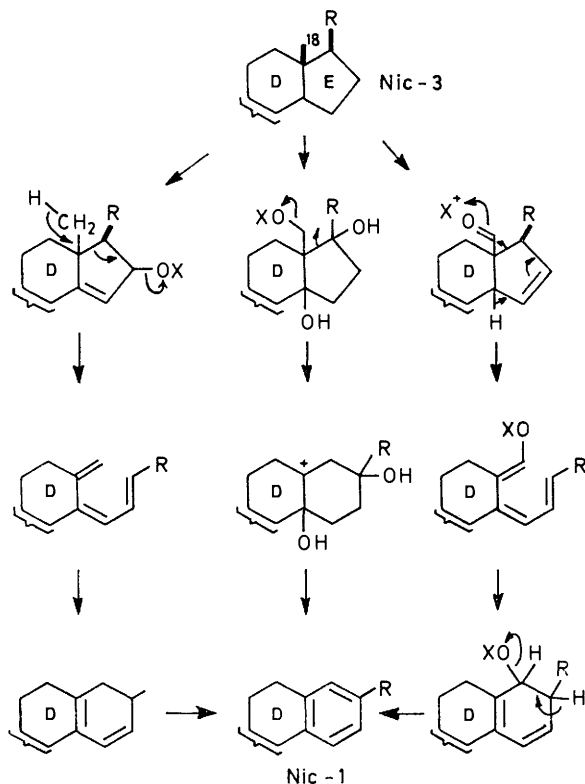
*Nic-1* [6 $\alpha$ ,7 $\alpha$ :22,26:24,25-triepoxy-5,26-dihydroxy-17(13  $\rightarrow$  18)abeo-5 $\alpha$ -ergosta-2,13,15,17-tetraen-1-one] (4) crystallised from benzene-chloroform, m.p. 117 °C, (precipitation from ethanol by light petroleum gave m.p.

d,  $J$  7 Hz, 26-H), 5.48 (1 H, s, 5-OH), 8.73 (3 H, s, 19- $H_3$ ), 8.76 (3 H, d,  $J$  7 Hz, 21- $H_3$ ), 8.86 (6 H, 27- $H_3$ , 28- $H_3$ );  $[M]^{25}_{272\text{ nm}} + 5\ 900^\circ$  (min.),  $[M]^{25}_{307\text{ nm}} + 6\ 800^\circ$  (max.), and  $[M]^{25}_{364\text{ nm}} 2\ 700^\circ$  (min.).

*Crystallographic Analysis of Nic-10.*—Nic-10 was crystallised from ethanol. For intensity measurement a crystal of dimensions ca. 0.6  $\times$  0.4  $\times$  0.4 mm was mounted about the  $a$  axis on a Hilger and Watts four-circle diffractometer. The experimental procedures described in the previous paper were adopted. Intensity data were collected with Cu- $K_\alpha$  radiation for  $2\theta \leq 154^\circ$  by use of an  $\omega$ – $2\theta$  scan.

1 610 Independent observed reflections, having  $I > 3\sigma(I)$  were used in the structure refinement. No absorption corrections were made. Data reduction and subsequent crystallographic calculations were performed by use of the 'X-Ray 70' system of programs. Normalised structure factors were calculated using the SAP programs of National Research Council (Ottawa). Atomic scattering factors were taken from ref. 5.

**Crystal data.**  $C_{21}H_{22}O_4$ ,  $M = 338.386$ . Orthorhombic,  $a = 6.448 \pm 0.002$ ,  $b = 9.589 \pm 0.002$ ,  $c = 27.838 \pm 0.005$  Å,  $U = 1721.2$  Å<sup>3</sup>,  $Z = 4$ ,  $D_c = 1.31$ ,  $F(000) = 720$ . Space group  $P2_12_12_1$  uniquely from systematic absences. Cu- $K_\alpha$  radiation,  $\lambda = 1.54178$  Å;  $\mu(\text{Cu-}K_\alpha) = 7.35$  cm<sup>-1</sup>.



SCHEME 2 Possible routes to ring D aromatisation at different levels of oxidation of C-18

The structure was determined by direct methods using the MULTAN program,<sup>6</sup> as described previously to yield a set of phases with a figure of merit of 1.059 3. A subsequent *E* map based on these phases revealed the positions of all 25 non-hydrogen atoms within the top 30 peaks on the map. Oxygen atoms were assigned on chemical grounds and verified in the subsequent refinement.

Initially, four cycles of block-diagonal least-squares refinement of atomic positions and isotropic temperature factors were carried out with all the data and unit weights. After the fourth cycle  $R$  was 0.127 and in subsequent refinement the atomic temperature factors were allowed to vary anisotropically. Two further cycles reduced  $R$  to 0.097. A difference-Fourier synthesis was then calculated which revealed the approximate positions of all hydrogen atoms which were then included in the subsequent refine-

ment with isotropic temperature factors: four further cycles reduced  $R$  to 0.071. A weighting scheme was then adopted of the form  $w = 1$  for  $|F_o| \leq 10.0$  and  $w = (10.0/F_o)^2$  for  $|F_o| > 10.0$ . Eight more cycles of least-squares refinement now reduced  $R$  to 0.057. Finally four further cycles of full-matrix least-squares refinement lowered  $R$  to 0.055 after a total of 22 cycles; the largest parameter shifts were then of the order  $0.5\sigma$ . A final difference map showed no peaks or depressions  $> 0.25$  eÅ<sup>-3</sup>. Final atomic coordinates are listed in the Table. Observed and calculated

Atomic co-ordinates, with their standard deviations in parentheses

Atom	<i>x</i>	<i>y</i>	<i>z</i>
C(1)	-0.2481(6)	0.2883(4)	0.3255(1)
C(2)	-0.2709(8)	0.2737(5)	0.2727(2)
C(3)	-0.1306(9)	0.3207(5)	0.2426(1)
C(4)	0.0606(8)	0.3939(4)	0.2580(1)
C(5)	0.0404(6)	0.4468(3)	0.3097(1)
C(6)	0.2431(7)	0.5110(4)	0.3261(1)
C(7)	0.3108(6)	0.5033(4)	0.3764(1)
C(8)	0.1893(5)	0.4217(3)	0.4125(1)
C(9)	-0.0301(5)	0.3804(3)	0.3960(1)
C(10)	-0.0316(5)	0.3270(3)	0.3436(1)
C(11)	-0.1073(6)	0.2766(4)	0.4343(1)
C(12)	-0.1424(7)	0.3517(5)	0.4815(1)
C(13)	0.0276(6)	0.4531(4)	0.4946(1)
C(14)	0.1821(5)	0.4901(3)	0.4620(1)
C(15)	0.3304(6)	0.5875(4)	0.4761(1)
C(16)	0.3279(7)	0.6459(4)	0.5215(1)
C(17)	0.1772(6)	0.6071(4)	0.5544(1)
C(18)	0.0289(6)	0.5111(4)	0.5403(1)
C(19)	0.1018(6)	0.1949(3)	0.3380(1)
C(20)	0.1790(8)	0.6682(4)	0.6039(1)
C(21)	0.0500(10)	0.6075(5)	0.6423(1)
O(1)	-0.3935(5)	0.2637(4)	0.3515(1)
O(5)	-0.1188(5)	0.5509(3)	0.3082(1)
O(6)	0.2174(5)	0.6296(2)	0.3583(1)
O(20)	0.2818(8)	0.7727(5)	0.6110(1)
H(2)	-0.389(8)	0.229(6)	0.261(2)
H(3)	-0.165(7)	0.306(4)	0.209(2)
H(4a)	0.187(7)	0.333(5)	0.255(1)
H(4b)	0.081(7)	0.478(5)	0.238(2)
H(6)	0.362(7)	0.532(5)	0.301(2)
H(7)	0.464(5)	0.518(3)	0.383(1)
H(8)	0.274(6)	0.337(4)	0.415(1)
H(9)	-0.121(6)	0.460(4)	0.398(1)
H(11a)	-0.226(7)	0.239(5)	0.427(1)
H(11b)	0.017(7)	0.193(5)	0.436(1)
H(12a)	-0.162(8)	0.289(6)	0.509(2)
H(12b)	-0.291(9)	0.417(5)	0.483(2)
H(15)	0.436(6)	0.616(4)	0.455(1)
H(16)	0.435(7)	0.717(5)	0.531(2)
H(18)	-0.075(5)	0.487(3)	0.563(1)
H(19a)	0.2381(8)	0.211(5)	0.342(2)
H(19b)	0.042(9)	0.123(6)	0.360(2)
H(19c)	0.094(8)	0.157(5)	0.304(2)
H(21a)	-0.110(8)	0.631(6)	0.631(2)
H(21b)	0.099(17)	0.660(11)	0.676(3)
H(21c)	-0.010(21)	0.524(14)	0.651(4)
H(05)	-0.116(6)	0.584(4)	0.333(1)

structure factors, and temperature factors are listed in Supplementary Publication No. SUP 21581 (12 pp., 1 microfiche).\*

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\* 'International Tables for Crystallography,' Kynoch Press, Birmingham, 1962.

\* G. Germain, P. Main, and M. M. Woolfson, *Acta Cryst.*, 1971, **A27**, 368.

\* See Notice to Authors No. 7 in *J.C.S. Perkin I*, 1974, Index Issue.