

MOLECULAR STRUCTURE AND ABSOLUTE CONFIGURATION OF  
2,3-DIDEHYDRO-1 $\beta$ ,10 $\beta$ -EPOXYPSILOSTACHYIN B (ISOPAULITIN)

J. GONZALO RODRIGUEZ\*

*Departamento de Química, C1, Facultad de Ciencias, Universidad Autónoma de Madrid,  
Canto Blanco, 28049 Madrid, Spain*

and AUREA PERALES

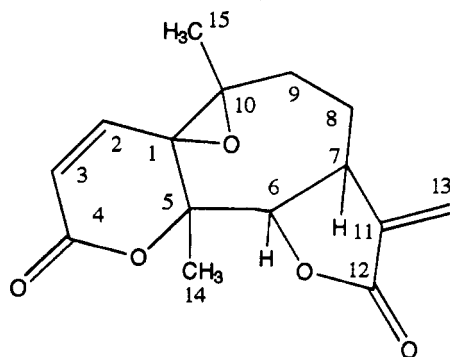
*Departamento de Cristalografía, Instituto Rocasolano, Serrano 119, 28996 Madrid, Spain*

**ABSTRACT.**—The molecular structure, conformational analysis, and absolute configuration of isopaulitin (**1**), a sesquiterpene lactone, have been determined by single-crystal X-ray diffraction analysis. The correlation between the position and stereochemistry of the  $\gamma$ -lactone fusion to the cycloheptane ring and the sign of the Cotton effect of the  $n\text{-}\pi^*$  transition of the  $\text{C}=\text{C}=\text{O}$  chromophore in **1** has been analyzed.

Isopaulitin (**1**) has been isolated from leaves of *Ambrosia cumanensis* HBK. (Compositae) (EtOH extract), a medicinal plant known as "altamisa" in El Salvador, where it was collected, and which has antirheumatic, vermifuge, and antitumor properties (1). An X-ray diffraction study of **1** was undertaken to determine its absolute configuration, which will be used as reference data to compare the stereochemistry in related psilostachylinolides such as altamisin, paulitin, and lagosin, other sesquiterpene dilactones isolated from *A. cumanensis* (1-4).

Isopaulitin (**1**) [mp 147-150°,  $[\alpha]_D^{20}$  -20° (CHCl<sub>3</sub>)] has a molecular formula of C<sub>15</sub>H<sub>16</sub>O<sub>5</sub>, as determined by elemental analysis and mass spectroscopy. The compound showed a uv band consistent with the presence of an  $\alpha,\beta'$ -unsaturated- $\gamma$ -lactone ( $\lambda$  max 225 nm,  $\epsilon$  23500). Evi-

dence of  $\gamma$ -lactone and  $\delta$ -lactone rings in the molecule of **1** was obtained from the absorption bands in the ir spectrum at 1770 cm<sup>-1</sup> and at 1725 cm<sup>-1</sup>, respectively. The <sup>1</sup>H-nmr spectrum recorded in CDCl<sub>3</sub> provided further information about the  $\alpha,\beta'$ -unsaturated  $\gamma$ -lactone of the psilostachylinolide type (5,6), e.g., the signals at  $\delta$  4.76 (1H, d,  $J=9$  Hz) for H-6 which suggested a cis-fused five membered ring; at  $\delta$  5.54 and 6.22 as two doublets,  $J=3$  Hz for the exocyclic methylene at C-11; the AB system at  $\delta$  6.28 (1H, d,  $J=10$  Hz) and 6.39 (1H, d,  $J=10$  Hz) for the  $\delta$ -lactone conjugated olefin protons; and, finally, two tertiary methyl signals at  $\delta$  1.46 and 1.59 for the methyls at C-5 and C-10, respectively. The <sup>13</sup>C-nmr spectrum of **1** in CDCl<sub>3</sub> exhibited two signals at  $\delta$  61.9 and 70.9 for the C-1 and C-10 carbons, respectively, of the



oxacyclopropane ring. The former is normal for a  $\beta$ -carbon atom relative to the methyl group at C-5 (7).

Crystallographic analysis of **1** was carried out to determine unambiguously its absolute configuration and by inference, those of biogenetically related compounds (4,7). Crystals of **1** are orthorhombic, space group  $P2_12_12_1$ , with  $a=13.918(1)$ ,  $b=10.526(1)$ ,  $c=9.220(1)$  Å, and  $V=1350.7(2)$  Å<sup>3</sup>, using  $\text{CuK}\alpha$  radiation ( $\lambda=1.5418$  Å). The molecular formula is  $\text{C}_{15}\text{H}_{16}\text{O}_5$ , mol wt 276.29,  $D_c=1.359(2)$  gcm<sup>-3</sup>,  $Z=4$ ,  $F(000)=586$ . Correction by absorption was neglected,  $\mu=8.11$  cm<sup>-1</sup>.

Fractional atomic co-ordinates and equivalent temperature factors are given in Table 1; bond lengths and angles are given in Tables 2 and 3, respectively; and

tion, absolute configuration, and numbering scheme are shown in Figure 1.<sup>1</sup>

From the values of the torsion angles (Table 4), it may be interpreted that the cycloheptane ring conformation is closer to the  $D_2$  twist-chair than to the  $D_3$  chair form, and the asymmetric parameters (8) show that the dominant symmetry is the rotational one  $D_2$  through C-7. The C-C bond distances and angles for this ring (Tables 2 and 3) show the expected values for carbon-oxygen bonds and C-C bonds, the last ranging within 1.50 to 1.54 Å, except for C-1-C-2=1.477(3) which agrees well with a C-sp<sup>3</sup>-C-sp<sup>2</sup> bond and C-1-C-5=1.552(3), probably due to steric hindrance (9) between C-14 and the oxygen of the oxacyclopropane ring. C-2-C-3=1.317(3) shows a short double-bond value. The bond distances and angles

TABLE 1. Atomic Parameters ( $\times 10^4$ ) for **1** [ $\text{C}_{15}\text{H}_{16}\text{O}_5$ , and equivalent temperature factors ( $\text{Å}^2$ ) ( $\times 10^3$ ) with e.s.d.'s in parentheses.<sup>a</sup>]

Atom	$x/a$	$y/b$	$z/c$	Ueq
O-1	8471 (1)	8778 (1)	8678 (2)	52 (0)
O-2	9995 (1)	8979 (2)	9195 (2)	74 (0)
O-3	7192 (1)	6048 (1)	6860 (1)	52 (0)
O-4	5919 (1)	10794 (2)	10406 (2)	76 (0)
O-5	6985 (1)	10192 (1)	8735 (2)	54 (0)
C-1	7865 (1)	7099 (2)	6994 (2)	41 (0)
C-2	8853 (2)	6598 (2)	7117 (2)	50 (0)
C-3	9538 (2)	7150 (2)	7875 (2)	54 (0)
C-4	9378 (2)	8348 (2)	8620 (2)	52 (0)
C-5	7607 (1)	8112 (2)	8153 (2)	41 (0)
C-6	7046 (1)	9265 (2)	7563 (2)	42 (0)
C-7	5997 (1)	9120 (2)	7062 (2)	43 (0)
C-8	5723 (1)	7891 (2)	6323 (2)	48 (0)
C-9	6430 (2)	7544 (2)	5129 (2)	50 (0)
C-10	7329 (1)	6860 (2)	5608 (2)	46 (0)
C-11	5443 (1)	9469 (2)	8394 (2)	46 (0)
C-12	6089 (2)	10215 (2)	9310 (2)	52 (0)
C-13	4547 (2)	9240 (2)	8756 (3)	58 (1)
C-14	7162 (2)	7490 (2)	9476 (2)	52 (1)
C-15	7858 (2)	6288 (3)	4332 (3)	66 (1)

$$^a \text{Ueq} = (1/3) \sum_i \sum_j U_{ij} a_i^* a_j^* \cos(a_i a_j)$$

torsion angles are given in Table 4. The X-ray analysis confirms that **1** is a sesquiterpene lactone wherein the  $\delta$ -lactone and the  $\alpha$ -methylene- $\gamma$ -lactone rings are cis-fused to a cycloheptaneoxacyclopropane bicyclic system whose constitu-

<sup>1</sup>Hydrogen coordinates, thermal parameters, bond distances and angles, and observed and calculated structure factors have been deposited with the Cambridge Crystallographic Data Centre and can be obtained upon request from Dr. Olga Kennard, University Chemical Laboratory, 12 Union Road, Cambridge CB2 1EZ, UK.

TABLE 2. Bond Lengths (Å) for **1** [C<sub>15</sub>H<sub>16</sub>O<sub>3</sub> (e.s.d.'s in parentheses)].

O-1-C-4 .....	1.342 (3)	C-3-C-4 .....	1.453 (3)
O-1-C-5 .....	1.474 (2)	C-5-C-6 .....	1.542 (3)
O-2-C-4 .....	1.208 (3)	C-5-C-14 .....	1.517 (3)
O-3-C-1 .....	1.455 (2)	C-6-C-7 .....	1.539 (2)
O-3-C-10 .....	1.449 (2)	C-7-C-8 .....	1.511 (3)
O-4-C-12 .....	1.204 (3)	C-7-C-11 .....	1.496 (2)
O-5-C-6 .....	1.458 (3)	C-8-C-9 .....	1.521 (3)
O-5-C-12 .....	1.355 (3)	C-9-C-10 .....	1.510 (3)
O-1-C-2 .....	1.477 (3)	C-10-C-15 .....	1.513 (3)
O-1-C-5 .....	1.552 (3)	C-11-C-12 .....	1.462 (3)
C-1-C-10 .....	1.501 (2)	C-11-C-13 .....	1.313 (3)
C-2-C-3 .....	1.317 (3)		

for the oxacyclopropane ring show the geometrically expected values for a tetrasubstituted derivative: thus, C-1-C-10=1.501(2), C-1-O-3=1.455(2), and C-10-O-3=1.449(2) Å, while the angles C-1-O-3-C-10=62.3(1), O-3-C-1-C-10=58.7(1), and O-3-C-10-C-1=59.1(1) Å.

The  $\delta$ -lactone ring shows a distorted boat conformation and the conjugate system C-2=C-3-C-4=O-2 is planar, with a maximum deviation of 0.053(2) Å, as is the olefinic double bond plane C-1-C-2=C-3-C-4, with the maximum deviation of 0.011(3) Å. The dihedral angle between both planes is 3°. This conformation is observed in a similar compound (10). The torsion angles in the

cycloheptane ring show a C<sub>2</sub> axis of symmetry and the  $\Sigma_2$  calculation for the dihedral angles (8) is approximately 0°, which is in agreement with the twist-chair stable conformation for cycloheptane rings (11,12). The  $\alpha$ -methylene- $\gamma$ -lactone ring adopts an envelope conformation and is cis-fused at C-6-C-7 to the cycloheptane ring.

A number of terpenoids containing the  $\alpha,\beta$ -methylene- $\gamma$ -lactone ring have been subjected to X-ray examination and the results show that the most of these lactone rings deviate significantly from the ideal envelope conformation (10).

The  $\gamma$ -lactone ring in **1** has the C=O and the exocyclic methylene groups slightly deviating from coplanarity. More-

TABLE 3. Bond Angles (°) for **1** [C<sub>15</sub>H<sub>16</sub>O<sub>3</sub> (e.s.d.'s in parentheses)].

C-4-O-1-C-5 .....	126.4 (2)	O-5-C-6-C-5 .....	107.1 (1)
C-1-O-3-C-10 .....	62.3 (1)	O-5-C-6-C-7 .....	103.5 (1)
C-6-O-5-C-12 .....	110.8 (2)	C-5-C-6-C-7 .....	120.5 (2)
O-3-C-1-C-2 .....	109.5 (2)	C-6-C-7-C-8 .....	117.4 (2)
O-3-C-1-C-5 .....	115.6 (1)	C-6-C-7-C-11 .....	102.6 (1)
O-3-C-1-C-10 .....	58.7 (1)	C-8-C-7-C-11 .....	116.8 (2)
C-2-C-1-C-5 .....	114.1 (2)	C-7-C-8-C-9 .....	111.6 (2)
C-2-C-1-C-10 .....	117.9 (2)	C-8-C-9-C-10 .....	116.0 (2)
C-5-C-1-C-10 .....	125.9 (2)	O-3-C-10-C-1 .....	59.1 (1)
C-1-C-2-C-3 .....	123.9 (2)	O-3-C-10-C-9 .....	113.9 (2)
C-2-C-3-C-4 .....	121.5 (2)	O-3-C-10-C-15 .....	116.7 (2)
O-1-C-4-O-2 .....	117.8 (2)	C-1-C-10-C-9 .....	125.5 (2)
O-1-C-4-C-3 .....	117.1 (2)	C-1-C-10-C-15 .....	119.1 (2)
O-2-C-4-C-3 .....	125.1 (2)	C-9-C-10-C-15 .....	111.5 (2)
O-1-C-5-C-1 .....	111.4 (1)	C-7-C-11-C-12 .....	106.8 (2)
O-1-C-5-C-6 .....	98.9 (1)	C-7-C-11-C-13 .....	130.8 (2)
O-1-C-5-C-14 .....	105.9 (2)	C-12-C-11-C-13 .....	122.4 (2)
C-1-C-5-C-6 .....	114.5 (1)	O-4-C-12-O-5 .....	121.2 (2)
C-1-C-5-C-14 .....	110.6 (2)	O-4-C-12-C-11 .....	129.5 (2)
C-6-C-5-C-14 .....	114.6 (2)	O-5-C-12-C-11 .....	109.3 (2)

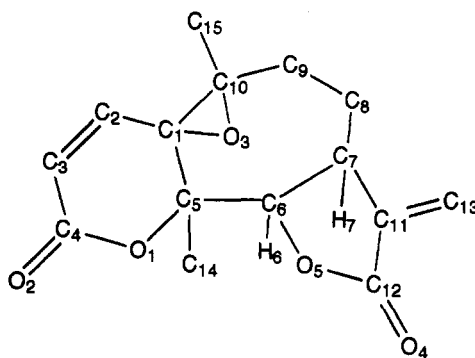
TABLE 4. Dihedral Angles ( $^{\circ}$ ) for **1** [ $C_{15}H_{16}O_5$ , (e.s.d.'s in parentheses)].

O-1-C-4-C-3-C-2.....	9.2 (3)	C-1-C-10-C-9-C-8.....	-36.2 (3)
O-1-C-5-O-1-O-3.....	154.4 (1)	C-2-C-1-O-3-C-10.....	-111.5 (2)
O-1-C-5-C-1-C-2.....	26.1 (2)	C-2-C-1-C-5-C-6.....	137.3 (2)
O-1-C-5-C-1-C-10.....	-137.0 (2)	C-2-C-1-C-5-C-14.....	-91.4 (2)
O-1-C-5-C-6-O-5.....	-53.6 (2)	C-2-C-1-C-10-C-9.....	-164.3 (2)
O-1-C-5-C-6-C-7.....	-171.4 (2)	C-2-C-1-C-10-C-15.....	-8.4 (3)
O-2-C-4-O-1-C-5.....	-174.8 (2)	C-3-C-2-C-1-C-5.....	-16.2 (3)
O-2-C-4-C-3-C-2.....	-171.2 (2)	C-3-C-2-C-1-C-10.....	148.3 (2)
O-3-C-1-C-2-C-3.....	-147.5 (2)	C-3-C-4-O-1-C-5.....	4.8 (3)
O-3-C-1-C-5-C-6.....	-94.4 (2)	C-4-O-1-C-5-C-6.....	-143.1 (2)
O-3-C-1-C-5-C-14.....	36.9 (2)	C-4-O-1-C-5-C-14.....	98.0 (2)
O-3-C-1-C-10-C-9.....	98.8 (2)	C-5-C-1-O-3-C-10.....	118.0 (2)
O-3-C-1-C-10-C-15.....	-105.4 (2)	C-5-C-1-C-10-C-9.....	-1.8 (3)
O-3-C-10-C-1-C-2.....	96.9 (2)	C-5-C-1-C-10-C-15.....	154.0 (2)
O-3-C-10-C-1-C-5.....	-100.6 (2)	C-5-C-6-O-5-C-12.....	-105.4 (2)
O-3-C-10-C-9-C-8.....	31.8 (2)	C-5-C-6-C-7-C-8.....	-35.8 (2)
O-4-C-12-O-5-C-6.....	170.4 (2)	C-5-C-6-C-7-C-11.....	93.7 (2)
O-4-C-12-C-11-C-7.....	171.8 (2)	C-6-O-5-C-12-C-11.....	-10.0 (2)
O-4-C-12-C-11-C-13.....	-6.3 (4)	C-6-C-5-C-1-C-10.....	-25.8 (2)
O-5-C-6-C-5-C-1.....	-172.1 (1)	C-6-C-7-C-8-C-9.....	-48.9 (2)
O-5-C-6-C-5-C-14.....	58.5 (2)	C-6-C-7-C-11-C-12.....	20.9 (2)
O-5-C-6-C-7-C-8.....	-155.4 (2)	C-6-C-7-C-11-C-13.....	-161.4 (2)
O-5-C-6-C-7-C-11.....	-25.9 (2)	C-7-C-6-O-5-C-12.....	23.0 (2)
O-5-C-12-C-11-C-7.....	-7.8 (2)	C-7-C-6-C-5-C-14.....	-59.2 (2)
O-5-C-12-C-11-C-13.....	174.2 (2)	C-7-C-8-C-9-C-10.....	83.5 (2)
C-1-O-3-C-10-C-9.....	-118.4 (2)	C-8-C-7-C-11-C-12.....	150.7 (2)
C-1-O-3-C-10-C-15.....	109.5 (2)	C-8-C-7-C-11-C-13.....	-31.5 (3)
C-1-C-2-C-3-C-4.....	-2.4 (4)	C-8-C-9-C-10-C-15.....	166.4 (2)
C-1-C-5-O-1-C-4.....	-22.3 (2)	C-9-C-8-C-7-C-11.....	-171.4 (2)
C-1-C-5-C-6-C-7.....	70.1 (2)	C-10-C-1-C-5-C-14.....	105.5 (2)

over, the related bond lengths C-11-C-13=1.313(3), C-12-O-4=1.204(3), C-11-C-12=1.462(3), and O-5-C-12=1.355(3) confirm the absence of conjugation through C=O and the double bond. In contrast, the C=O and the C-2=C-3 double bond groups in the  $\delta$ -lactone ring show some con-

jugation effect.

From measurements of the sign of the Cotton effect of the  $n-\pi^*$  transition of the  $\alpha$ -methylene- $\gamma$ -lactone chromophore in a number of sesquiterpene lactones (13), it has been established that, with some exceptions, the C-6-C-7 cis-fused lactones give a positive Cotton effect,

FIGURE 1. Molecule of isopaulitin-**1** showing the atomic labeling.

whereas a negative Cotton effect is found when the  $\gamma$ -lactone is trans-fused across the C-6-C-7 bond. On the other hand, the sign of the Cotton effect of the  $\alpha$ -methylene- $\gamma$ -lactone chromophore has been related (11) to the chirality of the C=C-C=O group which permits determination of the absolute configuration of the molecule. For **1** with a cis-fused C-6-C-7 lactone ring the experimentally determined Cotton effect is positive,  $\lambda$  max (EtOH) (log  $\epsilon$ ) 262 nm (1.3), which is in agreement with the above predictions. Moreover, the torsion angle of the C=C-C=O group is  $-6^\circ$ , which is paired with that of the endocyclic torsion angle about the C-6-C-7 bond ( $-26^\circ$ ) and with the latter angle about three times greater than the former. All these results are in good agreement with the correlations established.

Figure 1 shows the absolute configuration of isopaulitin [**1**] which was determined by comparing the more relevant Bijvöet pairs. The absolute configurations of the five chiral centers in isopaulitin are given by the torsion angles, being: C-1, *R*, C-5, *S*, C-6, *R*, C-7, *S*, and C-10, *S*.

## EXPERIMENTAL

**PLANT MATERIAL.**—Isopaulitin [**1**] has been isolated from an extract of the leaves of *Ambrosia cumanensis* H.B.K., a species which belongs to the tribe Heliantheae (Compositae). It is known as "altamisa" in the Republic of El Salvador, where the plant was collected (7).

**X-RAY CRYSTALLOGRAPHIC ANALYSIS OF ISOPAULITIN [1].**—Colorless crystals of **1** for X-ray analysis were obtained from slow evaporation of a dilute EtOAc solution. A crystal of 0.20×0.16×0.30 mm dimensions was used for the crystallographic study. Accurate cell dimensions were determined by least-squares analysis of setting angles of 34 reflections ( $10 < \theta < 32^\circ$ ), using graphite monochromated CuK $\alpha$  radiation automatically located and centered on a Philips PW 1100 diffractometer. Two reflections were measured every 90 reflections to ascertain crystal stability, and no significant variation was observed. All the reflections were corrected for Lorentz and polarization effects, but no absorption correction was applied. For the intensity measurement, reflections were surveyed in the range  $2\theta \leq 65$ , from the 2292 reflections measured as indepen-

dent Friedel pairs, 2080 were considered as observed, satisfying the criterion  $I \geq 2\sigma(I)$ , and were used in the subsequent calculations.

The crystal structure was partially elucidated by direct methods (15). After preliminary adjustment of the positions of the C and O atoms, the H atoms were positioned from difference Fourier maps. Thereafter, several cycles of full-matrix least-squares calculations were carried out with anisotropic thermal parameters for C and O atoms and the H atoms were included as fixed contributors, and convergence was reached at  $R=0.044$  and  $R_w=0.048$  with a weighting scheme (16) to prevent trends in  $w\Delta^2 F$  vs  $\langle F_o \rangle$  and vs  $\langle \sin\theta/\lambda \rangle$  including both  $\pm hkl$  pairs. The final difference synthesis showed no peaks exceeding  $0.40 \text{ e}\text{\AA}^{-3}$ , and the value of the parameters shift error was less than 0.01.

The absolute configuration (17) was determined by comparison of the 76 more relevant Bijvöet pairs, considering reflections with  $F_o > 10\sigma(F_o)$ , with  $\Delta F_c > 0.07$ . The following discrepancy indexes were found:  $R_1 = \sum\{[F_o(h)] - [F_o(-h)]\} - \{[F_o(h)] - [F_o(-h)]\} / N = 0.219$  (0.280 for the reversed enantiomer);  $R_2 = 1 + \sum\{[R_o - R_c] - 1 / N\} = 1.031$  (1.035 for the reversed enantiomer);  $R_3 = \sum\{[\Delta I_o - \Delta I_c] / \sum(\Delta I_o)\} = 0.916$  (1.242 for the reversed enantiomer); where  $R_o = [F_o(h)] / [F_o(-h)]$ ,  $R_c = [F_c(h)] / [F_c(-h)]$ ,  $\Delta I_o = [F_o(h)]^2 - [F_o(-h)]^2$ ,  $\Delta I_c = [F_c(h)]^2 - [F_c(-h)]^2$ .

The atomic scattering factors and the anomalous dispersion correction were taken from the literature (18).

## ACKNOWLEDGMENTS

We dedicate this paper to the memory of our colleague and friend Professor J. Borges del Castillo. We thank Professor Gavilanes, Biochemistry Department of the UCM, for the circular dichroism data.

## LITERATURE CITED

1. P.C. Stanley and S. Calderon, "Lista Preliminar de las Plantas de El Salvador," Ediciones Culturales de la Universidad de El Salvador, San Salvador, 1944.
2. J.A. Lagos, "Pólenes Anemófilos de El Salvador," Editorial Universitaria, San Salvador, 1975.
3. W. Herz and J.F. Blount, *J. Org. Chem.*, **43**, 4888 (1978), and references cited therein.
4. J. Borges, M.T. Manresa, J.L. Martín, C. Pascual, and P. Vázquez, *Tetrahedron Lett.*, 1513 (1978).
5. T.J. Mabry, H.E. Millar, H.B. Kagan, and W. Renold, *Tetrahedron*, **22**, 1128 (1966).
6. T.J. Mabry, H.E. Millar, H.B. Kagan, and W. Renold, *Tetrahedron*, **22**, 1943 (1966).
7. J. Borges, M.T. Manresa, J.L. Martín, F. Rodríguez, and P. Vázquez, *An. Quím.*, **77**, 52 (1981).

8. W.L. Duax, C.M. Weeks, and D.C. Rohrer, "Topics in Stereochemistry," Wiley-Interscience, New York, 1979, Vol. 9, pp. 229-238.
9. J. López de Lerma, S. García-Blanco, and J.G. Rodríguez, *Tetrahedron Lett.*, **21**, 1273 (1980).
10. A.F. Beechman, *Tetrahedron*, **28**, 5543 (1972).
11. J.B. Hendrickson, *J. Am. Chem. Soc.*, **83**, 4537 (1961).
12. J.B. Hendrickson, *Tetrahedron*, **19**, 1387 (1963).
13. A.T. McPhail and G.A. Sim, *Tetrahedron*, **29**, 1751 (1973).
14. W. Stocklin, T.G. Waddell, and T.A. Geissman, *Tetrahedron*, **26**, 2397 (1970).
15. P. Main, S.J. Fiske, S.E. Hull, L. Lessinger, G. Germain, J. Declerck, and M.M. Woolfson, "MULTAN 80. A System of Computer Programs for the Automatic Solution of Crystal Structures from X-Ray Diffraction Data," University of York, England, 1980.
16. M. Martinez-Ripoll and F.H. Cano, "PESOS Program," Instituto Rocasolano, CSIC, 28006-Madrid, Spain, 1975.
17. M. Martinez-Ripoll and J. Fayos, "CONFAB Program," Instituto Rocasolano, CSIC, 28006 Madrid, Spain, 1977.
18. "International Tables for X-Ray Crystallography," Knoch Press, Birmingham, UK, 1974, Vol. IV.

Received 12 May 1994