Studies on the Ionophorous Antibiotics. Part 14.[1] Crystal and Molecular Structure of the Thallium Salt of Carriomycin

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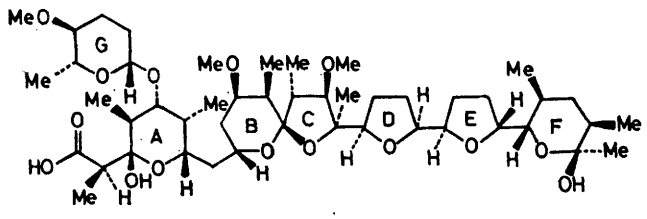
Carriomycin (I) is a new polyether antibiotic produced by *Streptomyces hygroscopicus*. The crystal and molecular structure of its thallium salt has been determined from three-dimensional *X*-ray analysis by the heavy-atom method from diffractometer data. Crystals are monoclinic, space group $P2_1$. a = 15.416(4), b = 12.325(5), c = 14.442(5) Å, β = 105.42(4)°, Z = 2. The structure was refined by block-diagonal least-squares methods to R 0.60 for 3 763 reflections. The carriomycin molecule contains seven ring systems: three five-membered rings take an envelope conformation, while the remaining four six-membered rings adopt a chair conformation. The molecule wraps around the thallium ion.

CARRIOMYCIN,[2] $C_{47}H_{80}O_{15}$ (I) is a new antibiotic isolated from culture broth of *Streptomyces hygroscopicus* T–42082 and shows inhibitory activity against gram-positive bacteria, mycobacteria, some species of fungi, yeasts, and mycoplasma. It is also effective in the treatment of coccidial infections in poultry.



(I)

In view of both its biological activity and physicochemical properties, carriomycin was thought to be a new naturally occurring ionophore of the polyether family.[3,4] It is remarkably less toxic (LD_{50} > 125 mg kg^{-1}, in mice, i.p.) than the other members of this family, although it shows rather stronger antibacterial activity.

As part of a series of our investigations on the ionophorous antibiotics,[5–9] the crystal structure of carriomycin has been determined from its corresponding thallium complex. A preliminary communication has appeared.[10]

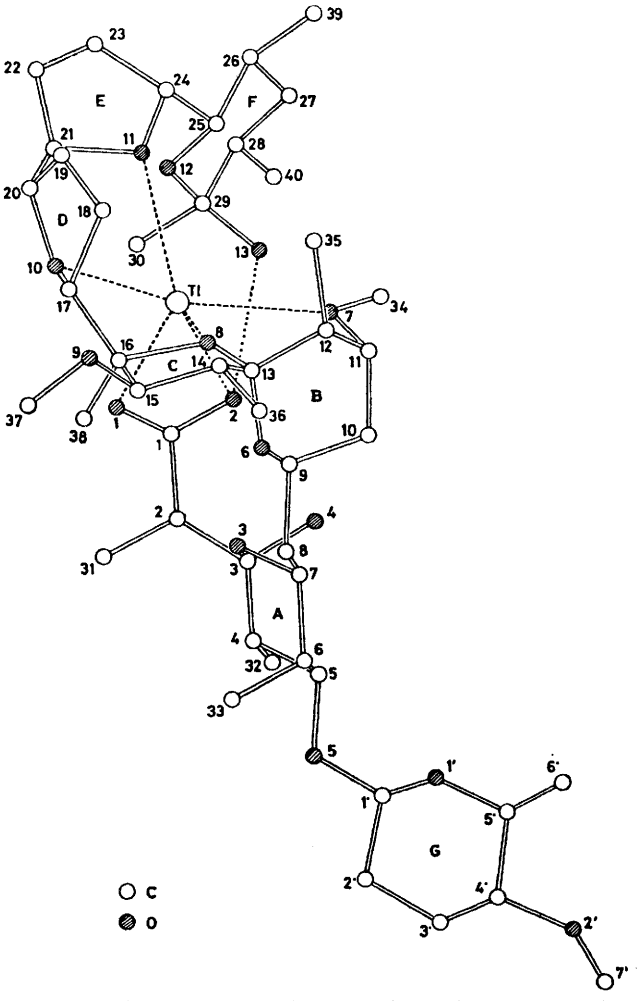
EXPERIMENTAL

Instruments.—M.p.s were measured with a Yanagimoto apparatus. ^{1}H N.m.r. spectra were taken with a JNM 4H 100 spectrometer, ^{13}C n.m.r. spectra with JEOL FX-100 spectrometer. *X*-Ray diffraction intensities were measured on a Philips automated four-circle diffractometer and calculations were carried out on our HITAC 8700/8800.

Preparation of the Thallium Salt of Carriomycin.—To a solution of the sodium salt of carriomycin (100 mg) in dioxan (20 ml) was added dropwise an aqueous solution of thallium acetate (30 mg) in redistilled water (5 ml) and the mixture placed in the dark for a week.

Crystals (m.p., with browning, 178.5—180.5 °C), grown by evaporation from a methanol solution, were needles, elongated along the *b* axis. The molecular formula, $C_{47}H_{79}O_{15}Tl$, was confirmed by elementary analyses as well as by comparison of the ^{13}C n.m.r. spectra of the sodium salt with that of the free acid.[2]

The ^{1}H n.m.r. data for the thallium salt revealed that the molecule contains three methoxy-groups at δ 3.35—3.50 p.p.m. and a number of methyl groups attached to second-



Projection of the structure of the thallium salt (I) viewed along the *b* axis

ary or tertiary carbons which overlap each other in the range δ 0.80 to δ 1.70.

Crystal Data.—$C_{47}H_{79}O_{15}Tl$, M = 1 088.5. Monoclinic, a = 15.416(4), b = 12.325(5), c = 14.442(5), Å, β =

105.42(4)°, U = 2 639.3 Å^3, D_m = 1.37, Z = 2, D_c = 1.38 g cm^{-3}, Mo-K_α radiation, λ = 0.7107 Å; μ(Mo-K_α) = 31.5 cm^{-1}. Space group $P2_1$.

TABLE 1

Atom parameters

Tl	1 988(0.5)	5 000(1.9)	3 710(0.5)
C(1)	2 333(13)	3 341(17)	5 402(12)
C(2)	2 683(13)	3 027(17)	6 502(12)
C(3)	3 590(11)	3 533(14)	6 970(11)
C(4)	3 946(12)	3 301(14)	8 085(11)
C(5)	4 807(12)	4 009(14)	8 516(10)
C(6)	4 593(12)	5 240(13)	8 333(11)
C(7)	4 254(12)	5 342(12)	7 215(11)
C(8)	4 021(12)	6 501(13)	6 912(12)
C(9)	3 787(11)	6 630(13)	5 788(11)
C(10)	4 595(11)	6 481(14)	5 413(12)
C(11)	4 319(11)	6 655(15)	4 324(12)
C(12)	3 774(12)	7 724(16)	4 064(13)
C(13)	3 046(10)	7 913(13)	4 585(12)
C(14)	2 668(11)	9 062(14)	4 530(14)
C(15)	1 811(12)	8 861(14)	4 851(14)
C(16)	1 497(11)	7 705(14)	4 462(14)
C(17)	700(11)	7 710(15)	3 554(14)
C(18)	854(12)	8 260(17)	2 656(14)
C(19)	155(15)	7 677(19)	1 793(17)
C(20)	−71(13)	6 611(20)	2 252(17)
C(21)	166(13)	5 569(17)	1 811(14)
C(22)	−371(17)	5 433(19)	746(15)
C(23)	206(16)	4 617(17)	412(13)
C(24)	1 178(11)	4 965(30)	983(12)
C(25)	1 843(15)	3 971(14)	1 406(17)
C(26)	2 020(16)	3 306(18)	563(13)
C(27)	2 593(17)	2 342(19)	1 049(15)
C(28)	2 143(16)	1 684(16)	1 666(15)
C(29)	1 954(12)	2 428(15)	2 433(11)
C(30)	1 331(14)	1 876(18)	2 959(16)
C(31)	1 952(13)	3 357(27)	6 990(13)
C(32)	4 210(16)	2 084(16)	8 266(16)
C(33)	3 884(15)	5 714(17)	8 834(14)
C(34)	4 281(13)	4 918(31)	3 617(19)
C(35)	3 356(14)	7 770(19)	2 911(13)
C(36)	3 266(11)	9 969(31)	5 105(13)
C(37)	605(18)	9 902(49)	5 050(25)
C(38)	1 295(14)	6 995(16)	5 262(15)
C(39)	2 610(28)	3 913(28)	−3(23)
C(40)	2 676(22)	651(21)	2 094(18)
C(1′)	5 906(12)	4 080(16)	10 058(12)
C(2′)	5 987(14)	4 058(17)	11 145(12)
C(3′)	6 976(15)	4 211(19)	11 700(13)
C(4′)	7 555(13)	3 415(19)	11 368(13)
C(5′)	7 385(14)	3 453(19)	10 256(12)
C(6′)	7 918(18)	2 605(31)	9 875(18)
C(7′)	9 036(20)	3 030(32)	12 463(30)
O(1)	1 619(9)	3 681(14)	5 072(8)
O(2)	2 941(9)	3 189(12)	4 960(9)
O(3)	3 450(7)	4 712(9)	6 849(7)
O(4)	4 256(8)	3 217(9)	6 520(8)
O(5)	5 010(8)	3 799(10)	9 560(8)
O(6)	3 421(7)	7 693(9)	5 586(8)
O(7)	3 748(8)	5 737(10)	3 829(9)
O(8)	2 296(7)	7 245(9)	4 225(8)
O(9)	1 161(8)	9 683(11)	4 451(12)
O(10)	472(8)	6 613(10)	3 258(9)
O(11)	1 092(9)	5 602(11)	1 791(9)
O(12)	1 440(9)	3 338(10)	1 979(8)
O(13)	2 746(8)	2 781(11)	3 034(9)
O(1′)	6 461(8)	3 260(11)	9 821(8)
O(2′)	8 514(11)	3 653(15)	11 778(11)

Crystals *ca.* 0.3 × 0.4 × 0.4 mm were used for data collection. Since the salt deteriorated during exposure to X-rays, three crystals were used for intensity measurement. Intensity data were collected on an automated four-circle diffractometer up to 2θ 55° by use of graphite-monochromated Mo-K_α radiation (λ = 0.7107 Å), and the θ—2θ scan technique. In this way 3 763 independent reflections were recorded and used for the structure determination; intensities were corrected for the effects of crystal deterioration

TABLE 2

Bond lengths (Å), with estimated standard deviations in parentheses

C(1)–C(2)	1.58(3)	C(17)–C(18)	1.54(3)
C(1)–O(1)	1.16(3)	C(17)–O(10)	1.43(2)
C(1)–O(2)	1.28(3)	C(18)–C(19)	1.59(3)
C(2)–C(3)	1.52(3)	C(19)–C(20)	1.55(4)
C(2)–C(31)	1.53(4)	C(20)–C(21)	1.52(3)
C(3)–C(4)	1.58(3)	C(20)–O(10)	1.47(3)
C(3)–O(3)	1.47(2)	C(21)–C(22)	1.55(3)
C(3)–O(4)	1.41(2)	C(21)–O(11)	1.44(2)
C(4)–C(5)	1.57(3)	C(22)–C(23)	1.50(4)
C(4)–C(32)	1.56(3)	C(23)–C(24)	1.57(4)
C(5)–C(6)	1.56(3)	C(24)–C(25)	1.61(4)
C(5)–O(5)	1.48(2)	C(24)–O(11)	1.44(4)
C(6)–C(7)	1.57(3)	C(25)–C(26)	1.55(4)
C(6)–C(33)	1.58(3)	C(25)–O(12)	1.40(3)
C(7)–C(8)	1.51(3)	C(26)–C(27)	1.53(4)
C(7)–O(3)	1.44(2)	C(26)–C(39)	1.57(5)
C(8)–C(9)	1.57(2)	C(27)–C(28)	1.50(4)
C(9)–C(10)	1.50(2)	C(28)–C(29)	1.53(3)
C(9)–O(6)	1.43(2)	C(28)–C(40)	1.55(4)
C(10)–C(11)	1.53(3)	C(29)–C(30)	1.53(3)
C(11)–C(12)	1.56(3)	C(29)–O(12)	1.43(2)
C(11)–O(7)	1.49(2)	C(29)–O(13)	1.37(2)
C(12)–C(13)	1.53(3)	C(34)–O(7)	1.39(4)
C(12)–C(35)	1.62(3)	C(37)–O(9)	1.40(6)
C(13)–C(14)	1.53(3)	C(1′)–C(2′)	1.54(3)
C(13)–O(6)	1.43(2)	C(1′)–O(5)	1.42(2)
C(13)–O(8)	1.40(2)	C(1′)–O(1′)	1.42(2)
C(14)–C(15)	1.53(3)	C(2′)–C(3′)	1.53(3)
C(14)–C(36)	1.54(4)	C(3′)–C(4′)	1.49(3)
C(15)–C(16)	1.56(3)	C(4′)–C(5′)	1.56(3)
C(15)–O(9)	1.43(3)	C(4′)–O(2′)	1.47(3)
C(16)–C(17)	1.54(3)	C(5′)–C(6′)	1.52(5)
C(16)–C(38)	1.55(3)	C(5′)–O(1′)	1.42(3)
C(16)–O(8)	1.48(2)	C(7′)–O(2′)	1.34(5)

TABLE 3

Bond angles (°), with estimated standard deviations in parentheses

C(2)–C(1)–O(1)	122.4 (2.0)	C(17)–C(16)–C(38)	111.4 (1.6)
C(2)–C(1)–O(2)	111.4 (1.8)	C(17)–C(16)–O(8)	109.3 (1.5)
O(1)–C(1)–O(2)	126.2 (2.1)	C(38)–C(16)–O(8)	107.6 (1.5)
C(1)–C(2)–C(3)	112.0 (1.6)	C(16)–C(17)–C(18)	116.9 (1.6)
C(1)–C(2)–C(31)	107.4 (1.9)	C(16)–C(17)–O(10)	109.1 (1.5)
C(3)–C(2)–C(31)	112.9 (1.9)	C(18)–C(17)–O(10)	104.2 (1.5)
C(2)–C(3)–C(4)	114.1 (1.5)	C(17)–C(18)–C(19)	103.8 (1.7)
C(2)–C(3)–O(3)	105.3 (1.4)	C(18)–C(19)–C(20)	103.3 (1.8)
C(2)–C(3)–O(4)	112.3 (1.5)	C(19)–C(20)–C(21)	115.4 (2.0)
C(4)–C(3)–O(3)	107.4 (1.3)	C(19)–C(20)–O(10)	106.7 (1.8)
C(4)–C(3)–O(4)	108.8 (1.4)	C(21)–C(20)–O(10)	106.1 (1.8)
O(3)–C(3)–O(4)	108.6 (1.3)	C(20)–C(21)–C(22)	112.5 (1.8)
C(3)–C(4)–C(5)	109.3 (1.4)	C(20)–C(21)–O(11)	109.6 (1.7)
C(3)–C(4)–C(32)	110.4 (1.6)	C(22)–C(21)–O(11)	104.8 (1.7)
C(5)–C(4)–C(32)	108.2 (1.6)	C(21)–C(22)–C(23)	100.7 (1.9)
C(4)–C(5)–C(6)	110.8 (1.4)	C(22)–C(23)–C(24)	102.3 (2.2)
C(4)–C(5)–O(5)	103.5 (1.4)	C(23)–C(24)–C(25)	114.5 (2.6)
C(6)–C(5)–O(5)	108.7 (1.4)	C(23)–C(24)–O(11)	107.1 (2.5)
C(5)–C(6)–C(7)	104.5 (1.4)	C(25)–C(24)–O(11)	107.1 (2.5)
C(5)–C(6)–C(33)	115.1 (1.5)	C(24)–C(25)–C(26)	109.4 (2.2)
C(7)–C(6)–C(33)	111.2 (1.5)	C(24)–C(25)–O(12)	108.2 (2.1)
C(6)–C(7)–C(8)	111.3 (1.5)	C(26)–C(25)–O(12)	111.6 (1.9)
C(6)–C(7)–O(3)	110.8 (1.4)	C(25)–C(26)–C(27)	104.5 (2.0)
C(8)–C(7)–O(3)	106.8 (1.4)	C(25)–C(26)–C(39)	113.9 (2.4)
C(7)–C(8)–C(9)	111.5 (1.4)	C(27)–C(26)–C(39)	105.4 (2.4)
C(8)–C(9)–C(10)	112.1 (1.4)	C(26)–C(27)–C(28)	112.9 (2.1)
C(8)–C(9)–O(6)	105.9 (1.3)	C(27)–C(28)–C(29)	108.3(2.0)
C(10)–C(9)–O(6)	111.1 (1.3)	C(27)–C(28)–C(40)	113.6 (2.2)
C(9)–C(10)–C(11)	108.9 (1.4)	C(29)–C(28)–C(40)	112.8 (2.1)
C(10)–C(11)–C(12)	110.6 (1.5)	C(28)–C(29)–C(30)	111.0 (1.7)
C(10)–C(11)–O(7)	110.7 (1.4)	C(28)–C(29)–O(12)	109.2 (1.6)
C(12)–C(11)–O(7)	108.3 (1.4)	C(28)–C(29)–O(13)	109.9 (1.6)
C(11)–C(12)–C(13)	115.6 (1.6)	C(30)–C(29)–O(12)	103.4 (1.5)

TABLE 3 (*Continued*)

C(11)–C(12)–C(35)	108.6 (1.6)	C(30)–C(29)–O(13)	113.5 (1.6)
C(13)–C(12)–C(35)	111.0 (1.6)	O(12)–C(29)–O(13)	109.5 (1.4)
C(12)–C(13)–C(14)	115.9 (1.5)	C(2′)–C(1′)–O(5)	108.4 (1.6)
C(12)–C(13)–O(6)	108.4 (1.4)	C(2′)–C(1′)–O(1′)	110.0 (1.6)
C(12)–C(13)–O(8)	111.7 (1.4)	O(5)–C(1′)–O(1′)	105.7 (1.5)
C(14)–C(13)–O(6)	105.9 (1.4)	C(1′)–C(2′)–C(3′)	109.3 (1.7)
C(14)–C(13)–O(8)	105.1 (1.4)	C(2′)–C(3′)–C(4′)	110.4 (1.9)
O(6)–C(13)–O(8)	109.5 (1.3)	C(3′)–C(4′)–C(5′)	111.2 (1.9)
C(13)–C(14)–C(15)	100.6 (1.5)	C(3′)–C(4′)–O(2′)	111.3 (1.9)
C(13)–C(14)–C(36)	118.6 (2.0)	C(5′)–C(4′)–O(2′)	106.5 (1.8)
C(15)–C(14)–C(36)	113.2 (2.0)	C(4′)–C(5′)–C(6′)	113.2 (2.2)
C(14)–C(15)–C(16)	104.4 (1.6)	C(4′)–C(5′)–O(1′)	109.0 (1.8)
C(14)–C(15)–O(9)	109.3 (1.6)	C(6′)–C(5′)–O(1′)	107.5 (2.1)
C(16)–C(15)–O(9)	112.5 (1.6)	C(3)–O(3)–C(7)	113.8 (1.2)
C(15)–C(16)–C(17)	113.8 (1.6)	C(5)–O(5)–C(1′)	112.8 (1.4)
C(15)–C(16)–C(38)	110.7 (1.6)	C(9)–O(6)–C(13)	114.4 (1.2)
C(15)–C(16)–O(8)	103.6 (1.4)	C(11)–O(7)–C(34)	110.4 (1.9)
C(13)–O(8)–C(16)	110.2 (1.3)		
C(15)–O(9)–C(37)	111.5 (2.8)		
C(17)–O(10)–C(20)	109.0 (1.5)		
C(21)–O(11)–C(24)	107.9 (1.9)		
C(25)–O(12)–C(29)	115.5 (1.5)		
C(1′)–O(1′)–C(5′)	111.8 (1.5)		
C(4′)–O(2′)–C(7′)	121.7 (2.5)		

by comparison with standard reflections, and for Lorentz and polarization factors, but not for absorption.

The structure was solved by the heavy-atom method. Refinement was carried out by a block-diagonal least-squares program with anisotropic thermal parameters. Convergence was attained at R 0.060 for the 3 763 reflections. At the final stage of the refinement all the shifts of the atomic co-ordinates were smaller than one seventh of their standard deviations. Final atom parameters are presented in Table 1.

The absolute configuration was determined by the use of the anomalous scattering effect of the thallium atom for Cu-K_α radiation. An equi-inclination Weissenberg photograph was taken of the first and second layers around the a axis with Cu-K_α radiation. Differences between the intensities of some reflections were clearly discernible. All differences were confirmed by measuring the intensities of the same reflections on a diffractometer by use of Mo-K_α radiation. The set of atom co-ordinates given in Table 1 correctly represents the absolute configuration of the molecule shown in (I) when referred to the right-handed set of axes. Final observed and calculated structure factors, thermal parameters, and details of intensities used in the determination of the absolute configuration are listed in Supplementary Publication No. SUP 22382 (27 pp., 1 microfiche).*

RESULTS AND DISCUSSION

The structure and the absolute configuration of carriomycin is established to be as depicted in (I) and a projection of the structure along the b axis in the Figure. Bond lengths and angles are given in Tables 2 and 3, respectively. Carriomycin is thus a new member of the class of glycoside polyether antibiotics closely related to septamycin [11] and A–204 A.[12] Carriomycin contains seven ring systems: three (C, D, and E) are five-membered and have an envelope conformation, while the remaining four are six-membered (A, B, F, and G) and adopt a chair conformation.

Like nigericin,[13] grisorixin,[14] and lonomycin,[7] it has a thirty carbon-atom backbone as fundamental framework; however, an additional sugar unit (ring G) which occurs identically in dianemycin [15] and septamycin,[11] is attached at C(5) of ring A through a glycosidic linkage. With respect to the stereochemistry of ring A, C(32) and C(5) of the glycosyl group are equatorial, while the C(33) methyl group and the C(4) hydroxy-group are axial. In ring B, the C(35) methyl is equatorial, while the O(7) methoxy-group is axial. In ring F, however, the C(30), C(39), and C(40) methyl groups are equatorial, while the O(13) hydroxy-group is axial. In ring G, both the C(6′) methyl and the O(2′) methoxy-group are equatorial.

The entire molecule takes a circular conformation (see Figure) and the thallium(I) ion is located in the central cavity formed by ' fastening ' the ends of the chain by a hydrogen bond between the carboxylate oxygen atoms of ring A and the hydroxy-group on ring F. The thallium atom is co-ordinated to six oxygen atoms with Tl–O distances ranging from 2.72(6) to 3.00(4) Å. As a result, most of the oxygen atoms are located in the interior of the molecule, rendering the exterior surface hydrophobic, as is the case in the structures of other metal-containing polyethers so far elucidated. Like lonomycin,[7] carriomycin contains two hemiacetal groups on rings A and F, which take up an axial conformation and the spiroacetal ring system formed by rings B and C which also occurs in a number of members of the polyether family including monensin,[16] nigericin,[13] and lonomycin.[7]

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* See Notice to Authors No. 7 in *J.C.S. Perkin II*, 1978, Index issue.

REFERENCES

[1] Part 13, N. Ôtake, H. Seto, and M. Koenuma, *Agric. and Biol. Chem. (Japan)*, 1978, **42**, 1879.
[2] A. Imada, Y. Nozaki, T. Hasegawa, E. Mizuta, S. Igarashi, and M. Yoneda, *J. Antibiot.*, 1977, **30**.
[3] J. W. Westley, *Ann. Reports, Medicin. Chem.*, 1975, **10**, 246.
[4] N. Ôtake, *Farmashia*, 1977, **13**, 367.
[5] H. Kinashi, N. Ôtake, H. Yonehara, S. Sato, and Y. Saito, *Acta Cryst.*, 1975, **B31**, 2411.
[6] N. Ôtake, M. Koenuma, H. Kinashi, S. Sato, and Y. Saito, *J.C.S. Chem. Comm.*, 1975, 92.
[7] N. Ôtake, M. Koenuma, H. Miyamae, S. Sato, and Y. Saito, *J.C.S., Perkin II*, 1977, 494.
[8] H. Seto, T. Yahagi, Y. Miyazaki, and N. Ôtake, 1977, *J. Antibiot.*, **30**, 530.
[9] D. E. Dorman, J. W. Pasohal, W. M. Nakatsukasa, L. L. Huckstep, and N. Neuss, *Helv. Chim. Acta*, 1976, **59**, 2625.
[10] N. Ôtake, H. Nakayama, H. Miyamae, S. Sato, and Y. Saito, *J.C.S. Chem. Comm.*, 1977, 590.
[11] T. J. Petcher, and H. P. Weber, *J.C.S. Chem. Comm.*, 1974, 697.
[12] N. D. Jones, M. O. Chaney, J. W. Chamberlin, R. L. Hamill, and S. Chen, *J. Amer. Chem. Soc.*, 1973, **95**, 3399.
[13] L. K. Steinrauf, M. Pinkerton, and J. W. Chamberlin, *Biochem. Biophys. Res. Comm.*, 1968, **33**, 29.
[14] P. Gachon, A. Kergomard, H. Veschambre, C. Esteve, and T. Staron. *Chem. Comm.*, 1970, 1421.
[15] E. W. Czerwinski, and L. K. Steinrauf, *Biochem. Biophys. Res. Comm.*, 1971, **45**, 1284.
[16] A. Agtarap, J. W. Chamberlin, M. Pinkerton, and L. K. Steinrauf, *J. Amer. Chem. Soc.*, 1967, **33**, 29.