angles from 120° was found at C6, C12, C18 and C24 with a compensatory enlargement [0·7 (4) to 1·7 (4)°] at C7, C11, C13, C17, C19, C23, C25 and C29. This effect may be connected with the π-electron-releasing effect of the double bond on the phenyls at C1 and C5 (Domenicano & Vaciago, 1979). However, a similar effect is observed for both phenyls at C3. A more realistic explanation (valid for all four rings) is the effect of thermal libration and rotation. This accounts for the shortening of the outer two phenyl bonds in each ring, which range from 1·349 (6) to 1·374 (6) Å [⟨C—C⟩ = 1·362 (7) Å], whilst other phenyl bonds lie in the range 1·370 (4)–1·396 (4) [⟨C—C⟩ = 1·382 (8) Å]. Similar effects are observed in other structures and will be reviewed elsewhere.

Molecular packing (see Fig. 2) and the conformation of the phenyl rings are controlled only by van der Waals forces. The shortest intermolecular distance between non-H atoms is C14—C31^{i} (i = $1-x$, $1-y$, $-z$) [3·351 (1) Å]. No changes in the diffraction pattern were observed during coloration, either in the single crystal or in the powdered material. Probably only a small part of the molecule undergoes photochromic excitation and/or coloration causes insignificant geometrical changes. Differences in photochomic behaviour between compounds may be attributed to differences in planarity of the 1,4-dihydropyridine rings, related to the packing effects of the phenyl groups in positions C1 and C5, and to steric interactions of the benzyl group in the 4-benzyl-1-methyl derivative (see Table 3) (Iwasaki, Watanabe & Maeda, 1987).

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Structure of the Cyclohexapeptide Cleromyrine II Trihydrate

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Abstract. $C_{29}H_{40}N_6O_7.3H_2O$, $M_r = 638·7$, trigonal, $P3_121$, $a = 14·190$ (2), $c = 29·833$ (4) Å, $V = 5202$ (1) Å^3, $Z = 6$, $D_x = 1·22$ g cm^{-3}, Cu $K\alpha$, $\lambda = 1·54178$ Å, $\mu = 7·8$ cm^{-1}, $F(000) = 2052$, $T = 291$ K, $R = 0·069$ for 1942 observed reflections. The new cyclohexapeptide cleromyrine II was isolated from *Clerodendrum myricoides*. Its structure was established by spectroscopic and X-ray diffraction methods as *cyclo*(-Gly-Tyr-Gly-Pro-Leu-Pro-). The conformation essentially consists of two β-turns including the Pro residues and one central very short antiparallel β-sheet stabilized by two intramolecular hydrogen bonds: N(Tyr2)···O(Leu5) = 2·94 (2) Å and N(Leu5)···O(Tyr2) = 3·02 (2) Å.

Introduction. Cleromyrine I and II are two new homodetic cyclohexapeptides isolated from *Clerodendrum myricoides* (Verbenaceae) as described in a preceding communication (Bashwira, Hootelé, Tourwé, Pepermans, Laus & van Binst, 1989); the

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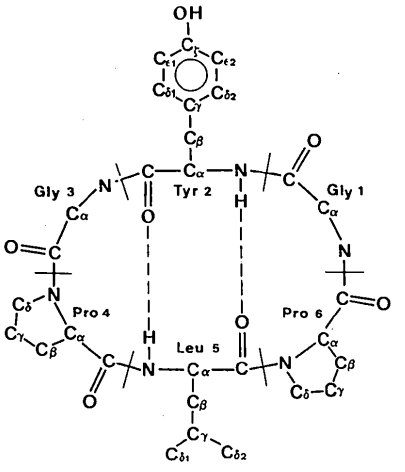
structure of cleromyrine I, the major compound, has been established as *cyclo*(-Pro-Ile-Val-Phe-Ala-Gly-) by chemical and spectroscopic methods. We now report the structure determination of cleromyrine II.

The high-resolution mass spectrum of cleromyrine II showed a strong molecular ion at *m/z* 584, corresponding to the formula $C_{29}H_{40}N_6O_7$. The cyclohexapeptidic nature of the new compound was immediately apparent from the inspection of the ^1H and ^{13}C NMR and IR spectra. As the crystals of cleromyrine II appeared suitable, the complete structure was determined by a single-crystal X-ray diffraction study and established as *cyclo*(-Gly-Tyr-Gly-Pro-Leu-Pro-).



The amino-acid composition was confirmed and the absolute configuration established after 6*N* DCl hydrolysis (Liardon & Lederman, 1984) of cleromyrine II. Gas chromatography of the *N*-trifluoroacetyl-isobutylesters on a Chirasil-Val column (Nicholson, Frank & Bayer, 1982) revealed the presence of Gly, L-Tyr, L-Pro and L-Leu. As cleromyrine I, cleromyrine II therefore belongs to the small group of natural cyclopeptides made exclusively of the protein-derived L-amino acids (Ovchinnikov & Ivanov, 1982).

Experimental. Mass spectra were obtained on a Micromass 7070 spectrometer. The NMR spectra were recorded on a Bruker WM 250 in $CDCl_3$ with TMS as internal standard.

Cleromyrine II was isolated (Bashwira *et al.*, 1989) in minute amounts from *Clerodendrum myricoides* (0·002% from the dried plant) and was obtained as a crystalline compound, m.p. 470–472 K (EtOH), $[\alpha]_D^{22°C} = -18°$[MeOH,$c = 1$]; MS: 584 (M^+, 71%, $C_{29}H_{40}N_6O_7$, found: 584·2918, calc.: 584·2958), 528 (34, $C_{25}H_{32}N_6O_7$, found: 528·2347, calc.: 528·2332), 478 (20, $C_{22}H_{34}N_6O_6$, found: 478·2503, calc.: 478·2540), 429 (17), 414 (76), 413 (34), 386 (68), 382 (24), 368 (20), 323 (24), 301 (100), 300 (46), 290 (44); ^1H NMR (CD_3OD): δ 1·00 and 1·03 [2*d*, J = 6 Hz, $CH(CH_3)_2$], 6·68 and 7·03 (*AA'XX'*syst.); ^{13}C NMR (CD_3OD): δ 21·5, 23·9 and 26·5 [$CD(CH_3)_2$]; 25·2, 26·8, 29·6, 30·8, 38·4, 40·7, 43·3, 44·4, 48·2 and 48·8 (10 × CH_2); 50·6, 57·6, 62·4 and 63·2 (4 × CH); 116·1, 129·6, 131·5, 157·2 (—C_6H_4OH); 170·6, 171·8, 172·5, 172·9, 174·3 and 175·3 (6 × —CO—N).

Table 1. *Atomic coordinates* ($\times 10^4$) *and equivalent isotropic temperature factors* (Å^2)

$B_{eq} = (8\pi^2/3)\sum_i\sum_j U_{ij}a_i^*a_j^*\mathbf{a}_i.\mathbf{a}_j.$

Residue		x	y	z	B_{eq}
Gly 1	N	5690 (5)	5478 (5)	718 (2)	5·7 (1)
	Cα	5199 (7)	4344 (7)	585 (3)	6·4 (2)
	C	3961 (8)	3708 (8)	553 (2)	6·1 (2)
	O	3512 (5)	2705 (5)	526 (2)	6·7 (1)
Tyr 2	N	3417 (6)	4241 (6)	577 (2)	5·9 (1)
	Cα	2220 (6)	3599 (7)	557 (3)	6·1 (2)
	C	1811 (6)	4376 (7)	414 (3)	5·8 (2)
	O	1599 (5)	4885 (5)	688 (2)	6·7 (1)
	Cβ	1762 (7)	3057 (8)	1010 (3)	6·8 (2)
	Cγ	587 (7)	2161 (8)	967 (3)	6·9 (2)
	Cδ1	−279 (8)	2359 (8)	1046 (3)	8·0 (2)
	Cδ2	335 (9)	1168 (9)	813 (5)	10·8 (3)
	Cε1	−1339 (8)	1514 (9)	998 (3)	7·9 (2)
	Cε2	−737 (10)	305 (10)	786 (6)	11·3 (3)
	Cζ	−1551 (9)	515 (10)	879 (4)	9·2 (3)
	OH	−2624 (6)	−334 (6)	836 (3)	10·9 (2)
Gly 3	N	1674 (6)	4435 (6)	−27 (2)	6·7 (2)
	Cα	1315 (8)	5144 (8)	−201 (3)	7·4 (2)
	C	2137 (9)	6291 (8)	−117 (3)	6·8 (2)
	O	3091 (6)	6682 (6)	−224 (2)	8·6 (2)
Pro 4	N	1788 (6)	6911 (7)	116 (2)	7·0 (2)
	Cα	2564 (8)	8065 (8)	229 (3)	7·6 (2)
	C	3220 (7)	8168 (8)	662 (3)	6·5 (2)
	O	3955 (7)	9058 (5)	757 (2)	9·9 (2)
	Cβ	1826 (12)	8545 (12)	314 (4)	10·9 (4)
	Cγ	791 (11)	7555 (13)	499 (6)	11·9 (4)
	Cδ	669 (9)	6597 (10)	240 (4)	8·8 (3)
Leu 5	N	2948 (5)	7284 (5)	895 (2)	5·7 (1)
	Cα	3586 (6)	7311 (7)	1279 (3)	6·0 (2)
	C	4448 (7)	7026 (6)	1113 (3)	5·6 (2)
	O	4341 (5)	6579 (5)	746 (2)	6·5 (1)
	Cβ	2843 (7)	6400 (7)	1625 (3)	6·0 (2)
	Cγ	1904 (9)	6471 (10)	1788 (4)	8·6 (3)
	Cδ1	1248 (10)	5512 (9)	2104 (4)	11·0 (3)
	Cδ2	2092 (13)	7439 (13)	1962 (6)	14·1 (4)
Pro 6	N	5306 (6)	7256 (6)	1371 (2)	6·1 (2)
	Cα	6110 (7)	6962 (7)	1218 (3)	6·3 (2)
	C	5564 (7)	5736 (7)	1120 (3)	5·8 (2)
	O	5020 (5)	5053 (5)	1409 (2)	7·1 (1)
	Cβ	6897 (7)	7298 (8)	1619 (3)	7·2 (2)
	Cγ	6790 (8)	8160 (9)	1843 (3)	8·4 (2)
	Cδ	5626 (8)	7826 (8)	1810 (3)	7·2 (2)
Ow1		5951 (4)	287 (6)	1093 (2)	10·7 (1)
Ow2		5285 (5)	7964 (5)	−14 (2)	10·9 (2)
Ow3		6579 (8)	7073 (8)	−26 (4)	7·9 (2)
Ow4		8627 (11)	7427 (13)	−14 (5)	12·8 (4)

Crystals of cleromyrine II trihydrate obtained by crystallization from ethanol. D_m not measured. Parallelepiped crystal with dimensions 0·07 × 0·1 × 0·2 mm. Lattice parameters refined using 17 reflections in the range $7 \le 2\theta \le 50°$. Huber 424 + 511 four-circle diffractometer and Rigaku RU200 rotating-anode generator, graphite-monochromated Cu $K\alpha$ radiation. ω–θ scans. 7047 reflections collected ($0 \le h \le 14$, $0 \le k \le 14$, $-35 \le l \le 35$) with $\sin\theta/\lambda < 0{\cdot}60$ Å^{-1}, 3523 independent ($R_{int} = 0{\cdot}075$), of which 1942 had $I \ge 2{\cdot}5\sigma(I)$. Standard reflection ($3\bar{1}1$) checked every 50 reflections: no significant deviation. Structure solved by direct methods using

*SHELXS*86 (Sheldrick, 1985). H atoms at C, N in computed positions (C—H, N—H = 1·08 Å). Two of the water molecules (O*w*3 and O*w*4) were included with half occupation factors, because of the proximity of the twofold axis: the equivalent positions generated by this axis cannot exist simultaneously. Anisotropic least-squares refinement (*SHELX*76; Sheldrick, 1976) on F; H atoms isotropic with common refined temperature factor ($B = 11$ Å^2) and allowed to ride on their bonded heavier atoms. $w = 1/(\sigma^2 + 0{\cdot}0027F^2)$, $R = 0{\cdot}069$, $wR = 0{\cdot}077$, $S = 1{\cdot}6$ for 1942 observed reflections. Final maximum shift to e.s.d. = 0·2. Maximum and minimum heights in final difference Fourier synthesis = 0·20 and $-0{\cdot}24$ e Å^{-3}. Atomic scattering factors from *International Tables for X-ray Crystallography* (1974).

Discussion. The atomic parameters are given in Table 1.* Fig. 1 is a stereoscopic view of the molecule, showing the numbering scheme of the amino acids (*PLUTO*; Motherwell & Clegg, 1978). Table 2(*a*) gives the bond distances in the main chain and 2(*b*) the conformational angles in the polypeptide. The conformation of the cyclic hexapeptide essentially consists of two β-turns and one central very short β-sheet. In the Pro residues, the closure of the five-membered ring imposes a φ value of approximately $-60°$. It is quite usual to observe Pro residues in β-turns, and in the present analysis both Pro appear in such conformations: Pro 6–Gly 1, where the φ and ψ conformational angles (-57, 122, 68, 10°) are close to the standard values of a β-turn II (-60, 120, 80, 0°), and Gly 3–Pro 4 (65, -124, -83, -7°), comparable to the β-turn II′ (60, -120, -80, 0°).

The relative dispositions of the two remaining residues (Tyr 2 and Leu 5) can be compared to a distorted antiparallel β-sheet characterized by the intramolecular hydrogen bonds [N(2)···O(5) 2·94 Å] and [N(5)···O(2) 3·02 Å]. Their conformational angles (-159,92° and -92,163°) bear only an approximate similarity to the expected values (-139,135°). The three-dimensional structure is stabilized by numerous intermolecular hydrogen bonds given in Table 2(*c*).

Table 2. *Molecular geometry in cleromyrine* II

(*a*) Bond distances in the main chain (Å)

	N—Cα	Cα—C	C=O	C—N*
Gly 1	1·45 (1)	1·52 (1)	1·24 (1)	1·33 (2)
Tyr 2	1·47 (1)	1·54 (2)	1·22 (1)	1·34 (1)
Gly 3	1·43 (2)	1·47 (1)	1·22 (1)	1·39 (2)
Pro 4	1·48 (1)	1·56 (1)	1·20 (1)	1·31 (1)
Leu 5	1·45 (1)	1·55 (1)	1·23 (1)	1·33 (1)
Pro 6	1·47 (1)	1·54 (1)	1·24 (1)	1·29 (1)

(*b*) Conformational angles in cleromyrine II (°)

	φ	ψ	ω	χ^1	χ^2
Gly 1	68	10	−178	–	–
Tyr 2	−159	92	−179	−164	−94
Gly 3	65	−124	178	–	–
Pro 4	−83	−7	174	31	−87
Leu 5	−92	163	178	−58	178
Pro 6	−57	122	180	−26	37

(*c*) Hydrogen bonds X—H···Y; residue numbers are in brackets; the symmetry operations apply to the second atom

	$X\cdots Y$ (Å) ($\sigma \approx 0{\cdot}02$ Å)	H···Y (Å)	X—H···Y (°)	Symmetry
Intramolecular bonds				
N(2)···O(5)	2·94	1·87	170	
N(5)···O(2)	3·02	2·06	146	
Intermolecular bonds				
N(3)···O(1)	2·83	1·82	154	$y,x,-z$
OH(2)···O*w*1†	2·69			$x-1,y,z$
OH(2)···O*w*4	2·88			$y-1,x-1,-z$
O*w*1···O(4)	2·67			$x,y-1,z$
O*w*1···O(6)	2·77			$1-x,y-x,1/3-z$
O*w*2···O(3)	2·78			x,y,z
O*w*2···O(5)	2·86			x,y,z
N(1)···O*w*3‡	2·96	1·92	160	x,y,z
N(1)···O*w*3‡	2·74	1·75	150	$y,x,-z$
O*w*3···O*w*2	2·70			x,y,z
O*w*4···O*w*2	2·70			$y,x,-z$
O*w*4···O*w*3	2·69			x,y,z

* N of the next residue.

† The coordinates of the H atoms of the water molecules and of the hydroxyl group of Tyr 2 were not determined.

‡ Due to the statistical disorder affecting O*w*3, only one of the bonds N(1)···O*w*3 exists at a time.

* Lists of structure factors, anisotropic thermal parameters, H-atom parameters and a list of bond lengths and angles have been deposited with the British Library Document Supply Centre as Supplementary Publication No. SUP 52663 (15 pp.). Copies may be obtained through The Technical Editor, International Union of Crystallography, 5 Abbey Square, Chester CH1 2HU, England.

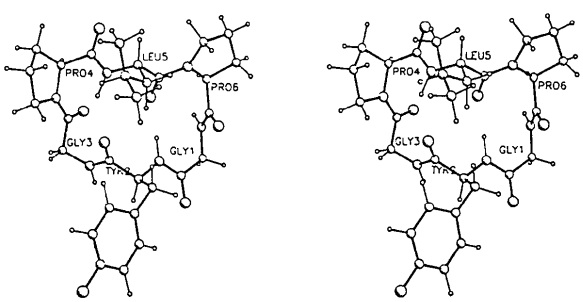


Fig. 1. Stereoscopic view of the molecule sharing the atomic numbering scheme.

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Structure of the 4-Oxo-2-butenoic Acid Alkyl Ester Moiety. I. Structures of Diethyl 4,4′-(1,4-Piperazinediyl)bis(4-oxo-2-butenoate) and Dimethyl 4,4′-(2,5-Dioxo-1,4-piperazinediyl)bis(4-oxo-2-butenoate)

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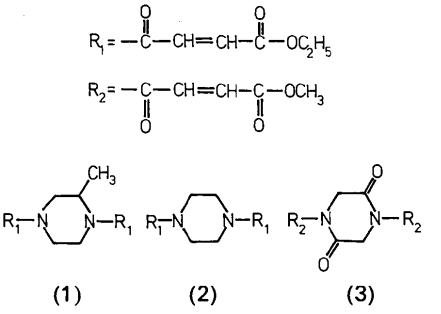
Abstract. $C_{16}H_{22}N_2O_6$, $M_r = 338{\cdot}4$, monoclinic, $P2_1/c$, $a = 8{\cdot}282$ (1), $b = 9{\cdot}357$ (1), $c = 11{\cdot}288$ (2) Å, $\beta = 93{\cdot}09$ (1)°, $V = 873{\cdot}5$ Å^3, $Z = 2$, $D_x = 1{\cdot}286$ Mg m^{-3}, λ(Cu $K\alpha$) = 1·54178 Å, $\mu = 0{\cdot}74$ mm^{-1}, $F(000) = 360$, room temperature, $R = 0{\cdot}050$ for 1369 reflections. $C_{14}H_{14}N_2O_8$, $M_r = 338{\cdot}3$, monoclinic, $P2_1/c$, $a = 24{\cdot}350$ (6), $b = 6{\cdot}973$ (1), $c = 9{\cdot}212$ (1) Å, $\beta = 91{\cdot}74$ (1)°, $V = 1563{\cdot}4$ Å^3, $Z = 4$, $D_x = 1{\cdot}437$ Mg m^{-3}, λ(Cu $K\alpha$) = 1·54178 Å, $\mu = 0{\cdot}99$ mm^{-1}, $F(000) = 704$, room temperature, $R = 0{\cdot}055$ for 2037 reflections. The fumaramate groups are approximately planar in both structures with the ester group *syn* to the double bond in the first and *anti* in the second structure. Substitution of 1,4-piperazine by the 2,5-dioxo-1,4-piperazine ring affects not only the ring conformation but also conjugation at the adjacent amide groups.

Introduction. 4,4′-(2-Methyl-1,4-piperazinediyl)bis(4-oxo-2-butenoic acid deithyl ester) (1) has been shown to inhibit the growth of transplantable neoplasms in mice: leukemias L1210 and P388, and sarcoma Sa180 (Graczyk, Pakulska, Groszkowski & Najman, 1980; Groszkowski & Najman, 1983). It has been suggested that the —CH=CH—C=O fragment may be responsible for antimitotic and cytostatic properties of compound (1) (Groszkowski & Najman, 1979) and cytostatic active acrylates (Lee, Kim, Piantadosi, Huang & Geissman, 1974; Loeffler, Sajadi & Hall, 1977). Consequently, a series of other piperazides, 2-methylpiperazides, 2,5-dimethylpiperazides and piperazide-2,5-diones of α,β-unsaturated carboxylic acids and their esters have been synthetized and tested against leukemias (Groszkowski, Najman & Sienkiewicz, 1972; Groszkowski & Najman, 1972; Andrzejewska-Golec, Broda & Najman, 1977; Groszkowski & Najman, 1979; Graczyk, Pakulska, Groszkowski & Najman, 1980; Groszkowski & Najman, 1983; Groszkowski & Najman, 1986). The investigations showed that even a slight change in the chemical structure of the 'carrier' piperazine unit affects antileukemic properties of the agent and therefore our X-ray studies attempt to determine the conformation and electronic structure of 4,4′-(1,4-piperazinediyl)bis(4-oxo-2-butenoic acid diethyl ester) (2) and 4,4′-(2,5-dioxo-1,4-piperazinediyl)bis-(4-oxo-2-butenoic acid dimethyl ester) (3). The only known structures containing a similar 'active' —N—C(O)—CH=CH—C(O)— moiety are α- and β-funaltrexamines (Griffin, Larson & Porthoghese, 1986).



(1) (2) (3)

Experimental. Colourless crystals were grown by slow evaporation of a methanol–water (2) or an ethanol–chloroform (3) solution. Crystals of dimensions 0·22 × 0·15 × 0·12 mm (2) and 0·32 × 0·25 × 0·21 mm (3) were used for data collection on an Enraf–Nonius CAD-4 diffractometer fitted with an

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