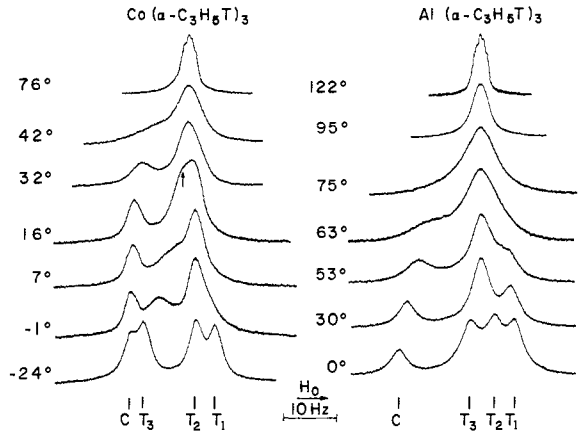

Figure 1. Methyl pmr spectra (100 MHz) of $Co(\alpha\text{-}C_3H_5T)_3$ in $CDCl_3$ and $Al(\alpha\text{-}C_3H_5T)_3$ in 1,1,2,2-tetrachloroethane. The arrow indicates the coalesced T_1 and T_3 signals.

taneous exchange broadening of all four resonances (C, T_1, T_2, T_3) and coalescence at higher temperatures to a single feature. Utilizing a total line-shape analysis,[8,14] methyl resonance averaging can be fit to a single kinetic process characterized by $\Delta H^{\pm} = 18.3 \pm 1.1$ kcal/mol, $\Delta S^{\pm} = 1.3 \pm 3.1$ eu, $E_a = 18.8 \pm 1.2$ kcal/mol, $\log A = 13.6 \pm 1.1$, and $k = 1/\tau = 0.4$ sec^{-1} (25°). The simultaneous exchange of all four resonances is similar to the coalescence pattern of unsymmetrical Al(III) β-diketonates.[2,8]

The methyl spectrum of $Co(\alpha\text{-}C_3H_5T)_3$ in $CDCl_3$ below −20° shows the presence of C and T isomers, with $\Delta F = -0.8 \pm 3.0 \times 10^{-4}T$ and K_{eq} (−24°) = 4.4. Slow-exchange chemical shifts are 1.79–1.93 ppm downfield from TMS. The pattern of line-shape changes at higher temperatures clearly proves that the molecule is stereochemically nonrigid on the pmr time scale.[15] Two essentially distinct kinetic processes are operative. In the low-temperature process (LTP, −20 to 20°), signals T_1 and T_3 exchange broaden and coalesce. At higher temperatures, all resonances broaden and average to a single resonance (HTP). By a total line-shape analysis the following kinetic parameters [LTP, HTP (T → C)] were obtained: $\Delta H^{\pm} = 16.2 \pm 1.1$, 16.5 ± 1.1 kcal/mol; $\Delta S^{\pm} = 5.4 \pm 3.9$, -2.9 ± 3.5 eu; $E_a = 16.7 \pm 0.9$, 16.9 ± 1.2 kcal/mol, $\log A = 14.4 \pm 1.0$, 12.5 ± 1.0, $k(25°) = 100$, 1.5 sec^{-1}. Recent analyses of isomerization and inversion mechanisms for $M(A\text{–}B)_3$ complexes[3,8] show that the observed site interchange between two of the three inequivalent T sites without exchange with the C site (which has not been observed previously in $M(A\text{–}B)_3$ complexes) may be accommodated by a non-bond-rupture pathway traversed by a twist around the pseudo-threefold axis of the T isomer as illustrated in Figure 7 of ref 8. Inasmuch as this site interchange can be achieved[8] otherwise only by highly selective bond-rupture mechanisms in the T form with no simultaneous bond breaking in the C form, and cannot be accommodated by twists about imaginary C_3 axes of C and T, the p-C_3 or "trigonal" twist[16] is deduced to be the most probable mechanism for the LTP of *trans*-$Co(\alpha\text{-}C_3H_5T)_3$.

Both $Al(\alpha\text{-}C_3H_7T)_3$[10] and $Co(\alpha\text{-}C_3H_7T)_3$ have also been shown to be stereochemically nonrigid by similar experiments. The exchange-broadened methyl region of the latter occurs from −10 to 75° (100 MHz) in $CDCl_3$. A line-shape analysis[17] of the former complex in 1,1,2,2-tetrachloroethane indicates that below *ca.* 30° the C and T isomers invert by twists about the real and p-C_3 axes, respectively.

The above results reveal that Co(III) tropolonates invert *ca.* 10^{10} times faster than β-diketonates[2–5] at 25°. The physical basis for this remarkable rate enhancement and for the operation of a twist mechanism, presumably involving a trigonal-prismatic (TP) transition state, is not presently understood. Distortions from octahedral toward TP, found in FeT_3[18] and expected for CoT_3 and AlT_3, might facilitate rotations about the C_3 axes and enhance the probability of twist mechanisms. However, such distortion does not necessarily lead to stereochemical nonrigidity of Co(III) chelates on the pmr time scale.[19] An apparent correlation of distorted structures and inversion by a twist pathway has recently been found for $Fe(R_1R_2\text{-dtc})_2(S_2C_2R_2)$ complexes.[20]

Acknowledgment. This work was supported by NSF Grants No. GP-7576X and GP-18978X.

(14) Chemical shifts were observed over a *ca.* 40° range in the slow-exchange region, plotted *vs.* temperature, and extrapolated through the intermediate-exchange region. Weighted averages of extrapolated shifts agreed well with experimental averages.

(15) A possible cause of this remarkable behavior could be electron-transfer catalysis of the intramolecular rearrangements by Co(II) impurity species. Although this possibility cannot be unequivocally disproven, it is considered improbable on the basis of the following observations: (i) line shapes at ambient temperature were the same for separate preparations of the complex and for varying degrees of purity; (ii) $Co(\alpha\text{-}C_3H_7T)_2$, prepared separately, is strongly absorbed on alumina and not eluted with chloroform, and therefore would be removed in the preparation of $Co(\alpha\text{-}RT)_3$; and (iii) methyl line shapes of $Co(\alpha\text{-}C_3H_5T)_3$ at 32° in $CDCl_3$ were unchanged by the addition of up to ~20 mol % $Co(\alpha\text{-}C_3H_7T)_2$.

(16) The trigonal twist is a conceptually simple but not unique way of describing the exchange pathway and produces different site interchanges than do the bond rupture processes analyzed elsewhere.[8]

(17) J. R. Hutchison, S. S. Eaton, R. H. Holm, and E. L. Muetterties, manuscript in preparation.

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(19) See footnote 54 of ref 18.

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(21) NSF Predoctoral Fellow, 1969–1971.

S. S. Eaton,[21] R. H. Holm*

Department of Chemistry, Massachusetts Institute of Technology Cambridge, Massachusetts 02139

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Eupacunin, a Novel Antileukemic Sesquiterpene Lactone from *Eupatorium cuneifolium*[1,2]

Sir:

We wish to report on the isolation and structural elucidation of eupacunin (**1**), a novel germacranolide from *Eupatorium cuneifolium* (Tourn.) L.[3] Eupacunin has significant antileukemic and tumor inhibitory prop-

(1) Tumor Inhibitors. LXVII. Part LXVI: S. M. Kupchan and A. J. Liepa, *Chem. Commun.*, 599 (1971).

(2) Supported by grants from the National Cancer Institute (CA-04500 and CA-11718), American Cancer Society (T-275), and Science Research Council, and a contract with Chemotherapy, National Cancer Institute (NIH 71-2099).

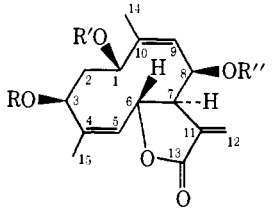
(3) Whole plant collected in Florida in 1966 and 1969. We thank Dr. Robert E. Perdue, Jr., USDA, Beltsville, Md., for supplying the plant material.

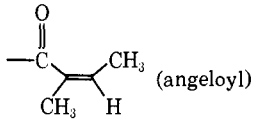
Table I. Nmr Data

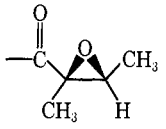
Compd	Position C-1	C-2	C-3	C-5	C-6	C-7	C-8	C-9	C-13	C-14	C-15
10[a]	4.60 m	7.24 d of d of d (2.5, 10, 14) 7.62 d of d of d (4, 7, 14)	4.80 d of d (2.5, 4)	4.82 d of d (1.5, 11)	4.22 d of d (2.5, 11)	6.7 m	4.36 d of d (1, 3)	5.54 d (3)	3.76 d (2.5) 4.04 d (2)	8.03 s	8.20 d (1.5)
6[b]	6.07 d of d (2, 11)	7.55 d of d of d (2, 5, 15) 8.30 d of d of d (3, 11, 15)	4.7 m	4.67 d of d (1.5, 11)	4.38 d of d (8, 11)	6.7 m	4.36 d (3.5)		3.63 d (3) 4.21 d (3)	8.53 s	8.11 d (1.5)
9[a]	5.61 d of d (2, 5)	7.9–8.3 m	4.19 d of d (2.5, 11)	4.92 br d of d (10)	3.56 t (10)	7.8 m	5.49 br d of d (5)	4.90 d of d (1.5, 5)	6.37 m	8.35 m	8.35 m

[a] τ values (100 MHz) in acetone-d_6 (TMS). [b] τ values (100 MHz) in $CDCl_3$ (TMS).

erties,[4] and appears to be the first recognized germacranolide *cis,cis*-diene.



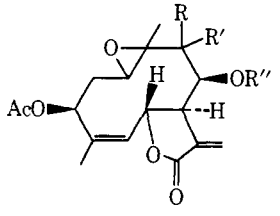
1, R = Ac; R′ = H; R″ =  (angeloyl)

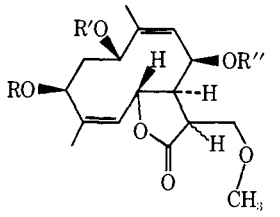


2, R = Ac; R′ = H; R″ =

3, R = R′ = Ac; R″ = angeloyl

4, R = R′ = H; R″ = angeloyl





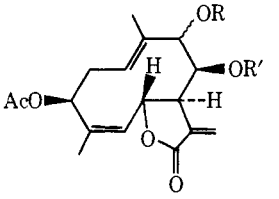
5, R = H; R′ = OH; R″ = angeloyl

6, R, R′ = O; R″ = angeloyl

7, R = R′ = H; R″ = angeloyl

8, R = R′ = Ac; R″ = angeloyl

9, R = R′ = R″ = H



10, R = H; R′ = angeloyl

Fractionation of the alcoholic extract was guided by the KB assay.[4] Successive solvent partitions and silicic acid chromatography yielded the new cytotoxic lactones eupacunin (**1**), eupacunoxin (**2**), and eupatocunin (**10**).

Eupacunin [**1**; $C_{22}H_{28}O_7$;[5] mp 166–167°; $[\alpha]^{25}D$ +55° (*c* 1.24); uv max 211 nm (ϵ 23,900); ir (KBr) 2.77, 5.70, 5.75, 5.84, 6.10, and 8.02 μ; *m/e* 345 [M − 59 (CH_3CO_2)], 305 [M − 99 ($CH_3CH{=}C(CH_3)CO_2$)], and 229 (M − 99 − 60 − 18); nmr ($CDCl_3$) τ 8.18 [3 H, d, J = 2 Hz, $-C(CH_3){=}CH-$], 8.30 [3 H, d, J = 2 Hz, $-C(CH_3){=}CH-$]] gave an acetate [**3**; $C_{24}H_{30}O_8$; mp 198–199°; $[\alpha]^{27}D$ −12° (*c* 0.90); nmr showed no D_2O-exchangeable proton]. Alkaline treatment products were: deacetyleupacunin [**4**, from 2% NaOH in aqueous dioxane; $C_{20}H_{26}O_6$; mp 155–156°; $[\alpha]^{25}D$ +114° (*c* 1.50)], deacetyl-13-methoxydihydroeupacunin [**7**, from 2% NaOH in aqueous MeOH; $C_{21}H_{30}O_7$; mp 150–151°; $[\alpha]^{26}D$ +76°; acetylated to diacetate **8**; $C_{25}H_{34}O_9$; mp 136–137°], and deacetyldeangeloyl-13-methoxydihydroeupacunin [**9**, from NaOMe in MeOH; $C_{16}H_{24}O_6$; mp 202–203°]. Oxidation of eupacunin (**1**) with Jones' reagent afforded two epoxy derivatives, one an epoxy alcohol [**5**; $C_{22}H_{28}O_8$; mp 195–196°; $[\alpha]^{26}D$ +54° (*c* 0.90); nmr ($CDCl_3$) τ 8.66 (s, 3 H, epoxide methyl)] and the other an epoxy ketone [**6**; $C_{22}H_{26}O_8$; mp 200–201°; $[\alpha]^{26}D$ +66° (*c* 1.11)]. The epoxy ketone **6** was also obtained from oxidation of eupatocunin (**10**).

Eupatocunin [**10**; $C_{22}H_{28}O_7$; mp 163–164°; $[\alpha]^{26}D$ −129° (*c* 1.36)] is isomeric with eupacunin (**1**), and the similarities in spectral data indicated a close structural relationship. The 100-MHz nmr spectrum of **10** (Table I) showed many more detailed couplings than that of **1**, and spin-decoupling experiments supported postulation of structures **10** and **1** (apart from stereochemistry). The nmr spectrum of **6** is similar to that of **10**, the only significant differences being the absence of a signal for a vinyl methyl and a multiplet signal for an olefinic proton (τ 8.03 and 4.60, respectively, in the spectrum of **10**) and the appearance of a methyl singlet at τ 8.53 and a new doublet of doublets at τ 6.07 (J = 11, 2 Hz), indicative of the presence of an

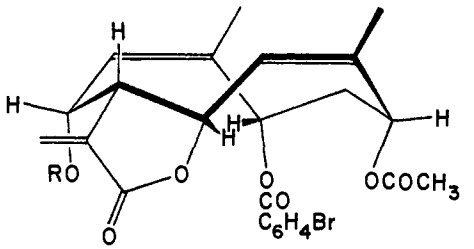
(4) Eupacunin showed confirmed activity against P-388 leukemia and WM-256 intramuscular carcinosarcoma. Cytotoxicity (KB) and *in vivo* activity were assayed by the procedures described in *Cancer Chemother. Rep.*, **25**, 1 (1962).

(5) All crystalline compounds have been characterized by concordant elemental and spectral (ir, uv, nmr, mass spectral) analyses. Unless otherwise stated, optical rotations were measured in acetone and uv spectra in methanol.

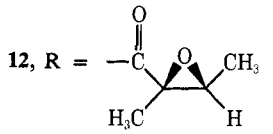
epoxide ring at C-1–C-10 in **6**. Evidence for a C-9 carbonyl in **6** was found in the disappearance of the doublet at τ 5.54 (J = 3 Hz, proton on carbon bearing oxygen), assigned to the C-9 proton signal in the spectrum of **10**, and the appearance of a new doublet at τ 4.36 (J = 3.5 Hz), which corresponds to a doublet of doublets at τ 4.36 (J = 3, 1 Hz) in the spectrum of **10**. This indicated that eupacunin has a C-1 hydroxyl group which undergoes allylic rearrangement,[6] on oxidation, to give the 1,10-epoxy 9-ketone grouping. Methanolysis of **10** (NaOMe) led to selective loss of the angeloyl group. This indicated that the angeloyl group in **10** is vicinal[7] to the C-9 hydroxyl group and is located at C-8 (in **10** *and* **1**).

Eupacunoxin [**2**; $C_{22}H_{28}O_8$; mp 171–172°; $[\alpha]^{25}D$ +27° (*c* 1.00); nmr (acetone-d_6) τ 6.95 (q, J = 5.5 Hz), 8.33 (s), 8.82 (d, J = 5.5 Hz) (epoxybutanoate)] was methanolyzed to methyl α-methyl-*trans*-α,β-epoxybutanoate and triol **9**, indicative that **2** differed from **1** solely in the ester function.

Unequivocal proof of the structure, stereochemistry, and absolute configuration of eupacunin was achieved by X-ray crystallographic analysis of eupacunin *o*-bromobenzoate (**11**), $C_{29}H_{31}BrO_8$, mp 184–186°, and eupacunoxin *m*-bromobenzoate (**12**), $C_{29}H_{31}BrO_9$, mp



11, R = angeloyl



191–192°. Eupacunin *o*-bromobenzoate (**11**) crystallized in the monoclinic space group $P2_1$ with a = 9.380 (4), b = 9.129 (4), and c = 17.309 (5) Å, β = 94.36 (5)°, and z = 2. The X-ray diffraction data were recorded on a Hilger and Watts' computer-controlled four-circle diffractometer with Cu Kα irradiation; 1769 significant independent intensities were obtained by the diffractometer measurements. The crystal structure was elucidated by Patterson and Fourier methods, and the atomic coordinates were subsequently adjusted by least-squares calculations incorporating corrections for anomalous dispersion; R is 11.8%. Eupacunoxin *m*-bromobenzoate crystallized in the orthorhombic space group $P2_12_12$ with a = 10.185 (5), b = 31.736 (12), c = 9.324 (5) Å, and z = 4. The X-ray diffraction data were collected and treated as above, except that Mo Kα radiation was used, and 1770 independent significant reflections were obtained; R is 11.3% for **12**.

Although a considerable number of sesquiterpene lactones have been found to show significant cytotoxicity toward KB carcinoma cell culture, eupacunin is one of the few which shows significant *in vivo* tumor inhibitory activity.[8] In view of the recent demonstration of the potential importance of nucleophilic additions of biologically important sulfhydryl groups for the tumor inhibitory activity of related compounds,[9] and of the inhibition of the sulfhydryl enzyme phosphofructokinase by eupacunin,[10] investigations are in progress to determine the significance of various structural features in relation to the biological activity of eupacunin.

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(9) S. M. Kupchan, D. C. Fessler, M. A. Eakin, and T. J. Giacobbe, *Science*, **168**, 376 (1970).
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S. Morris Kupchan,* M. Maruyama, R. J. Hemingway
J. C. Hemingway, S. Shibuya, T. Fujita
Department of Chemistry, University of Virginia
Charlottesville, Virginia 22901

P. D. Cradwick, A. D. U. Hardy, G. A. Sim
Chemistry Department, University of Glasgow
Glasgow W-2, Scotland

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Liatrin, a Novel Antileukemic Sesquiterpene Lactone from *Liatris chapmanii*[1,2]

Sir:

In the course of a continuing search for tumor inhibitors from plant sources, it was found that chloroform extracts of *Liatris chapmanii* (Compositae)[3] showed significant inhibitory activity against cells derived from human carcinoma of the nasopharynx (KB) carried *in vitro*.[4] We report herein the isolation and structural elucidation of an antileukemic sesquiterpene lactone, liatrin (**1**).[5] Liatrin numbers among a very small group of sesquiterpene lactones which show significant *in vivo* tumor inhibitory activity,[6] and appears to be but the second recognized naturally occurring germacranolide *cis,cis*-diene (*cf.* ref 1).

The active principle was isolated from *L. chapmanii* by fractionation involving successive solvent partitions and alumina and silicic acid chromatography, guided at each step by the KB assay.[4] Liatrin (**1**) [$C_{22}H_{26}O_8$; mp 130–132°; $[\alpha]^{24}D$ −142° (*c* 1.93, $CHCl_3$); uv end absorption (EtOH) 220 nm (ϵ 19,420); ir (KBr) 2.92 (OH), 5.67 (α,β-unsaturated γ-lactone), 5.76 (ester), 5.84 (α,β-unsaturated ester), and 6.03 μ (C=C); *m/e* 418 (M^+), 400 [M − 18 (H_2O)], 375 [M − 43 ($COCH_3$)], 358 [M − 60 (CH_3COOH)], 343 [M − 75

(1) Tumor Inhibitors. LXVIII. Part LXVII: S. M. Kupchan, M. Maruyama, R. J. Hemingway, J. C. Hemingway, S. Shibuya, T. Fujita, P. D. Cradwick, A. D. U. Hardy, and G. A. Sim, *J. Amer. Chem. Soc.*, **93**, 4914 (1971).
(2) Supported by grants from the National Cancer Institute (CA-11718 and CA-11760) and American Cancer Society (T-275), and a contract with Chemotherapy, National Cancer Institute (NIH 71-2099).
(3) Whole plant gathered in Florida in 1962. The authors acknowledge with thanks receipt of the dried plant material from Dr. Robert E. Perdue, Jr., U. S. Department of Agriculture (USDA), Beltsville, Md., in accordance with the program developed with the USDA by the Cancer Chemotherapy National Service Center (CCNSC).
(4) Cytotoxicity (KB) and *in vivo* activity were assayed under the auspices of the CCNSC, by the procedures described in *Cancer Chemother. Rep.*, **25**, 1 (1962).
(5) Liatrin showed significant tumor inhibitory activity against P-388 lymphocytic leukemia in mice.
(6) *Cf.* J. L. Hartwell and B. J. Abbott, *Advan. Pharmacol. Chemother.*, 7, 117 (1969).