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ERYCRISTIN, A NEW ANTIMICROBIAL PETROCARPAN FROM *ERYTHRINA CRISTA-GALLI*

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Key Word Index—*Erythrina crista-galli*; Leguminosae; stem bark; erycristin; sandwicensin; erythrabyssin-II; pterocarpan metabolites; antimicrobial agents.

Abstract—Bioassay-directed fractionation of ethanolic extracts of the stem bark of *Erythrina crista-galli* resulted in the isolation of a new pterocarpan, erycristin, and two previously known pterocarpans, sandwicensin and erythrabyssin-II. The structure of erycristin was determined by spectroscopic examination and by chemical transformation of sandwicensin. The absolute stereochemistry of erycristin was established by circular dichroism measurements. Erythrabyssin-II diacetate was converted to erycristagallin diacetate, supporting the previous proposed structure of erycristagallin.

INTRODUCTION

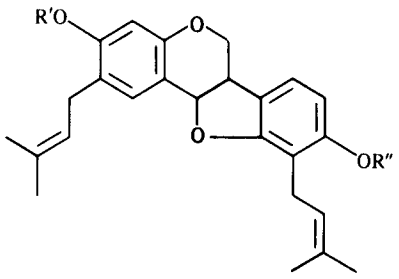
There has been a recent increase in research effort on the non-alkaloidal secondary metabolites of the genus *Erythrina* [1–4]. Our interest was stimulated when, in our hands, ethanolic extracts of the stem and root barks of *E. crista-galli* showed reproducible antimicrobial activity against *Mycobacterium smegmatis* (ATCC 607) and *Staphylococcus aureus* (ATCC 13709). *Erythrina crista-galli* (Leguminosae) is widely distributed in tropical and subtropical regions of the American continent. It is variously known as the Bolivian coral tree, Ceibo (in Spanish) and chilichi (in Quechua). Previous investigators have isolated and identified several alkaloids [5, 6], flavonoids [7] and phytoalexins [8] from the plant. In a preliminary report, we have briefly described the isolation and structure determination of a new antimicrobial pterocarpene, erycristagallin (**4**) from the root bark [9]. In the present paper we wish to describe the isolation from the stem bark and structure elucidation of a new pterocarpan, erycristin (**1**), in addition to two previously reported phytoalexins and antimicrobial pterocarpans, erythrabyssin-II (**3**) and sandwicensin (**2**) [10, 11].

RESULTS AND DISCUSSION

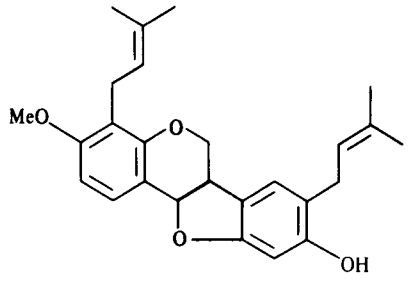
The dried and powdered stem bark of *E. crista-galli* was extracted with ethanol in a Soxhlet apparatus and the extract concentrated to dryness. The residue showed antimicrobial activity against *S. aureus* and *M. smegmatis* at < 1000 μg/ml in the agar dilution-streak assay [12]; fractionation was performed essentially as described previously [13]. The antimicrobial activity was found to reside exclusively in the methanol soluble polar lipid and phenol fractions. Extensive chromatography over silica gel afforded pure erycristin (**1**), sandwicensin (**2**) and erythrabyssin-II (**3**).

Erycristin (**1**) has the empirical formula, $C_{26}H_{30}O_4$. Its UV absorption was similar to that of sandwicensin (**2**) [11] suggesting an analogous substituent pattern. ^{1}HNMR signals at δ3.53 (1H, *ddd*, J = 5.1, 6.3, 11.4 Hz), 3.62 (1H, *dd*, J = 10.8 Hz), 4.22 (1H, *dd*, J = 5.1, 10.8 Hz) and 5.45 (1H, *d*, J = 6.3 Hz) were assigned to the H_{6a}, H_{6eq}, H_{6ax} and H_{11a} protons of the pterocarpan ring, suggesting a *cis* arrangement of the 6a and 11a protons [14]. ^{13}CNMR signals at δ40.12, 66.57 and 78.04 were in agreement with the signals assigned to the C-6, C-6a, and C-11a carbons of the pterocarpan ring in reference substances [15]. A pair of *ortho* coupled protons at δ6.40 (*d*, J = 7.8 Hz) and 7.01 (*d*, J = 7.8 Hz), and a pair of *para* coupled protons at δ6.41 (*s*), 7.27 (*s*) were clearly seen. Additional signals at δ3.81 (*s*, 3H) and 8.51 (*s*, 1H) were assigned to methoxyl and hydroxyl groups, respectively. The hydroxyl signal was D_2O exchangeable. The remaining signals at δ1.67 (*s*, 3H), 1.77 (*s*, 3H), 1.80 (*s*, 6H), 3.30 (*br*, 2H) 3.37 (*d*, 2H) 5.25 (*br*, 1H) and 5.32 (*t*, 1H) together with certain peaks in the mass spectrum ($[M-43]^+$, $[M-55]^+$ and $[M-111]^+$) indicate the presence of two separate isoprenyl groups [16, 17]. Erycristin (**1**) was acetylated with pyridine and acetic anhydride to give erycristin monoacetyl ester (**5**) in which one of the *para* coupled aromatic protons was shifted downfield from δ6.41 to 6.64 ($\Delta\delta$ = 0.23) suggesting that the *para* coupled proton was adjacent to the hydroxyl group [18]. Thus, two possible structures (**1**, **1a**) can be written for erycristin based upon this data. Erycristin monomethyl ether (**6**) was prepared by refluxing erycristin with dimethyl sulphate and base, and was found to be identical to erythrabyssin-II dimethyl ether prepared under the same conditions from erythrabyssin-II (**6**). This sequence favoured structure **1** over **1a** for erycristin. The chemical conversion of sandwicensin (**2**) to erycristin (**1**) now to be described, further confirms the proposed structure.

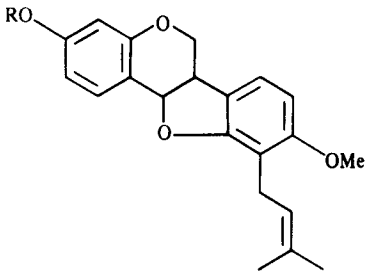
Sandwicensin (**2**), on reflux with 3-chloro-3-methylbutyne, potassium carbonate and potassium iodide in acetone, gave ether **7** [19]. Catalytic hydrogenation of **7** with 10% palladium on barium sulphate poisoned by quinoline gave olefin **8** [20]. Claisen rearrangement of **8**



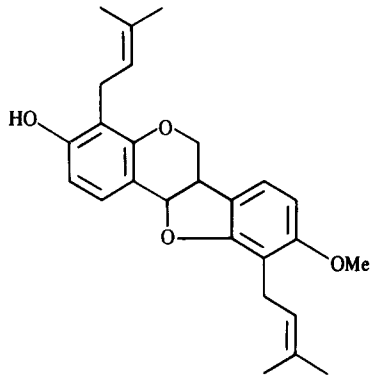
1 R^1 = H, R^2 = Me
3 R^1 = R^2 = H
5 R^1 = Ac, R^2 = Me
6 R^1 = R^2 = Me
10 R^1 = R^2 = Ac



1a



2 R = H
7 R = $Me_2CC{\equiv}CH$
8 R = $Me_2CH{=}CH_2$



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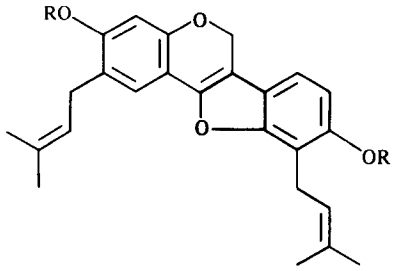

4 R = H
11 R = Ac

was accomplished by refluxing in *N,N*-diethylaniline to afford a mixture of isoerycristin (**9**) and erycristin (**1**) [20]. Isoerycristin was identified from the four *ortho*-coupled protons in its ^{1}H NMR spectrum.

Nakanishi has assigned the absolute configuration for erythrabyssin-II based on circular dichroism (CD) spectroscopy [10]. We obtained a CD spectrum for erycristin (**1**) which coincides closely to that which was obtained by Nakanishi for **3**. Furthermore, erycristin gives a large negative optical rotation at the sodium D line and this finding is in agreement with the findings of Pelter *et al* [21]. Thus we assigned the absolute configuration of erycristin to be 6a-*R* and 11a-*R*.

Further chromatography led to the isolation of crystalline sandwicensin (**2**) [11] and erythrabyssin-II (**3**) [10]. These compounds were easily identified by comparing their physical and spectroscopic properties to those previously reported in the literature. The availability of erythrabyssin-II allowed for a mutually supportive structural interconversion with erycristagallin. Thus, erythrabyssin-II diacetate (**10**) [10] was dehydrogenated with 2,3-dichloro-5,6-dicyano-1,4-benzoquinone in dry ben-

zene [22] to give erycristagallin diacetate (**11**) identical to a sample prepared from the natural product. These spectroscopic properties and chemical transformations further support the structures of erycristin, sandwicensin, erythrabyssin-II and erycristagallin whose assignments were previously based primarily upon spectral properties and biogenetic grounds.

Erycristin (**1**), sandwicensin (**2**) and erythrabyssin-II (**3**) were tested *in vitro* for their antimicrobial activities using an agar dilution-streak method [12]. Both erycristin and erythrabyssin-II are active against *S. aureus* and *M. smegmatis* (Table 1). Their potency is sufficient to account for the potency of the stem bark extracts and the phenolic portions. Sandwicensin was found to be inactive in our experiments.

EXPERIMENTAL

Plant material. E. crista-galli was collected near Pairumani, Department de Cochabamba, Bolivia, in January 1983, and a specimen is on deposit at the Kansas Biological Survey Herbarium in Lawrence.

Extraction. Stem bark powder (2.4 kg) was percolated to exhaustion with 95% EtOH. The ext was concd to a residue (178 g) which was partitioned between 5% aq. HCl and CH_2Cl_2. The CH_2Cl_2 layer was filtered and evapd to dryness to give 45 g of a dark viscous liquid. This was partitioned between 90% MeOH and *n*-hexane to give 35.5 g of MeOH sol biologically active polar materials. The residue was dissolved in CH_2Cl_2 and chromatographed on 300 g silica gel. Elution with C_6H_6–EtOAc (9:1) (7 ml fractions) produced 5.28 g of residue from fractions 7–20 containing two major compounds. The residue obtained was rechromatographed on silica gel (180 g) and eluted with C_6H_6. Examination by TLC showed that fractions 11–18 contained a single substance (240 mg). Repeated crystallization from cyclohexane gave 229 mg of erycristin (**1**), mp 120–121°; $[\alpha]_D = -140$ (MeOH: *c* 1.964) CD $[\theta]_{max}^{MeOH}$ nm (ε): 325 (+17,650), 286 (+30,490), 215 (−216600); UV λ_{max}^{MeOH} nm (log ε): 325 (3.56), 288 (3.86), 227 (415); $\lambda_{max}^{MeOH-HCl}$ 328 (3.56), 288 (3.81), 231 (4.10); $\lambda_{max}^{MeOH-NaOH}$ 374 (3.70), 299 (sh.), 291 (3.83), 249 (3.95); $\lambda_{max}^{MeOH-AlCl_3}$:326 (3.59), 288 (3.85), 232 (4.12); $\lambda_{max}^{MeOH-AlCl_3-HCl}$ 332 (3.60), 288 (3.82), 232 (4.10); IR ν_{max}^{KBr} cm^{-1}: 3445, 1622, 1485, 1468, 1356, 1271, 1165, 1116, 1084, 1030; ^1H NMR ($CDCl_3$): δ 1.67 (3H, *s*), 1.77 (3H, *s*), 1.80 (6H, *s*), 3.30 (2H, *br*), 3.37 (2H, *d*, *J* = 7.2 Hz), 3.53 (1H, *ddd*, *J* = 5.1, 6.3, 11.4 Hz), 3.62 (1H, *dd*, *J* = 10.8 Hz), 3.81 (3H, *s*), 4.22 (1H, *dd*, *J* = 5.1, 10.8 Hz), 5.25 (1H, *br*), 5.32 (1H, *t*, *J* = 7.2 Hz), 5.45 (1H, *d*, *J* = 6.3 Hz), 6.40 (1H, *d*, *J* = 7.8 Hz), 6.41 (1H, *s*), 7.01 (1H, *d*, *J* = 7.8 Hz), 7.27 (1H, *s*), 8.51 (1H, *s*, D_2O exch); ^{13}C NMR ($CDCl_3$): 17.80, 17.93, 22.98, 25.88, 29.14, 29.78, 40.12, 55.98, 66.57, 78.04, 103.04, 103.85, 112.71, 113.29, 119.48, 121.11, 121.64, 122.04, 122.39, 131.52, 132.11, 134.73, 154.97, 155.58, 158.61 ppm. MS *m/z* (rel. int. %): 404.2145 ($[M]^+$, calc for $C_{26}H_{30}O_4$: 406.2142 (100), 350 (23.6), 295 (22.3), 201 (10), 175 (10), 168 (11), 161 (26), 149 (11), 147 (13), 115 (13), 91 (18.6), 77 (13.2), 69 (39.8) and 55 (21.5).

Erycristin monoacetate (**5**). Erycristin (**1**) (30 mg) was dissolved in pyridine (1.5 ml) and Ac_2O (1 ml) was added. The reaction mixt. was kept at room temp. overnight and then poured into cold H_2O (20 ml). The ppt obtained was filtered, washed with H_2O, dried and chromatographed on silica gel using *n*-hexane–C_6H_6 (1:4) to produce the pure monoacetate of **5** as a white solid (21 mg) mp 91–92°; IR ν_{max}^{KBr} cm^{-1}: 2934, 2924, 1765, 1622, 1491, 1458, 1441, 1378, 1265, 1200, 1156, 1113, and 1086. ^1H NMR ($CDCl_3$): 1.66 (3H, *s*), 1.72 (3H, *s*), 1.76 (6H, *s*), 2.30 (3H, *s*), 3.21 (2H, *d*, *J* = 7.8 Hz), 3.26 (2H, *br*), 3.55 (1H, *m*), 3.60 (1H, *dd*, *J* = 10.4 Hz), 3.81 (3H, *s*), 4.22 (1H, *dd*, *J* = 4.5, 10.4 Hz), 5.23 (2H, *br*), 5.46, (1H, *d*, *J* = 6.8 Hz), 6.41 (1H, *d*, *J* = 8.4 Hz), 6.64 (1H, *s*), 7.01 (1H, *d*, *J* = 8.4 Hz), 7.39 (1H, *s*); MS *m/z* (rel. int. %): 448.2252, $[M]^+$ (29.7), calc. for $C_{28}H_{32}O_5$: 448.22749), 407 (16), 406 (47), 350 (15), 295 (14), 203 (14), 175 (14), 161 (21), 149 (23), 147 (11), 128 (12), 115 (12), 98 (15), 91 (21), 84 (14), 84 (26), 79 (12), 77 (15), 69 (70), 68 (26), 55 (21), 49 (37) and 43 (100).

Erycristin monomethyl ether (**6**). Erycristin (**1**) (50 mg), Me_2SO_4 (200 mg), Me_2CO (15 ml) and K_2CO_3 (200 mg) were refluxed for 7 hr, filtered, the residue washed with Me_2CO and the filtrate and washings evapd under red. pres. The residue obtained was dissolved in Et_2O (20 ml), washed well with H_2O (3 × 10 ml), dried and the solvent removed. The residue obtained was further purified on a silica gel column (15 g) using *n*-hexane–Et_2O (7:1) to give pure erycristin monomethyl ether (**6**) (35 mg) as a white solid: mp 119–120°; IR ν_{max}^{KBr} cm^{-1}: 2928, 2916, 1496, 1487, 1446, 1442, 1200, 1084; ^1H NMR ($CDCl_3$): δ 1.67 (3H, *s*), 1.74 (3H, *s*), 1.77 (6H, *s*), 3.39 (4H, *br*), 3.51 (1H, *m*, *J* = 4.8, 6.6, 11.2 Hz), 3.64 (1H, *dd*, *J* = 10.8 Hz), 3.81 (6H, *s*), 4.44 (1H, *dd*, *J* = 4.8, 10.8 Hz), 5.25 (1H, *t*, *J* = 7.2 Hz), 5.34 (1H, *t*, *J* = 7.2 Hz), 5.44 (1H, *d*, *J* = 6.6 Hz), 6.40 (1H, *d*, *J* = 7.8 Hz), 6.43 (1H, *s*), 7.01 (1H, *d*, *J*

Table 1. Antimicrobial activity of *Erythrina crista-galli* components

	Microorganism						
Sample	1	2	3	4	5	6	7
E. crista-galli (Stem bark)	1000 μg/ml	—	—	—	100	—	—
Phenolic fractions	12.5	—	—	—	12.5	—	—
Erycristin	6.25	—	—	—	6.25	—	—
Erythrabyssin-II	3.12	—	—	—	0.78	—	—
Sandwicensin	—	—	—	—	—	—	—
Streptomycin sulphate	5	5	50	2.5	1.25	—	—

Microorganisms: (1) *Staphylococcus aureus* ATCC 13709, (2) *Escherichia coli* ATCC 9637, (3) *Salmonella gallinarum* ATCC 9184, (4) *Klebsiella pneumoniae* ATCC 10031, (5) *Mycobacterium smegmatis* ATCC 607, (6) *Candida albicans* ATCC 10231, (7) *Pseudomonas aeruginosa* ATCC 27853.

= 7.8 Hz), 7.27 (1H, *s*); MS *m/z* 420.22980 ([M]$^+$ (100)), calc for $C_{28}H_{32}O_4$: 420.22988, 364 (18), 363 (15), 201 (9), 161 (18), 128 (6), 115 (6), 91 (9), 69 (27) and 43 (23).

Isolation of sandwicensin (**2**). Fractions 19–25 from the second column were combined, the solvent removed and the residue obtained crystallized from cyclohexane to give **2** as white needles (350 mg): mp 88.5–89.5°; $[\alpha]_D$ −185 (*c* 2 mg/ml) UV λ_{max}^{MeOH} nm (log ε): 287 (3.80), 381 (3.77), 231 (sh) (4.21), 214 (4.57), 212 (4.46) and 210 (4.51); $\lambda_{max}^{EtOH-NaOH}$ 289 (3.91), 251 (4.12), 221 (sh) (4.61), 219 (4.66), 214 (4.57), 213 (4.12), 211 (4.57) and 206 (4.06); IR ν_{max}^{KBr} cm^{-1}: 3474, 1601, 1507, 1485, 1468, 1356, 1242, 1226, 1200, 1152, 1030 and 787; ^{1}H NMR ($CDCl_3$): δ1.64 (3H, *s*), 1.73 (3H, *s*), 3.27 (2H, *br*), 3.48 (1H, *m*), 3.63 (1H, *dd*, *J* = 11 Hz), 3.78 (3H, *s*), 4.20 (1H, *dd*, *J* = 4.8, 11 Hz), 5.21 (1H, *t*, *J* = 7.2 Hz), 5.44 (1H, *d*, *J* = 6.9 Hz), 6.39 (1H, *s*), 6.40 (1H, *d J* = 7.8 Hz), 6.55 (1H, *d*, *J* = 8.4 Hz), 6.98 (1H, *d*, *J* = 7.8 Hz) and 7.38 (1H, *d*, *J* = 8.4 Hz); MS *m/z* (rel. int. %): 338 [M]$^+$ ($C_{21}H_{22}O_4$; 100), 323 (10.5), 295 (20.4), 283 (19), 282 (55), 281 (54), 267 (20), 253 (25), 201 (10.5), 161 (21), 128 (12), 123 (23), 115 (15), 91 (19), 77 (24), 69 (16.5), 55 (15), 53 (13), 43 (32) and 41 (38.5).

Isolation of erythrabyssin-II (**3**). Fractions 53–59 from the first column were combined, concd and the residue obtained crystallized from cyclohexane to give erythrabyssin-II (**3**) as a white solid (1.3 g), mp 152–153°; IR ν_{max}^{KBr} cm^{-1}: 3403, 2926, 2912, 2860, 1622, 1527, 1511, 1217, 1192, 1115, 1019; MS *m/z* (rel. int. %): 392 [M]$^+$ ($C_{25}H_{28}O_4$; 100), 393 (29), 337 (16), 336 (34), 335 (15), 282 (11), 281 (13), 280 (22), 189 (9), 161 (25), 147 (31), 115 (11), 91 (13), 77 (12), 69 (30), 55 (14) and 41 (40).

Erythrabyssin-II diacetate (**10**). Erythrabyssin-II (**3**) (50 mg) was dissolved in pyridine (2 ml) and Ac_2O (3 ml) was added. The reaction mixt was kept at room temp overnight and worked-up as described above. The residue obtained was chromatographed on silica gel (10 g) using *n*-hexane–EtOAc (7:1) to produce pure erythrabyssin-II diacetate (**10**) as white solid (35 mg), mp 80–81°; IR ν_{max}^{KBr} cm^{-1}: 2978, 2928, 1757, 1624, 1603, 1492, 1442, 1338, 1269, 1213, 1113, 1088, 1034, 1018, 981 and 789; ^{1}H NMR ($CDCl_3$): δ1.67 (3H, *s*), 1.72 (3H, *s*), 1.73 (3H, *s*), 1.76 (3H, *s*), 2.28 (3H, *s*), 2.30 (3H, *s*), 3.20 (4H, *d*, *J* = 7.4 Hz), 3.60 (1H, *m*), 3.65 (1H, *dd*, *J* = 11 Hz), 4.20 (1H, *dd*, *J* = 11 Hz), 5.1 (1H, *t*, *J* = 7.4 Hz), 5.25 (1H, *t*, *J* = 7.4 Hz), 5.50 (1H, *d*, *J* = 6 Hz), 6.58 (1H, *d*, *J* = 7.5 Hz), 6.62 (1H, *s*), 7.09 (1H, *d*, *J* = 7.5 Hz) and 7.4 (1H, *s*); MS *m/z* (rel. int. %): 476.21988 ([M$^+$]), calc for $C_{29}H_{32}O_6$: 476.2197; 435 (2), 434 (6), 420 (2), 417 (2), 392 (4), 379 (2), 378 (3), 336 (10), 281 (8), 161 (5), 115 (4), 69 (13) and 43 (100).

Erythrabyssin-II dimethyl ether (**6**). Erythrabyssin-II (**3**) (50 mg), Me_2SO_4 (200 mg), Me_2CO (5 ml) and K_2CO_3 (200 mg) were refluxed for 7 hr and worked-up as described for **5**. The residue obtained was chromatographed on silica gel using *n*-hexane–Et_2O (7:1) to give erythrabyssin-II dimethyl ether (**6**) (32 mg). It showed identical physical and spectral properties when compared to erycristin monomethyl ether (**6**).

DDQ dehydrogenation of erythrabyssin-II diacetate (**10**) *to erycristagallin diacetate* (**11**). Erythrabyssin-II diacetyl ester (**10**) (50 mg) was dissolved in dry C_6H_6 (degassed and saturated with Ar) and DDQ (25 mg) added. The reaction mixt was stirred at room temp under Ar for 24 hr. The solvent was evapd under red. pres. and the residue obtained was chromatographed on silica gel (15 g) using *n*-hexane–Et_2O (4:1) to give pure erycristagallin diacetyl ester (**11**) (8.3 mg) as a white solid identical to the diacetyl ester of the natural compound in its physical and spectral properties.

Conversion of sandwicensin (**2**) *to erycristin* (**1**). *Sandwicensin-3-O-α,α-dimethyl propargyl ether* (**7**). Sandwicensin (**2**) (300 mg), 3-chloro-3-methylbutyne (200, μl), K_2CO_3 (3 g), KI (2 g) and Me_2CO (20 ml) were refluxed for 24 hr under Ar. The reaction mixt was filtered and the residue washed with Me_2CO (10 ml). The filtrate and washings were combined and the solvent removed under red. pres. The residue obtained was chromatographed on silica gel (10 g) using C_6H_6 to give sandwicensin-*O*,α,α-dimethyl propargyl ether (**7**) (180 mg) as a colourless oil: ^{1}H NMR ($CDCl_3$): δ1.64 (9H, *s*), 1.76 (3H, *s*) 2.59 (1H, *s*), 3.28 (2H, *br*), 3.52 (1H, *m*), 3.60 (1H, *dd*, 10.8 Hz), 3.79 (3H, *s*), 4.23 (1H, *dd*, *J* = 4.8, 10.8 Hz), 5.23 (1H, *t*, *J* = 5 Hz), 5.45 (1H, *d J* = 7 Hz), 6.40 (1H, *d*, *J* = 7.8 Hz), 6.87 (1H, *s*), 6.89 (1H, *d*, *J* = 8.4 Hz), 7.01 (1H, *d*, *J* = 8.4 Hz) and 7.43 (1H, *d*, *J* = 7.8 Hz); MS *m/z* (rel int. %): 404.19967 ([M$^+$] (29)), calc for $C_{26}H_{28}O_4$: 404.19860; 338 (29), 295 (9), 283 (13), 282 (38), 281 (45), 267 (110), 253 (12), 161 (15), 147 (16), 128 (12), 123 (11), 115 (12), 91 (15), 77 (17), 67 (100), 55 (13) and 51 (20).

Sandwicensin-3-O-α,α-dimethyl allyl ether (**8**). Sandwicensin-*O*-α,α-dimethyl propargyl ether (**7**) (100 mg) was hydrogenated in MeOH (20 ml) at atmospheric pressure using a Pd-C–$BaSO_4$ catalyst (16.11 mg) poisoned by quinoline (175 mg). The reaction mixt. was filtered and the residue washed with MeOH (3 × 5 ml). The filtrate and washings were combined, the solvent removed and the residue obtained chromatographed on silica gel (10 g) using C_6H_6 to give sandwicensin-3-*O*-α,α-dimethyl allyl ether (**8**) (98 mg) as a viscous liquid; ^{1}H NMR ($CDCl_3$): δ1.29 (2H, *d*, *J* = 9.5 Hz), 1.47 (6H, *s*), 1.70 (3H, *s*), 1.76 (3H, *s*), 3.29 (2H, *br*), 3.51 (1H, *m*), 3.62 (1H, *dd*, *J* = 10.8 Hz), 3.80 (3H, *s*), 4.20 (1H, *dd*. *J* = 4.8, 10.8 Hz), 5.15 (1H, *d*, *J* = 10.2 Hz), 5.24 (1H, *br*), 5.43 (1H, *d*, *J* = 7.0 Hz), 6.15 (1H, *dd*, *J* = 10.2, 18.3 Hz), 6.39 (1H, *d*, *J* = 8 Hz), 6.60 (1H, *s*), 6.7 (1H, *d*, *J* = 8 Hz), 7.00 (1H, *d*, *J* = 8 Hz) and 7.38 (1H, *d*, *J* = 8 Hz); MS *m/z* (rel. int. %): 406.21489 ([M$^+$] (21)), calc for $C_{26}H_{30}O_4$: 406.21425, 338 (54), 295 (11), 283 (15), 282 (48), 281 (31), 253 (12), 161 (13), 147 (13), 123 (8), 115 (7), 91 (12), 69 (60). 55 (11), 43 (13) and 41 (100).

Claisen rearrangement of sandwicensin-3-O-α,α-dimethyl allyl ether (**8**). Sandwicensin-3-*O*-α,α-dimethyl allyl ether (**8**) (50 mg) and *N,N*-diethyl aniline (2 ml) were refluxed for 1 hr. The reaction mixt was dil with cold 5% H_2SO_4 and brine. The residue obtained (50 mg) after removal of solvent was chromatographed on silica gel (15 g) using C_6H_6 and fractions of 4 ml were collected. Fractions 9–12 were combined and the solvent removed to give isoerycristin (**9**) as a gummy residue (44 mg); IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3740, 3050, 1700, 1610, 1520, 1480, 1420, 1220, 1080, and 930; ^{1}H NMR ($CDCl_3$): δ1.66 (3H, *s*), 1.76 (6H, *s*), 1.81 (3H, *s*), 3.30 (2H, *br*), 3.41 (2H *d*, *J* = 6 Hz), 3.52 (1H, *m*), 3.61 (1H, *dd*, *J* = 10.8 Hz), 3.80 (3H, *s*), 4.27 (1H, *dd*, *J* = 4.2, 10.8 Hz), 5.23 (2H, *m*), 5.45 (1H, *d*, *J* = 6 Hz), 6.39 (1H, *d*, *J* = 8 Hz), 6.54 (1H, *d*, *J* = 8 Hz), 7.00 (1H, *d*, *J* = 8 Hz) and 7.29 (1H, *d*, *J* = 8 Hz); MS *m/z* (rel. int.%): 406.21497 ([M]$^+$ (100)), calc for $C_{26}H_{30}O_4$: 406.21424, 407 (36), 351 (10.5), 350 (26), 335 (20), 307 (21), 295 (21), 294 (40), 201 (11), 189 (12), 175 (10), 161 (25), 147 (18), 135 (14), 91 (14), 77 (14), 69 (19), 43 (34) and 41 (42).

Fractions 33–40 were combined and the solvent removed. The residue obtained was crystallized from cyclohexane to give erycristin (**1**) (8.2 mg) as white crystals identical with the natural compound in its physical and spectral properties.

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REFERENCES

1. Fomum, Z. T., Ayafor, J. F. and Wandji, J. (1985) *Phytochemistry* **24**, 3075.

2. Fomum, Z. T., Ayafor, J. F., Wandji, J., Fomban, W. G. and Nkengfack, A. E. (1986) *Phytochemistry* **25**, 757.
3. Fomum, Z. T., Ayafor, J. F., Ifeadike, P. N., Nkengfack, A. E. and Wandji, J. (1986) *Planta Med.* 341.
4. Fomum, Z. T. and Ayafor, J. F. (1986) *J. Chem. Soc. Perkin Trans. I*, 33.
5. Ito, K., Haruna, N., Furukawa, H., Haruna, H. and Ito, M. (1973) *Yakugaku Zasshi* **93**, 1674.
6. Ito, K., Haruna, N., Jinno, Y. and Furukawa, H. (1976) *Chem. Pharm. Bull.* **24**, 52.
7. Imamura, H., Ito, M. and Ohhashi, H. (1981) *Res. Bull. Fac. Agr. Gifu Univ.* **45**, 77.
8. Ingham, J. L. and Markham, K. R. (1980) *Phytochemistry* **19**, 1203.
9. Mitscher, L. A., Ward, J. A., Drake, S. and Rao, G. S. (1984) *Heterocycles* **22**, 1673.
10. Kamat, V. S., Chuo, F. V., Kubo, I. and Nakanishi, K. (1981) *Heterocycles* **15**, 1163.
11. Ingham, J. L. (1980) *Z. Naturforsch.* **35c**, 384.
12. Mitscher, L. A., Leu, R. P., Bathala, M. S., Wu, W. N., Beal J. L. and White, R. (1972) *J. Nat. Prod.* **35**, 157.
13. Mitscher, L. A. (1977) in *Isolation, Separation and Purification of Antibiotics* (Weinstein, G. and Wagman, G., eds) p. 463. Elsevier, Amsterdam.
14. Pachler, L. G. R. and Underwood, W. G. E. (1967) *Tetrahedron* **23**, 1817.
15. Chalmers, A. A., Rall, G. J. H. and Oberholzer, M. E. (1977) *Tetrahedron* **33**, 1735.
16. Jackman, L. M. (1965) *Prog. Chem. Org. Nat. Prod.* **23**, 315.
17. Stout, G. F., Krahn, M. M., Yates, P. and Bhat, H. B. (1968) *Chem. Comm.*. 211.
18. Brown, K. S., Cameron, D. W. and Weiss, U. (1969) *Tetrahedron Letters* 471.
19. Hlubucek, J., Ritchie, E. and Taylor, W. C. (1971) *Aust. J. Chem.* **24**, 2347.
20. Hlubucek, J., Ritchie, E. and Taylor, W. C. (1971) *Aust. J. Chem.* **24**, 2355.
21. Pelter, A. and Amenechi, P. I. (1969) *J. Chem. Soc.* (*C*), 887.
22. Cardillo, G., Cricchio, R. and Merlini, L. (1968) *Tetrahedron* **24**, 4825.