# SESQUITERPENE LACTONES OF *Cephalophora aromatica* (HOOK.) SCHRADER AND THEIR DETERRENT ACTIVITY. THE STEREOSTRUCTURE OF GEIGERININ*

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From species *Cephalophora aromatica* (HOOK.) SCHRADER the authors isolated a series of previously described sesquiterpene lactones *I*—*V* and newly the lactone geigerinin (*VI*). Its structure was confirmed by X-ray analysis and its absolute configuration was determined on the basis of the CD spectra of its complex with $Pr(thd)_3$. The lactones *I*—*VII* were tested for antifeedant activity against main crop pests.

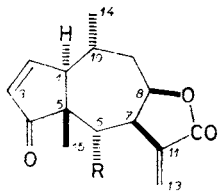
Within the framework of our studies on sesquiterpene lactones of the *Compositae* family we reinvestigated the *Cephalophora aromatica* (HOOK.) SCHRADER species (synonym *Helenium aromaticum* (HOOK.) L.H. BAILEY, *Graemia aromatica* HOOK.) of the *Helianthae tribe*. This species was investigated many years ago by Mexican authors[1] who found helenalin (*I*), aromatin (*II*), aromaticin (*III*) and mexicanin I (*IV*). More recently, we isolated and identified[2] helenalin (*I*) and linifolin A (*V*)**.

We performed a new detailed analysis of the mentioned species (obtained as described in our preceding paper[2]). Along with already described sesquiterpene lactones helenalin (*I*), mexicanin I (*IV*), linifolin A (*V*) and a mixture of aromatin (*II*) and aromaticin (*III*), we isolated and identified also geigerinin (*VI*).

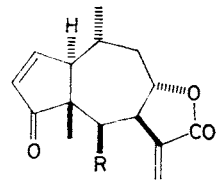
* Part CCIC in the series On Terpenes; Part CCXCVIII: Collect. Czech. Chem. Commun. *54*, 473 (1989)).

** Review articles on sesquiterpene lactones[3,4] describe linifolin A as having *cis*-fused five-membered and seven-membered rings, although it has been reported[5,6] that linofolin A is identical with acetylmexicanin I and thus has the stereostructure V with *trans*-fusion of the homocycles.

In connection with identification of compounds *I*–*VI* we also measured their $^1H$ and $^{13}C$ NMR spectra. Since most of the previously published data represent more or less incomplete sets obtained at lower frequencies, we have summarized



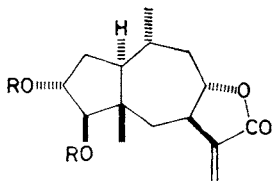
*I*, R = OH
*II*, R = H



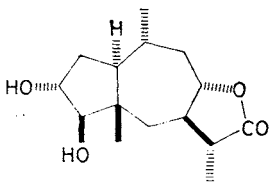
*III*, R = H
*IV*, R = OH
*V*, R = OAc

the $^1H$ NMR data of *I*–*VI* (Tables I and II). The $^{13}C$ NMR spectra of compounds *I* and *IV*–*VI* (Table III) are new, except for helenalin (*I*) for which they are in very good accord with those in refs[7,8], except for the C-5 carbon signal ($\delta$ 57·88) for which a markedly lower chemical shift has been reported[7,8] ($\delta$ 47·8; probably a typographical error).

Geigerinin (*VI*) was first isolated from *Geigeria aspera* HARV. species (tribe *Inulae*) and its originally published[9] constitution was later revised[10]. Our present paper concerns the so far unsolved stereostructure of *VI* (configuration of carbon atoms 1, 3, 4, 5, 8 and 10). Along with the native compound *VI* we also studied its dihydro derivative *VII* and diacetate *VIII*.



*VI*, R = H
*VIII*, R = Ac



*VII*

The 200 MHz $^1H$ NMR spectrum of geigerinin (*VI*) enabled us to discriminate and assign all the proton signals, except the H-1, H-2α, H-2β and H-10 signals which appeared as a non-analysable multiplet in the region $\delta$ 1·7–2·0. A similar distribution of signals was exhibited by the di(trichloroacetylcarbamoyl) (TAC) derivative, (prepared by in situ acylation of *VI* with trichloroacetyl isocyanate (TAI)) as well as the diacetate *VIII*. Still more complicated was the situation with the 11,13-dihydro compound *VII* and its di-TAC derivative, where the difficultly interpretable spectral

region moreover involved the H-7 and H-11 proton signals. The accessible $^1$H NMR (200 MHz) data for all the mentioned compounds are summarized in Table II.

TABLE I

Proton NMR parameters of sesquiterpene lactones *I*— *V* in $CDCl_3$

| Proton | *I*[a] | *II* | *III* | *IV*[b] | *V*[c] |
|---|---|---|---|---|---|
| | | | Chemical shifts, ppm | | |
| H-1 | 3·08 ddd | 2·40 ddd | 2·75 ddd | 2·70 ddd | 2·79 dt |
| H-2 | 7·69 dd | 7·52 dd | 7·63 dd | 7·65 dd | 7·58 dd |
| H-3 | 6·08 dd | 6·11 dd | 6·13 dd | 6·13 dd | 6·12 dd |
| H-6α | — | 1·70 dd | 1·64 dd | 4·52 d | 5·95 d |
| H-6β | 4·47 bdd | 2·47 dd | 2·50 dd | — | — |
| H-7 | 3·56 ddt | 3·24 m | 2·90 m | 3·11 m | 3·27 m |
| H-8 | 4·98 ddd | 4·82 ddd | 4·50 ddd | 4·81 ddd | 4·80 ddd |
| H-9α | 1·82 ddd | —[d] | 1·45 dt | 1·39 dt | 1·43 dt |
| H-9β | 2·27 ddd | —[d] | 2·54 ddd | 2·54 ddd | 2·57 ddd |
| H-10 | 2·07 m | —[d] | —[d] | 2·21 m | 2·24 m |
| H-13 | 6·38 d | 6·30 d | 6·19 d | 6·38 d | 6·28 d |
| H-13′ | 5·80 d | 5·68 d | 5·52 d | 5·69 d | 5·69 d |
| H-14 | 1·27 d | 1·26 d | 1·26 d | 1·25 d | 1·26 d |
| H-15 | 1·00 s | 1·16 s | 1·19 s | 1·24 s | 1·23 s |
| | | | Coupling constants, Hz | | |
| *J*(1,2) | 1·9 | 1·8 | 1·9 | 1·9 | 2·0 |
| *J*(1,3) | 2·9 | 2·9 | 2·9 | 2·8 | 2·8 |
| *J*(1,10) | 11·6 | 12·3 | 10·5 | [d] | 10·6 |
| *J*(2,3) | 6·0 | 6·1 | 6·0 | 6·1 | 6·0 |
| *J*(6α,6β) | — | 15·2 | 14·4 | — | — |
| *J*(6α,7) | — | 6·0 | 7·0 | 5·0 | 4·8 |
| *J*(6β,7) | 1·9 | 11·5 | 11·3 | — | — |
| *J*(7,8) | 7·7 | 7·7 | 9·6 | 9·4 | 9·3 |
| *J*(7, 13) | 3·1 | 2·7 | 3·5 | 3·7 | 3·6 |
| *J*(7, 13′) | 2·8 | 2·3 | 3·2 | 3·2 | 3·2 |
| *J*(8,9) | 8·7 | 3·5 | 3·2 | 3·1 | 3·1 |
| *J*(8,9′) | 2·5 | 11·4 | 11·7 | 11·9 | 11·9 |
| *J*(9α,9β) | 14·7 | —[d] | 13·1 | 13·1 | 13·1 |
| *J*(9,10) | 4·5 | —[d] | 4·4 | 5·0 | 4·9 |
| *J*(9′,10) | 6·2 | —[d] | 11·7 | —[d] | 11·5 |
| *J*(10,14) | 6·7 | 6·5 | 6·8 | 6·5 | 6·6 |

[a] OH: 2·63 d, *J*(OH, 6) 4·4 Hz; [b] mixture of $CDCl_3$ with 10% $CD_3SOCD_3$ used as solvent; [c] OAc: 2·07 s; [d] value of parameter could not be determined.

The missing NMR parameters of protons H-1, H-2α, H-2β and H-10, important for solving the stereochemistry, were obtained at higher frequency (400 MHz) using

TABLE II
Proton NMR parameters of compounds *VI*– *VIII* in $CDCl_3$

| Proton | *VI*[a] | *VII*[b] | *VIII*[c] |
|---|---|---|---|
| | Chemical shifts (TAI-acylation shifts), ppm | | |
| H-3 | 3·93 ddd (1·20) | 3·94 ddd (1·20) | 4·96 dt |
| H-4 | 3·68 d (1·64) | 3·62 bd (1·63) | 5·23 d |
| H-6α | 1·41 dd ([d]) | 1·26 dd (0·05) | 1·28 dd |
| H-6β | 2·26 dd (0·12) | 2·10 dd ([d]) | 2·29 dd |
| H-7 | 2·86 m (0·05) | [d] | 2·86 m |
| H-8 | 4·25 ddd (−0·03) | 4·23 ddd (−0·03) | 4·21 ddd |
| H-9α | 1·36 q ([d]) | 1·25 ddd (0·05) | 1·38 q |
| H-9β | 2·39 ddd (0·02) | 2·34 ddd (0·03) | 2·39 ddd |
| H-13 | 6·17 d (−0·02) | 1·20 d (0·01) | 6·16 d |
| H-13′ | 5·45 d (0·00) | — | 5·41 d |
| H-14 | 0·96 d (0·05) | 0·94 d (0·04) | 0·98 d |
| H-15 | 0·93 s (0·19) | 0·92 s (0·19) | 1·01 s |
| | Coupling constants, Hz | | |
| *J*(2α,3) | 3·4 | 3·4 | 3·0 |
| *J*(2β,3) | 9·5 | 9·4 | 9·2 |
| *J*(3,4) | 7·0 | 7·0 | 6·9 |
| *J*(6α,6β) | 14·5 | 14·2 | 14·6 |
| *J*(6α,7) | 6·1 | 5·8 | 5·9 |
| *J*(6β,7) | 11·9 | 11·3 | 11·8 |
| *J*(7,8) | 9·2 | 9·5 | 9·2 |
| *J*(7, 13) | 3·4 | — | 3·4 |
| *J*(7,13′) | 3·1 | — | 3·1 |
| *J*(8,9α) | 11·7 | 11·6 | 11·7 |
| *J*(8,9β) | 3·1 | 3·4 | 3·1 |
| *J*(9α,9β) | 13·0 | 13·0 | 13·0 |
| *J*(9α,10) | 11·9 | 11·2 | 12·0 |
| *J*(9β,10) | 4·3 | 3·8 | 4·2 |
| *J*(10,14) | 6·5 | 6·6 | 6·5 |

Additional parameters: [a] H-1: 1·95 m, H-2α 1·71 m, H-2β: 1·94 m, H-10: 1·75 m, *J*(1, 2α)10·0, *J*(1,2β) 8·0, *J*(1,10) 11·3, *J*(2α,2β) 15·5 (in *VI*; data obtained from 400 MHz 1D and 2D NMR spectra); NH: 8·63 s, 8·58 s (in TAC-derivative of *VI*); [b] *J*(11,13) 6·9 Hz, NH: 8·40 s and 8·45 s (in TAC-deriative of *VII*); [c] OAc: 2·04 s and 2·12 s; [d] value of the parameter could not be determined.

the homocorrelated 2D-COSY and 2D-J-resolved spectra of geigerinin (*VI*) (for these data see notes in Table II). The large vicinal coupling constant $J(7, 8)$ (9·2 Hz) and allylic long-range coupling constants ($J(7, 13) = 3{\cdot}4$ Hz and $J(7, 13') = 3{\cdot}1$ Hz) prove the *trans*-fusion of the lactone ring in *VI* (see the "lactone rule" in ref.[11]). Also the relative configuration of protons H-1 and H-10 should be trans, according to the high value of $J(1, 10)$ (11·3 Hz). Selective saturation of methyl protons H-15 resulted in an NOE enhancement of the H-6β (4%), H-8 (8%), H-3 (6%) and H-10 (5%) signals, whereas no effect on the H-1 signal was observed. This indicates *cis*-orientation of the methyl on the C-5 atom relative to the H-8, H-1 and H-3 atoms and *trans*-annelation of the five- and seven-membered homocycles. Under assumption of β-configuration on C-7, this means the relative configurations 8α-OR, 10α-$-CH_3$, 1α-H, 3α-OH and 5β-$CH_3$. Thus, the only undetermined configuration was that of the hydroxyl in position 4 of the five-membered ring. Since the coupling constant $J(4, 3) = 7{\cdot}0$ Hz. gave no unequivocal decision, the conformation of the five-membered ring had to be considered first. The *trans*-annelation limits its conformation approximately to the region ${}^3E \leftrightarrow E_3$ of the pseudorotation trajectory which is further considerably reduced by the vicinal coupling constants of protons

TABLE III

$^{13}$C NMR Chemical shifts of helenalin (*I*), mexicanin I (*IV*), linifolin A (*V*) and geigerinin (*VI*)

| Carbon | *I*[a] ($CDCl_3$) | *IV* ($CDCl_3$ + 10% $CD_3OD$) | *V* ($CDCl_3$) | *VI*[b] ($CD_3OD$) |
|---|---|---|---|---|
| C-1 | 51·36 (1·83) | 53·03[c] | 51·92 | 46·12[c] |
| C-2 | 164·10 (−1·57) | 163·05 | 160·58 | 45·27 |
| C-3 | 129·83 (−0·52) | 130·23 | 130·28 | 83·40 |
| C-4 | 212·34 (−3·80) | 215·19 | 210·70 | 90·74 |
| C-5 | 57·88 (−2·33) | 57·08 | 55·30 | 45·97 |
| C-6 | 74·06 (7·68) | 65·09 | 65·52 | 39·42[d] |
| C-7 | 50·84 (−3·75) | 53·44[c] | 53·28 | 46·46[c] |
| C-8 | 78·31 (−0·19) | 76·08 | 75·58 | 75·28 |
| C-9 | 39·46 (0·69) | 44·35 | 44·04 | 36·89[d] |
| C-10 | 26·11 (−0·05) | 27·21 | 27·14 | 31·12 |
| C-11 | 137·91 (−0·73) | 135·15 | 134·05 | 143·02 |
| C-12 | 169·89 (−0·63) | 166·57 | 169·01 | 172·24 |
| C-13 | 123·08 (2·43) | 122·98 | 122·99 | 119·75 |
| C-14 | 20·15 (−0·30) | 20·89 | 20·74 | 20·87 |
| C-15 | 18·67 (−0·34) | 19·80 | 19·69 | 19·05 |

[a] TAI acylation induced shift values are given in parentheses; [b] acetyl group: 169·38 (C = 0), 21·43 ($CH_3$); [c,d] assignment of signals with the same symbols can be interchanged.

in positions 1, 2 and 3 into the narrow region ${}_4E \leftrightarrow E^5$ where the torsion angles between the H-3 and H-4 protons can be about 25–45° for the *cis*-orientation (3α,4α-OH) or about 145–165° for the *trans*-orientation (3α,4β-OH). The latter alternative (4β-OH) obviously better fits the found coupling constant ($J(3, 4) = 7{\cdot}0$ Hz), explaining at the same time the absence of NOE for the H-4 signal when the C(5)-$CH_3$ signal was saturated (vide supra). According to the $^1$H NMR data, the relative configuration of geigerinin is depicted by formula *VI*.

The absolute configuration of *VI* was derived from the CD spectrum of its complex with tris(dipivaloylmethanato)praseodyme. The found negative helicity in the CD spectrum of the complex (314 nm; $\Delta\varepsilon$ −0·2) indicates the *R*-confiugration at the C(3) and C(4) carbon atoms. The low value of $\Delta\varepsilon$ is obviously due to the conformation of the five-membered homocycle: the torsion angle O(3)—C(3)—C(4)—O(4) in crystal has been found to be 80° (vide infra) and vicinal coupling constants $J$(H, H) show a similar spatial arrangement of compound *VI* in solution (Table IV). The γ-lactone group with conjugated exomethylene double bond gives rise to a Cotton

TABLE IV

Comparison of interproton torsion angles found in crystal (X-ray *A* and *B* mean different conformers) and calculated using observed values of the corresponding $^3J$(H, H) from $^1$H NMR spectrum in $CDCl_3$ solution

| Atoms | X-ray (crystal) $\Phi(H_i, H_j)$ | | $^1$H NMR (solution) | |
|---|---|---|---|---|
| | *A* | *B* | $J$(obs.) | $\Phi$(calc.)[a] |
| H(1)–C(1)–C(2)–H(2α) | −23·0 | −21·1 | 10·0 | −24 |
| H(1)–C(1)–C(2)–H(2β) | −143·4 | −141·9 | 8·0 | −142 |
| H(2α)–C(2)–C(3)–H(3) | −126·3 | −126·7 | 3·4 | −123 |
| H(2β)–C(2)–C(3)–H(3) | −5·6 | −5·6 | 9·5 | −19 |
| H(3)–C(3)–C(4)–H(4) | 149·4 | 154·9 | 7·0 | 149 |
| H(6α)–C(6)–C(7)–H(7) | 49·9 | 54·2 | 6·1 | 45 |
| H(6β)–C(6)–C(7)–H(7) | 167·9 | 172·4 | 11·9 | 166 |
| H(7)–C(7)–C(8)–H(8) | 139·3 | 139·4 | 9·2 | 157 |
| H(8)–C(8)–C(9)–H(9α) | −169·1 | −172·2 | 11·7 | 180 |
| H(8)–C(8)–C(9)–H(9β) | 72·7 | 70·1 | 3·1 | 57 |
| H(9α)–C(9)–C(10)–H(10) | −174·2 | −174·5 | 11·9 | 180 |
| H(9β)–C(9)–C(10)–H(10) | −56·3 | −56·7 | 4·3 | −53 |
| H(10)–C(10)–C(1)–H(1) | −166·0 | −163·5 | 11·3 | −165 |

[a] From $J$(obs.) using Karplus-type equation adjusted for cyclohexane (ref.[25]) and common corrections for substituent electronegativities ($J$(corr.) = $J$(obs)/(1−0·1 $\Delta E$)). The $\Phi$-values closest to those from X-ray are given only.

effect (CE) at 262 nm ($\Delta\varepsilon$ +0·14) in the CD spectrum of geigerinin and at 258 nm ($\Delta\varepsilon$ +0·33) in the spectrum of its diacetate *VIII*. For the *trans*-annelated C(8)--lactone, these values are compatible with the *R*-configuration of C(7) when interpreted according to the rule of Geissman and collaborators[12]. These data, together with the relative configuration, derived from the NMR spectra and the X-ray diffraction (vide infra), lead to formula *VI* as the actual stereostructure of geigerinin. The absolute configuration of the native compound *VI* and the CD spectrum of dihydro derivative of geigerinin have also determined the absolute configuration at the C(11) atom in this dihydro derivative, depicted by formula *VII*.

### X-*Ray Structure Determination of Geigerinin* (VI)

In the solid state the structure of geigerinin (*VI*) contains two crystallographically distinct but chemically equivalent molecules (hereafter molecules A and B). The final atomic parameters are collected in Tables V and VI. The overall conformation and the absolute configuration of both molecules is illustrated in Fig. 1 and is also

TABLE V
Final fractional coordinates of carbon and oxygen atoms in geigerinin (*VI*)

| Atom | *x* | *y* | *z* | *x* | *y* | *z* |
|---|---|---|---|---|---|---|
| | | Molecule *A* | | | Molecule *B* | |
| C(1) | −0·1533(3) | −0·0396(6) | −1·2701(2) | −0·3306(3) | −0·5654(6) | −0·7613(3) |
| C(2) | −0·2154(3) | −0·2056(6) | −1·2905(3) | −0·3088(3) | −0·7067(6) | −0·8368(3) |
| C(3) | −0·2849(3) | −0·2311(6) | −1·2040(2) | −0·3408(3) | −0·6347(6) | −0·9374(3) |
| C(4) | −0·2506(3) | −0·0934(6) | −1·1329(2) | −0·3940(2) | −0·4708(6) | −0·9148(2) |
| C(5) | −0·2195(2) | 0·0591(6) | −1·1967(2) | −0·3361(3) | −0·3947(6) | −0·8241(2) |
| C(6) | −0·1669(3) | 0·1968(6) | −1·1316(3) | −0·3954(3) | −0·2443(6) | −0·7823(2) |
| C(7) | −0·0685(3) | 0·2707(6) | −1·1697(2) | −0·4143(3) | −0·2526(6) | −0·6734(2) |
| C(8) | −0·0770(3) | 0·3327(6) | −1·2739(3) | −0·3176(3) | −0·2734(7) | −0·6105(3) |
| C(9) | −0·0499(3) | 0·2018(7) | −1·3498(3) | −0·2895(3) | −0·4574(7) | −0·5866(3) |
| C(10) | −0·1248(3) | 0·0540(7) | −1·3642(3) | −0·2553(3) | −0·5679(7) | −0·6725(3) |
| C(11) | −0·0305(3) | 0·4266(6) | −1·1164(3) | −0·4609(3) | −0·0956(7) | −0·6301(3) |
| C(12) | 0·0114(3) | 0·5433(7) | −1·1890(4) | −0·4177(4) | −0·0800(9) | −0·5304(3) |
| C(13) | −0·0351(3) | 0·4660(7) | −1·0225(3) | −0·5267(4) | 0·0144(8) | −0·6666(3) |
| C(14) | −0·0776(4) | −0·0737(8) | −1·4366(3) | −0·2395(4) | −0·7507(8) | −0·6348(4) |
| C(15) | −0·3140(3) | 0·1382(7) | −1·2478(3) | −0·2296(3) | −0·3378(7) | −0·8517(3) |
| O(3) | −0·2740(2) | −0·4022(5) | −1·1669(2) | −0·4054(2) | −0·7469(5) | −0·9946(2) |
| O(4) | −0·3248(2) | −0·0457(5) | −1·0642(2) | −0·3996(2) | −0·3567(5) | −0·9963(2) |
| O(8) | −0·0062(2) | 0·4785(0) | −1·2790(2) | −0·3392(2) | −0·1884(6) | −0·5183(2) |
| O(12) | 0·0551(3) | 0·6794(6) | −1·1770(3) | −0·4441(3) | −0·0112(7) | −0·4650(2) |

described in terms of the torsion angles listed in Table VII. As seen in Fig. 1, the stereostructure of the molecule of geigerinin (*VI*) is as follows. The C(5)-methyl group is β-oriented and *trans* with respect to the α-hydrogen atom at C(1), indicating *trans*-junction of the cyclopentanediol and cycloheptane rings. The C(7) α-hydrogen is *trans* to the C(8) β-hydrogen, forming a *trans*-fused α,β-unsaturated γ-lactone ring. Thus the three ring system of pseudoguaianolide skeleton is of the *trans-anti-trans* configuration. The methyl group at C(10) is α-oriented and *cis* to the C(1)—αH, a feature characteristic of the helenanolide subgroup of pseudoguaianolides. The hydroxyl group at C(4) is β-oriented and is *cis* to the C(5)-β$CH_3$ and *trans* to the α-oriented hydroxyl group at C(3).

Two crystallographically independent molecules of geigerinin are conformationally similar but not the same. They differ mainly in the mode of puckering of the α,β--unsaturated γ-lactone ring. While in molecule A the conformation is intermediate

TABLE VI

Final fraction coordinates (.$10^3$) of hydrogen atoms in geigerinin (*VI*)

| Atom | *x* | *y* | *z* | *x* | *y* | *z* |
|---|---|---|---|---|---|---|
| | | Molecule *A* | | | Molecule *B* | |
| H(1) | −78 | −60 | −1 238 | −401 | −582 | −724 |
| H(2α) | −165 | −314 | −1 297 | −352 | −821 | −821 |
| H(2β) | −260 | −192 | −1 357 | −229 | −737 | −835 |
| H(3) | −365 | −219 | −1 222 | −276 | −618 | −982 |
| H(4) | −189 | −140 | −1 087 | −473 | −491 | −899 |
| H(6α) | −150 | 139 | −1 061 | −468 | −238 | −820 |
| H(6β) | −219 | 302 | −1 123 | −353 | −128 | −796 |
| H(7) | −18 | 162 | −1 160 | −465 | −362 | −672 |
| H(8) | −155 | 364 | −1 290 | −255 | −221 | −650 |
| H(9α) | 23 | 147 | −1 329 | −355 | −519 | −556 |
| H(9β) | −44 | 268 | −1 419 | −228 | −455 | −532 |
| H(10) | −195 | 108 | −1 392 | −186 | −515 | −699 |
| H(13a) | −61 | 395 | −963 | −571 | 1 | −722 |
| H(13b) | 3 | 589 | −1 000 | −561 | 119 | −635 |
| H(14a) | −129 | −173 | −1 462 | −204 | −826 | −690 |
| H(14b) | −13 | −132 | −1 399 | −309 | −812 | −614 |
| H(14c) | −53 | 0 | −1 498 | −189 | −740 | −572 |
| H(15a) | −350 | 44 | −1 295 | −186 | −446 | −876 |
| H(15b) | −294 | 250 | −1 290 | −190 | −279 | −790 |
| H(15c) | −366 | 176 | −1 192 | −238 | −245 | −910 |
| O(3)H | −315 | −394 | −1 105 | −369 | −844 | −1 018 |
| O(4)H | −337 | −148 | −1 026 | −466 | −304 | −1 009 |

TABLE VII

Torsion angles of non-hydrogen atoms with standard deviations in parentheses

| Atoms | Angle, deg | |
|---|---|---|
| | molecule *A* | molecule *B* |
| C(5)–C(1)–C(2)–C(3) | −22·4(4) | −20·3(3) |
| C(2)–C(1)–C(5)–C(4) | 41·4(3) | 39·9(3) |
| C(2)–C(1)–C(5)–C(6) | 158·5(4) | 157·3(3) |
| C(2)–C(1)–C(5)–C(15) | −74·2(4) | −75·7(3) |
| C(10)–C(1)–C(2)–C(3) | −150·0(4) | −148·3(4) |
| C(2)–C(1)–C(10)–C(9) | −170·7(4) | −166·8(4) |
| C(2)–C(1)–C(10)–C(14) | −49·5(4) | −45·7(4) |
| C(10)–C(1)–C(5)–C(4) | 165·8(3) | 165·1(3) |
| C(10)–C(1)–C(5)–C(6) | −77·0(4) | −77·5(4) |
| C(10)–C(1)–C(5)–C(15) | 50·3(4) | 49·5(4) |
| C(5)–C(1)–C(10)–C(9) | 69·8(5) | 73·0(5) |
| C(5)–C(1)–C(10)–C(14) | −169·0(4) | −165·9(4) |
| C(1)–C(2)–C(3)–C(4) | −6·9(4) | −8·8(4) |
| C(1)–C(2)–C(3)–O(3) | −128·9(4) | −129·9(4) |
| C(2)–C(3)–C(4)–C(5) | 34·2(3) | 35·3(4) |
| C(2)–C(3)–C(4)–O(4) | 157·5(4) | 160·1(3) |
| O(3)–C(3)–C(4)–C(5) | 153·4(3) | 158·7(3) |
| O(3)–C(3)–C(4)–O(4) | −83·2(4) | −76·5(3) |
| C(3)–C(4)–C(5)–C(1) | −47·4(3) | −47·1(3) |
| C(3)–C(4)–C(5)–C(6) | −170·9(3) | −169·9(3) |
| C(3)–C(4)–C(5)–C(15) | 69·3(4) | 69·2(4) |
| O(4)–C(4)–C(5)–C(1) | −172·3(3) | −170·7(3) |
| O(4)–C(4)–C(5)–C(6) | 64·3(4) | 66·6(4) |
| O(4)–C(4)–C(5)–C(15) | −55·6(4) | −54·4(4) |
| C(1)–C(5)–C(6)–C(7) | 22·2(4) | 17·9(4) |
| C(4)–C(5)–C(6)–C(7) | 133·7(4) | 129·0(4) |
| C(15)–C(5)–C(6)–C(7) | −106·1(4) | −109·8(4) |
| C(5)–C(6)–C(7)–C(8) | 50·7(4) | 55·5(4) |
| C(5)–C(6)–C(7)–C(11) | 168·4(4) | 173·1(4) |
| C(6)–C(7)–C(8)–C(9) | −92·2(4) | −92·7(4) |
| C(6)–C(7)–C(8)–O(8) | 148·0(4) | 150·1(4) |
| C(6)–C(7)–C(11)–C(12) | −141·9(4) | −145·8(4) |
| C(6)–C(7)–C(11)–C(13) | 35·0(5) | 33·9(6) |
| C(11)–C(7)–C(8)–C(9) | 143·5(4) | 141·9(4) |
| C(11)–C(7)–C(8)–O(8) | 23·7(4) | 24·7(4) |
| C(8)–C(7)–C(11)–C(12) | −17·9(4) | −22·0(4) |
| C(8)–C(7)–C(11)–C(13) | 159·0(5) | 157·8(5) |
| C(7)–C(8)–C(9)–C(10) | 70·5(4) | 68·6(4) |
| C(7)–C(8)–O(8)–C(12) | −22·1(4) | −19·3(5) |
| O(8)–C(8)–C(9)–C(10) | −171·7(4) | −175·3(4) |

TABLE VII
(*Continued*)

| Atoms | Angle, deg | |
|---|---|---|
| | molecule *A* | molecule *B* |
| C(9)–C(8)–O(8)–C(12) | −146·1(4) | −141·7(4) |
| C(8)–C(9)–C(10)–C(1) | −51·3(4) | −52·2(4) |
| C(8)–C(9)–C(10)–C(14) | −174·3(4) | −175·2(4) |
| C(7)–C(11)–C(12)–O(8) | 5·2(4) | 11·5(5) |
| C(7)–C(11)–C(12)–O(12) | −175·4(5) | −168·0(6) |
| C(13)–C(11))–C(12)–O(8) | −171·9(4) | −168·3(5) |
| C(13)–C(11)–C(12)–O(12) | 7·5(6) | 12·2(7) |
| C(11)–C(12)–O(8)–C(8) | 10·6(4) | 5·1(5) |

between C(7)α, C(8)β half-chair and C(8)β envelope, in molecule B it is intermediate between C(7)α, C(8)β half-chair and C(7)α envelope. The lactone rings in both molecules are flattened, the average internal torsion angle being 16·0° and 15·4° for molecules A and B, respectively. The conformation of the five-membered carbocyclic ring is the same in both molecules and is intermediate between C(5)β envelope and C(5)β, C(4)α half-chair. The ring is puckered with average endocyclic torsion angle 30·4° and 30·3°, for molecules A and B, respectively. The seven-membered ring

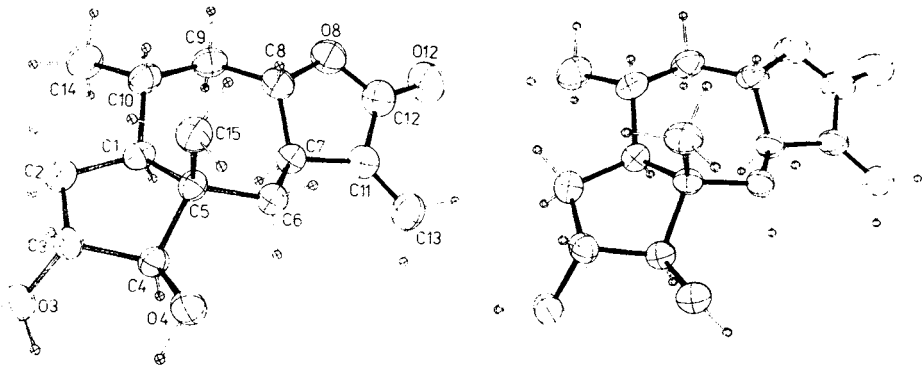
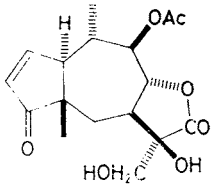



FIG. 1

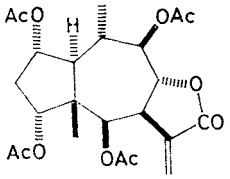
A perspective view of two molecules of geigerinin (*VI*) present in the asymmetric part of the unit cell. The numbering system is the same in both independent molecules and is given for molecule A. Non-hydrogen atom ellipsoids were drawn at the 40% probability level. Hydrogen atom spheres are on arbitrary scale

conformation is also similar in both molecules and approximates to a twist-chair form with the $C_2$ axis passing through C(6) and the midpoint of the C(10—C(9) bond. This type of conformation was also found in other helenanolides with a *trans* lactone ring closed at C(8), namely amblyodiol (*IX*) (ref.[13]), diacetylspathulin (*X*) (ref.[14]) and the nor-derivative of carpesiolin (*XI*) (ref.[15]). However, the seven--membered ring in geigerinin shows considerable deviation from the ideal $C_2$ form.

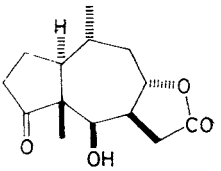
The molecules A and B also differ in the orientation of their O—H bonds as a result of their different involvement in the hydrogen bond formation (vide infra). This is illustrated in Fig. 2 which shows a superposition of the two molecules by the least--squares method. The molecules were fitted together using a routine described by Nyburg[16].




*IX*




*X*




*XI*

The molecular packing is illustrated in Fig. 3. This shows a distinct separation of hydrophobic and hydrophilic regions within the cell. In the hydrophilic region there are several hydrogen bonds of the O—H···O type linking molecules A and B into infinite channels along the *y* axis. The geometrical details of the hydrogen bonding together with the symmetry code are given in Table VIII. Each molecule of type A donates both its hydroxyl hydrogens to the hydroxyl oxygen O(4B) of the B molecule, and accepts one of the protons O(3B)—H from the B molecule translated over *y*. Moreover, the hydrogen bond O(4B)—H···O(3B) links the molecules of type B into

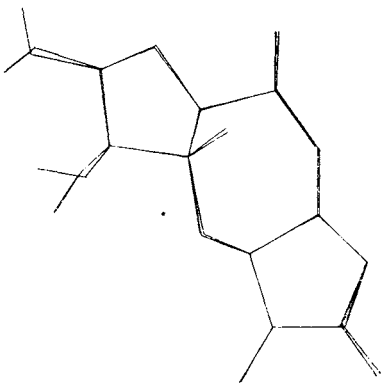


FIG. 2

Least-squares fitting of two crystallographically distinct molecules of geigerinin. The molecules differ in orientation of their O—H bonds as a result of their different involvement in hydrogen bond formation

helices, along the $2_1$ screw axis. In this way each molecule of type A is hydrogen--bonded to two translationally equivalent molecules of type B, while each molecule

TABLE VIII

Distances (in Å) and angles (in °) in the hydrogen bonding system

| D—H···A acceptor position | D—H | H···A | D···A | <D—H···A |
|---|---|---|---|---|
| O(3A)—H(O3A)···O(4B) *x, y, z* | 1·02 | 1·92 | 2·934(4) | 173 |
| O(4A)—H(O4A)···O(4B) *x, y, z* | 0·97 | 1·87 | 2·786(5) | 156 |
| O(3B)—H(O3B)···O(4A) *x*, −1 + *y*, *z* | 0·96 | 1·80 | 2·741(5) | 169 |
| O(4B)—H(O4B)···O(3B) −1 − *x*, 1/2 + *y*, −2 − *z* | 0·98 | 1·76 | 2·714(4) | 164 |

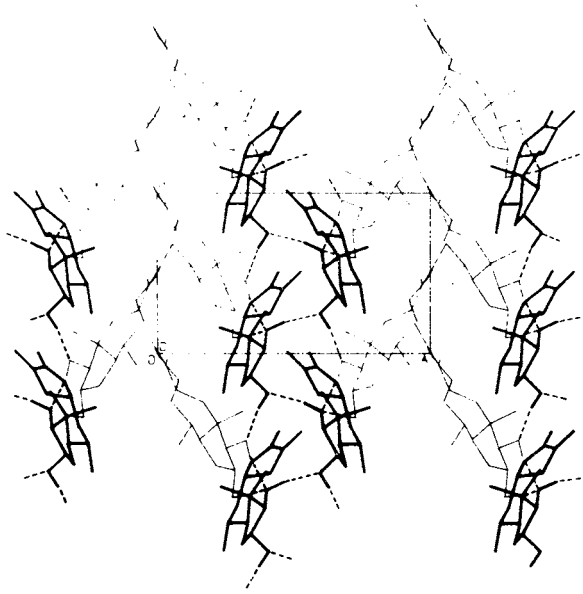


FIG. 3

Packing of the molecules of geigerinin in the crystal, as viewed along the *c*-axis. Thin lines denote molecules A, thick lines denote molecules B. For details of hydrogen bonds (broken lines) see Table VIII

of type B is hydrogen-bonded to four nearest neighbours: two of type A and two of type B. In this hydrogen-bonding pattern, an infinite hydrogen bond chain, composed exclusively of hydroxyl groups, can be distinguished. Such endless chain of OH···OH··· bonds is energetically more favourable than the sum of the individual bonds because the cooperative effect comes into play[17]. Due to this cooperativity the oxygen of a hydroxyl group that accepts a hydrogen bond is a stronger hydrogen--bond donor, and this effect is clearly seen in the crystal structure of geigerinin. Of the four hydroxyl groups two are symmetrical donor–acceptor groups (O(4A)—H and O(3B)—H,) one is a single donor and a double acceptor (O(4B)—H), and one acts exclusively as a proton donor (O(3A)—H). The latter hydroxyl group forms the longest hydrogen bond in the structure indicative of its lower proton-donor ability caused by the lack of the hydrogen-bond conjugation.

*Biological Testing of Lactones* I–IV, VI *and* VII *for Antifeedant Activity*

We have examined the feeding deterrency of mexicanin I (*IV*), geigerinin (*VI*), 11β*H*, 13-dihydrogeigerinin (*VII*), mixture of aromatin and aromaticin (*II* and *III*), and helenalin (*I*) as a standard according to the method described in the previous communications[18,19]. All these substances showed feeding deterrent activity – with the exception of 11β*H*,13-dihydrogeigerinin – against selected storage pests, i.e. adults of *Sitophilus granarius* L. and *Tribolium confusum* Duv. and larvae of *T. confusum* and *Trogoderma granarium* Ev. (Table IX). Very high activity (the value of „total coefficients" in Table IX between 150–200) was exhibited against

Table IX

Deterrent activity of compounds *I*, *IV*, *VI* and *VII* and of the mixture of compounds *II* and *III*

| Compound | Coefficients | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | rel. | abs. | tot. | rel. | abs. | tot. | rel. | abs. | tot. | rel. | abs. | tot. |
| | Adults of *Sithophilus gran.* | | | Adults of *Tribolium conf.* | | | Larvae of *Tribolium conf.* | | | Larvae of *Trogoderma gran.* | | |
| *I* | 100 | 100 | 200 | 100 | 43 | 143 | 100 | 86 | 186 | 98 | 74 | 172 |
| *IV* | 100 | 92 | 192 | 86 | 45 | 131 | 90 | 66 | 156 | 100 | 94 | 194 |
| *VI* | 84 | 33 | 117 | 100 | 46 | 146 | 100 | 83 | 183 | 94 | 69 | 163 |
| *VII* | 66 | −2 | 64 | —[a] | —[a] | —[a] | 84 | 51 | 135 | 42 | −12 | 30 |
| *II* + *III* | | | 149 | | | 162 | | | 157 | | | 136 |

[a] Not determined.

adults of *Sitophilus granarius* by mexicanin I (*IV*) and against larvae of *Trogoderma granarium* and *Tribolium confusum* by mexicanin I (*IV*) and geigerinin (*VI*). 11β*H*,13-Dihydrogeigerinin (*VII*) showed in general lower antifeedant activity than the mentioned native sesquiterpene lactones with the exomethylene double bond in their γ-lactone ring and, on the contrary, it showed feeding attractivity for adults of Sitophilus granarius and larvae of *Trogoderma granarium*.

## EXPERIMENTAL

The melting points were determined on a Kofler block and are not corrected. Silica gel for column chromatography was prepared according to Pitra and Štěrba (30–60 mμ, deactivated by addition of 11% of water). Thin-layer chromatography was carried out on silica gel G according to Stahl (Merck). The IR spectra were determined in chloroform on a Perkin–Elmer PE 580 spectrophotometer. The $^1$H and $^{13}$C NMR spectra (at 200 MHz or 50·3 MHz, resp) were measured on a Varian XL-200 instrument. Chemical shifts and coupling constants of protons were obtained by first order analysis from the expanded spectra with resolution enhancement processing of FID. 2D NMR spectra of geigerinin *VI* (homocorrelated 2D-COSY and homonuclear 2D-J-resolved) were measured on a Varian VXR-400 instrument (at 400 MHz in $CDCl_3$) using standard Varian pulse sequences COSY and HOM2DJ. The mass spectra were measured on an AEI MS 902 spectrometer. Optical rotations were determined in methanol on an objective polarimeter Perkin–Elmer 141; the CD spectra on a Roussel-Jouan CD 185 dichrographe in methanol.

### Isolation of Helenalin (*I*), Aromatin (*II*), Aromaticin (*III*), Mexicanin I (*IV*), Linifolin A (*V*) and Geigerinin (*VI*)

The aerial part of species *Cephalophora aromatica* (Hook.) Schrader (*Compositae* family, *Helianthae* tribe) was collected in August 1984 (voucher No 235/84 is deposited in the Herbarium of Medical Plants, Medical Academy, Poznań, Poland), dried and the material (540 g) was processed as described[2].

The chloroform extract (12·5 g) was chromatographed on a column of silica gel (200 g); elution with chloroform afforded a mixture of aromatin (*II*) and aromaticin (*III*). Further elution with chloroform–ethyl acetate (9 : 1) gave linifolin *A* (V), m.p. 200–202°C, $[\alpha]_D$ +30° (*c* 1·0), identical in all respects with an authentic sample[2]. Elution with chloroform–ethyl acetate (85 : 15) furnished a mixture of helenalin (*I*) and mexicanin I (*IV*). This mixture was rechromatographed on silica gel column (80 g) in chloroform–acetone (85 : 15) to afford helenalin (*I*; 1·2 g), m.p. 172°C, $[\alpha]_D$ −102° (*c* 1·0), identical in all respects with a standard[2]. Further fractions gave mexicanin I (*IV*; 0·8 g), m.p. 255–258°C, $[\alpha]_D$ +40° (*c* 1·0) (reported[1] m.p. 260–263°C, $[\sigma]_D$ +57°).

Continuation of the original chromatography in chloroform–ethyl acetate (70 : 30) gave geigerinin (*VI*; 0·35 g), m.p. 202–203°C, $[\alpha]_D$ −9° (*c* 1·0) (reported[9] m.p. 202–203°C, $[\alpha]_D$ −10·7°). CD spectrum: 262 nm, Δε +0·14. CD spectrum of the geigerinin–Pr(thd)$_3$ complex: 314 nm, Δε −0·2.

### 11β*H*,13-Dihydrogeigerinin (*VII*)

A suspension of sodium borohydride (0·1 g) in ethanol (5 ml) was added to a solution of geigerinin (*VI*; 0·1 g) in ethanol (100 ml) and the mixture was set aside at room temperature for

1 h. Water was added, the mixture was acidified to pH 6 with 5% $H_2SO_4$ and the product was extracted with chloroform. The combined chloroform extracts were worked up in the usual manner. The residue after evaporation of the solvent afforded 11β*H*,13-dihydrogeigerinin (*VII*; 65 mg), m.p. 157–160°C (methanol–ether) (reported[9] m.p. 152–154°C). CD spectrum: 217 nm, $\Delta\varepsilon$ −1·3.

3,4-Diacetylgeigerinin (*VIII*)

Acetic anhydride (0·5 ml) was added to a solution of geigerinin (*VI*; 70 mg) in pyridine (1 ml). After standing at room temperature for 24 h, the mixture was diluted with water, acidified with 5% $H_2SO_4$ and extracted with ether. The combined ethereal extracts were worked up in the usual manner. After evaporation of the solvent, the residue gave 3,4-diacetylgeigerinin (*VIII*; 62 mg), m.p. 158–160°C, $[\alpha]_D$ −82·8° (*c* 1·0) (reported[9] m.p. 158°C, $[\alpha]_D$ −33·9°). CD spectrum: 258 nm, $\Delta\varepsilon$ +0·3.

X-Ray Structure Determination

Single crystals of geigerinin were obtained by slow crystallization from 1-propanol. They are monoclinic, space group $P2_1$, with $a = 13{\cdot}208(1)$, $b = 7{\cdot}782(1)$, $c = 13{\cdot}737(2)$ Å, $\beta = 91{\cdot}765(8)°$, $d_m = 1{\cdot}25$ g/cm$^3$ for $z = 4$. The intensity data were measured on a Syntex $P2_1$ diffractometer using graphite monochromated $CuK_\alpha$ radiation ($\lambda = 1{\cdot}5418$ Å). Of the 3 804 reflections, measured in four octants (according to the point group 2), 3 542 had $I \geqq 1{\cdot}96\sigma(I)$ and were considered observed. The background and integrated intensity for each reflection were evaluated from a profile analysis according to the Lehmann and Larsen method[20] using the PRARA program[21]. Lorentz and polarization corrections were applied but no absorption corrections were made ($\mu(CuK_\alpha)$ 0·65 mm$^{-1}$]. The solution and refinement of the structure was based, at first, on the averaged set of reflections (according to the Laue group *2/m*), with no corrections for anomalous dispersion effects. After enantiomorph definition (vide infra) the refinement was carried out on the full data set with oxygen and carbon atoms allowed for dispersion.

The solution of the structure was not straightforward. Initial attempts to determine the structure with the MULTAN program package[22] was unsuccessful. A recognizable molecular fragment found on one of the E-maps was used as input to the DIRDIF[23] program which gave all the nonhydrogen atoms present in the asymmetric part of the unit cell. The usual sequence of isotropic and anisotropic full matrix least-squares was followed. All non-hydrogen atoms were refined anisotropically. The hydrogen atoms were placed at calculated positions and were subjected to constrained refinement, with the exception of the hydroxyl and methylene hydrogens which were located from a difference Fourier map and were kept fixed during the refinement. In the final stages of the refinement an empirical isotropic extinction parameter $x$ was introduced to correct the calculated structure factors by multipyling them by a factor $1 - xF_c/\sin\Theta$ and it refined to a value $1{\cdot}9(1) \cdot 10^{-6}$. The function minimized was $\sum w(|F_o| - |F_c|)^2$ with $w = 1/[\sigma^2(F_o) + 0{\cdot}0003F_o]$, where $\sigma$ is the standard deviation of the observed amplitudes, based on counting statistics. Convergence attained at $R = 0{\cdot}054$ ($wR = 0{\cdot}075$) for 3 541 observed reflections. The refinement was carried out using the SHELX-76 program[24].

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