

EUPONIN: A NEW EPOXY SESQUITERPENE LACTONE
 INHIBITING INSECT DEVELOPMENT FROM *EUPATORIUM JAPONICUM*¹⁾

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Euponin, a new naturally occurring guaianolide inhibiting insect development, was isolated from the leaves of *Eupatorium japonicum* Thunb. (Compositae). Its structure was elucidated on the basis of chemical transformations and full spectral analysis.

As described previously,²⁻⁴⁾ our novel "Drosophila test" proved to be a convenient bioassay method for searching insect development inhibitors in extracts of higher plants.

One of these extracts tested, the methanol extract of *Eupatorium japonicum* Thunb. was of special interest because of the observation that larvae of the fruit-fly could not grow in rearing medium containing the extract. This fact suggested that it contained growth inhibitors and/or antifeedants against the insect. We have examined the extract of *E. japonicum* and isolated a sesquiterpene lactone as an active principle.

In this communication we wish to report the isolation and

structure elucidation of the new epoxy sesquiterpene lactone, which we have named euponin.

Euponin $C_{20}H_{24}O_6$ (I) (M^+ , m/z 360), mp. 148-150°C, $[\alpha]_D^{24}$ -69° (c 1.0, EtOH), UV(EtOH) 210 nm(end absorption, $\epsilon=19000$), was isolated from the methanol extract of *E. japonicum* leaves by successive solvent partitions and chromatography.

Euponin contains a hydroxyl group (3510 cm^{-1}) and an α -methylene- γ -lactone grouping (δ 6.2 and 5.5, 1H each, doublets, $J=3$ Hz; 1770 and 1650 cm^{-1}).

The ^{13}C -NMR spectrum⁵⁾ of euponin revealed that it has a tetracyclic structure.

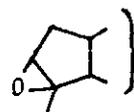
The IR spectrum⁵⁾ of euponin also showed the presence of an ester carbonyl group (1700 cm^{-1}). The ^1H -NMR spectrum⁵⁾ displayed the typical signals of an angeloyl group (one olefin proton multiplet at δ 6.06, vinyl methyl multiplets at δ 1.76 and 1.90). The presence of the angeloyl function was further shown by the high-resolution mass spectrum, which had peaks corresponding to $M-C_5H_7O$ and $M-C_5H_8O_2$, and a base peak at m/z 83 (C_5H_7O).

The ^1H -NMR spectrum of euponin showed 3H multiplets overlapped near δ 4. These signals were ascribed to two carbinyll protons and one lactonic proton. These carbinyll proton signals were shifted downfield by 0.38 ppm and appeared as a pair of doublets (δ 4.29 and 4.43, $J=12$ Hz) in the spectrum of the acetate (II) of euponin, suggesting that this carbinol is primary. Manganese dioxide oxidation of euponin gave an aldehyde (III), the ^1H -NMR spectrum (C_6D_6) of which exhibited a sharp singlet (δ 9.49) due

to an aldehyde proton, but no carbonyl proton signals in the region of δ 4. Its UV absorption (EtOH) λ_{max} 241 nm ($\epsilon=12600$) suggested that the aldehyde group was conjugated with a tetra-substituted carbon-carbon double bond. This suggestion was confirmed by its $^1\text{H-NMR}$ spectrum which displayed no olefinic protons other than those of the γ -lactone and angeloyl moiety. Hence, euponin is deduced to have a primary carbinol attached to a tetrasubstituted double bond.

Two $^{13}\text{C-NMR}$ signals due to two species of carbon carrying oxygen (δ 63.6, $-\text{O}-\overset{|}{\text{C}}\text{H}$ and 66.6, $-\text{O}-\overset{|}{\text{C}}-$) suggested the presence of an oxide ring as the sixth oxygen function.

A sharp 3H singlet (δ 1.70) and a broad 1H singlet (δ 3.41) seemed to be ascribed to the methyl and the proton attached to the oxide ring.



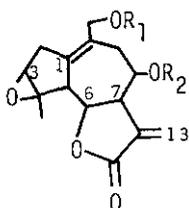
(A)

Inspection of literatures⁶⁻⁹ revealed that a set of these signals were characteristic of the moiety (A) in some guaianolides, particularly these signals in berlandin (IV)⁷ resembled those of euponin very closely in chemical shift and form. This regarded euponin as a guaianolide with C-3/C-4 epoxy grouping.

Location of angeloxy and the lactone ether oxygen on the seven membered ring remained to be solved. In the $^1\text{H-NMR}$ spectrum of euponin, irradiation at δ 3.1 (H-7) collapsed the multiplet at δ 5.7 ($-\overset{|}{\text{C}}\text{H}-\text{OAng}$) as well as the H-13a and H-13b doublets and the lactonic proton triplet. On the other hand, in the $^1\text{H-NMR}$ spectrum (C_6D_6) of III, irradiation of the proton ($-\overset{|}{\text{C}}\text{H}-\text{OAng}$) at

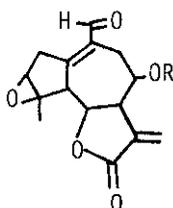
δ 5.4 collapsed the H-9 double doublet (δ 3.4, $J=15$ and 6Hz) to a doublet ($J=15$ Hz), and irradiation at δ 2.3 (H-9' and H-7) collapsed not only the H-13a and H-13b doublets to singlets but also the multiplet at δ 5.4 to a doublet ($J=6$ Hz). These facts indicate that the proton ($-\overset{|}{\text{C}}\text{H}-\text{OAng}$) is located at C-8.

The data mentioned above lead to the conclusion that the plane structure of eupoinin is represented by I.

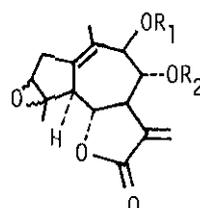


I, $R_1 = \text{H}$; $R_2 = \text{Angeloyl}$

II, $R_1 = \text{Ac}$, $R_2 = \text{Angeloyl}$



III, $R = \text{Angeloyl}$



IV, $R_1 = \text{Angeloyl}$

$R_2 = \text{Ac}$

In the $^1\text{H-NMR}$ spectrum of the acetate (II), the splitting pattern (triplet, $J=10$ Hz) of the lactonic proton signal could be clearly observed. The splitting pattern shows the *trans*-diaxial disposition of the protons at C-5(α), C-6(β) and C-7(α), a feature common to the most of the C-6 lactone in known guaianolides.⁶⁻¹⁰⁾

The stereochemistry of eupoinin is under investigation.

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Part III. See References 3 and 4.
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