

CHROM. 11,698

**Note**

**Glass capillary gas chromatographic separation of naturally occurring phloroglucinols**

**II. Investigation of some acylfilicinic acid derivatives**

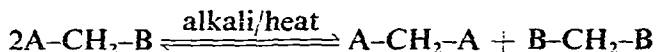
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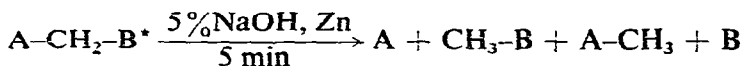
The decomposition reactions of naturally occurring polycyclic phloroglucinol derivatives can be divided into the following three steps.

(1) The so-called rottlerone change, which is a typical behaviour of methylene-bis-polyhydroxyphenols (polyhydroxydiphenylmethanes) and proceeds according to Scheme 1<sup>1,2</sup>:



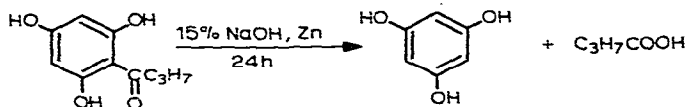
This reaction type has been observed in both alkaline and acidic media and by gentle heating of polycyclic phloroglucinols in their melting range<sup>3,4</sup>. Moreover, it also occurs in the ionization chamber of the mass spectrometer at high vacuum<sup>2,5</sup>.

(2) Cleavage of the methylene bridge between the rings (Scheme 2):



This reaction occurs on heating polycyclic phloroglucinols for a short period with dilute lime under reductive conditions and leads to the formation of several different monocyclic acylphloroglucinol derivatives<sup>5-9</sup>. Some rottlerone change may also occur as a side-reaction<sup>5-9</sup>.

(3) Cleavage of the acyl side-chain of the phloroglucinol rings (Scheme 3)<sup>6,7</sup>:



This reaction occurs on heating acylphloroglucinols with more concentrated lime under reductive conditions for several hours. It leads to the formation of substituted or unsubstituted phloroglucinols and fatty acids.

\* A and B are acylphloroglucinol rings.

In Part I of this series we described the analysis by high-resolution glass capillary gas-liquid chromatography (GLC) of several monocyclic acylphloroglucinols, 3-methylacylphloroglucinols and also their corresponding mono- and dimethyl ethers<sup>10</sup>. In this work we have analysed the separation of geminally substituted filicinic acids (1-6) and 3-methylfilicinic acids (7-10) by using similar glass capillary GLC methods. The acylfilicinic acids (1-6) are particularly common decomposition products of naturally occurring phloroglucinols<sup>5-9</sup>. We have also studied the direct separation of naturally occurring polycyclic phloroglucinols and the occurrence of the decomposition reactions illustrated above (Schemes 1-3) with our improved methods. For paper chromatographic (PC) and thin-layer chromatographic methods (TLC), see refs. 11-14.

## EXPERIMENTAL

### *Compounds investigated*

Table I lists the compounds studied. The synthesis and properties of compounds 1-7 and 10 have been described previously<sup>7,14</sup>.

Compounds 8 and 9 were synthesized in an analogous manner, and the resulting mixtures from the hydrogen carbonate fractions were separated by column chromatography on silica gel with *n*-hexane and benzene and their mixtures as eluents<sup>7</sup>. 3-Methylacetylfilicinic acid(8) consisted of colourless needles/prisms recrystallized from benzene (m.p. 158°, ref. 15; m.p. 160-161°; MS, M<sup>+</sup> at *m/e* = 210); 3-methylpropionylfilicinic acid(9) consisted of colourless needles recrystallized from benzene (m.p. 103°; MS, M<sup>+</sup> at *m/e* = 224).

### *Reductive alkaline cleavage*

This was performed by heating a mixture of 20 mg of *p*-aspidin BB (11), 40 mg of zinc dust and 20 ml of 5% sodium hydroxide solution for 5 min on a boiling water-bath (cleavage A, p. 1216 in ref. 7).

### *Preparation of trimethylsilyl ethers*

A 10-mg amount of the compound was dissolved in 200  $\mu$ l of a mixture of 0.5 ml N,O-bis(trimethylsilyl)acetamide (BSA) (Supelco, Bellefonte, Pa., U.S.A.), and 9.5 ml of dry benzene and allowed to stand at 60° for 15 min. The BSA and benzene were evaporated in a flow of nitrogen and the solid residue was dissolved in 1 ml of dry benzene.

### *GLC experiments*

The GLC experiments were carried out immediately after the silylations. Several types of glass capillary GLC columns were tested, the acidity, length and polarity of which were changed. The most suitable for the detection of silylated phloroglucinol derivatives were columns coated with OV-17 liquid stationary phase, which were constructed by the method described in detail by Grob and co-workers<sup>16,17</sup>.

In the GLC experiments a splitless injection technique was used, and the temperature of the glass-coated injection oven was 200°. A 1- $\mu$ l volume of the solution was injected and the GLC oven was temperature programmed from 50 to 240° at 8°

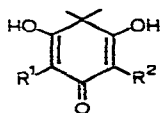
min. Hydrogen at 2 ml/min was used as the carrier gas and the instrument (Carlo-Erba 2300) was equipped with a flame-ionization detector (FID).

## RESULTS AND DISCUSSION

In Table I the retention times of the silylated filicinic acids (1-6) and 3-methylfilicinic acids (7-10) are given. All of the compounds studied, even those differing only in the branching of the side-chain, could be well separated in the form of their silyl ethers by high-resolution glass capillary GLC. In spite of considerable efforts with numerous GLC liquid phases, it is not possible to detect the filicinic acids without derivatization.

TABLE I

SEPARATION OF FILICINIC ACIDS (1-6) AND 3-METHYLFILICINIC ACIDS (7-10) AS SILYL ETHERS BY HIGH-RESOLUTION GLASS CAPILLARY GLC



Compound	R <sup>1</sup>	R <sup>2</sup>	Name	Relative retention time
1	H	H	Filicinic acid	0.90
2	H	COCH <sub>3</sub>	Acetylfilicinic acid	0.75
3	H	COC <sub>2</sub> H <sub>5</sub>	Propionylfilicinic acid	0.81
4	H	COC <sub>3</sub> H <sub>7</sub>	Butyrylfilicinic acid	0.89
5	H	COCH(CH <sub>3</sub> ) <sub>2</sub>	Isobutyrylfilicinic acid	0.87
6	H	COC <sub>4</sub> H <sub>9</sub>	Valerylfilicinic acid	1.00
7	CH <sub>3</sub>	H	3-Methylfilicinic acid	0.79
8	CH <sub>3</sub>	COCH <sub>3</sub>	3-Methylacetylfilicinic acid	0.80
9	CH <sub>3</sub>	COC <sub>2</sub> H <sub>5</sub>	3-Methylpropionylfilicinic acid	0.84
10	CH <sub>3</sub>	COC <sub>3</sub> H <sub>7</sub>	3-Methylbutyrylfilicinic acid	0.95

The GLC detection of the "normal types" of naturally occurring bicyclic and tricyclic phloroglucinol derivatives with one or two filicinic acid moieties in their molecules<sup>8,9</sup> was also studied by GLC. However, it was impossible to detect these polycyclic phloroglucinol derivatives either when silylated or ethylated (*cf.*, ref. 10). In spite of this, the GLC method can be used for structural determinations on this type of compounds, as illustrated by Fig. 1, in which *p*-aspidin BB (11) served as a model.

After reductive alkaline cleavage of the methylene bridge between the rings and silylation of the products formed, butyrylfilicinic acid (4) and aspidinol B (12) were detected. This is in agreement with earlier PC and TLC experiments of the same non-silylated phloroglucinol mixture<sup>18,19</sup>.

When *p*-aspidin BB (11) was silylated as such and injected into the chromatograph, decomposition according to Scheme 2 mainly occurred. 3-Methylbutyrylfilicinic acid (10) and aspidinol B (12) were observed among the decomposition products. However, butyrylfilicinic acid (4) was not observed, a distinct difference from the former experiment.

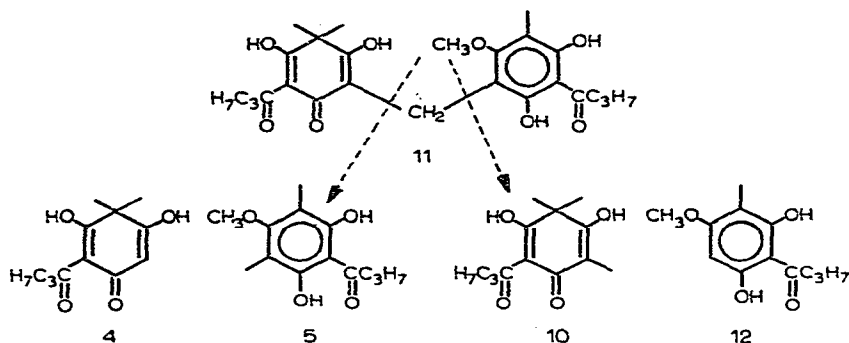


Fig. 1. Monocyclic acylphloroglucinols formed from *p*-aspidin BB<sup>11</sup> under two different sets of conditions.

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