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# PHLEBOTRICHIN, A PHENOLIC COMPOUND FROM THE FRESH LEAVES OF *VIBURNUM PHLEBOTRICHUM*

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**Key Word Index**—*Viburnum phlebotrichum*; Caprifoliaceae; phenolic glucoside; arbutin; phlebotrichin; 2,6-dimethyl-6(*S*)-hydroxyl-2-*trans*-2,7-octadienoic acid; linalool-1-oic acid.

**Abstract**—Arbutin and a new phenolic glucoside were isolated from the fresh leaves of *Viburnum phlebotrichum*. The structure of the new compound named phlebotrichin was established as 1-*O*-(4-hydroxyphenyl)-6-*O*-[(2*E*, 6*S*)-2,6-dimethyl-6-hydroxy-2,7-octadienoyl]-*β*-D-glucopyranoside.

## INTRODUCTION

We were interested in the well-known fact that the leaves of plants belonging to the genus *Viburnum* are liable to go brown when they are heated or pressed between papers for preparing dried specimens. This report deals with the isolation of arbutin (**1**), the isolation and structural determination of a new phenolic compound, phlebotrichin (**2**), and the quantitative comparison of the phenolic compounds in the fresh and brown leaves of *Viburnum phlebotrichum* Sieb. et Zucc.

## RESULTS AND DISCUSSION

The fresh leaves were treated as described in the Experimental to yield **1** and **2**. Compound **1**, arbutin, was identified by direct comparison with an authentic sample. Compound **2**, phlebotrichin, white powder-like crystals, mp 141–143°, $C_{22}H_{30}O_9$, $[\alpha]_D^{27} = -32.6°$. The IR spectrum showed the presence of carbonyl and hydroxyl groups whilst the $^1H$ NMR spectrum showed the presence of two methyl groups, one vinyl group, $A_2B_2$-type aromatic protons and an aromatic hydroxyl group. Hydrolysis of **2** with 0.5 N sodium hydroxide gave **3** and **4**. Compound **3**, a pale-yellow oil, $C_{10}H_{16}O_3$ was soluble in 5% aq. sodium bicarbonate. Its structure was established as (*E*)-linalool-1-oic acid (2,6-dimethyl-6-hydroxyl-2-*trans*-2,7-octadienoic acid) by comparison of its IR, NMR and mass spectra with those recently described for the natural [1] and synthetic [2] forms of (*E*)-linalool-1-oic acid. Compound **4**, colourless needles, mp 199–200°, $C_{12}H_{16}O_7$ was identified as arbutin by IR and mixed fusion.

The $^1H$ NMR data of **2** were compared with those of **1** and **3** (Table 1). The signals of C-5 and C-6 in **1** were shifted +3.1 and −2.6 ppm respectively compared to those of **2** and the signal of $\delta$168.1 in **2** indicates that C-1″ in **3** was attached to C-6 in **1** by an ester linkage. The signs of specific rotation of **3** ($[\alpha]_D^{25}$ +19.3°) and molecular rotations of **1** (−174.3°), **2** (−142.9°) and *d*-linalool (+29.8°) suggested that **2** and **3** had a *S*-configuration at C-6″ (Fig. 1).

Table 1. $^{13}C$ NMR data for compounds **1**–**3** ($C_5H_5N$)

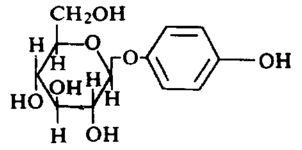
| Carbon No. | **2** | **1** | **3** |
|---|---|---|---|
| 1 | 103.6 | 103.6 | — |
| 2 | 74.9 | 75.1 | — |
| 3 | 78.5 | 78.8 | — |
| 4 | 71.7 | 71.3 | — |
| 5 | 75.5 | 78.6 | — |
| 6 | 65.0 | 62.4 | — |
| 1′ | 154.1 | 154.0 | — |
| 2′ | 116.8 | 116.9 | — |
| 3′ | 118.9 | 118.8 | — |
| 4′ | 151.7 | 151.8 | — |
| 5′ | 118.9 | 118.8 | — |
| 6′ | 116.8 | 116.9 | — |
| 1″ | 168.1 | — | 170.5 |
| 2″ | 127.7 | — | 128.7 |
| 3″ | 143.6 | — | 142.4 |
| 4″ | 24.1 | — | 24.1 |
| 5″ | 41.5 | — | 41.7 |
| 6″ | 72.2 | — | 72.1 |
| 7″ | 146.5 | — | 146.4 |
| 8″ | 111.8 | — | 111.6 |
| 9″ | 12.5 | — | 12.7 |
| 10″ | 28.7 | — | 28.4 |

From the available data, the structure of **2** was established to be 1-*O*-(4-hydroxyphenyl)-6-*O*-[(2*E*, 6*S*)-2,6-dimethyl-6-hydroxy-2,7-octadienoyl]-*β*-D-glucopyranoside. The contents of **1** and **2** in methanol extracts of fresh and brown leaves were assayed by using a dual wavelength TLC scanner. The results showed that the brown leaves contained only *ca* 0.04% of **1** and 0.008% of **2**, while the fresh leaves contained *ca* 0.11% and 0.08%, respectively. They suggest that the change in colour of the leaves from green to brown is related to the breakdown of **1** and **2**.

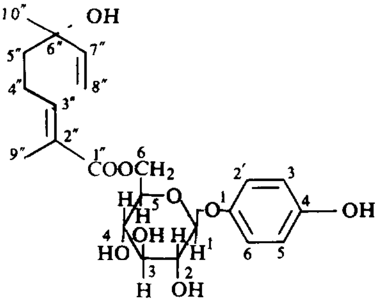
## EXPERIMENTAL

Mps were uncorr. $^1H$ NMR 100 MHz, TMS as int. standard. Plant material was collected at Ohme in Tokyo, 1979.

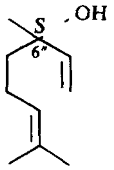
*Extraction and isolation.* Fresh leaves (190 *g*) were extracted



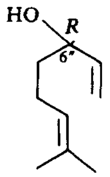
**1** $[\phi]_{184}^{25}$ −174 3°



**2** $[\phi]_{438}^{27}$ − 142 9°



d − Linalool $[\phi]_{154}^{20}$ + 29 8°



l − Linalool $[\phi]_{154}^{20}$ −31 0°

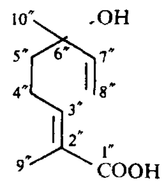


Fig 1 Structures and molecular rotations of **1**, **2** and D- and L-linalool

with MeOH (60l) The extracts were concd *in vacuo* and diluted with $H_2O$ The soln was extracted with EtOAc and then *n*-BuOH

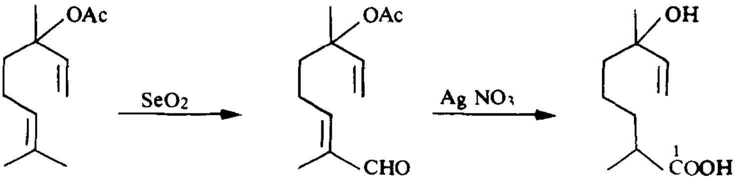
*Purification of arbutin* (**1**) *and phlebotrichin* (**2**) The *n*-BuOH extract (4 5 g) was subjected to Rotation Locular Counter Current Chromatography (RLCCC) [3] [EtOAc-EtOH-$H_2O$ (10 3 6), upper layer (mobile phase, 242 ml), lower layer (stationary phase, 373 ml), locular 800, flow rate 0 32 ml/min, 1 fraction (fr ), 50 ml] Frs 16–38 were collected, taken to dryness and chromatographed over Si gel using EtOAc-MeOH-$H_2O$ (21 4 3) to give arbutin (415 mg) The EtOAc extract (4g) was chromatographed on RLCCC [EtOAc-hexane-EtOH-$H_2O$ (10 5 6 5), upper layer (247 ml), lower layer (390 ml), locular 800, flow rate 0 32 ml/min, 1 fr, 50 ml] Frs 25–40 were collected, taken to dryness and chromatographed over Si gel using EtOAc to afford phlebotrichin (400 mg)

*Arbutin* (1) was recrystallized (EtOAc) to give colourless needless, mp 199–200° High resolution MS (70 eV) Found 272 0849, calc for $C_{12}H_{16}O_7$ 272 2542, UV $\lambda_{max}^{EtOH}$ nm (log ε) 285 (3 42), IR $\nu_{max}^{KBr}$ $cm^{-1}$ 3300, 1515, 1210, 830, $^1H$ NMR ($Me_2CO$-$d_6$) δ6 73 (*d*), 6 95 (*d*), 8 02 (*s*)

*Phlebotrichin* (**2**) was recrystallized (*n*-hexane–EtOAc) to give white powder-like crystals, mp 141–143°, $[\alpha]_D^{27}$ − 32 6° (EtOH, *c*5) High resolution MS (70 eV) Found 438 1861, calc for $C_{22}H_{30}O_9$ 438 4736, UV $\lambda_{max}^{EtOH}$ nm (logε) 285 (2 7), IR $\nu_{max}^{KBr}$ $cm^{-1}$ 3400, 1695, 1640, 1510, 1215, 815, $^1H$ NMR ($Me_2CO$-$d_6$) δ1 28 (3H, *s*, H-10″), 1 83 (3H, *s*, H-9″), 5 01 (1H, *dd*, *J* = 2, 10 Hz, H-8″), 5 23 (1H, *dd*, *J* = 2, 17 Hz, H-8″), 5 98 (1H, *dd*, *J* = 10, 17 Hz, H-7″), 6 73 (2H, *d*, *J* = 9 Hz, H-2′ or H-6′, H-3′ or H-5′), 6 95 (2H, *d*, *J* = 9 Hz, H-5′ or H-3′, H-6′ or H-2′), 8 02 (1H, *s*, OH-4′), $^{13}C$ NMR See Table 1

*Hydrolysis of phlebotrichin by alkali* A soln of **2** (50 mg) in 0 5 N NaOH (5 ml) was stirred for 10 min at room temp The soln was acidified (dil HCl) and extracted with $Et_2O$ The extract was evaporated and chromatographed on Si gel using $C_6H_6$–EtOAc (5 1) to afford **3** (20 mg) The acidic soln was neutralized (dil aq NaOH) and extracted with EtOAc The extract was evaporated *in vacuo* to give **4** (25 mg)

Compound **3** was obtained as a pale yellow oil $[\alpha]_D^{25}$ + 19 3° ($CHCl_3$, *c*0 15) High resolution MS (70 eV) Found 184 1126, calc for $C_{10}H_{16}O_3$ 184 2346, IR $\nu_{max}^{capillary}$ $cm^{-1}$ 3400, 1680, 1640, 995, $^1H$ NMR ($CDCl_3$) δ1 32 (3H, *s*, H-10″), 1 64 (2H, *dd*, *J* = 7, 17 Hz, H-5), 1 82 (3H, *s*, H-9″), 2, 25 (2H, *dd*, *J* = 7, 17 Hz, H-4″), 5 08 (1H, *dd*, *J* = 2, 10 Hz, H-8″), 5 24 (1H, *dd*, *J* = 2, 17 Hz, H-8″), 5 92 (1H, *dd*, *J* = 10, 17 Hz, H-7″), 6 89 (1H, *t*, H-3″), $^{13}C$ NMR See Table 1 There spectral properties were identical to those of synthetic (*E*)-linalool-1-oic acid



**5** **6** **3**

Fig 2 Chemical synthesis of linalool-1-oic acid

Compound **4** was recrystallized from EtOAc to give colourless needles, and identified with arbutin by IR and mixed fusion

*Synthesis of* (E)-*linalool-1-oic acid* (**3**) Compound **3** was synthesized from linalool acetate (**5**) (20 g) by a modification of the method (Fig 2) used for the synthesis of D-2,6-dimethyl-8, 8-ethylendioxyoct-2-enioc acid from the cyclic acetal of D-citronellal [2] The aldehyde (**6**) (15 g) was obtained as a colourless oil (bp 124–125°/5 mm) Compound **3** (1 5 g), also obtained as an oil, was purified by chromatography on Si gel in $C_6H_6$–EtOAc (7 3) **3** pale yellow oil, $C_{10}H_{16}O_3$ (MS), IR $\nu_{\max}^{\text{capillary}}$ $\text{cm}^{-1}$ 3400 (OH), 1680 (conj COOH), 1640, 1410, 1365, 1280, 1150, 1100, 1065, 995, 920 ($CH = CH_2$), 800, 740, 685, 640, 550

*Quantitative analysis of arbutin and phlebotrichin in fresh and brown leaves* Fresh leaves (90 g) and brown leaves which had been prepared from fresh leaves (90 g) by treatment for 3 hr at 30° in a drying apparatus, were extracted with MeOH (6 l) and the extracts reduced (*in vacuo*) in vol to 250 ml Each extract (1 $\mu$l) was then assayed for **1** and **2** using a dual wavelength TLC scanner (Shimazu CS-910) (TLC, Si gel 60F 254, EtOAc–MeOH–$H_2O$ (21 4 3), $R_f$ arbutin 0 62, phlebotrichin 0 86, 22°, wavelength $\lambda_1 = 290$ nm, $\lambda_2 = 340$ nm) Fresh leaves arbutin 48 4 mg (0 11 %), phlebotrichin 37 9 mg (0 08 %), brown leaves arbutin 18 5 mg (0 08 %), phlebotrichin 3 6 mg (0 008 %)

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