# Resin Glycosides. V.[1] Identification and Characterization of the Component Organic and Glycosidic Acids of the Ether-Soluble Crude Resin Glycosides ("Jalapin") from Rhizoma Jalapae Braziliensis (Roots of *Ipomoea operculata*)

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**Alkaline hydrolysis of the ether-soluble resin glycoside ("jalapin") fraction of the roots of *Ipomoea operculata* (Convolvulaceae) gave five glycosidic acids, operculinic acids A (1), B (2), C (3), D and E, along with *n*-decanoic and *n*-dodecanoic acids. Operculinic acids A, B and C were characterized to be 11*S*-jalapinolic acid 11-*O*-β-D-glucopyranosyl-(1→3)-*O*-[α-L-rhamnopyranosyl-(1→4)]-*O*-α-L-rhamnopyranosyl-(1→4)-*O*-α-L-rhamnopyranosyl-(1→2)-β-D-fucopyranoside, 11*S*-jalapinolic acid 11-*O*-β-D-glucopyranosyl-(1→3)-*O*-[α-L-rhamnopyranosyl-(1→4)]-*O*-α-L-rhamnopyranosyl-(1→4)-*O*-α-L-rhamnopyranosyl-(1→2)-β-D-glucopyranoside and 11*S*-jalapinolic acid 11-*O*-α-L-rhamnopyranosyl-(1→4)-*O*-α-L-rhamnopyranosyl-(1→4)-*O*-α-L-rhamnopyranosyl-(1→2)-β-D-fucopyranoside, respectively, on the basis of chemical and spectroscopic data.**

***n*-Decanoic and *n*-dodecanoic acids found in this study are the first component organic acids of the resin glycosides so far studied.**

**Keywords** resin glycoside; Rhizoma Jalapae Braziliensis; *Ipomoea operculata*; jalapin; organic acid; *n*-decanoic acid; *n*-dodecanoic acid; glycosidic acid; operculinic acid; *S*-jalapinolic acid glycoside

Rhizoma Jalapae Braziliensis (Brazilian jalap), the root of *Ipomoea operculata* (GOMES) MART. (Convolvulaceae) is a laxative crude drug, and it is used as a substitute for Mexican jalap (Vera Cruz Jalap, the root of *I. purga* HAYNE). Its active ingredient, so-called resin glycoside, is known to be composed of ether-soluble ("jalapin") and -insoluble ("convolvulin") fractions.[2] In 1929, Votocek and Valentin[3] reported that rhamnoconvolvulin obtained as a "convolvulin" from the resin afforded, on hydrolysis with methanolic HCl, a hydroxyfatty acid, which was synthetically characterized as 3,12-dihydroxypalmitic acid by Graf and Buhle.[4] Graf, *et al.*[5] reported that, on alkaline hydrolysis, the "jalapin" afforded acetic, tiglic, *n*-valeric, trimethylacetic, 2-methylbutyric, isovaleric and propionic acids as component organic acids, while the "convolvulin" gave, besides those of "jalapin", 4-oxodecanoic acid and exogonic acid (3,6:6,9-diepoxydecanoic acid) which are characteristic of Brazilian resin, and hence are used as markers to distinguish between Brazilian and Mexican resins. Wagner and Kazmaier[6] isolated a major glycosidic acid, operculinic acid (rhamnoconvolvulinic acid), 3,12-dihydroxypalmitic acid 12-*O*-α-D-glucopyranosyl-(1→4)-*O*-[α-L-rhamnopyranosyl-(1→6)]-*O*-α-D-glucopyranosyl-(1→3)-*O*-α-L-rhamnopyranosyl-(1→2)-*O*-[β-D-glucopyranosyl(1→3)-β-D-glucopyranoside, from the "convolvulin."

In the course of our studies on the resin glycosides, which are characteristic of Convolvulaceae plants, we reexamined the components of the "jalapin" fraction and obtained five new glycosidic acids along with *n*-decanoic acid and *n*-dodecanoic acid. This paper deals with the detailed structural studies on the three new component glycosidic acids, operculinic acids A, B and C.

The MeOH extractive (6.9%) of the crude drug was partitioned between ether and $H_2O$. The ether soluble fraction was chromatographed over MCI gel CHP 20P and LH-20 to afford "jalapin" fraction, pale yellow powder, mp 65—78 °C (yield; 1.04%). The $H_2O$ soluble fraction was partitioned between *n*-BuOH and $H_2O$ to give "convolvulin" fraction (yield; 1.64%). The alkaline hydrolysis product of the "jalapin" was fractionated into organic and glycosidic acid fractions. The former fraction was methylated with diazomethane and then examined by gas chromatography–mass spectrometry (GC-MS); it exhibited no peaks corresponding to the organic acids reported by Graf, *et al.*,[5] but two peaks identical to those of authentic methyl *n*-decanoate and methyl *n*-dodecanoate were seen.

The glycosidic acid fraction gave, on acidic hydrolysis, an aglycone and a sugar mixture. Methylation of the former with diazomethane furnished needles, mp 43—44 °C, $[\alpha]_D$ +0.9°. This product was identical with an authentic sample of methyl (+)-11*S*-jalapinolate (11-hydroxypalmitate).[1,7]

Silica gel chromatography and subsequent preparative high performance liquid chromatography (HPLC) of the glycosidic acid fraction provided five homogeneous methyl esters of glycosidic acids named operculinic acids A—E.

Operculinic acid A methyl ester (**1**), white powder, mp 172—173 °C (dec.), $[\alpha]_D$ −72.5°, $C_{47}H_{84}O_{24}$, gave, on acidic hydrolysis, jalapinolic acid as an aglycone and D-glucose ($[\alpha]_D$ +46.8°), L-rhamnose ($[\alpha]_D$ +10.4°) and D-fucose ($[\alpha]_D$ +74.1°) in the ratio of about 1:3:1 (HPLC). Saponification of **1** furnished operculinic acid A (**1'**), mp 172—174 °C (dec.), $[\alpha]_D$ −63.6°, $C_{46}H_{82}O_{24}$, which showed the $(M-H)^-$ ion peak at *m/z* 1017 along with fragment peaks at *m/z* 871 (1017−146 ($C_6H_{10}O_4$, 6-deoxyhexose unit))$^-$, 855 (1017−162 ($C_6H_{10}O_5$, hexose unit))$^-$, 709 (871−162 and/or 855−146)$^-$, 563 (709−146), 417 (563−146) and 271 (417−146, jalapinolic acid−H)$^-$ in the negative ion fast atom bombardment mass spectrum (negative FAB-MS), suggesting that **1'** is a glycoside composed of one mol each of jalapinolic acid, D-glucose and D-fucose, and three mol of L-rhamnose, and its sugar moiety forms a branched chain with terminal glucose and either rhamnose or fucose units.

In the proton nuclear magnetic resonance ($^1$H-NMR) spectrum, each of the ester methyl, terminal methyl and equivalent methylene signals attributable to methyl jalapinolate was observed, as well as five anomeric proton signals. All the proton signals owing to the sugar moieties of **1** and its peracetate (**4**), white powder, mp 83—86 °C, $[\alpha]_D$

TABLE I. $^1$H-NMR Spectral Data for **1**, **2**, **3′**, **4** and **5** (in Pyridine-$d_5$)

| H atom | | **1** | **4** | **2** | **5** | **3′** |
|---|---|---|---|---|---|---|
| Fuc | -1 | 4.77 (d, 7.9) | 4.94 (d, 7.5) | | | 4.74 (d, 7.9) |
| | 2 | 4.47 (dd, 7.9, 9.5) | 4.30 (dd, 7.5, 10.0) | | | 4.46 (dd, 7.9, 9.4) |
| | 3 | 4.12 (dd, 9.5, 3.4) | 5.46 (dd, 10.0, 3.5) | | | 4.11 (dd, 9.4, 3.4) |
| | 4 | 3.93 (d, 3.4) | 5.55 (dd, 3.5, 1.0) | | | 3.92 (d, 3.4) |
| | 5 | 3.78 (q, 6.4) | 4.00 (dq, 1.0, 6.5) | | | 3.76 (q, 6.4) |
| | 6 | 1.51 (d, 6.4) | 1.25 (d, 6.5) | | | 1.50 (d, 6.4) |
| Glc | -1 | 5.20 (d, 7.7) | 5.20 (d, 7.5) | 4.90 (d, 7.6) | 5.10 (d, 7.7) | |
| | 2 | 3.98 (dd, 7.7, 8.8) | 5.33 (dd, 7.5, 9.5) | 4.14 (dd, 7.6, 9.0) | 4.11 (dd, 7.7, 9.5) | |
| | 3 | 4.16 (dd, 8.8, 8.8) | 5.69 (dd, 9.5, 9.5) | 4.17[a] | 5.82 (dd, 9.5, 9.5) | |
| | 4 | 4.08 (dd, 8.8, 8.8) | 5.47 (dd, 9.5, 9.5) | 4.04 (dd, 9.0, 9.0) | 5.40 (dd, 9.5, 9.5) | |
| | 5 | 3.94[a] | 4.07[a] | 3.81 (ddd, 3.0, 6.0, 9.0) | 4.17[a] | |
| | 6 | 4.26 (dd, 6.1, 11.9) | 4.50 (dd, 2.0, 12.5) | 4.26 (dd, 11.0, 6.0) | 4.44 (dd, 2.2, 12.5) | |
| | | 4.51 (dd, 2.4, 11.9) | 4.73 (dd, 3.5, 12.5) | 4.41[a] | 4.59 (dd, 5.2, 12.5) | |
| Glc′ | -1 | | | 5.12 (d, 7.9) | 5.22 (d, 7.9) | |
| | 2 | | | 3.91 (dd, 7.9, 9.0) | 5.38 (dd, 7.9, 9.5) | |
| | 3 | | | 4.08 (dd, 9.0, 9.0) | 5.73 (dd, 9.5, 9.5) | |
| | 4 | | | 4.00 (dd, 9.0, 9.0) | 5.52 (dd, 9.5, 9.5) | |
| | 5 | | | 3.86 (ddd, 3.0, 5.5, 9.0) | 4.06[a] | |
| | 6 | | | 4.18 (dd, 5.5, 11.5) | 4.53 (dd, 2.2, 12.5) | |
| | | | | 4.43 (dd, 3.0, 11.5) | 4.80 (dd, 3.3, 12.5) | |
| Rha | -1 | 6.20 (d, 1.5) | 5.47 (d, 1.8) | 6.20 (br s) | 5.42 (br s) | 6.21 (d, 1.2) |
| | 2 | 4.65 (dd, 1.5, 3.4) | 5.59 (dd, 1.8, 3.5) | 4.58 (dd, 1.5, 3.0) | 5.58[a] | 4.63 (dd, 1.2, 3.4) |
| | 3 | 4.59 (dd, 3.4, 9.2) | 5.71 (dd, 3.5, 10.0) | 4.53 (dd, 3.0, 9.0) | 5.70 (dd, 4.0, 9.2) | 4.58 (dd, 3.4, 9.1) |
| | 4 | 4.20 (dd, 9.2, 9.2) | 4.14 (dd, 10.0, 10.0) | 4.16 (dd, 9.0, 9.0) | 4.18 (dd, 9.2, 9.2) | 4.28 (dd, 9.1, 9.1) |
| | 5 | 4.84 (dq, 9.2, 6.1) | 4.80 (dq, 10.0, 6.0) | 4.81 (dq, 9.0, 6.1) | 4.76 (dq, 9.2, 6.1) | 4.82 (dq, 9.1, 5.8) |
| | 6 | 1.59 (d, 6.1) | 1.68 (d, 6.0) | 1.66 (d, 6.1) | 1.69 (d, 6.1) | 1.56 (d, 5.8) |
| Rha′ | -1 | 5.86 (d, 1.8) | 5.32 (d, 2.0) | 5.78 (d, 1.8) | 5.35 (br s) | 6.18 (d, 1.8) |
| | 2 | 5.14 (dd, 1.8, 3.0) | 5.48 (dd, 2.0, 4.0) | 5.06 (dd, 1.8, 3.0) | 5.50[a] | 4.74 (dd, 1.8, 3.3) |
| | 3 | 4.70 (dd, 3.0, 9.1) | 4.56 (dd, 4.0, 9.5) | 4.63 (dd, 3.0, 8.5) | 4.57 (dd, 9.5, 9.5) | 4.53 (dd, 3.3, 8.9) |
| | 4 | 4.47 (dd, 9.1, 9.1) | 4.19 (dd, 9.5, 9.5) | 4.40 (dd, 8.5, 8.5) | 4.26[a] | 4.39 (dd, 8.9, 8.9) |
| | 5 | 4.38 (dq, 9.1, 6.1) | 4.24[a] | 4.34[a] | 4.26[a] | 4.32 (dq, 8.9, 6.1) |
| | 6 | 1.58 (d, 6.1) | 1.58 (d, 5.8) | 1.54 (d, 6.1) | 1.60 (d, 5.5) | 1.56 (d, 6.1)[b] |
| Rha″ | -1 | 6.17 (d, 1.5) | 5.67 (d, 1.6) | 6.07 (br s) | 5.72[a] | 6.26 (d, 1.5) |
| | 2 | 4.86 (dd, 1.5, 3.5) | 5.74 (dd, 1.6, 3.5) | 4.77 (dd, 1.5, 3.5) | 5.80 (br d, 3.5) | 4.78 (dd, 1.5, 3.0) |
| | 3 | 4.40 (dd, 3.5, 9.2) | 5.66 (dd, 3.5, 10.0) | 4.33 (dd, 3.5, 9.0) | 5.72[a] | 4.43 (dd, 3.0, 9.1) |
| | 4 | 4.20 (dd, 9.2, 9.2) | 5.53 (dd, 10.0, 10.0) | 4.12 (dd, 9.0, 9.0) | 5.58 (dd, 10.0, 10.0) | 4.23 (dd, 9.1, 9.1) |
| | 5 | 4.28 (dq, 9.2, 6.1) | 4.25[a] | 4.23 (dq, 9.0, 5.8) | 4.28 (dq, 10.0, 6.1) | 4.32 (dq, 9.1, 6.1) |
| | 6 | 1.56 (d, 6.1) | 1.34 (d, 6.0) | 1.58 (d, 5.8) | 1.34 (d, 6.1) | 1.58 (d, 6.1)[b] |
| $OCH_3$ | | 3.62 (s) | 3.63 (s) | 3.61 (s) | 3.65 (s) | |
| Ag | -2 | 2.33 (t, 7.4) | 2.38 (t, 8.0) | 2.32 (t, 7.6) | 2.40 (t, 7.3) | 2.50 (t, 7.3) |
| | 11 | 3.97[a] | 4.05[a] | 4.01[a] | 4.04[a] | 3.92[a] |
| | 16 | 0.92 (t, 7.0) | 0.92 (t, 7.2) | 0.90 (t, 7.0) | 0.96 (t, 7.0) | 0.92 (t, 6.8) |

*a*) Signals are overlapping. *b*) These assignments may be interchanged. $\delta$ in ppm from TMS (coupling constants ($J$) in Hz are given in parentheses); Fuc, fucopyranose; Glc, glucopyranose; Rha, rhamnopyranose; Ag, aglycone (11*S*-jalapinolic acid). All assignments are based on the $^1$H–$^1$H COSY and NOESY spectral data.

$-36.4°$, were assigned with the aid of the $^1$H–$^1$H shift correlated 2D-NMR (COSY) method and nuclear Overhauser effect 2D-NMR (NOESY) (Table I). The coupling constants of the anomeric and methine proton signals indicated that all the monosaccharides are of pyranose form, and further, the mode of glycosidic linkage of rhamnose units is $\alpha$ in $^1C_4$ conformation and those of fucose and glucose are $\beta$ in $^4C_1$. Moreover, comparing the chemical shifts of signals due to the sugar moieties between **1** and **4**, the signals at 3- and 4-H of fucose (Fuc), 2- and 3-H of rhamnose (Rha), 2-H of the second (Rha′), 2-, 3-, 4- and 6-$H_2$ of glucose (Glc) and 2-, 3- and 4-H of the third rhamnose (Rha″) were shifted downfield by 0.34—1.62 ppm suggesting that the sugar linkages of **1** are located at 2-OH of Fuc, 4-OH of Rha and 3- and 4-OH of Rha′. This suggestion was supported by the glycosylation shifts observed at $C_2$ of Fuc (+3.3 ppm), $C_4$ of Rha (+8.5) as well as $C_3$ and $C_4$ of Rha′ (+10.6 and +4.8)[8] in the $^{13}$C-NMR spectrum of **1**, these signals were assigned by the $^1$H–$^{13}$C heteronuclear shift-correlated 2D-NMR (HETCOR) method (Table II).

In order to determine the sequence of the sugar moiety, the NOESY spectrum of **1** was recorded. Two of four cross peaks observed were unambiguously assigned as those between 1-H of Glc and 3-H of Rha′, and 1-H of Rha′ and 4-H of Rha, while the counterparts due to 1-H of Rha and 1-H of Rha″ could not be defined because the signals of 2-H of Fuc and 4-H of Rha′ appeared at the same chemical shift ($\delta$ 4.47) (Fig. 1). However, taking the fact that **1** is a monodesmoside of methyl jalapinolate into account, it could be concluded that Rha is linked at 2-OH of Fuc, and Rha″ at 4-OH of Rha′.

Accordingly, the structure of operculinic acid A (**1′**) was concluded to be 11*S*-jalapinolic acid 11-*O*-$\beta$-D-glucopyranosyl-(1→3)-*O*-[$\alpha$-L-rhamnopyranosyl-(1→4)]-*O*-$\alpha$-L-rhamnopyranosyl-(1→4)-*O*-$\alpha$-L-rhamnopyranosyl-(1→2)-$\beta$-D-fucopyranoside.

Operculinic acid B methylester (**2**), white powder, mp 157—159 °C (dec.), $[\alpha]_D$ $-83.5°$, $C_{47}H_{84}O_{25}$, afforded, on complete acidic hydrolysis, jalapinolic acid, L-rhamnose

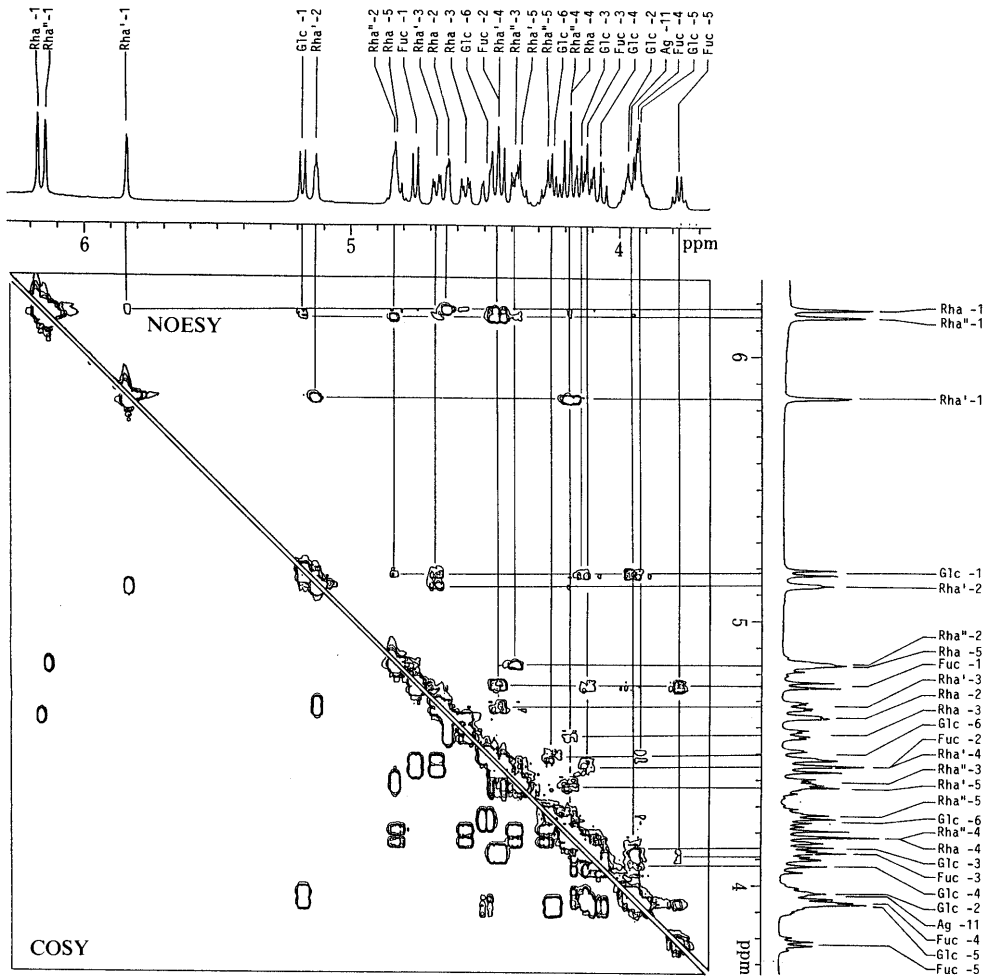


Fig. 1. COSY and NOESY Spectra of **1** (in Pyridine-$d_5$, 400 MHz)

TABLE II $^{13}$C-NMR Spectral Data for the Sugar Moieties of **1**, **2** and **3′** (in Pyridine-$d_5$)

| C atom | 1 | 2 | 3′ | C atom | 1 | 2 | 3′ |
|---|---|---|---|---|---|---|---|
| Fuc -1 | 101.2 | | 101.5 | Rha -1 | 101.5 | 101.4 | 101.5 |
| 2 | 75.3 | | 75.1 | 2 | 72.7 | 72.5 | 72.7 |
| 3 | 76.5 | | 76.6 | 3 | 72.6 | 72.5 | 73.2 |
| 4 | 73.4 | | 73.4 | 4 | 82.3 | 82.4 | 80.6 |
| 5 | 71.2 | | 71.2 | 5 | 67.6 | 67.7 | 67.2 |
| 6 | 17.1 | | 17.2 | 6 | 18.7 | 18.8 | 19.0 |
| Glc -1 | 105.4 | 101.0 | | Rha′-1 | 103.3 | 103.3 | 102.9 |
| 2 | 75.0 | 77.5 | | 2 | 71.9 | 71.9 | 73.1 |
| 3 | 78.4 | 79.5 | | 3 | 82.7 | 82.7 | 73.4 |
| 4 | 71.6 | 72.3 | | 4 | 78.6 | 78.7 | 79.6 |
| 5 | 78.4 | 77.9 | | 5 | 68.5 | 68.5 | 68.3 |
| 6 | 62.9 | 63.0 | | 6 | 18.9 | 18.9 | 18.8 |
| Glc′-1 | | 105.4 | | Rha″-1 | 103.0 | 102.9 | 103.3 |
| 2 | | 75.0 | | 2 | 72.4 | 72.5 | 72.4 |
| 3 | | 78.4 | | 3 | 72.8 | 72.8 | 72.8 |
| 4 | | 71.8 | | 4 | 73.9 | 74.0 | 73.9 |
| 5 | | 78.3 | | 5 | 70.4 | 70.3 | 70.2 |
| 6 | | 62.9 | | 6 | 18.3 | 18.3 | 18.4 |

$\delta$ in ppm from TMS. All the chemical shifts are based on the HETCOR spectral data.

($[\alpha]_D$ +10.1°) and D-glucose ($[\alpha]_D$ +52.9°). The saponification product of **2**, operculinic acid B (**2′**), white powder, mp 169—171 °C (dec.), $[\alpha]_D$ −80.9°, $C_{46}H_{82}O_{25}$, showed an $(M-H)^-$ ion peak at *m/z* 1033 and fragment peaks at *m/z* 887 (1033−146 (6-deoxyhexose unit))$^-$, 871 (1033−162 (hexose unit))$^-$, 725 (887−162 and/or 871−146)$^-$, 579 (725−146), 433 (579−146) and 271 (433−162, jalapinolic acid−H)$^-$ in the negative FAB-MS, indicating that **2′** is a pentaglycoside having a branched chain structure which consists of 1 mol of jalapinolic acid, 2 mol of glucose and 3 mol of rhamnose. Moreover, the fact that the $(M-H)^-$ and fragment ions except for the last one (*m/z* 271) are 16 mass units higher than those of **1′** suggested its sugar moiety to be identical to that of **1′** except that the fucose directly bonded to aglycone in **1′** is displaced by Glc in **2′**.

Inspection of the $^1$H- and $^{13}$C-NMR spectra of **2** and its peracetate (**5**), powder, mp 72—74 °C (dec.), $[\alpha]_D$ −33.6° (Tables I and II) indicated that the glycosidic linkages of **2** are placed at 2-OH of Glc, 4-OH of Rha, 3- and 4-OH of Rha′ and 11-OH of the jalapinolic acid moiety, and that the linkages of the rhamnose units are $\alpha$ in $^1C_4$ conformation and those of the glucose units are $\beta$ in $^4C_1$. The arrangement of sugar linkages was determined from the NOESY spectrum of **2**, that is, four cross peaks between 1-H of Rha and 2-H of Glc, 1-H of Rha′ and 4-H of Rha, 1-H of the second glucose (Glc′) and 3-H of Rha′, and 1-H of Rha″ and 4-H of Rha′ were exhibited.

Consequently, the structure of operculinic acid B (**2′**) was defined as 11*S*-jalapinolic acid 11-*O*-$\beta$-D-glucopyranosyl-(1→3)-*O*-[$\alpha$-L-rhamnopyranosyl-(1→4)]-*O*-$\alpha$-L-rhamnopyranosyl-(1→4)-*O*-$\alpha$-L-rhamnopyranosyl-(1→2)-$\beta$-D-glucopyranoside, as shown in Fig. 2.

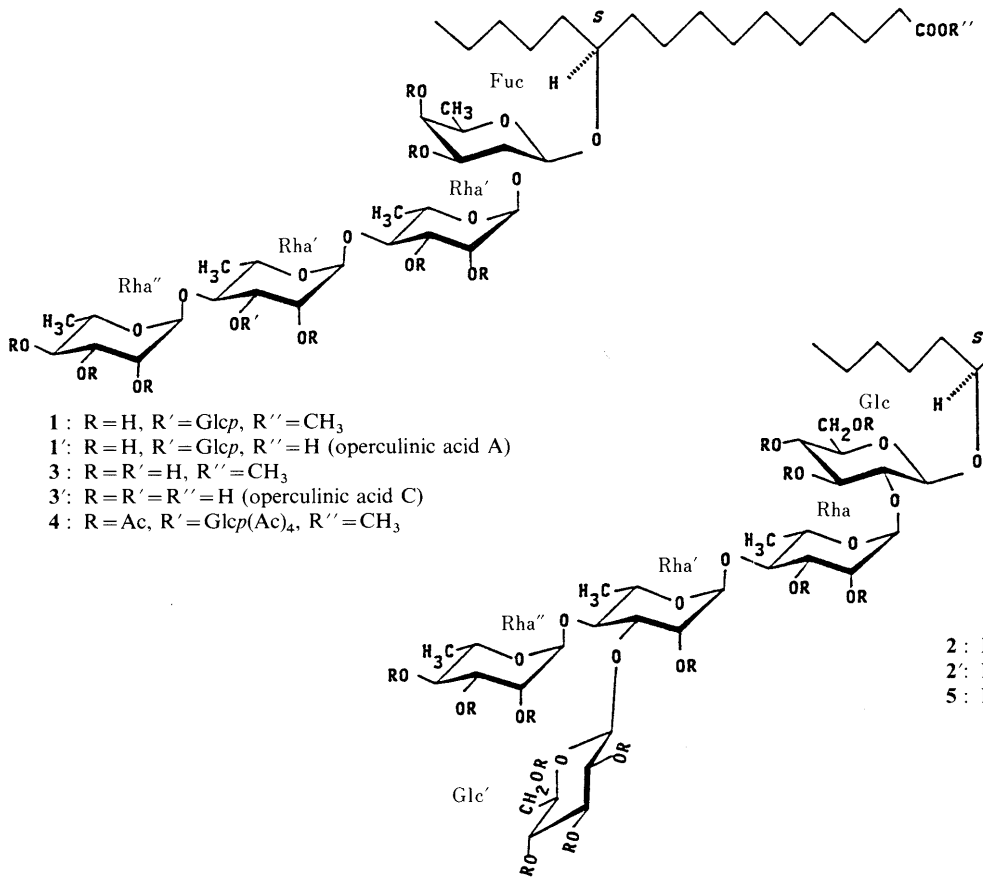


Fig. 2

Operculinic acid C methyl ester (**3**), white powder, mp 99—103 °C (dec.), $[\alpha]_D$ −91.1°, negative FAB-MS *m/z*: 869 $[(M-H)^-]$, $C_{41}H_{74}O_{19}$, furnished, on acidic hydrolysis, jalapinolic acid, L-rhamnose ($[\alpha]_D$ +10.4°) and D-fucose ($[\alpha]_D$ +78.7°). The alkaline hydrolysis product of **3**, operculinic acid C (**3′**), white powder, mp 116—120 °C, $[\alpha]_D$ −84.0°, $C_{40}H_{72}O_{19}$, revealed a $(M-H)^-$ ion at *m/z* 855 and fragment ion peaks at *m/z* 709 (855−146 (6-deoxyhexose unit)), 563 (709−146), 417 (563−146) and 271 (417−146) in the negative FAB-MS. From these data and the fact of coexistence of **1**, it was presumed that the sugar moiety of **3′** might be lacking the terminal glucose from **1′**. In addition to the results of detailed analyses of $^1$H- and $^{13}$C-NMR spectra of **3′** (Tables I and II), the appearanc of three cross peaks between 1-H of Rha and 2-H of Fuc, 1-H of Rha′ and 4-H of Rha, and 1-H of Rha″ and 4-H of Rha′ in the NOESY spectrum of **3′** led us to conclude the structure of operculinic acid C (**3′**) to be 11*S*-jalapinolic acid 11-*O*-α-L-rhamnopyranosyl-(1→4)-*O*-α-L-rhamnopyranosyl-(1→4)-*O*-α-L-rhamnopyranosyl-(1→2)-β-D-fucopyranoside, as shown in Fig. 2.

Contrary to the report of Graf, *et al.*,[5] none of acetic, tiglic, *n*-valeric, trimethylacetic, 2-methylbutyric, isovaleric and propionic acids could be detected in the organic acid fraction obtained by saponification of the "jalapin". To our knowledge, this is the first report of the presence of *n*-decanoic and *n*-dodecanoic acids as component organic acids of the resin glycosides.

The structures of the minor glucosidic acids, operculinic acids D and E will be reported elsewhere.

## Experimental

**General Procedures** All melting points (mp) were determined on a Yanaco MP-S3 apparatus and are uncorrected. IR spectra were taken on a JASCO A 302 spectrometer. $^1$H- and $^{13}$C-NMR spectra were recorded on a JEOL JNM GX-400 spectrometer. Spectra were taken as 1—2% (w/v) solutions in pyridine-$d_5$ at 26 °C with tetramethylsilane as an internal reference. COSY spectra were obtained by the use of a $^1$H–$^1$H shift correlation sequence with a 45° mixing pulse and N-type peak selection (1—2% (w/v) solution). The $^1$H–$^{13}$C shift-correlated spectra (HETCOR) were measured for 10—16% solution in pyridine-$d_5$ using a usual sequence. MS and GC-MS were obtained on a JEOL JMS DX-300 spectrometer equipped with a gas chromatograph and a JMA 3500 data system (EI-MS: ionization voltage, 30 eV; accelerating voltage, 3 kV. Negative ion FAB-MS: accelerating voltage, 1.5—2.0 kV; matrix, triethanolamine; collision gas, Xe). The analytical GC was carried out with a Shimadzu gas chromatograph GC-8A with a flame ionizing detector. Optical rotations were determined with a JASCO DIP-140 polarimeter. HPLC separation was run on a JASCO TWINCLE equipped with a Shodex SE-11 differential refractometer, and Shimadzu NUCLEOSIL 5C8 (20 mm i.d. × 250 mm) or NUCLEOSIL 5NH$_2$ (10 mm i.d. × 300 mm). Thin layer chromatography (TLC) was carried out on Silica gel precoated Al sheets (Merck, Art 5554), while for analysis of saccharides, Silica gel 60 HPTLC plates (Merck, Art 5556) were used. For column chromatography, Silica gel 60 (Merck, Art 7743), MCI gel CHP 20P (100—200 mesh, Mitsubishi Chemical Industries) and Sephadex LH-20 (25—100 μm, Pharmacia Fine Chemicals) were used.

**Extraction and Preparation of "Jalapin"** The powdered roots of *I. operculata* (0.96 kg) purchased from Paul Mugenburg GmbH & Co., Hannover, FRG were extracted with MeOH (5.5 l) at room temperature and the extract was evaporated under reduced pressure to afford a brown powder (66.68 g). This was suspended in water (125 ml) and then extracted with ether (3 × 100 ml). The ether extractive (21.79 g) was applied to an MCI gel CHP 20P column (45 mm i.d. × 230 mm; 90% MeOH (2 l)→MeOH (2 l)→ acetone (2 l)) to give three fractions, 90% MeOH (6.68 g), MeOH (12.65 g) and acetone (1.86 g). Chromatography of the MeOH fraction on an LH-20 column (MeOH) furnished a pale yellow

powder, mp 65—78 °C ("jalapin," 10.0 g). The $H_2O$ layer was partitioned between *n*-BuOH (100 ml) and $H_2O$ (125 ml) and the organic phase was evaporated *in vacuo* to give a pale yellow solid ("convolvulin," 15.84 g).

**Alkaline Hydrolysis of "Jalapin"** The "jalapin" (10.0 g) was dissolved in 1% KOH (100 ml) and heated at 95 °C for 1 h. After cooling, the reaction mixture was adjusted to pH 4.0 with 1 N HCl and shaken with ether (150, 100, 100 ml). The ether layer was dried over $MgSO_4$ and evaporated under reduced pressure to give an oil (organic acid fraction, 2.64 g). Aliquots of this fraction were methylated with diazomethane in the usual way and examined by GC-MS (column, 5% SE-30 (60—80 mesh, 3 mm i.d. × 4 m); column temperature, 170 °C; carrier gas, He (20 ml/min); EI mode), exhibiting two peaks: $t_R$ (min): 3.30 (*m/z*: 186 ($M^+$)), 6.57 (*m/z*: 214 ($M^+$)), which were identical with authentic methyl *n*-decanoic acid and methyl *n*-dodecanoic acid, respectively. The $H_2O$ layer was applied to an MCI gel column (4.5 cm i.d. × 23 cm), then eluted with $H_2O$ (1 l), 50% acetone (1 l) and acetone (0.5 l). The 50% acetone eluate was evaporated to dryness to afford a powder (6.27 g, glycosidic acid fraction).

**Acidic Hydrolysis of Glycosidic Acid Fraction** A solution of the glycosidic acid fraction (0.80 g) in 2 N $H_2SO_4$ (10 ml) was heated at 95 °C for 2 h. The reaction mixture was extracted with ether and the ether layer was washed with $H_2O$ and dried over $MgSO_4$. It was treated with diazomethane and then the mixture was evaporated to afford a solid (144 mg). The solid was purified over silica gel (benzene–AcOEt, 5:1) followed by crystallization from *n*-hexane–AcOEt to furnish methyl jalapinolate (100 mg), colorless needles, mp 43—44 °C, $[\alpha]_D^{21}$ +0.9° (*c* = 5.6, $CHCl_3$). This product was identical with an authentic methyl jalapinolate obtained from the root of *I. orizabensis* (mp 46—47 °C)[9] by mixed melting point determination, and GC and TLC comparisons.

**Isolation of Methyl Esters of Operculinic Acids A(1), B(2), C(3), D and E** The glycosidic acid fraction (6.27 g) in MeOH (50 ml) was methylated with diazomethane. The concentrated reaction mixture was chromatographed over silica gel (3.8 cm i.d. × 34 cm, $CHCl_3$–MeOH–$H_2O$, 8:2:0.2 (1000 ml)→7:3:0.5 (1600 ml)→MeOH (500 ml)) to afford fr. 1 (100 mg), fr. 2 (610 mg), fr. 3 (427 mg), fr. 4 (673 mg), fr. 5 (125 mg), fr. 6 (2.90 g), fr. 7 (900 mg) and fr. 8 (214 mg). Fraction 2 was subjected to preparative HPLC (NUCLEOSIL 5C8, 70% MeOH) to give **3** (361 mg). Fraction 3 and fr. 4 were chromatographed repeatedly over Fuji-gel ODS G3 (3.6 cm i.d. × 22 cm, 75% MeOH, 1.5 l) and Kusano CIG Si-gel (2.2 cm i.d. × 10 cm, $CHCl_3$–MeOH–$H_2O$, 6:4:0.7) to give operculinic acid E (113 mg). HPLC of fr. 6 under the same condition as for fr. 2 furnished **1** (1.88 g) and operculinic acid D (75 mg). Fraction 7 gave **1** (450 mg), operculinic acid D (106 mg) and **2** (90 mg). **1**: White powder (MeOH–$H_2O$), mp 172—173 °C (dec.), $[\alpha]_D^{18}$ −72.5° (*c* = 3.0, MeOH). IR (KBr) $cm^{-1}$: 3350 (br, OH), 1725 (C=O). Negative FAB-MS *m/z*: 1031 [(M−H)$^-$]. *Anal.* Calcd for $C_{47}H_{84}O_{24}$: C, 54.64; H, 8.19. Found: C, 54.90; H, 8.34. $^1$H-NMR $\delta$: see Table I. $^{13}$C-NMR $\delta$: see Table II. **2**: White powder (MeOH–$H_2O$), mp 157—159 °C (dec.). $[\alpha]_D^{23}$ −83.5° (*c* = 1.0, MeOH). IR (KBr) $cm^{-1}$: 3400 (br, OH), 1725 (C=O). Negative FAB-MS *m/z*: 1047 [(M−H)$^-$]. *Anal.* Calcd for $C_{47}H_{84}O_{25}$: C, 53.81; H, 8.07. Found: C, 54.05; H, 8.19. $^1$H-NMR $\delta$: see Table I. $^{13}$C-NMR $\delta$: see Table II. **3**: White powder (MeOH–$H_2O$), mp 99—103 °C (dec.). $[\alpha]_D^{23}$ −91.1° (*c* = 1.0, MeOH). IR (KBr) $cm^{-1}$: 3350 (br, OH), 1720 (C=O). Negative FAB-MS *m/z*: 869 [(M−H)$^-$]. *Anal.* Calcd for $C_{41}H_{74}O_{19}$: C, 56.54; H, 8.56. Found: C, 56.81; H, 8.43. Operculinic acid D methyl ester: white powder (MeOH–$H_2O$), mp 170—172 °C (dec.). $[\alpha]_D^{23}$ −81.7° (*c* = 1.0, MeOH). IR (KBr) $cm^{-1}$: 3400 (br, OH), 1725 (C=O). Negative FAB-MS *m/z*: 1017 [(M−H)$^-$]. *Anal.* Calcd for $C_{46}H_{82}O_{24}$: C, 54.21; H, 8.11. Found: C, 54.19; H, 7.93. Operculinic acid E methyl ester: white powder (MeOH–$H_2O$), mp 99—102 °C (dec.), $[\alpha]_D^{18}$ −65.0° (*c* = 1.2, MeOH). IR (KBr) $cm^{-1}$: 3400 (br, OH), 1725 (C=O). Negative FAB-MS *m/z*: 885 [(M−H)$^-$]. *Anal.* Calcd for $C_{41}H_{74}O_{20}$: C, 55.52; H, 8.41. Found: C, 55.75; H, 8.32.

**Acidic Hydrolysis of 1, 2 and 3** Compounds **1** (322 mg), **2** (50 mg) and **3** (50 mg) were separately dissolved in 2 N $H_2SO_4$ (3 ml) and heated at 95 °C for 1 h. The reaction mixture was diluted with $H_2O$ (5 ml) and extracted with ether (2 × 10 ml), and the ether layer was washed with $H_2O$ and dried over $MgSO_4$ and then treated with diazomethane in ether. The mixture was evaporated and the residue was crystallized from *n*-hexane–AcOEt to furnish methyl jalapinolate, colorless needles (50 mg from **1**, 9 mg from **2** and 16 mg from **3**), mp 45.5—46.5 °C. The $H_2O$ layer was neutralized with 3% KOH followed by desalting (LH-20, MeOH) to afford a sugar mixture, which was subjected to HPLC (NUCLEOSIL 5$NH_2$, 85% acetonitrile) to give L-rhamnose (53 mg), syrup, $[\alpha]_D^{24}$ +10.3° (*c* = 1.3, $H_2O$), D-fucose (23 mg), syrup, $[\alpha]_D^{24}$ +74.1° (*c* = 0.8, $H_2O$) and D-glucose (25 mg), syrup, $[\alpha]_D^{24}$ +46.8° (*c* = 1.6, $H_2O$) from **1**, L-rhamnose (10 mg), syrup, $[\alpha]_D^{21}$ +10.1° (*c* = 0.6, $H_2O$) and D-glucose (6 mg), syrup, $[\alpha]_D^{21}$ +52.9° (*c* = 0.3, $H_2O$) from **2**, and L-rhamnose (10 mg), syrup, $[\alpha]_D^{21}$ +10.4° (*c* = 1.1, $H_2O$) and D-fucose (2 mg), syrup, $[\alpha]_D^{21}$ +78.7° (*c* = 0.3, $H_2O$) from **3**.

**Acetylation of 1 and 2** Compounds **1** (20 mg) and **2** (20 mg) in $Ac_2O$–pyridine (1:1, 3 ml) were separately left to stand at room temperature overnight. The mixture was poured into ice-water (50 ml), and the precipitates were collected by filtration. **4** (25 mg): White powder, mp 83—86 °C. $[\alpha]_D^{18}$ −36.4° (*c* = 1.4, MeOH). IR (KBr) $cm^{-1}$: no OH absorption, 1745 (C=O). *Anal.* Calcd for $C_{71}H_{108}O_{36}$: C, 55.46; H, 7.08. Found: C, 55.29; H, 7.15, $^1$H-NMR $\delta$: see Table I. **5** (22 mg): White powder, mp 72—74 °C. $[\alpha]_D^{24}$ −33.6° (*c* = 2.2, MeOH). IR (KBr) $cm^{-1}$: no OH absorption, 1750 (C=O). *Anal.* Calcd for $C_{73}H_{110}O_{38}$: C, 54.95; H, 6.95. Found: C, 54.77; H, 6.94, $^1$H-NMR $\delta$: see Table I.

**Alkaline Hydrolysis of 1, 2 and 3** Solutions of **1** (27 mg), **2** (52 mg) and **3** (50 mg) in 3% KOH (6 ml) were each heated at 95 °C for 1 h. The reaction mixture were adjusted to pH 4.0 with 1 N HCl and desalted by using chromatography over MCI-gel CHP 20P to afford **1′** (24 mg), **2′** (35 mg) and **3′** (45 mg), respectively. **1′**: White powder (MeOH–$H_2O$), mp 172—174 °C. $[\alpha]_D^{18}$ −63.6° (*c* = 1.0, MeOH). IR (KBr) $cm^{-1}$: 3400 (OH), 1705 (C=O). Negative FAB-MS *m/z*: 1017 [base peak, (M−H)$^-$], 871, 855, 709, 563, 417, 271. *Anal.* Calcd for $C_{46}H_{82}O_{24}$: C, 54.21; H, 8.11. Found: C, 54.16; H, 8.18. **2′**: White powder (MeOH–$H_2O$), mp 169—171 °C. $[\alpha]_D^{24}$ −80.9° (*c* = 2.6, MeOH). IR (KBr) $cm^{-1}$: 3400 (OH), 1700 (C=O). Negative FAB-MS *m/z*: 1033 [base peak, (M−H)$^-$], 887, 871, 725, 579, 433, 271. *Anal.* Calcd for $C_{46}H_{82}O_{25}$: C, 53.38; H, 7.98. Found: C, 53.30; H, 7.97. **3′**: White powder (MeOH–$H_2O$), mp 116—120 °C. $[\alpha]_D^{18}$ −84.0° (*c* = 1.9, MeOH). IR (KBr) $cm^{-1}$: 3400 (OH), 1700 (C=O). Negative FAB-MS *m/z*: 855 [base peak, (M−H)$^-$], 709, 563, 417, 271. *Anal.* Calcd for $C_{40}H_{72}O_{19}$: C, 56.06; H, 8.47. Found: C, 56.09; H, 8.35. $^1$H-NMR $\delta$: see Table I. $^{13}$C-NMR $\delta$: see Table II.

**Acknowledgment** The authors are indebted to Dr. H. Lorenz of Paul Mugenburg GmbH & Co. Hannover, FRG for microscopical identification of the crude material. Thanks are also due to Mr. Masatoshi Nishi and Mrs. Noriko Shima of this university for measurements of the NMR spectra, MS and elemental analyses. This work was supported by a Grant-in-Aid for General Scientific Research (No. 61571022) from the Ministry of Education, Science and Culture, Japan.

**References and Notes**

1) *a*) Part IV: N. Noda, M. Nishi, K. Miyahara and T. Kawasaki, *Chem. Pharm. Bull.*, **36**, 1707 (1988); *b*) A part of this work was reported in a preliminary communication (M. Ono, K. Kubo, K. Miyahara and T. Kawasaki, *ibid.*, **37**, 241 (1989)).
2) There has been a confusion in nomenclature. That is, ether-soluble and -insoluble fractions are called in Great Britain, contrary to the terminology on the continent, "convolvulin" and "jalapin" respectively. In this paper the names are used according to the report by Shellard (E. J. Shellard, *Planta Med.*, **9**, 102 (1961)).
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7) Previously the absolute configuration at $C_{11}$ of methyl (+)-jalapinolate was determined[9,10] to be *R* by Horeau's method (A. Horeau, *Tetrahedron Lett.*, **1961**, 506; A. Horeau and H. B. Kagan, *Tetrahedron*, **20**, 2431 (1964)), but Kitagawa, *et al.* have recently revised it to *S* on the basis of a direct comparison with synthetic (*via* Sharpless epoxidation) *R* and *S* compounds.[11] We obtained the same result as reported by Kitagawa *et al.*, by application of Dale and Mosher's method (J. A. Dale and H. S. Mosher, *J. Am. Chem. Soc.*, **95**, 512 (1973)).[1b])
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