Profiling of the Resin Glycoside Content of Mexican Jalap Roots with Purgative Activity†

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Received June 24, 2006

Mexican Jalap roots, a prehispanic medicinal plant complex still considered to be a useful laxative, can be found as an ingredient in some over-the-counter products sold by herbalists in contemporary Mexico. The drug is prepared from the dried roots of several morning glories, all of which have been identified as members of the genus *Ipomoea*. Analysis of several commercial samples was assessed by generating HPLC and ^{13}C NMR spectroscopic profiles of the glycosidic acids obtained through saponification of the resin glycoside contents. These profiles distinguish the three Mexican jalaps currently in frequent use and can serve as analytical tools for the authentication and quality control of these purgative herbal drugs. *Ipomoea purga*, the authentic "jalap root", yielded two new hexasaccharides of convolvulinic and jalapinolic acids, purgic acids A (**1**) and B (**2**), respectively. Scammonic acid A (**3**), a tetrasaccharide, was produced from *Ipomoea orizabensis*, the Mexican scammony or false jalap. Operculinic acid B (**4**), a pentasaccharide, was identified in *Ipomoea stans*. Semipreparative HPLC was performed to obtain pure samples of new compounds **1** and **2** in sufficient quantity to elucidate their structure by high-field NMR spectroscopy. Purgic acid A (**1**) was identified as (11*S*)-hydroxytetradecanoic acid 11-*O*-*β*-D-quinovopyranosyl-(1→2)-*O*-*β*-D-glucopyranosyl-(1→3)-*O*-[*β*-D-fucopyranosyl-(1→4)]-*O*-α-L-rhamnopyranosyl-(1→2)-*O*-*β*-D-glucopyranosyl-(1→2)-*O*-*β*-D-quinovopyranoside, while purgic acid B (**2**) was characterized with (11*S*)-hydroxyhexadecanoic acid as its aglycon but having the same glycosidation sequence in the oligosaccharide core.

"*Cacamótic tlanoquiloni*",[1] the Aztec term for purgative remedies that consisted of diverse kinds of tuber-shaped roots, including "*tlalantlacacuitlapilli*" ("purgative root of Michoacán"),[2] varied in morphological characteristics, habitat, and potency of their physiological effects. In the literature, the plant members of this medicinal plant complex (MPC)[3] have been identified as belonging to the genus *Ipomoea* (Convolulaceae) and are currently recognized as *I. purga* (Wender) Hayne, *I. orizabensis* (Pelletan) Lebed. ex Steud., *I. stans* Cav., *I. jalapa* (L.) Pursh, *I. batatas* (L.) Lam., and *I. simulans* Hanbury, along with others less often used.[4] These perennial, herbaceous bindweeds with cathartic, acrid-tasting, and resin-producing roots attracted the attention of the Spanish colonists, since their purgative properties were important to 16th century European galenic medicine.[5]

The MPC of Mexican purgative roots was readily accepted as a New World succedaneum of the skammonia or scammony[6] MPC that had been used since pre-Christian times (e.g., Syrian or purging bindweed in 17th century England).[7] The most important of these remedies was the "root of Michoacán" known in English herbals as Indian Rhubarb, which was illustrated in the *Codex de la Cruz-Badiano* (an Aztec herbal of 1552) and prescribed by Aztec healers for purgative purposes.[8] The precise identification of this root was much disputed, although it is now agreed that it is *I. purga*.[9] In recognition of its important benefits, the colonists bestowed the vernacular name "Jalapa" on this signature species ("officinal jalap" or "Rhizoma Jalapa"), which they found in abundance in the tropical region of Xalapa, in the state of Veracruz. A second purgative root likewise restricted to the tropical areas in the Gulf of Mexico, "Orizaba jalap", identified as *I. orizabensis*, is often referred to as "false jalap", "light jalap", or "Mexican scammony" and used as a substitute for jalap root. A third morning glory with laxative qualities, but from the Mexican highlands, is now known to be the dry root of *I. stans* ("tumbavaquero" roots); it was used in the treatment of kidney inflammation, bile disorders, and epileptic attacks[10] and currently, in combination with other medicinal plants, is employed for nervous disorders.[11] In addition to their use as purgatives, the MPC of Mexican jalaps has been employed as an antihelmintic and galactogogue and in the treatment of abdominal fever, dysentery, epilepsy, hydrocephaly, skin spots, meningitis, and tumors.[12] A decoction of the root of *I. purga* is normally prepared using a 2 cm section of root to a liter of water. The usual recommendation is to drink one cup of the cold decoction, before bedtime.[13] Contemporary data on commercialization of jalap root are scarce.[14] Today, the three species are easily found in the numerous Mexican herbal markets or as an ingredient in over-the-counter products sold in health food stores in Mexico and the United States.[15] Most of the ethnobotanical, anatomical, and chemical descriptions of these purgative roots found in the literature are confusing and not scientifically traceable. There is an obvious question as to whether much of the exportation from Mexico during more than 450 years was actually jalap root or a mixture of the roots of different species of *Ipomoea* (closely related to *I. purga* or not).[16]

As part of a continuing effort to elucidate the structural diversity found in the convolvulaceous resin glycosides,[17] the present study focuses on the chemical differences found among the Mexican jalap roots, *I. purga* (Figure S1, Supporting Information), the authentic jalap root, and the substitute species, *I. orizabensis*[18] (Figure S2, Supporting Information) and *I. stans* (Figure S3, Supporting Information). This research involved the generation of HPLC and ^{13}C NMR spectroscopic profiles of their respective glycosidic acid components, which are the saponification derivatives of the resin glycosides. In the case of the new purgic acids A (**1**) and B (**2**), multiple semipreparative HPLC collections were performed to obtain pure samples in sufficient quantities to elucidate their structures.

† This work was taken in part from the Ph.D. theses of E. Escalante-Sánchez and B. Hernández-Carlos.

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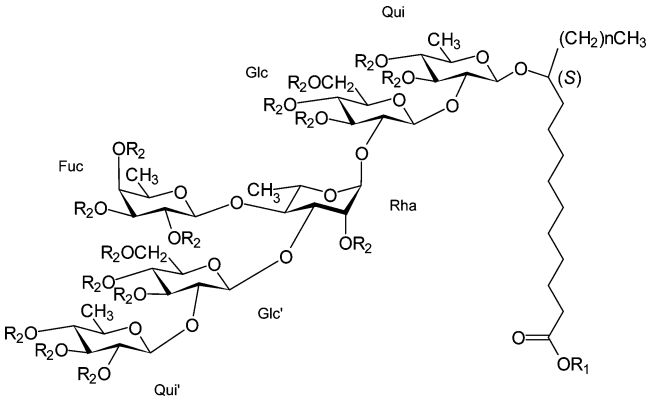
10.1021/np060295f CCC: $33.50
Published on Web 10/11/2006



	R_1	R_2	n
1	H	H	2
2	H	H	4
5	CH_3	H	2
6	CH_3	H	4
7	CH_3	Ac	2
8	CH_3	Ac	4

Results and Discussion

Convolvulaceous resin glycosides are a challenging group of molecules to separate and purify. Difficulties encountered during the purification of these intact molecules are caused by the presence of isomeric structures with differing esters, determining the sites and extent of esterification, as well as variations in the sequence of glycosylation of the oligosaccharide cores.[17] In many cases their high molecular weights and amphiphilic properties have prevented the HPLC isolation of pure samples needed as chromatographic standards to assess the quality of some commercial products.[19] A common feature of all resin glycosides, either $CHCl_3$-soluble ("jalapin" or nonpolar fraction) or MeOH-soluble ("convolvulin" or polar fraction) residues,[17] is the liberation of a major glycosidic acid upon hydrolysis. The same H_2O-soluble product is yielded from both fractions.[20] The structure of this product is distinctive enough for chromatographic and spectroscopic differentiation of the resin glycoside mixture in each species of *Ipomoea*. For the identification of these polar glycosidic acids in the three Mexican jalaps analyzed, $CHCl_3$ and MeOH extracts were prepared from authenticated samples and then subsequently submitted to saponification. The dried neutral aqueous phases from this hydrolysis were directly analyzed by ^{13}C NMR spectroscopy (Figure 1) to generate data to be used as fingerprint tools for the pattern recognition of each jalap. The characteristic anomeric signals were easily distinguishable in the spectra and constituted a good starting point for structure elucidation. These resonances around δ 95−110, outside the bulk region (δ 60−80), permitted an immediate estimation of the number of different monosaccharide residues in each glycosidic acid and were used as structural "reporter" signals for each resin: a tetrasaccharide (**3**) for *I. orizabensis*, a pentasaccharide (**4**) for *I. stans*, and a hexasaccharide for *I. purga*. Each concentrate from these mixtures was analyzed by negative FABMS to identify the pseudomolecular ion $[M - H]^-$.[17]

A carbohydrate HPLC separation (Figure 2) was performed by application of hydrophilic-interaction chromatography using an aminopropyl silane-base column,[21] which, under isocratic conditions (CH_3CN-H_2O, 3:2), resolved the major oligosaccharide present in each analyzed commercial sample.[22] This method allowed the efficient differentiation of each glycosidic acid with the following retention times (t_R): 6.6 min for purgic acid A (**1**) from authenticated jalap root (*I. purga*, sample code IP); 5.6 min for scammonic acid (**3**) from the "Mexican scammony" (*I. orizabensis*, sample code IO); and 11.8 min for operculinic acid B (**4**) from *I. stans* (sample code IS). These profiles were compared with those generated from commercial samples (Figures S4 and S5, Supporting Information).

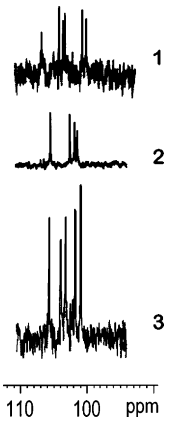


Figure 1. Expansion at 90−110 ppm of ^{13}C NMR spectra of saponified crude extracts from authenticated Mexican jalap root samples: **1**, *Ipomoea purga* ("Rhizoma Jalapae"); **2**, *Ipomoea orizabensis* ("Mexican scammony"); **3**, *Ipomoea stans* ("tumbavaquero" root, a substitute or false jalap).

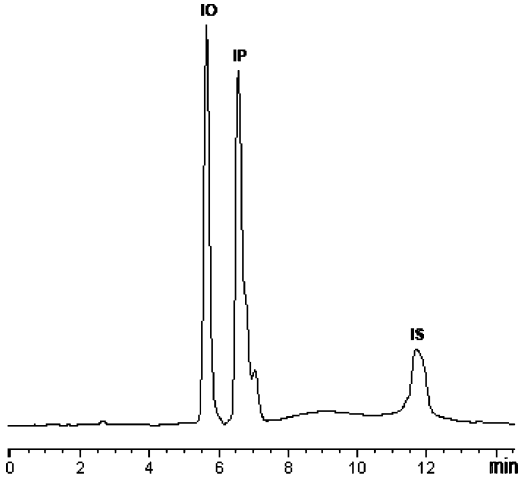
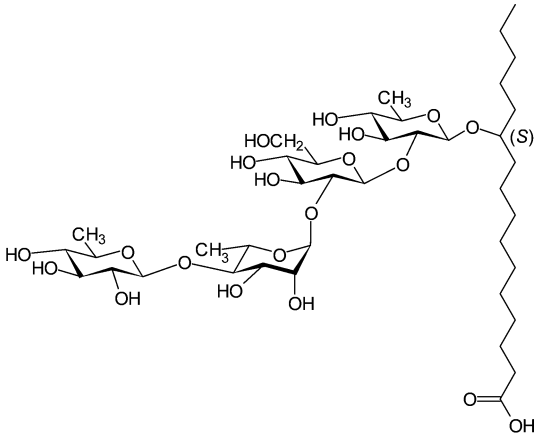


Figure 2. HPLC coelution profile of saponified crude MeOH extracts from authenticated Mexican jalap root samples. Instrumental conditions: stationary phase, standard column for carbohydrate analysis (3.9 × 300 mm, 10 μm; Waters); mobile phase, CH_3CN-H_2O (3:2); flow rate, 0.5 mL/min; injection volume, 20 μL (sample concentration: IP = 0.25 mg; IO = 0.125 mg; IS = 0.15 mg); detection: refractive index. Abbreviations used: IP, *Ipomoea purga*; IO, *Ipomoea orizabensis*; and IS, *Ipomoea stans*.

I. purga and *I. orizabensis* samples were also submitted to methylation (CH_2N_2 treatment) in order to reduce the polarity of the analytes and decrease the amount of water in the mobile phase (CH_3CN-H_2O, 4:1) in the HPLC analysis. Chromatograms exhibited no substantial difference in composition (Figure S4, Supporting Information) for the commercial powder (sample code IP-3), the two commercial root samples (codes IP-1 and IP-2), and the collected wild sample of the jalap root (code IP). Neither was there a significant difference displayed between the powdered commercial sample of false jalap (code IO-1) with that of the "Mexican scammony" crude resin (Figure S4, Supporting Information). However, the chromatogram for the commercial roots of "tumbavaquero" (*I. stans*, sample code IS-2) indicated adulteration with "Mexican scammony" (Figure S5, Supporting Information). Although "Mexican scammony" and "tumbavaquero" roots are so similar in appearance to the non-professionally trained eye as to be indistinguishable when cut into small sections (Figure S3, Supporting Information), our results proved that there is a significantly greater yield of resin glycosides in the "Mexican scammony" (Figure S6, Supporting Information).[23] The previous chemical



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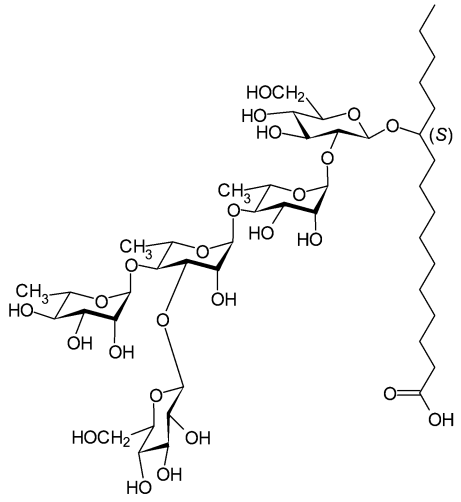

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studies on *I. stans*[24] are consistent with our results for *I. orizabensis*,[18] raising the question as to whether the previously analyzed samples were correctly identified or were in reality "Mexican scammony" or an adulterated sample.[25]

Semipreparative HLPC was performed to obtain pure compounds from each saponified sample. Scammonic acid A (**3**)[26] and operculinic acid B (**4**)[27] were identified by comparison of NMR data with published values. New glycosidic acids **1** and **2** from *I. purga* were obtained in sufficient quantity to elucidate their structure (Figure S7, Supporting Information). The main approaches involved were the use of degradative chemical reactions to break up the large oligosaccharides and obtain smaller, more manageable molecules, together with the use of a combination of high-field NMR spectroscopy and high-resolution FABMS applied directly to the glycosidic acids as well as to their methylated and peracetylated derivatives. Compounds **1** and **2** were hydrolyzed in acid, and their Et_2O-soluble extract was methylated and then silylated. GC-MS analysis of the aglycon of **1** showed that the mass spectrum corresponded to that of the trimethylsilyl derivative of methyl 11-hydroxytetradecanoate because of the diagnostic α-cleavage ions at *m*/*z* 287 and 145. Compound **2** yielded methyl 11-hydrohexadecanoate (*m*/*z* 287 and 173) as the aglycon. The HPLC analysis of the aqueous phase of the acid hydrolyzate led to the identification of rhamnose, quinovose, fucose, and glucose in the approximate ratio 1:2:1:2. Coelution experiments with authentic samples confirmed these assignments and ratios.

High-resolution negative FABMS of compound **1** showed the $[M - H]^-$ ion at *m*/*z* 1151.5338, indicating a molecular formula of $C_{50}H_{87}O_{29}$ and accounting for the presence of a 11-hydroxytetradecanoic acid moiety glycosidically linked to a hexasaccharide consisting of four deoxyhexose units (2 × quinovose, 1 × rhamnose, and 1 × fucose) and two hexoses (glucose), with these results confirmed by HPLC of the acid hydrolysis mixture. The peak at *m*/*z* 1005 represented a mass difference of 146 (methylpentose unit) from the $[M - H]^-$ ion, indicating the loss of a terminal deoxyhexose. Cleavage at the next monosaccharide unit corresponded to the loss of glucose, affording the peak at *m*/*z* 843 $[1005 - 162$ (hexose unit)$]^-$. The next intense peak at *m*/*z* 551 was 292 mass units lower and represented the loss of two methylpentose units, which indicated that the oligosaccharide core is branched at one of these two deoxyhexoses.[28] This was confirmed by additional minor peaks at *m*/*z* 859 $[M - H - 146 \times 2$ (two methylpentose units) $]^-$ and 697 $[843 - 146$ (methylpentose unit)$]^-$. The fragment ion at *m*/*z* 389 represented the loss of a glucose unit from the peak at *m*/*z* 551. The further loss of a methylpentose unit gave a peak at *m*/*z* 243, corresponding to the aglycon anion, 11-hydroxytetradecanoic acid (convolvulinic acid). Negative-ion FABMS of compound **2** was performed and provided an intense pseudomolecular $[M - H]^-$ ion at *m*/*z* 1179. The difference of 28 mass units (two methylene groups) between compounds **1** and **2** as well as the production of the same general fragmentation pattern by glycosidic cleavage of each sugar moiety at *m*/*z* 1033, 887, 871, 725, 579, 417, and 271 confirmed the similar branched hexasaccharide core in both acids and the presence of 11-hydroxyhexadecanoic acid (jalapinolic acid) as the aglycon for **2**.

The ^1H (Figure S8, Supporting Information) and ^{13}C NMR data (Table 1) of the methyl ester derivatives **5** and **6** showed features generally similar to those of the known convolvulaceous resin glycosides,[17,20] especially the hexasaccharide from cowvine (*I. lonchophylla*).[28] In the low-field region of the HMQC NMR spectrum, six anomeric signals were confirmed at δ 4.7 (1H, d, qui-1; δ_C 102.2); 5.7 (1H, d, glc-1; δ_C 102.4); 6.4 (1H, brs, 1.5, rha-1; δ_C 100.1); 6.2 (1H, d, glc′-1; δ_C 100.6); 5.1 (1H, d, qui′-1; δ_C 102); and 5.8 (1H, d, fuc-1; δ_C 102.9). Therefore, six separate spin systems for sugar skeletons were readily distinguished in the $^1H-^1H$ COSY and TOCSY spectra. This allowed for the assignment of chemical shift values for C-bonded protons in the six individual monosaccharide moieties, permitting the identification of each sugar unit. Carbon signals of each fragment were assigned by HMQC experiments. The inter-glycosidic connectivities were established on the basis of detailed long-range heteronuclear coupling correlations ($^3J_{CH}$) by HMBC studies.[17] For example, the following key correlations were observed in compounds **5** and **6**: (a) a connectivity between H-1 on the internal quinovose (δ 4.78) and C-11 of the fatty acid (δ 80.1); (b) H-1 on glucose (δ 5.7) with C-2 on the inner quinovose (δ 80.9); (c) H-1 on the external quinovose (δ 5.09) with C-2 on the external glucose′ (δ 100.6). A significant signal overlap in the proton region δ 3.7−4.6 hampered complete assignments for the glycosidation sequence. To enhance signal dispersion, it was decided to prepare their peracetylated derivatives **7** and **8** (Figure S9, Supporting Information). COSY (Figure S10, Supporting Information), TOCSY, and HMQC (Figure S11, Supporting Information) were used to assign the important ^1H and ^{13}C chemical shifts of each sugar unit. ROESY (Figure S12, Supporting Information) and HMBC correlations completed the linkage within the hexasaccharide core. The anomeric configuration in each sugar unit was deduced from a 2D $^1J_{CH}$ NMR experiment.[17] For D-sugars in the $^4C_1$ conformation, the α-anomeric configuration (β-equatorial C−H bond) has a $^1J_{CH}$ value of 170 Hz, which is 10 Hz higher than that (ca. 160 Hz) for the β-anomer (α-axial C−H bond).[29] From the completely resolved multiplets for the six anomeric signals in the ^{13}C NMR spectra of derivatives **5** and **6**, the $^1J_{CH}$ values for qui = 157 Hz, glc = 156 Hz, glc′ = 162 Hz, fuc = 159, and qui′ = 162 Hz supported their β-anomeric configurations, with the assumption made that all monosaccharides were in their naturally occurring forms, i.e., as D-sugars. The α-configuration was deduced for the L-rhamnopyranosyl unit ($^1J_{CH}$ =171 Hz).[17] Therefore, the

Table 1. Assignments of ^{1}H and ^{13}C NMR Signals for Compounds **5** and **6** in C_5D_5N[a]

	5		**6**	
position[b]	δ H	δ C	δ H	δ C
qui-1	4.78 d (7.5)	102.4	4.76 d (7.5)	102.4
2	4.18−4.12*	80.9	4.18−4.12*	80.9
3	4.63 dd (9.0, 9.0)	78.5	4.62 dd (8.0, 8.0)	78.5
4	3.52 dd (9.0, 9.0)	77.1	3.52 dd (8.5, 8.5)	77.1
5	3.70 dq (9.0, 6.5)	72.1	3.68 dq (9.0, 6.0)	72.2
6	1.45 d (6.5)	18.3	1.44 d (6.0)	18.3
glu-1	5.75 d (8.0)	102.3	5.74 d (7.5)	102.3
2	4.35−4.23*	77.8	4.32−4.24*	77.8
3	4.35−4.23*	76.4	4.32−4.24*	76.4
4	4.05−3.97*	72.5	4.04−3.97*	72.5
5	3.80−3.74*	77.5	3.80−3.74*	77.5
6a	4.44 dd (11.0, 3.0)	62.8	4.43 dd (12.0, 2.5)	62.8
6b	4.25 dd (11.0, 6.0)		4.24 dd (12.0, 5.5)	
rha-1	6.45 d (1.5)	100.1	6.44 d (2.0)	100.1
2	4.85 dd (1.5, 3.0)	71.7	4.85**	71.8
3	5.30 dd (9.5, 3.0)	78.2	5.30 dd (9.5, 3.0)	78.2
4	4.81 dd (10.0, 9.5)	78.9	4.88 dd (9.5, 9.5)	78.9
5	5.20 dq (10.0, 6.0)	67.7	5.19 dq (9.5, 6.0)	67.7
6	1.95 d (6.0)	19.0	1.94 d (6.0)	19.0
glu′-1	6.23 d (7.5)	100.6	6.23 d (7.5)	100.6
2	4.09−3.97*	84.7	4.04−3.97*	84.8
3	4.52 dd (9.0, 9.0)	76.8	4.53 dd (8.5, 8.5)	76.7
4	4.05−3.97*	72.2	4.04−3.97*	72.3
5	4.18−4.12*	79.5	4.18−4.12*	79.5
6a	4.56−4.50*	63.3	4.55−4.50*	63.3
6b	4.18−4.12*		4.18−4.12*	
qui′-1	5.09 d (8.0)	104.8	5.09 d (8.0)	104.8
2	4.06 dd (9.0, 8.0)	76.1	4.06 dd (9.0, 8.0)	76.1
3	4.04−3.97*	77.5	4.04−4.00*	77.4
4	3.61 dd (9.5, 9.5)	76.8	3.62 dd (9.5, 9.0)	76.8
5	3.74 dq (9.5, 6.5)	73.6	3.74 dq (9.5, 6.0)	73.7
6	1.66 d (6.5)	18.6	1.66 d (6.0)	18.6
fuc-1	5.78 d (7.5)	102.9	5.77 d (7.5)	102.9
2	4.35−4.23*	73.4	4.32 dd (9.0, 7.5)	73.5
3	4.35−4.23*	74.9	4.32−4.24*	75.0
4	4.04−3.97*	72.8	4.04−3.97*	72.9
5	4.18−4.12*	71.0	4.18−4.12*	71.1
6	1.54 d (6.0)	16.9	1.53 d (6.5)	17.0
con-1		174.0		
2	2.32 t (7.0)	34.1		
11	3.81 m	80.1		
14	0.86 t (7.0)	14.3		
jal-1				174.0
2			2.32 t (8.0)	34.1
11			3.80 m	80.5
16			0.91 t (7.0)	14.2
MeO	3.40 s	51.2	3.63 s	51.6

[a] 500 MHz for ^{1}H and 125.7 MHz for ^{13}C NMR. Chemical shifts (δ) are in ppm relative to TMS. The spin coupling (*J*) is given in parentheses (Hz). Chemical shifts marked with an asterisk (*) indicate overlapped signals. Spin-coupled patterns are designated as follows: s = singlet, d = doublet, t = triplet, m = multiplet, dq = doublet quartet. All assignments are based on ^{1}H−^{1}H COSY, TOCSY, HMQC, and HMBC experiments. [b]Abbreviations: qui = quinovose; glu = glucose; rha = rhamnose; fuc = fucose; jal = 11-hydroxyhexadecanoyl; con = 11-hydroxytetradecanoyl.

structure of purgic acid A (**1**) was characterized as (11*S*)-hydroxytetradecanoic acid 11-*O*-*β*-D-quinovopyranosyl-(1→2)-*O*-*β*-D-glucopyranosyl-(1→3)-*O*-[*β*-D-fucopyranosyl-(1→4)]-*O*-α-L-rhamnopyranosyl-(1→2)-*O*-*β*-D-glucopyranosyl-(1→2)-*O*-*β*-D-quinovopyranoside, while purgic acid B (**2**) was found to possess (11*S*)-hydroxyhexadecanoic acid as its aglycon but having the same glycosidation sequence as **1** for the oligosaccharide core.

The identification of the active principles found to be effective in local herbal drugs through the use of HPLC and/or NMR profiling facilitates the prediction for therapeutic alternatives when signature species are not available. The verification of both signature species and substitutes is important to validate and standardize "genuine" herbal drugs in pharmacopoeias.

Experimental Section

General Experimental Procedures. All melting points were determined on a Fisher-Johns apparatus and are uncorrected. Optical rotations were measured with a Perkin-Elmer model 241 polarimeter. ^{1}H (500 MHz) and ^{13}C (125.7 MHz) NMR experiments were conducted on a Bruker DMX-500 instrument. Negative-ion LRFABMS and HRFABMS were recorded using a matrix of triethanolamine on a JEOL SX-102A spectrometer. The instrumentation used for HPLC analysis consisted of a Waters 600E multisolvent delivery system equipped with a Waters 410 differential refractometer detector (Waters Corporation, Milford, MA). Control of the equipment, data acquisition, processing, and management of chromatographic information were performed by the Millennium 32 software program (Waters). GC-MS was performed on a Hewlett-Packard 5890-II instrument coupled to a JEOL SX-102A spectrometer. GC conditions: HP-5MS (5%-phenyl)−methylpolysiloxane column (30 m × 0.25 mm, film thickness 0.25 μm); He, linear velocity 30 cm/s; 50 °C isothermal for 3 min, linear gradient to 300 °C at 20 °C /min; final temperature hold, 10 min. MS conditions: ionization energy, 70 eV; ion source temperature, 280 °C; interface temperature, 300 °C; scan speed, 2 scans s^{-1}; mass range, 33−880 amu.

Plant Material. Roots of *Ipomoea purga* were collected in abandoned plantations in Coxmatla near Xico, Veracruz, Mexico, in November 1996. The plant material was identified by the botanist Alberto Linajes and then compared with previously deposited vouchers housed in the Herbarium of the Instituto de Ecología, A.C. (Ortega 380, 408, 475 XAL; Martínez-Vázquez 563, 613, 661, XAL). A voucher specimen was archived at the Departamento de Farmacia, Facultad de Química, UNAM (sample code IP). Two samples of the herbal drug "raíz de jalapa" (jalap root) and two of "tumbavaquero" (*I. stans*) were purchased in March 2002 and June 2004 at "Las Plantas Medicinales de América, S.A. de C.V.", a herbal shop in Mexico City. A small sample of each drug (150 g) was visually examined and archived at the Departamento de Farmacia, Facultad de Química, UNAM. Voucher specimens (R. Pereda 01−02; sample codes IP-1, IP-2, IS-1, and IS-2, respectively) were deposited in the Ethnobotanical Collection of the National Herbarium (MEXU), Instituto de Biología, UNAM. Powdered, commercial samples of *I. purga* (R. Bye 26909; sample code IP-3, January 1999) and *I. orizabensis* (R. Bye 17761: sample code IO, March 1990; 26909: sample code IO-1, January 1999) were obtained from Laboratorios Mixim, S.A. de C.V. Mexico, manufacturers of botanical extracts, as was a crude commercial sample of "Mexican scammonny" resin (R. Pereda 06: sample code IO-2), July 1997. The root of *I. stans* was collected in Tepeaculco, Hidalgo, Mexico, in February 2003. The plant was identified by two of the authors, R.B. and E.L. A voucher specimen (Bye & Linares 28184; sample code IS) was deposited at the Ethnobotanical Collection of the National Herbarium (MEXU), Instituto de Biología, UNAM.

Generation of HPLC Profiles. Samples for HPLC analysis were prepared as follows. In brief, each commercial herbal drug (20 g) was first extracted with $CHCl_3$ (500 mL) and then with MeOH (500 mL) for 24 h. The maceration was performed twice, and the solvents were removed at reduced pressure. A solution of each extract (200 mg) in 5% KOH−H_2O (5 mL) was refluxed at 95 °C for 2 h. The reaction mixture was acidified and extracted with $CHCl_3$ (20 mL) followed by EtOAc (20 mL). The aqueous phase was extracted with *n*-BuOH (10 mL), concentrated to dryness, filtered over Whatmann No. 1 paper after charcoal addition, and subsequently analyzed by HPLC.

The HPLC glycosidic acid profile for each saponified total extract or resin glycoside fraction was generated on a Waters standard column for carbohydrate analysis (aminopropylmethylsilyl-bonded amorphous silica; 3.9 × 300 mm, 10 μm) with an isocratic elution of CH_3CN−H_2O (3:2), a flow rate of 0.5 mL/min, and a filtered (syringe filter with nylon membrane; 13 mm, 0.45 μm) sample injection of 10 μL (sample concentration: 10 mg/mL). Coelution experiments with the test samples afforded the corresponding HPLC profiles (Figure 2): t_R 5.6 min (IO: compound **3**), 6.6 min (IP: compound **1**), and 11.8 min (IS: compound **4**). In order to decrease the polarity of the analytes, the crude saponification mixtures of *I. purga* and *I. orizabensis* (100 mg) were treated with CH_2N_2 and directly analyzed by HPLC: elution, CH_3CN−H_2O (4:1); a flow rate of 0.5 mL/min, and an injection volume

of 10 μL (sample concentration: 10 mg/mL): t_R 8.2 min (IO: compound **3** as a methyl ester) and 10.5 min (IP: compound **2** as a methyl ester).

Generation of ^{13}C NMR Profiles. The spectra were recorded on a Bruker DMX-500 spectrometer equipped with a multinuclear inverse detection probe with z-gradient, operating at 125.8 MHz. The spectra were measured at 298 K using C_6D_5N solutions (sample concentration: 200 mg/mL) in 5 mm NMR tubes and referencing to TMS. The spectra were acquired using the zgdc pulse sequence in 32K files, a 10.0 μs pulse (56 deg), and a relaxation delay of 800 ms. Typically, samples were measured using a spectral width of 31 500 Hz, an acquisition time of 160 ms, and a total of 82 000 scans.

Extraction and Isolation of Resin Glycoside Fractions. *Ipomoea purga* (IP): roots (1 kg) were powdered and defatted by maceration at room temperature with hexane. The residual material was extracted exhaustively with $CHCl_3$ and MeOH and after removal of the solvent afforded, in turn, a brownish extract (540 mg) and a dark brown syrup (170 g). The crude MeOH extract was subjected to column chromatography over silica gel (1 kg) in a gravity column using a mixture of MeOH−$CHCl_3$−Me_2CO (5:12:3). A total of 100 fractions (150 mL each) was collected, and these were partially recombined to give several pools containing resin glycosides (165 g). *Ipomoea orizabensis* (IO): powdered commercial roots (1 kg) were defatted with hexane and extracted exhaustively with $CHCl_3$ by maceration at room temperature to give, after removal of the solvent, a brown syrup (160 g). Then, the plant material was extracted with MeOH to yield a dark brown syrup (659 mg), which was rich in simple carbohydrates. The crude $CHCl_3$ resinous extract was subjected to column chromatography over silica gel (500 g) using a gradient of MeOH and $CHCl_3$. Eluates containing resin glycosides (R_f 0.58; TLC: silica gel 60 F_{254} sheets; $CHCl_3$−MeOH, 43:7) were combined (132 g). *Ipomoea stans* (IS): roots (3.5 kg) were powdered and defatted with hexane at room temperature. Then, the material was extracted with $CHCl_3$−MeOH to give, after removal of solvents, a brownish, oily residue (142 g). The crude extract was subjected to column chromatography over silica gel (500 g) using $CHCl_3$ in hexane followed by a $CHCl_3$−MeOH gradient. Elution with $CHCl_3$−MeOH (9:1) afforded fractions (1.1 g) containing resin glycosides (R_f 0.61).

Alkaline Hydrolysis of Resin Glycoside Fractions. A solution of each crude resin glycoside fractions (200 mg) in 5% KOH−H_2O (5 mL) was refluxed at 95 °C for 3 h. Each reaction mixture was acidified to pH 4.0 and extracted with $CHCl_3$ (30 mL) followed by EtOAc (30 mL). The organic layers were combined, washed with H_2O, dried over anhydrous Na_2SO_4, and evaporated under reduced pressure. The aqueous phase was extracted with *n*-BuOH (30 mL), concentrated to dryness, and analyzed by HPLC as described above. The residue from the organic phase was directly analyzed by GC-MS. *I. purga* afforded three peaks: 2-methylbutyric acid (t_R 5.06 min): *m/z* $[M]^+$ 102 (3), 87 (33), 74 (100), 57 (50), 41 (28), 39 (8); *n*-decanoic acid (t_R 9.65 min): *m/z* $[M]^+$ 172 (2), 155 (3), 143 (12), 129 (62), 115 (15), 112 (12), 87 (20), 73 (100), 60 (90), 57 (40), 55 (45), 43 (30), 41 (35), 39 (6); and *n*-dodecanoic acid (t_R 10.93 min): *m/z* $[M]^+$ 200 (15), 183 (2), 171 (18), 157 (40), 143 (10), 129 (48), 115 (20), 101 (15), 85 (33), 73 (100), 60 (80), 57 (30), 55 (47), 43 (44), 41 (30). *I. orizabensis* afforded three peaks: 2-methylbutyric acid (t_R 5.12 min); tiglic acid (t_R 6.95 min): *m/z* $[M]^+$ 100 (100), 85 (25), 55 (85), 39 (38); and 3-hydroxy-2-methylbutyric acid (t_R 7.95 min): *m/z* $[M]^+$ 118 (0.5), 103 (5.0), 100 (95), 83 (12), 73 (15), 58 (30), 55 (43), 45 (38), 44 (30), 43 (48), 41 (30), 39 (25), 31 (15), 29 (18), 27 (30). The analysis of the volatile acids from the resin glycoside fraction of *I. stans* resulted in the identification of three major peaks: *n*-octanic acid (t_R 8.22 min): *m/z* $[M]^+$ 144 (3), 127 (1), 115 (15), 101 (30), 85 (10), 73 (85), 60 (100), 55 (20), 43 (25), 43 (28), 41 (18), 39 (6); *n*-decanoic acid (t_R 9.63 min); and *n*-hexadecanoic acid (t_R 13.11 min): *m/z* $[M]^+$ 256 (15), 227 (4), 213 (8) 185 (2), 171 (5), 157 (4), 143 (1), 129 (18), 115 (9), 101 (2), 97 (10), 85 (18), 73 (90), 60 (80), 57 (30), 55 (23), 43 (100), 41 (50).

Semipreparative HPLC Separation of Glycosidic Acids. The analytical separation was scaled up and modified to semipreparative HPLC for the isolation of individual glycosidic acids. Each saponification mixture was subjected to preparative HPLC on a Waters μBondapak NH_2 column (7.8 × 300 mm; 10 μm). The elution was isocratic with CH_3CN−H_2O (4:1), using a flow rate of 3 mL/min and a sample injection of 500 μL (20−100 mg/mL). For *I. purga*, the crude saponification residue (20 mg) was purified to give compounds **1** (17.4 mg, t_R = 13.9 min) and **2** (2 mg, t_R = 6.6 min). For *I. orizabensis*, the saponification residue (100 mg) afforded compound **3** (87.2 mg, t_R = 5.7 min). Compound **4** (38.6 mg, t_R = 13.4 min) was obtained from the saponification mixture of resin glycosides (50 mg) of *I. stans*.

Purgic acid A (1): white powder; mp 162−164 °C; $[\alpha]_D$ −20 (*c* 0.2, MeOH); negative FABMS *m/z* 1151 $[M - H]^-$, 1005 [M − H − 146 (methylpentose unit)]$^-$, 859 [1005 − 146 (methylpentose unit)]$^-$, 843 [1005 − 162 (hexose unit)]$^-$, 697 [843 − 146 (methylpentose unit)]$^-$, 551 [697 − 146 (methylpentose unit)]$^-$, 389 [551 − 162 (hexose unit)]$^-$, 243 [389 − 146 (methylpentose unit); convolvulinic acid methyl ester − H]$^-$; HRFABMS *m/z* 1151.5338 $[M - H]^-$ (calcd for $C_{50}H_{87}O_{29}$ requires 1151.5333).

Purgic acid B (2): white powder; mp 132−134 °C; $[\alpha]_D$ −14 (*c* 0.14, MeOH); negative FABMS *m/z* 1179 $[M - H]^-$, 1033 [M − H − 146 (methylpentose unit)]$^-$, 887 [1033 − 146 (methylpentose unit)]$^-$, 871 [1033 − 162 (hexose unit)]$^-$, 725 [871 − 146 (methylpentose unit)]$^-$, 579 [725 − 146 (methylpentose unit)]$^-$, 417 [579 − 162 (hexose unit)]$^-$, 271 [389 − 146 (methylpentose unit); convolvulinic acid methyl ester − H]$^-$; HRFABMS *m/z* 1179.5650 $[M - H]^-$ (calcd for $C_{52}H_{91}O_{29}$ requires 1179.5646).

Scammonic acid A (3): white powder; mp 154−157 °C; $[\alpha]_D$ −51 (*c* 1.0, MeOH); negative FABMS *m/z* 871 $[M - H]^-$; ^{13}C NMR anomeric signals 104.9, 102.1, 101.5, 101.1; identified by comparison of NMR data with published values.[26]

Operculinic acid B (4): white powder; mp 170−171 °C; $[\alpha]_D$ −80 (*c* 1.0, MeOH); HRFABMS *m/z* 1033.5066 $[M - H]^-$ (calcd for $C_{46}H_{81}O_{25}$ requires 1033.5067); ^{13}C NMR anomeric signals 101.0, 101.5, 102.8, 103.5, 105.4; identified by comparison of spectral data with published values.[27]

Derivatization of 1 and 2. A sample of the saponification mixture of *I. purga* (150 mg) was methylated with CH_2N_2 to further perform the separations by HPLC of the methyl ester derivatives **5** (138 mg, t_R = 25.7 min) and **6** (12 mg, t_R = 23.4 min), using an isocratic elution with CH_3CN−H_2O (3:2) and a flow rate of 4 mL/min. Each residue (10 mg) was acetylated (Ac_2O−C_5H_5N, 2:1) and subjected to preparative HPLC on a silica gel column (ISCO, 21.2 × 250 mm, 10 μm,). The elution was isocratic with $CHCl_3$−Me_2CO (9:1) using a flow rate of 6 mL/min to afford peracetylated derivatives **7** (8.7 mg, t_R =17.6 min) and **8** (8.3 mg, t_R = 29.8 min).

Purgic acid A methyl ester (5): white powder; mp 152−154 °C; $[\alpha]_D$ −77 (*c* 1.0, MeOH); ^{1}H and ^{13}C NMR, see Table 1; negative FABMS *m/z* 1165 $[M - H]^-$, 1019 [M − H − 146 (methylpentose unit)]$^-$, 873 [1019 − 146 (methylpentose unit)]$^-$, 857 [1019 − 162 (hexose unit)]$^-$, 711 [857 − 146 (methylpentose unit)]$^-$, 565 [711 − 146 (methylpentose unit)]$^-$, 403 [565 − 162 (hexose unit)]$^-$, 257 [403 − 146 (methylpentose unit); convolvulinic acid methyl ester − H]$^-$; HRFABMS *m/z* 1165.5485 $[M - H]^-$ (calcd for $C_{51}H_{89}O_{29}$ requires 1165.5490).

Purgic acid B methyl ester (6): white powder; mp 146−148 °C; $[\alpha]_D$ −29 (*c* 1.0, MeOH); ^{1}H and ^{13}C NMR, see Table 1; negative FABMS *m/z* 1193 $[M - H]^-$, 1047 [M − H − 146 (methylpentose unit)]$^-$, 901 [M − H − 146 × 2 (two methylpentose units)]$^-$, 885 [1047 − 162 (hexose unit)]$^-$, 739 [885 − 146 (methylpentose unit)]$^-$, 593 [739 − 146 (methylpentose unit)]$^-$, 431 [593 − 162 (hexose unit)]$^-$, 285 [431 − 146 (methylpentose unit); jalapinolic acid methyl ester − H]$^-$; HRFABMS *m/z* 1193.5810 $[M - H]^-$ (calcd for $C_{53}H_{93}O_{29}$ requires 1193.5802).

Spectroscopic Characterization of 7: mp 98−100 °C; $[\alpha]_D$ −50 (*c* 1.0, $CHCl_3$); negative FABMS *m/z* 1795 $[M - H]^-$; ^{1}H NMR (C_5D_5N, 500 MHz) δ 4.71 (1H, d, *J* = 7.5 Hz, Qui-1), 4.22 (1H, dd, *J* = 9.5, 7.5 Hz, Qui-2), 5.78 (1H, dd, *J* = 9.5, 9.5 Hz, Qui-3), 5.07 (1H, dd, *J* = 9.5, 9.5 Hz, Qui-4), 4.13 (1H, dq, *J* = 9.5, 6.0 Hz, Qui-5), 1.34 (3H, d, *J* = 6.0 Hz, Qui-6), 5.12 (1H, d, *J* = 8.0 Hz, Glc-1), 4.22 (1H, dd, *J* = 9.5, 8.0 Hz, Glc-2), 5.67 (1H, dd, *J* = 10.0, 9.5 Hz, Glc-3), 5.33 (1H, dd, *J* = 10.0, 10.0 Hz, Glc-4), 4.09 (1H, ddd, *J* = 10.0, 5.0, 3.0 Hz, Glc-5); 4.64−4.58 (1H, m, Glc-6a), 4.35−4.26 (1H, m, Glc-6b), 5.45 (1H, d, *J* = 1.5 Hz, Rha-1), 5.66 (1H, m, Rha-2), 4.60 (1H, dd, *J* = 9.0, 3.4 Hz, Rha-3), 4.35−4.26 (1H, m, Rha-4), 4.55−4.50 (1H, m, Rha-5), 1.85 (3H, d, *J* = 6.5 Hz, Rha-6), 5.45 (1H, m, Glc′-1), 4.24 (1H, dd, *J* = 8.5, 7.5 Hz, Glc′-2), 5.57 (1H, dd, *J* = 9.0, 8.5 Hz, Glc′-3), 5.29 (1H, dd, *J* = 9.0, 9.0 Hz, Glc′-4), 4.35−4.26 (1H, m, Glc′-5), 4.55−4.50 (1H, m, Glc′-6a), 4.39 (1H, dd, *J* = 11.7, 2.0 Hz, Glc′-6b), 5.24 (1H, d, *J* = 8.3 Hz, Qui′-1), 5.50 (1H, dd, *J* = 9.0, 8.3 Hz, Qui′-2), 5.66 (1H, dd, *J* = 10.0, 9.0 Hz, Qui′-3), 5.46 (1H, dd, *J* = 10.0, 9.5 Hz, Qui′-4), 3.96 (1H, dq, *J* = 9.5, 6.0 Hz, Qui′-5),

1.44 (3H, d, $J = 6.0$ Hz, Qui′-6), 5.59 (1H, d, $J = 8.0$ Hz, Fuc-1), 5.80 (1H, dd, $J = 10.0$, 8.0 Hz, Fuc-2), 5.59 (1H, dd, $J = 10.0$, 3.0 Hz, Fuc-3), 5.90 (1H, d, $J = 3.0$ Hz, Fuc-4), 4.35−4.26 (1H, m, Fuc-5), 1.33 (3H, d, $J = 7.0$ Hz, Fuc-6), 2.34 (2H, t, $J = 7.4$ Hz, Con-2), 3.65 (1H, m, Con-11), 0.89 (3H, t, $J = 7.2$ Hz, Con-14), 3.56 (3H, s, OCH_3); ^{13}C NMR (125 MHz, C_5D_5N) δ 100.9 (CH, Qui-1), 77.1 (CH, Qui-2), 75.8 (CH, Qui-3), 74.5 (CH, Qui-4), 69.4 (CH, Qui-5), 18.0 (CH_3, Qui-6), 101.3 (CH, Glc-1), 72.7 (CH, Glc-2), 76.2 (CH, Glc-3), 70.0 (CH, Glc-4), 72.4 (CH, Glc-5), 63.4 (CH_2, Glc-6), 97.4 (CH, Rha-1), 73.9 (CH, Rha-2), 78.6 (CH, Rha-3), 77.1 (CH, Rha-4), 67.8 (CH, Rha-5), 19.2 (CH_3, Rha-6), 102.5 (CH, Glc′-1), 78.4 (CH, Glc′-2), 75.8 (CH, Glc′-3), 70.0 (CH, Glc′-4), 69.6 (CH, Glc′-5), 62.8 (CH_2, Glc′-6), 101.7 (CH, Qui′-1), 72.2 (CH, Qui′-2), 74.0 (CH, Qui′-3), 74.2 (CH, Qui′-4), 71.2 (CH, Qui′-5), 18.2 (CH_3, Qui′-6), 101.1 (CH, Fuc-1), 69.6 (CH, Fuc-2), 72.7 (CH, Fuc-3), 71.2 (CH, Fuc-4), 71.7 (CH, Fuc-5), 16.3 (CH_3, Fuc-6), 174.0 (CO, Con-1), 34.3 (CH_2, Con-2), 80.8 (CH, Con-11), 14.7 (CH_3, Con-14), 51.4 (OCH_3).

Spectroscopic Characterization of 8: mp 88−90 °C; $[\alpha]_D$ −9 (*c* 0.1, $CHCl_3$); ^{1}H NMR δ 4.65 (1H, d, $J = 7.5$ Hz, Qui-1), 4.14 (1H, dd, $J = 8.0$, 8.0 Hz, Qui-2), 5.54 (1H, dd, $J = 9.0$, 9.0 Hz, Qui-3), 5.02 (1H, dd, $J = 9.5$, 9.5 Hz, Qui-4), 4.01 (1H, dq, $J = 9.4$, 6.0 Hz, Qui-5), 1.27 (3H, d, $J = 6.2$ Hz, Qui-6), 5.06 (1H, d, $J = 8.0$ Hz, Glc-1), 4.14 (1H, dd, $J = 8.0$, 8.0 Hz, Glc-2), 5.62 (1H, dd, $J = 9.0$, 9.0 Hz, Glc-3), 5.28 (1H, dd, $J = 9.8$, 9.8 Hz, Glc-4), 4.00 (1H, ddd, $J = 10.0$, 6.0, 3.0 Hz, Glc-5); 4.60−4.55 (1H, m, Glc-6a), 4.25−4.21 (1H, m, Glc-6b), 5.40 (1H, brs, Rha-1), 5.61 (1H, m, Rha-2), 4.60−4.55 (1H, m, Rha-3), 4.30−4.21 (1H, m, Rha-4), 4.50−4.45 (1H, m, Rha-5), 1.78 (3H, d, $J = 6.0$ Hz, Rha-6), 5.41−5.40 (1H, m, Glc′-1), 4.17 (1H, dd, $J = 8.3$, 8.3 Hz, Glc′-2), 5.52 (1H, dd, $J = 8.6$, 8.6 Hz, Glc′-3), 5.24 (1H, dd, $J = 9.5$, 9.5 Hz, Glc′-4), 4.30−4.21 (1H, m, Glc′-5), 4.50−4.45 (1H, m, Glc′-6a), 4.34 (1H, dd, $J = 12.0$, 2.0 Hz, Glc′-6b), 5.18 (1H, d, $J = 8.3$ Hz, Qui′-1), 5.48 (1H, dd, $J = 9.0$, 9.0 Hz, Qui′-2), 5.61 (1H, dd, $J = 9.5$, 9.5 Hz, Qui′-3), 5.42 (1H, dd, $J = 9.4$, 9.4 Hz, Qui′-4), 3.91 (1H, dq, $J = 9.5$, 6.0 Hz, Qui′-5), 1.38 (3H, d, $J = 6.0$ Hz, Qui′-6), 5.53 (1H, d, $J = 8.0$ Hz, Fuc-1), 5.73 (1H, dd, $J = 10.0$, 8.0 Hz, Fuc-2), 5.54 (1H, dd, $J = 10.0$, 3.3 Hz, Fuc-3), 5.83 (1H, d, $J = 3.3$ Hz, Fuc-4), 4.30−4.21 (1H, m, Fuc-5), 1.33 (3H, d, $J = 6.0$ Hz, Fuc-6), 2.24 (1H, t, $J = 7.4$ Hz, Jal-2), 3.62 (1H, m, Jal-11), 0.90 (3H, t, $J = 7.0$ Hz, Jal-16), 3.56 (3H, s, OCH_3); ^{13}C NMR (125 MHz, C_5D_5N) δ 100.9 (CH, Qui-1), 76.9 (CH, Qui-2), 76.0 (CH, Qui-3), 74.2 (CH, Qui-4), 69.2 (CH, Qui-5), 17.7 (CH_3, Qui-6), 101.1 (CH, Glc-1), 72.6 (CH, Glc-2), 76.0 (CH, Glc-3), 69.8 (CH, Glc-4), 72.2 (CH, Glc-5), 63.2 (CH_3, Glc-6), 97.3 (CH, Rha-1), 73.8 (CH, Rha-2), 78.4 (CH, Rha-3), 76.9 (CH, Rha-4), 67.6 (CH, Rha-5), 19.0 (CH_3, Rha-6), 102.3 (CH, Glc′-1), 78.2 (CH, Glc′-2), 75.6 (CH, Glc′-3), 69.8 (CH, Glc′-4), 69.7 (CH, Glc′-5), 62.6 (CH_3, Glc′-6), 101.5 (CH, Qui′-1), 72.1 (CH, Qui′-2), 73.7 (CH, Qui′-3), 74.0 (CH, Qui′-4), 71.0 (CH, Qui′-5), 17.9 (CH_3, Qui′-6), 101.0 (CH, Fuc-1), 69.4 (CH, Fuc-2), 72.5 (CH, Fuc-3), 71.0 (CH, Fuc-4), 69.4 (CH, Fuc-5), 16.1 (CH_3, Fuc-6), 173.4 (CO, Jal-1), 34.1 (CH_2, Jal-2), 81.0 (CH, Jal-11), 14.2 (CH_3, Jal-16), 51.2 (OCH_3).

Sugar Analysis. Compound **1** (40 mg) in 4 N HCl (10 mL) was heated at 90 °C for 2 h. The reaction mixture was diluted with H_2O (5 mL) and extracted with Et_2O (30 mL). The organic layer was evaporated to dryness, dissolved in $CHCl_3$ (5 mL), and treated with CH_2N_2. The aqueous phase was neutralized with 1 N KOH, extracted with *n*-BuOH (30 mL), and concentrated to give a colorless solid. The residue was dissolved in CH_3CN-H_2O and directly analyzed by HPLC: Waters standard column for carbohydrate analysis (3.9 × 300 mm, 10 μm), using an isocratic elution of CH_3CN-H_2O (17:3), a flow rate of 1 mL/min, and a sample injection of 20 μL (sample concentration: 5 mg/mL). Coelution experiments with standard carbohydrate samples allowed the identification of L-rhamnose ($t_R = 6.9$ min), D-quinovose ($t_R = 7.3$ min), D-fucose ($t_R = 8.3$ min), and D-glucose ($t_R = 13.6$ min).

Identification of Aglycons. The organic layer residue obtained during the acid-catalyzed hydrolysis of purgic acid A (**1**) was submitted to normal-phase HPLC (ISCO, 21.2 × 250 mm, 10 μm), using an isocratic elution of *n*-hexane−$CHCl_3$−Me_2CO (6:3:1) and a flow rate of 6 mL/min to give 7.3 mg of methyl (11*S*)-hydroxytetradecanoate (convolvulinic acid methyl ester):[30] t_R 18.6 min; mp 27−29 °C; $[\alpha]_D$ +1.5 (*c* 2, $CHCl_3$); ^{13}C NMR 174.4, 71.7, 51.5, 39.6, 37.5, 34.1, 29.6, 29.5, 29.3, 29.2, 29.1, 25.6, 24.9, 18.8, 14.1. An aliquot of this pure sample (3 mg) was derivatized with Sigma Sil-A for 5 min at 70 °C. GC-MS analysis gave one peak (t_R 7 min): *m*/*z* $[M]^+$ 330 (0.3), 315 (3.5), 287 (66.8), 145 (100), 73 (35.4).

Acid hydrolysis of compound **2** (15 mg) as described above yielded 2 mg of methyl (11*S*)-hydroxyhexadecanoate[30] (jalapinolic acid methyl ester): t_R 16.4 min; mp 42−44 °C; $[\alpha]_D$ +7.3 (*c* 2, $CHCl_3$); ^{13}C NMR 174.4, 72.0, 51.4, 37.5, 37.4, 34.1, 31.9, 29.6, 29.5, 29.4, 29.2, 29.1, 25.6, 25.3, 24.9, 22.6, 14.1. This aglycon (1 mg) was derivatized by treatment with Sigma Sil-A and analyzed by GC-MS analysis (t_R 12.8 min): *m*/*z* $[M]^+$ 358 (0.3), 343 (0.5), 311 (10.5), 287 (59.7), 173 (100), 73 (46.3).

Acknowledgment. This research was partially supported by Consejo Nacional de Ciencia y Tecnología (CONACyT: 45861-Q). E.E.-S. and B.H.-C. are grateful to CONACyT for graduate student scholarships. The sequence of chemical degradations used for characterization of purgic acids was performed by H. Cruz and O. Yañez as part of their B.Sc. theses. Thanks are due to G. Duarte, M. Guzmán, and M. Gutierrez (USAI, Facultad de Química) for the recording of mass spectra and optical rotations. M. Trejo assisted in the study of herbarium specimens. J. Ebrard (General Director, Laboratorios Mixim, Mexico) kindly provided samples of "Mexican scammony" and its resin, as well as shared information on commercialization of jalaps.

Supporting Information Available: Photographs of herbal drugs: "Rhizoma Jalapae" (Figure S1), "Mexican scammony" (Figure S2), and "tumbavaquero" (Figure S3). HPLC profiles of crude drug MeOH extracts from commercial Mexican jalap samples (Figures S4 and S5). Extraction of resin glycosides (Figure S6). HPLC purification of purgic acids **1** and **2** (Figure S7). ^{1}H NMR spectra of compounds **5**−**8** (Figures S8 and S9). COSY, HMQC, and ROESY spectra of peracetylated derivatives (Figures S10−S12). This information is available free of charge via the Internet at http://pubs.acs.org.

References and Notes

(1) Dr. Francisco Hernández (1515−1587), King Phillip II's chief medical officer in the Spanish colonies in the New World (1570−1577), was the first trained scientist to gather ethnobotanical information directly from Aztec healers and to assess the medicinal usefulness of the natural resources found in the highland central area of Mexico. He described the properties of the "*cacamótic tlanoquiloni*" or purgative potatoes (Mexican jalap roots) as being "benign remedies" to purge the stomach "with wonderful gentleness and safety, and furthermore they remove bilious and other humors from the veins". Selective sections in English from his *The Natural History of New Spain* can be found in: Hernández, F. *The Mexican Treasury: the Writings of Dr. Francisco Hernández*; Varey, S., Ed.; Stanford University Press: Stanford, CA, 2000; pp 117−156.

(2) Hernández, F. *Historia Natural de Nueva España*; Universidad Nacional Autónoma de México: Mexico City, 1959; Vol. II, pp 133−135.

(3) A medicinal plant complex consists of an assemblage of herbal drugs that are taxonomically different at the specific, generic, and/or familial level but that share (a) one or more key morphological features; (b) a common name; (c) certain organoleptic characteristics; and (d) at least one therapeutic application. Within the complex, usually one species (called dominant or signature) is considered to have the most effective properties and is widely employed and distributed beyond its natural geographic range. Subordinate species are those considered to be of lesser quality and usually applied when the dominant species is not available, but they are not necessarily adulterants. For some examples in Mexico and the adjacent United States, see: Linares, E.; Bye, R. *J. Ethnopharmacol.* **1987**, *19*, 153−183.

(4) Valdés, J.; Flores, H. In *Francisco Hernández. Obras Completas*; Universidad Nacional Autónoma de México: Mexico City, 1985; Vol. VII, pp 9−222.

(5) Purgatives along with diaphoretics, diuretics, and emetics were basic attributes of prehispanic herbal remedies since the Aztecs viewed diseases as being caused by phlegm that needed to be discharged. For Aztec medicinal concepts and practices, see: Ortiz de Montellano, B. R. *Aztec Medicine, Health, and Nutrition*; Rutgers University Press: New Brunswick, NJ, 1990; pp 189−192.

(6) (a) Gunther, R. T. *The Greek Herbal of Dioscorides*; Hafner: New York, 1968; p 571. (b) von Bingen, H. *Hildegard's Healing Plants: from her Medieval Classic Physica*; Beacon: Boston, 2001; p 177.

(7) Gerard, J. *The Herbal or General History of Plants*; Dover: New York, 1975; pp 866−869.

(8) *Libellus de Medicinalibus Indorum Herbis* (*Little Book of Indian Medicinal Herbs*) is a treasury of Aztec herbal remedies and the first medical text known to have been written in the New World. The author of this valuable document was Martín de la Cruz, an Aztec

physician, who originally dictated it in Nahuatl, the language of the Aztecs, and which was later translated into Latin by the Aztec nobleman Juan Badiano in 1552. One illustration in this herbal (Folio 32 recto) represents a bindweed called "*Uelicpahtli*" (Nahuatl language, *uelic* = savory, *pahtli* = medicine). It appears as a red-flowered *Ipomoea* with a large tuberous root. The legend accompaning this miniature reads "*purgatio ventris*" (purging the abdomen). Its therapeutical descriptions is similar to that given by Dr. Francisco Hernández for the "cacamótic tlanoquiloni", suggesting it is the jalap root (*I. purga*). For transcription, translation, and contemporary botanical interpretations of this centuries-old Aztec remedy, see: (a) Emmart, E. W. *The Badianus Manuscript (Codex Barberini, Latin 241). An Aztec Herbal of 1552*; The Johns Hopkins Press: Baltimore, 1940; p 260. (b) Gates, W. *An Aztec Herbal. The Classic Codex of 1552*; Dover: New York, 2000; p 55. (c) Miranda, F.; Valdés, J. In *Libellus de Medicinalibus Indorum Herbis*; Instituto Mexicano del Seguro Social: Mexico City, 1964; pp 243−282.

(9) (a) Williams, L. O. *Econ. Bot.* **1970**, *24*, 399−401. (b) Comisión Permanente de la Farmacopea de los Estados Unidos Mexicanos. *Extrafarmacopea Herbolaria de los Estados Unidos Mexicanos*; Secretaría de Salud: Mexico City, 2001; pp 18−19.

(10) (a) Noriega, J. M. *Curso de Historia de Drogas*; Instituto Médico Nacional: Mexico City, 1902; pp 372−373. (b) Herrera, A. L. *Farmacopea Latino-Americana*; Herrero Hermanos: Mexico City, 1921; pp 658−659.

(11) The root of "tumbavaquero", a Spanish composite word that literally means "knock the cowboy over" or "cowboy stunner", is used in combination with orange flowers (*Citrus sinensis*), "*flor de manita*" (hand flower, *Chiranthodendron pectadactylon*), heart flower (*Talauma mexicana*), "palo brasil" (logwood, *Haematoxylum brasiletto*), "salvia de bolita" (butterfly bush, *Buddleja perfoliata*), and tila flower (*Ternstroemia* spp.); one cup of the infusion to be drunk in the morning and at night. For origin and description of this herbal drug, see: Linares, E.; Bye, R.; Flores, B. *Medicinal Plants of Mexico: Traditional Uses and Remedies*; Instituto de Biología, Universidad Nacional Autónoma de México: Mexico City, 1999; pp 118−119.

(12) Martínez, M. *Las Plantas Medicinales de México*; Ediciones Botas: Mexico City, 1989; pp 276−279.

(13) The recommended dosages of jalap root (to a liter of water) are 1−3 g if a powder, 0.2−0.4 g if an extract, 0.1−0.6 g if a resin, and 10 to 20 drops every 4 h if a tincture; if given in sugar or jelly, this remedy is a safe purge for children. A teaspoon of the root, cut small or granulated, to a cup of boiling water. For preparation of home remedies, see: http://electrocomm.tripod.com/jalapa-jalapa.html.

(14) In the 1940s, the Xico region in Veracruz cultivated and exported about 40 tons of dried jalap root (*I. purga)* to the United States per year, maintaining this level up to the 1990s, after which it dwindled to nearly zero. However, the use of wild *I. orizabensis* as a substitute for cultivated jalap root has never been documented, so no official trade records exist, although in pharmaceutical commerce it is currently available as liquid alcoholic extracts and resin. The commercial demand for jalap roots has declined in Mexico due to the use of other laxatives derived from psyllium fruits (*Plantago* spp.) and cassia leaves (*Senna* spp.) as well as the domination of the world market by Italian and German herbal drug traders who import resins of the Brazilian jalap (*I. operculata*). For the traditional production system of jalap root, see: Linajes, A.; Rico-Gray, V.; Carrión, G. *Econ. Bot.* **1994**, *48*, 84−89.

(15) Davidow, J. *Infusions of Healing. A Treasury of Mexican-American Herbal Remedies*; Fireside: New York, 1999; pp 134−135.

(16) The attempts of taxonomists to classify the diversity of Mexican jalaps has resulted in considerable confusion in both the commercial names and the scientific nomenclature. In addition, other plants were used as "jalap" and employed as substitutes or adulterants; they included *Phytolacca octandra* L. and *Mirabilis jalapa* L., species totally unrelated taxonomically to *Ipomoea*.

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(19) High-purity oligosaccharides are required for structure elucidation by NMR or MS and for use as substrates to determine their biological activity. For an example of the biological activities of oligosaccharides, see: Pereda-Miranda, R.; Kaatz, G. W.; Gibbons, S. *J. Nat. Prod.* **2006**, *69*, 406−409.

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(22) The chromatographic mode is partition, and retention time increases with the polarity of the analyte and decreases with the polarity of the mobile phase.

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NP060295F