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# HPLC Isolation and structural elucidation of diastereomeric niloyl ester tetrasaccharides from Mexican scammony root☆

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**Abstract**—The application of preparative-scale recycling HPLC is illustrated by describing the complete resolution of diastereomeric mixtures of niloyl esters involving both of the (2*R*,3*R*) and (2*S*,3*S*) enantiomers of 3-hydroxy-2-methylbutanoic acid bonded to a macrocyclic tetrasaccharide from the resin of the Mexican scammony root (*Ipomoea orizabensis*). The characterization of 13 new diastereomeric niloyl ester glycosides, orizabins IX–XXI, based on the same structure of scammonic acid A was performed by high field NMR spectroscopy. The absolute configuration of the stereogenic carbinol center in the saponification-liberated nilic acid residues esterifying each of the individual pure glycolipids has been determined by careful $^1$H NMR analysis of (*S*)- and (*R*)-Mosher's ester derivatives. 

Mexican scammony or false jalap, *Ipomoea orizabensis* (Pelletan) Ledebour ex Steudel, is part of the group of purgative jalaps from the Convolvulaceae family.[1] A resin with cathartic properties which provokes peristaltic movements in the small intestine can be extracted from the thick roots of these perennial herbaceous vines. Such morning-glories species were in the Aztec herbolaria[2] long before the 16th century Spanish colonizers introduced them as cathartic agents to Europe. The Spaniards, who paused in the then small village of Xalapa while journeying from the port of Veracruz in the Gulf of Mexico to the capital of the New Spain, Mexico City, named these medicinal plants 'jalapa' for they were abundant in the surroundings.[3] The cathartic crude drugs are derived from the roots which are rich in glycoresins (10–18%). Home remedies are prepared as decoction, normally a 2 cm section of root to a liter of water. Pharmaceutical products come in the form of liquid alcoholic extracts, root powders or resins consumed singly or in combination with other ingredients to modify the therapeutical effect. Today the demand for Mexican scammony and jalap (*Ipomoea purga*) has declined due to the use of other laxatives[3] derived from psyllium fruits (*Plantago* ssp.) and cassia leaves (*Senna* spp.) as well as the introduction of Brazilian grown jalap (*Operculina tuberosa*) in the world market by Italian and German herb traders.[4]

Prior to the application of HPLC and high-resolution spectroscopic techniques, the structures of the active ingredients in these resins were poorly known. In the systematic approach, the chemistry of these oligosaccharides was characterized by their alkaline hydrolysis products, i.e. the fatty acids ester-linked at the oligosaccharide cores and the glycosidic acids.[5] HPLC has permitted the isolation of intact oligosaccharides whose general structure turned out to be monohydroxy and dihydroxy $C_{14}$ and $C_{16}$ fatty acids glycosidically linked to a homo- or hetero-oligosaccharide core. These components proved to be individual macrocyclic esters and not the supposed oligomers[5] of high molecular weight formed by the condensation of several glycosidic acids. This advanced methodology for isolation revealed that crude resins represent complex mixtures of isomers involving a different substitution pattern for the lactonization and the position of esterification at the oligosaccharide core.[6,7]

Our ongoing investigations, directed toward the discovery of novel secondary metabolites with biomedical and ecological importance,[8,9] permit us to describe the application of preparative-scale recycling HPLC[10] for the isolation of orizabins IX–XXI (**1**–**13**). The foregoing mentioned macrocyclic glycolipids represent diastereomeric niloyl ester derivatives of scammonic acid A,[1] involving both of the *threo* nilic acid enantiomers, i.e. 2*R*,3*R*-(−)- and 2*S*,3*S*-(+)-3-hydroxy-2-methylbutanoic acids, as the esterifying moieties of the tetrasaccharide core. A previous study on the roots of *I. stans*[11,12] described the HPLC

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*Keywords*: chromatography; recycling HPLC; lipopolysaccharides; nilic acid; NMR; stereochemistry; biologically active compounds; plants; *Ipomoea orizabensis*.

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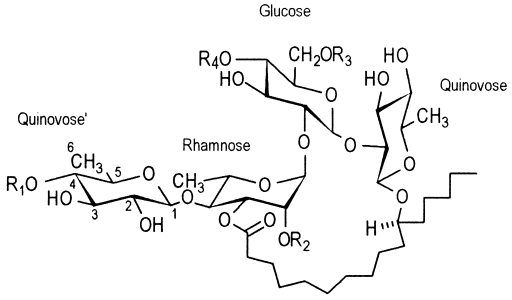
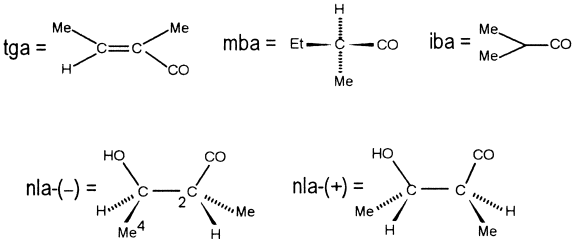
isolation of a tetrasaccharide with the same general structure as the one reported here for the diastereomeric pair composed by **12** and **13** but the stereochemistry for the ester residues was not established. In that investigation, a detailed analysis of the $^{13}C$ NMR spectral data suggested that the collected peak was actually an equimolecular mixture of two diastereomeric nilic esters derivatives.[11] In the present paper, we report the resolution of this type of mixtures by recycling HPLC as well as the establishment of the absolute configuration of the saponification-liberated nilic acid residues through a chiral derivatization with Mosher's reagent (methoxy(trifluoromethyl)phenylacetic acid or MTPA).[13,14]




| | $R_1$ | $R_2$ | $R_3$ | $R_4$ |
|---|---|---|---|---|
| **1** | tga | mba | H | nla-(–) |
| **2** | tga | nla-(–) | iba | H |
| **3** | tga | nla-(+) | iba | H |
| **4** | tga | iba | nla-(–) | H |
| **5** | tga | iba | nla-(+) | H |
| **6** | tga | nla-(–) | mba | H |
| **7** | tga | nla-(+) | mba | H |
| **8** | tga | mba | nla-(–) | H |
| **9** | tga | mba | nla-(+) | H |
| **10** | mba | nla-(–) | mba | H |
| **11** | mba | nla-(+) | mba | H |
| **12** | mba | mba | nla-(–) | H |
| **13** | mba | mba | nla-(+) | H |




## 1. Results and discussion

The cytotoxic fractions of Mexican scammony resin allowed for the isolation of thirteen additional tetrasaccharide glycolipids, orizabins IX–XXI (**1**–**13**), with a macrocyclic structure similar to those of the scammonic acid[15] A-based orizabins V–VIII.[1] The fractionation of this chloroform-soluble portion of the crude resin by reversed-phase HPLC ($C_{18}$) resulted in three subfractions

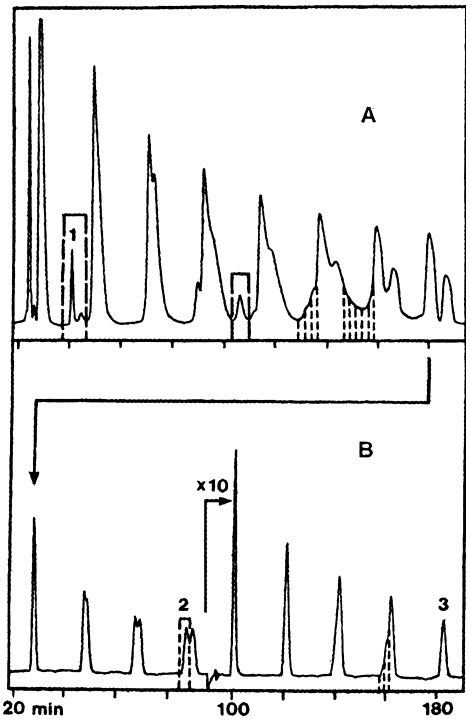



**Figure 1.** Recycling HPLC separation of the major constituents (**1**–**5**) from subfraction I. The areas marked with a large broken line indicate the fractions collected during the recycling process. Shaving of peaks, indicated by small broken lines, lessened the number of cycles needed for separation. Shown are chromatograms from (A) the separation of the two mixtures of diastereomeric niloyl ester pairs, **2/3** and **4/5**, and (B) the total resolution of the less polar peak into pure compounds **2** and **3**. The refractometer sensitivity was increased (10×) to optimize peak detection. Column: 150×19 mm μBondapak-amino; mobile phase: $CH_3CN$–$H_2O$, 95:5; flow rate=4 mL/min; sample injection: 10 mg/500 μL.

which were further separated by application of the preparative-scale recycling mode[10] through an aminopropyl column.[6,7] In the three analyzed subfractions, two major individual peaks were observed after 8–12 cycles (see Fig. 1(A)). Each of these mixtures were composed of two pairs of diastereomeric niloyl ester derivatives. Their final resolution was accomplished by peak shaving[16] and recycling through the same amino column. The upslope, center or downslope of a selected peak was collected and the rest was subjected to further recycling chromatography, as shown in Fig. 1(B). Saponification of these pure compounds (**2**–**13**) proved that each diastereomeric pair resulted from esterification of the oligosaccharide core by both of the *threo*[1] nilic acid enantiomers ($J_{2,3}$=7 Hz, H-2). The application of Mosher's method[13,14] determined the absolute configuration of each liberated nilic acid residue.

This acid was first isolated at the beginning of the 20th century as a saponification product from the resin of *Pharbitis nil*[17] and it is very frequently found as an esterifying moiety in natural products of plant origin. Although, very few phytochemical studies were carried through to the resolution of the stereochemistry of this acid,[18–21] as was done in this present investigation. Relevant proton NMR data for the (*R*)- and (*S*)-Mosher esters of the *threo* enantiomeric *RR*-(−) and *SS*-(+) nilic acids are summarized in Table 1. In the (*R*)-MTPA ester of the levoisomer, prepared from (*S*)-MTPA-Cl, the H-4 methyl group protons are

**Table 1.** [1]H NMR chemical shift data for diagnostic signals from the (*S*)- and (*R*)-ester derivatives of both enantiomeric *threo* nilic acids liberated from natural products **1**–**13**

| Nilic acid, $[\alpha]_D$ | MTPA-ester[a] configuration | Proton chemical shifts ($\Delta\delta_H=\delta_S-\delta_R$)[b] | | | | | | Configuration C-3 |
|---|---|---|---|---|---|---|---|---|
| | | H-4 | $\Delta\delta_H$ | H-5 | $\Delta\delta_H$ | H-2 | $\Delta\delta_H$ | |
| Negative | *S* (−8.6) | 1.44 | +0.08 | 1.24 | −0.07 | 2.93 | −0.03 | *R* |
| | *R* (+7.0) | 1.36 | | 1.31 | | 2.96 | | |
| Positive | *S* (−8.8) | 1.36 | −0.08 | 1.31 | +0.07 | 2.96 | +0.03 | *S* |
| | *R* (+8.8) | 1.44 | | 1.24 | | 2.93 | | |

[a] $[\alpha]_D$ values recorded in $CHCl_3$ at room temperature.
[b] Data registered in $CDCl_3$ at 300 MHz.

shielded relative to those in the derivative obtained from (*R*)-MTPA-Cl. Similarly, for the (*R*)-MTPA ester of the dextrorotatory isomer, the H-2 and H-5 protons (methyl group) are shielded relative to those in its diastereomeric ester. The chemical shift difference ($\Delta\delta=\delta_S-\delta_R$) between corresponding H-4 protons is positive for the MTPA esters of the levoisomer and negative for the dextrorotatory one. The opposite trend is observed for the H-2 and H-5 protons, i.e. a negative value for the levorotatory isomer and a positive sign for the dextroisomer. Application of the configurational correlation model proposed by Kakisawa[14] confirmed the (3*R*) absolute configuration for the [−]-*threo* nilic acid and therefore, its (2*R*,3*R*) stereochemistry. By identical reasoning, the absolute configuration of the enantiomeric residue (i.e. [+]-*threo* nilic acid) was deduced as (2*S*,3*S*).

The present investigation also proved that the isomerism between the two pairs of diastereomeric niloyl esters resulted through the interchange of the fatty acids residues at positions C-2 of the rhamnose unit and C-6 of glucose. Preparative-scale recycling HPLC of subfraction I afforded orizabin IX (**1**) and two major diastereomeric pairs which were resolved into pure compounds **2**–**5** (Fig. 1), whose FABMS supported a molecular formula of $C_{54}H_{90}O_{23}$. Saponification of orizabins X (**2**) and XI (**3**) yielded the levo- and dextrorotatory niloyl residues, respectively. Thus, they formed a positional isomeric pair in relation to orizabins XII (**4**) and XIII (**5**) by interchanging the position of the nilic and isobutyric acids. A residue of tiglic acid was present in these glycolipids as the third esterifying moiety at C-4 of the terminal quinovose. This chemical feature is also found in the next two pairs of isomeric glycolipids of orizabin IX (**1**), all of which have the molecular formula $C_{55}H_{92}O_{23}$, orizabins XIV–XVII (**6**–**9**). The isomerism for these two major pairs of diastereomeric nilic esters forming the second subfraction resulted as a consequence of the interchange of the nilate and methylbutyrate residues. The above mentioned situation was also observed for the third pair of isomers of molecular formula $C_{54}H_{94}O_{23}$, orizabins XVIII–XXI (**10**–**13**), where the position C-4 of the terminal quinovose unit was esterified by methylbutyric acid. While **2**–**13** exist as pairs of diastereoisomers, compound **1** is present in only one isomeric form due to the esterification of the oligosaccharide core by [−]-*threo* nilic acid as previously reported for orizabins V–VIII.[1]

Two-dimensional homonuclear NMR techniques (COSY and TOCSY)[6,7] were used to establish the pattern of substitution on individual saccharide units in **1**–**13**. The following common structural features for all the glycolipids were identified on the basis of NMR spectral data: (i) their macrocyclic lactone-type structure[1,6] was confirmed by the non-equivalent proton signals of the methylene group at C-2 in the aglycone moiety; (ii) the signals for the three short-chain fatty acid residues esterifying the oligosaccharide core were (a) H-2 for the saturated ones: a septet-like signal centered at $\delta$ 2.5 for the isobutyroyl (iba H-2), a sextet at $\delta$ 2.3–2.5 (mba H-2) for the methylbutyroyl[6] and a quintet at $\delta$ 2.8–2.9 (nla H-2) for the *threo*-isomer[22,23] of the niloyl residues; (b) the vinylic H-3 proton at $\delta$ 6.9–7.00 easily identified the tiglate[24,25] moiety; (iii) the site of lactonization at C-3 of rhamnose was established through the significant downfield shift observed in its geminal proton; (iv) the hydroxyl group on position C-2 of the rhamnose unit was obviously acylated ($\delta_H$ ca. 6.2; $\delta_C$ 68–70); (v) All the remaining esterified positions were identified by their deshielding effects which also indicated the location of the two additional ester linkages. Through HMBC-connectivity analysis,[6,7] it was possible to establish the location of the *O*-acyl groups in theses complex glycolipids. The differentiation of each individual carbonyl ester group was observed through its specific links with the corresponding vicinal methyne (or methylene for the aglycone) group resonance ($^2J_{CH}$) and also through the pyranose ring proton at the position of esterification ($^3J_{CH}$). The carbonyl resonance of the saturated esters most shielded ($\delta$ 173) was the lactone functionality by means of the $^2J$-coupling to each of the methylene protons $\delta_H$ ca. 2.6–2.9 on the adjacent C-2 position of the aglycone. The site of lactonization was corroborated at position C-3 of the rhamnose unit by the observed $^3J$-coupling to this proton ($\delta_H$ ca. 6.4). For compound **1**, the $^2J_{CH}$ coupling between the carbonyl resonance at $\delta$ 174.8 and the quintet-like proton at $\delta$ 2.8 were used to assign the nilate residue which identification then established its location at position C-4 of the glucose through the $^3J_{CH}$ coupling with the signal at $\delta$ 5.5. The carbonyl resonance of the methylbutyrate group ($\delta_C$ 175.3) at C-2 in the rhamnose unit was recognized through a similar analytical approach. In each pair of diastereomeric niloyl esters, the same $^{2,3}J_{CH}$ interactions were used to differentiate between the carbonyl resonances for the nilate and the isobutyrate or methylbutyrate substituents at positions C-2 of rhamnose and C-6 of glucose.

Finally, the cytotoxic potential of isolates **1**–**13** was evaluated against four human cancer cultured cell lines. According to the NCI guidelines[26] (active leads, $ED_{50}<$ 4 μg/mL), all compounds exhibited a weak cytotoxicity against colon carcinoma, squamoux cell cervix carcinoma

and ovarium cancer cell lines ($ED_{50}$ 4–20 μg/mL) but a stronger effect was observed when tested against oral epidermoid carcinoma (KB, $ED_{50}$ 1–5 μg/mL). The potency displayed for **1**–**13** was higher than the values reported for the polar isolates from Mexican scammony resin, i.e. orizabins V–VIII (KB, $ED_{50}$ 7–10 μg/mL), whose structures are composed of the same basic oligosaccharide described for **1**–**13** but which have one or two short-chain fatty acids rather than three.[1] Ongoing investigation in our laboratory suggests that the observed variation in activity[8] for this type of amphipatic oligosaccharides could be due to a possible perturbation in the target cell membrane induced by a non-selective pore formation.

## 2. Experimental

### 2.1. 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. Negative ion LRFABMS and HRFABMS were recorded using a matrix of triethanolamine on a JEOL SX102A spectrometer. A glycerol matrix was used for positive FABMS. $^1H$ (500 MHz) and $^{13}C$ (125.7 MHz) NMR experiments were registered in $C_5D_5N$ and conducted either on a Brucker AMX-500 or a Varian XL-500 instruments. The experimental procedures, including preparative HPLC instrumentation and NMR techniques,[6] along with handling of the plant material and extraction of the resin glycosides from *I. orizabensis*[1] were described in the preceding papers of this series. Cytotoxicity was conducted using cultured KB (nasopharyngeal carcinoma), HCT-15 (colon cancer), OVCAR (ovarian adenocarcinoma) and SQC-1 (squamus cell cervix carcinoma) cells according to previously described protocols.[1,27]

### 2.2. Recycling HPLC separation of compounds 1–13

A preliminary separation of the crude resins was performed by standard column chromatography.[1] The chloroform-soluble pool, fractions 150–274 (2.183 g; KB: $ED_{50}$ 4.4 μg/mol), was subjected to preparative HPLC on a Waters $C_{18}$ column (250×19 mm, Spherisorb, 10 μm). This preliminary separation of the cytotoxic fraction was performed to eliminate impurities appearing before and after the three selected peaks and before recycling each major collected fraction. The elution was isocratic with $CH_3CN$–$H_2O$ (88:12) and a flow rate of 8 mL/min. Subfractions I–III across the peaks with $t_R$ of 23.8 min (305 mg), 26.7 min (806 mg) and 33.4 min (659.3 mg) were collected by heart-cutting[16] and independently reinjected on a Waters aminopropylmethylsilyl amorphous Si gel column (150×19 mm, μBondapak, 10 μm). Elution was conducted isocratically with $CH_3CN$–$H_2O$ (95:5; flow rate=4 mL/min) and the HPLC system was operated in the recycle mode.[10] The separation of all components was monitored using a refractive index detector. Column overloading was used to increase the capacity for peak detection and also for preparative-scale recycling work by minimizing band broadening due to sample diffusion. Recycling separation of subfraction I afforded compound **1** (47 mg) from the first peak with $t_R$ of 22 min. The diastereomeric mixture of **2** (36 mg), **3** (49 mg), **4** (32 mg) and **5** (59 mg) was completely separated by shaving[16] and recycling the additional peak with $t_R$ of 30 min. Subfraction II ($t_R$= 20 min) was subjected to recycling in order to achieve the separation of the second diastereomeric mixture of **6** (62 mg), **7** (130 mg), **8** (130 mg) and **9** (136 mg). Finally, the glycolipids **10** (50 mg), **11** (120 mg), **12** (84 mg) and **13** (84 mg) were purified from subfraction III ($t_R$=18 min). Complete resolution for each compound was achieved after 8–12 consecutive cycles using the same aminopropyl column.

**2.2.1. Orizabin IX (1).** Obtained as a white amorphous powder; mp 124–127°C; $[\alpha]_D$=−29 (*c* 0.2, MeOH); $^1H$ NMR: 6.99 (1H, qq, *J*=7.5, 1.5 Hz, tga-3), 6.33 (1H, br s, Rha-1), 6.30 (1H, br s, Rha-2), 6.28 (1H, dd, *J*=10.0, 2.0 Hz, Rha-3), 5.89 (1H, d, *J*=7.5 Hz, Glu-1), 5.51 (1H, dd, *J*=9.5, 9.5 Hz, Glu-4), 5.35 (1H, dd, *J*=9.0, 9.0 Hz, Qui′-4), 5.08 (1H, d, *J*=8.0 Hz, Qui′-1), 4.99 (1H, dd, *J*= 11.5, 2.5 Hz, Glu-6), 4.99 (1H, d, *J*=8.0 Hz, Qui-1), 4.74 (1H, dq, *J*=10.0, 6.0 Hz, Rha-5), 4.37 (1H, dd, *J*=9.5, 9.5 Hz, Glu-3), 4.31 (m, nla-3), 4.29 (1H, dd, *J*=10.0, 10.0 Hz, Rha-4), 4.28 (1H, dd, *J*=9.5, 7.5 Hz, Glu-2), 4.25 (1H, dd, *J*=9.0, 8.0 Hz, Qui-2), 4.19 (1H, dd, *J*=9.0, 9.0 Hz, Qui′-3), 4.13 (1H, dd, *J*=12.0, 6.5 Hz, Glu-6′), 4.09 (1H, dd, *J*=9.0, 9.0 Hz, Qui-3), 3.97–3.94 (2H, m, Glu-5 and Jal-11), 3.89 (1H, dd, *J*=9.0, 8.0 Hz, Qui′-2), 3.75–3.68 (2H, m, Qui-5 and Qui′-5), 3.54 (1H, dd, *J*=9.0, 9.0 Hz, Qui-4), 2.86 (1H, ddd, *J*=15.0, 8.0, 3.0 Hz, Jal-2), 2.81 (1H, dq, *J*=7.0, 7.0 Hz, nla-2), 2.60 (1H, ddd, *J*=15.0, 10.0, 3.0 Hz, Jal-2′), 2.34 (1H, tq, *J*=7.0, 7.0 Hz, mba-2), 1.85 (3H, quintet, *J*=1.5 Hz, tga-5), 1.62 (3H, dq, *J*=7.5, 1.5 Hz, tga-4), 1.60 (3H, d, *J*=6.5 Hz, Rha-6), 1.58 (3H, d, *J*=6.0 Hz, Qui-6), 1.35 (3H, d, *J*=6.5 Hz, nla-4), 1.34 (3H, d, *J*=7.0 Hz, Qui′-6), 1.25 (3H, d, *J*=7.0 Hz, nla-5), 1.16 (3H, d, *J*=7.0 Hz, mba-5), 0.98 (3H, t, *J*=7.0 Hz, mba-4), 0.85 (3H, t, *J*=7.0 Hz, Jal-16); $^{13}C$ NMR: 175.3 (mba-1), 174.8 (nla-1), 173.2 (Jal-1), 167.5 (tga-1), 137.5 (tga-3), 129.1 (tga-2), 105.4 (Qui′-1), 101.9 (Glu-1), 100.8 (Qui-1), 96.2 (Rha-1), 79.6 (Qui-2), 79.1 (Glu-2), 78.9 (Rha-4 and Jal-11), 78.2 (Qui-3), 77.2 (Qui-4), 76.9 (Qui′-4), 75.7 (Glu-5), 75.3 (Qui′-3), 75.1 (Qui′-2), 74.5 (Glu-3), 73.4 (Glu-4), 72.5 (Qui-5), 71.3 (Rha-3), 70.4 (Qui′-5), 69.3 (nla-3), 69.0 (Rha-2), 68.7 (Rha-5), 62.9 (Glu-6), 48.7 (nla-2), 41.1, (mba-2), 34.4 (Jal-2), 21.0 (nla-4), 18.5 (Qui-6 and Rha-6), 18.0 (Qui′-6), 17.0 (mba-5), 14.3 (Jal-16 and tga-4), 13.2 (nla-5), 12.2 (tga-5), 11.8 (mba-4); negative FAB-MS *m/z* 1119 $[M-H]^-$, 1075 $[M-H-C_2H_4O]^-$, 1019 $[M-H-C_5H_8O_2]^-$, 973 $[1019-C_2H_4-H_2O]^-$, 919 $[1109-C_5H_6O-H_2O]^-$, 661, 617, 561, 417, 271; HRFAB-MS *m/z* 1119.5968 ($[M-H]^-$, calcd for $C_{55}H_{91}O_{23}$ +1.5 ppm error). $ED_{50}$ (μg/mL): 5.5 (KB), 12.0 (SQC-1), 9.1 (HCT-15), 15.8 (OVCAR).

**2.2.2. Orizabin X (2).** Obtained as a white amorphous powder; mp 118–120°C; $[\alpha]_D$=−54.5 (*c* 0.1, MeOH); $^1H$ NMR: 6.97 (1H, qq, *J*=7.0, 1.5 Hz, tga-3), 6.39 (1H, dd, *J*=10.5, 3.0 Hz, Rha-3), 6.28 (1H, d, *J*=1.5 Hz, Rha-1), 6.21 (1H, dd, *J*=3.0, 1.5 Hz, Rha-2), 5.91 (1H, d, *J*= 7.0 Hz, Glu-1), 5.34 (1H, dd, *J*=10.0, 9.0 Hz, Qui′-4), 5.03 (1H, d, *J*=8.0 Hz, Qui′-1), 5.01 (1H, d, *J*=8.0 Hz,

Qui-1), 4.97 (1H, dd, *J*=12.0, 2.5 Hz, Gluc-6), 4.83 (1H, dq, *J*=9.5, 5.5 Hz, Rha-5), 4.65 (1H, dd, *J*=12.0, 6.5 Hz, Glu-6′), 4.38 (1H, dq, *J*=7.0, 6.5 Hz, nla-3), 4.33 (1H, dd, *J*=10.5, 9.5 Hz, Rha-4), 4.30 (1H, dd, *J*=9.0, 8.0 Hz, Qui-2), 4.23 (1H, dd, *J*=9.0, 8.0 Hz, Glu-3), 4.20 (1H, dd, *J*=8.0, 7.0 Hz, Glu-2), 4.19 (1H, dd, *J*=9.5, 9.0 Hz, Qui′-3), 4.16 (1H, dd, *J*=9.0, 9.0 Hz, Qui-3), 3.99–3.94 (2H, m, Glu-5 and Jal-11), 3.91 (1H, dd, *J*=9.5, 8.0 Hz, Qui′-2), 3.89 (1H, dd, *J*=9.0, 8.0 Hz, Glu-4), 3.76–3.69 (2H, m, Qui-5 and Qui′-5), 3.61 (1H, dd, *J*=9.0, 9.0 Hz, Qui-4), 2.84 (1H, dq, *J*=7.0, 7.0 Hz, nla-2), 2.81 (1H, ddd, *J*=15.0, 8.5, 3.0 Hz, Jal-2), 2.64 (1H, ddd, *J*=15.0, 10.0, 3.0 Hz, Jal-2′), 2.54 (1H, septet, *J*=7.0 Hz, iba-2), 1.84 (3H, quintet, *J*=1.5 Hz, tga-5), 1.62–1.58 (9H, m, Qui-6, Rha-6 and tga-4), 1.36 (3H, d, *J*=6.5 Hz, nla-4), 1.32 (3H, d, *J*=6.0 Hz, Qui′-6), 1.28 (3H, d, *J*=7.0 Hz, nla-5), 1.24 (3H, t, *J*=7.0 Hz, iba-4), 1.16 (3H, d, *J*=7.0 Hz, iba-5), 0.87 (3H, t, *J*=7.0 Hz, Jal-16); $^{13}$C NMR: 175.9 (iba-1), 175.5 (nla-1), 173.3 (Jal-1), 167.5 (tga-1), 137.5 (tga-3), 129.1 (tga-2), 105.2 (Qui′-1), 101.1 (Glu-1), 100.8 (Qui-1), 96.2 (Rha-1), 79.2 (Glu-2), 79.0 (Qui-2, Rha-4 and Jal-11), 78.4 (Qui-3), 77.2 (Qui-4 and Glu-3), 76.9 (Qui′-4), 75.4 (Qui′-3), 75.1 (Qui′-2), 74.9 (Glu-5), 73.4 (Glu-4), 72.4 (Qui-5), 72.3 (Glu-4), 71.3 (Rha-3), 70.4 (Qui′-5), 69.8 (Rha-2), 69.0 (nla-3), 68.5 (Rha-5), 64.8 (Glu-6), 48.5 (nla-2), 34.4 (Jal-2), 34.1 (iba-2), 20.8 (nla-4), 19.4 (iba-3), 19.1 (iba-4), 18.5 (Qui-6 and Rha-6), 18.0 (Qui′-6), 14.3 (Jal-16), 14.1 (tga-4), 13.4 (nla-5), 12.2 (tga-5); negative FAB-MS *m/z* 1105 $[M-H]^-$, 1019 $[M-H-C_2H_4O-C_3H_6]^-$, 1005 $[M-H-C_5H_8O_2]^-$, 661, 617, 561, 417, 271; HRFAB-MS *m/z* 1105.5751 ($[M-H]^-$, calcd for $C_{54}H_{89}O_{23}$ −3.9 ppm error). $ED_{50}$ (μg/mL): 3.3 (KB), 19.1 (SQC-1), 10.0 (HCT-15), 8.3 (OVCAR).

**2.2.3. Orizabin XI (3).** Obtained as a white amorphous powder; mp 118–122°C; $[\alpha]_D$=−49 (*c* 0.26, MeOH); $^1$H NMR: 6.96 (1H, qq, *J*=7.0, 1.5 Hz, tga-3), 6.34 (1H, dd, *J*=10.0, 3.0 Hz, Rha-3), 6.22 (1H, d, *J*=1.5 Hz, Rha-1), 6.17 (1H, dd, *J*=3.0, 1.5 Hz, Rha-2), 5.83 (1H, d, *J*=7.0 Hz, Glu-1), 5.29 (1H, dd, *J*=9.0, 9.0 Hz, Qui′-4), 4.98 (1H, d, *J*=8.0 Hz, Qui′-1), 4.97 (1H, d, *J*=7.5 Hz, Qui-1), 4.94 (1H, dd, *J*=11.5, 2.5 Hz, Glu-6), 4.78 (1H, dq, *J*=9.5, 6.0 Hz, Rha-5), 4.62 (1H, dd, *J*=11.5, 6.5 Hz, Glu-6′), 4.34 (1H, dq, *J*=7.0, 6.5 Hz, nla-3), 4.30 (1H, dd, *J*=10.0, 10.0 Hz, Rha-4), 4.25 (1H, dd, *J*=9.0, 7.0 Hz, Qui-2), 4.19 (1H, dd, *J*=9.0, 9.0 Hz, Glu-3), 4.14 (1H, dd, *J*=9.0, 9.0 Hz, Qui′-3), 4.13 (1H, dd, *J*=9.0, 7.5 Hz, Glu-2), 4.12 (1H, dd, *J*=9.0, 9.0 Hz, Qui-3), 3.96–3.92 (2H, m, Glu-5 and Jal-11), 3.89 (1H, dd, *J*=9.0, 9.0 Hz, Glu-4), 3.85 (1H, dd, *J*=9.0, 8.0 Hz, Qui′-2), 3.74–3.66 (2H, m, Qui-5 and Qui′-5), 3.57 (1H, dd, *J*=9.0, 9.0 Hz, Qui-4), 2.82 (1H, dq, *J*=7.0, 7.0 Hz, nla-2), 2.76 (1H, ddd, *J*=15.5, 8.0, 3.5 Hz, Jal-2), 2.60 (1H, ddd, *J*=15.5, 9.5, 3.0 Hz, Jal-2′), 2.55 (1H, septet, *J*=7.0 Hz, iba-2), 1.83 (3H, quintet, *J*=1.5 Hz, tga-5), 1.60 (3H, dq, *J*=7.0, 1.5 Hz, tga-4), 1.58 (3H, d, *J*=6.0 Hz, Rha-6), 1.55 (3H, d, *J*=6.0 Hz, Qui-6), 1.35 (3H, d, *J*=6.5 Hz, nla-4), 1.31 (3H, d, *J*=6.0 Hz, Qui′-6), 1.28 (3H, d, *J*=7.0 Hz, nla-5), 1.23 (3H, t, *J*=7.0 Hz, iba-4), 1.16 (3H, d, *J*=7.0 Hz, iba-5), 0.87 (3H, t, *J*=7.0 Hz, Jal-16); $^{13}$C NMR: 175.9 (iba-1), 175.5 (nla-1), 173.3 (Jal-1), 167.5 (tga-1), 137.4 (tga-3), 129.2 (tga-2), 105.2 (Qui′-1), 101.2 (Glu-1), 100.9 (Qui-1), 96.4 (Rha-1), 79.3 (Glu-2 and Jal-11), 79.2 (Qui-2), 79.0 (Rha-4), 78.5 (Qui-3), 77.3 (Qui-4 and Glu-3), 77.0 (Qui′-4), 75.5 (Qui′-3), 75.3 (Qui′-2), 75.0 (Glu-5), 72.4 (Qui-5 and Glu-4), 71.5 (Rha-3), 70.5 (Qui′-5), 69.9 (Rha-2), 69.1 (nla-3), 68.7 (Rha-5), 64.9 (Glu-6), 48.5 (nla-2), 34.5 (Jal-2), 34.2 (iba-2), 20.8 (nla-4), 19.4 (iba-3), 19.1 (iba-4), 18.5 (Qui-6 and Rha-6), 18.0 (Qui′-6), 14.3 (Jal-16), 14.1 (tga-4), 13.4 (nla-5), 12.2 (tga-5); positive HRFAB-MS *m/z* 1107.5942 ($[M+H]^+$, calcd for $C_{54}H_{91}O_{23}$ −0.8 ppm error). $ED_{50}$ (μg/mL): 1.7 (KB), 10.0 (SQC-1), 6.3 (HCT-15), 7.9 (OVCAR).

**2.2.4. Orizabin XII (4).** Obtained as a white amorphous powder; mp 120–123°C; $[\alpha]_D$=−49 (*c* 0.2, MeOH); $^1$H NMR: 6.95 (1H, qq, *J*=7.0, 1.5 Hz, tga-3), 6.37 (1H, dd, *J*=9.5, 3.5 Hz, Rha-3), 6.23 (1H, d, *J*=1.5 Hz, Rha-1), 6.14 (1H, dd, *J*=3.5, 1.5 Hz, Rha-2), 5.90 (1H, d, *J*=7.5 Hz, Glu-1), 5.31 (1H, dd, *J*=9.5, 9.0 Hz, Qui′-4), 5.00 (1H, d, *J*=7.5 Hz, Qui′-1), 4.99 (1H, d, *J*=7.5 Hz, Qui-1), 4.98 (1H, dd, *J*=11.5, 2.0 Hz, Glu-6), 4.80 (1H, dq, *J*=9.5, 6.5 Hz, Rha-5), 4.58 (1H, dd, *J*=11.5, 6.5 Hz, Glu-6′), 4.34 (1H, dq, *J*=7.0, 6.5 Hz, nla-3), 4.31 (1H, dd, *J*=9.5, 9.5 Hz, Rha-4), 4.30 (1H, dd, *J*=9.0, 7.5 Hz, Qui-2), 4.21 (1H, dd, *J*=9.0, 9.0 Hz, Glu-3), 4.18 (1H, dd, *J*=9.0, 7.5 Hz, Glu-2), 4.16 (2H, m, Qui-3 and Qui′-3), 3.97–3.91 (2H, m, Glu-5 and Jal-11), 3.89 (1H, dd, *J*=9.0, 9.0 Hz, Glu-4), 3.87 (1H, dd, *J*=9.0, 8.0 Hz, Qui′-2), 3.72 (1H, dq, *J*=9.0, 6.0 Hz, Qui-5), 3.69 (1H, dq, *J*=9.5, 6.5 Hz, Qui′-5), 3.59 (1H, dd, *J*=9.0, 9.0 Hz, Qui-4), 2.82 (1H, dq, *J*=7.0, 7.0 Hz, nla-2), 2.78 (1H, ddd, *J*=15.5, 8.5, 3.0 Hz, Jal-2), 2.61 (1H, ddd, *J*=15.0, 10.0, 3.0 Hz, Jal-2′), 2.51 (1H, septet, *J*=7.0 Hz, iba-2), 1.81 (3H, quintet, *J*=1.5 Hz, tga-5), 1.57 (9H, m, tga-4, Rha-6 and Qui-6), 1.33 (3H, d, *J*=6.5 Hz, nla-4), 1.29 (3H, d, *J*=6.5 Hz, Qui′-6), 1.28 (3H, d, *J*=7.0 Hz, nla-5), 1.22 (3H, t, *J*=7.0 Hz, iba-4), 1.13 (3H, d, *J*=7.0 Hz, iba-5), 0.84 (3H, t, *J*=7.0 Hz, Jal-16); $^{13}$C NMR: 175.9 (iba-1), 175.4 (nla-1), 173.3 (Jal-1), 167.5 (tga-1), 137.5 (tga-3), 129.1 (tga-2), 105.3 (Qui′-1), 101.0 (Qui-1 and Glu-1), 96.2 (Rha-1), 79.3 (Glu-2), 79.2 (Jal-11), 79.0 (Rha-4), 78.8 (Qui-2), 78.4 (Qui-3), 77.3 (Qui-4 and Glu-3), 76.9 (Qui′-4), 75.4 (Qui′-3), 75.1 (Qui′-2), 74.9 (Glu-5), 72.4 (Qui-5), 72.3 (Glu-4), 71.3 (Rha-3), 70.4 (Qui′-5), 70.0 (Rha-2), 69.0 (nla-3), 68.6 (Rha-5), 64.6 (Glu-6), 48.7 (nla-2), 34.5 (Jal-2), 34.1 (iba-2), 20.9 (nla-4), 19.5 (iba-3), 19.1 (iba-4), 18.5 (Qui-6 and Rha-6), 18.0 (Qui′-6), 14.3 (Jal-16), 14.1 (tga-4), 13.3 (nla-5), 12.2 (tga-5); positive HRFAB-MS *m/z* 1107.5949 ($[M+H]^+$, calcd for $C_{54}H_{91}O_{23}$ −0.1 ppm error). $ED_{50}$ (μg/mL): 1.8 (KB), >20 (SQC-1, HCT-15, OVCAR).

**2.2.5. Orizabin XIII (5).** Obtained as a white amorphous powder; mp 123–125°C; $[\alpha]_D$=−49.4 (*c* 0.61, MeOH); $^1$H NMR: 6.98 (1H, qq, *J*=7.0, 1.5 Hz, tga-3), 6.40 (1H, dd, *J*=10.0, 3.0 Hz, Rha-3), 6.27 (1H, d, *J*=1.5 Hz, Rha-1), 6.18 (1H, dd, *J*=3.0, 1.5 Hz, Rha-2), 5.93 (1H, d, *J*=8.0 Hz, Glu-1), 5.35 (1H, dd, *J*=9.5, 9.5 Hz, Qui′-4), 5.03 (2H, br d, *J*=7.5 Hz, Qui-1 and Qui′-1), 5.00 (1H, dd, *J*=11.5, 2.0 Hz, Glu-6), 4.84 (1H, dq, *J*=9.5, 6.0 Hz, Rha-5), 4.61 (1H, dd, *J*=11.5, 6.5 Hz, Glu-6′), 4.37 (1H, dq, *J*=7.0, 6.0 Hz, nla-3), 4.34 (1H, dd, *J*=10.0, 9.5 Hz, Rha-4), 4.33 (1H, dd, *J*=9.5, 7.5 Hz, Qui-2), 4.25 (1H, dd, *J*=8.5, 8.5 Hz, Glu-3), 4.22–4.17 (3H, m, Qui-3, Glu-2 and Qui′-3), 4.01–3.95 (2H, m, Glu-5 and Jal-11), 3.94 (1H, dd,

$J$=8.5, 8.5 Hz, Glu-4), 3.90 (1H, dd, $J$=9.5, 7.5 Hz, Qui′-2), 3.77–3.70 (2H, m, Qui-5 and Qui′-5), 3.63 (1H, dd, $J$=9.0, 9.0 Hz, Qui-4), 2.86 (1H, dq, $J$=7.0, 7.0 Hz, nla-2), 2.81 (1H, ddd, $J$=15.0, 8.5, 3.0 Hz, Jal-2), 2.64 (1H, ddd, $J$=15.0, 10.0, 3.0 Hz, Jal-2′), 2.54 (1H, septet, $J$=7.0 Hz, iba-2), 1.84 (3H, quintet, $J$=1.5 Hz, tga-5), 1.61–1.59 (9H, m, Qui-6, Rha-6 and tga-4), 1.36 (3H, d, $J$=6.0 Hz, nla-4), 1.33 (3H, d, $J$=6.0 Hz, Qui′-6), 1.31 (3H, d, $J$=7.0 Hz, nla-5), 1.23 (3H, t, $J$=7.0 Hz, iba-4), 1.16 (3H, d, $J$=7.0 Hz, iba-5), 0.87 (3H, t, $J$=7.0 Hz, Jal-16); $^{13}$C NMR: 175.9 (iba-1), 175.4 (nla-1), 173.3 (Jal-1), 167.5 (tga-1), 137.5 (tga-3), 129.1 (tga-2), 105.3 (Qui′-1), 101.0 (Qui-1 and Glu-1), 96.2 (Rha-1), 79.3 (Glu-2), 79.2 (Jal-11), 79.0 (Rha-4), 78.8 (Qui-2), 78.4 (Qui-3), 77.3 (Qui-4 and Glu-3), 76.9 (Qui′-4), 75.4 (Qui′-3), 75.1 (Qui′-2), 74.9 (Glu-5), 72.5 (Qui-5), 72.3 (Glu-4), 71.3 (Rha-3), 70.4 (Qui′-5), 70.0 (Rha-2), 69.0 (nla-3), 68.6 (Rha-5), 64.6 (Glu-6), 48.7 (nla-2), 34.5 (Jal-2), 34.1 (iba-2), 20.9 (nla-4), 19.5 (iba-3), 19.1 (iba-4), 18.5 (Qui-6 and Rha-6), 18.0 (Qui′-6), 14.3 (Jal-16), 14.1 (tga-4), 13.3 (nla-5), 12.2 (tga-5); negative FAB-MS $m/z$ 1105 $[M-H]^-$, 1019 $[1105-C_2H_4O-C_3H_6]^-$, 1005 $[1105-C_5H_8O_2]^-$, 973 $[1105-C_2H_4O-C_4H_6O-H_2O]^-$, 919, 679, 661, 617, 561, 417, 271; HRFAB-MS $m/z$ 1105.5791 ($[M-H]^-$, calcd for $C_{54}H_{89}O_{23}$ −0.3 ppm error). $ED_{50}$ (μg/mL): 12.0 (KB), >20 (SQC-1), 15.8 (HCT-15), 8.7 (OVCAR).

**2.2.6. Orizabin XIV (6).** Obtained as a white amorphous powder; mp 119–122°C; $[\alpha]_D$=−35.2 ($c$ 0.2, MeOH); $^1$H NMR: 6.96 (1H, qq, $J$=7.0, 1.0 Hz, tga-3), 6.39 (1H, dd, $J$=10.0, 2.0 Hz, Rha-3), 6.25 (1H, d, $J$=1.5 Hz, Rha-1), 6.20 (1H, dd, $J$=3.0, 1.5 Hz, Rha-2), 5.88 (1H, d, $J$=7.0 Hz, Glu-1), 5.33 (1H, dd, $J$=9.5, 8.5 Hz, Qui′-4), 5.00 (1H, d, $J$=8.0 Hz, Qui′-1), 4.96 (1H, dd, $J$=11.5, 2.0 Hz, Glu-6), 4.98 (1H, d, $J$=9.0 Hz, Qui-1), 4.81 (1H, dq, $J$=9.5, 6.5 Hz, Rha-5), 4.64 (1H, dd, $J$=11.5, 7.0 Hz, Glu-6′), 4.36 (1H, dq, $J$=7.0, 6.0 Hz, nla-3), 4.32 (1H, dd, $J$=10.0, 9.5 Hz, Rha-4), 4.29 (1H, dd, $J$=9.0, 9.0 Hz, Qui-2), 4.22 (1H, dd, $J$=9.0, 9.0 Hz, Glu-3), 4.19 (1H, dd, $J$=9.0, 9.0 Hz, Qui-3), 4.18 (1H, dd, $J$=9.0, 7.0 Hz, Glu-2), 4.15 (1H, dd, $J$=8.5, 8.5 Hz, Qui′-3), 3.99–3.94 (2H, m, Glu-5 and Jal-11), 3.89 (1H, dd, $J$=9.5, 9.0 Hz, Glu-4), 3.88 (1H, dd, $J$=8.5, 8.0 Hz, Qui′-2), 3.75–3.70 (2H, m, Qui-5 and Qui′-5), 3.59 (1H, dd, $J$=9.5, 9.0 Hz, Qui-4), 2.82 (1H, ddd, $J$=15.0, 8.0, 2.0 Hz, Jal-2), 2.82 (1H, dq, $J$=7.0, 7.0 Hz, nla-2), 2.61 (1H, ddd, $J$=15.0, 9.5, 2.5 Hz, Jal-2′), 2.38 (1H, tq, $J$=7.0, 7.0 Hz, mba-2), 1.83 (3H, quintet, $J$=1.0 Hz, tga-5), 1.60 (3H, dq, $J$=7.5, 1.0 Hz, tga-4), 1.58 (3H, d, $J$=6.5 Hz, Rha-6), 1.57 (3H, d, $J$=6.5 Hz, Qui-6), 1.36 (3H, d, $J$=6.5 Hz, nla-4), 1.34 (3H, d, $J$=6.5 Hz, Qui′-6), 1.28 (3H, d, $J$=7.0 Hz, nla-5), 1.16 (3H, d, $J$=7.0 Hz, mba-5), 0.99 (3H, t, $J$=7.0 Hz, mba-4), 0.87 (3H, t, $J$=7.0 Hz, Jal-16); $^{13}$C NMR: 175.7 (nla-1), 175.6 (mba-1), 173.5 (Jal-1), 167.7 (tga-1), 137.7 (tga-3), 129.3 (tga-2), 105.4 (Qui′-1), 101.3 (Qui-1), 101.1 (Glu-1), 96.5 (Rha-1), 79.4 (Glu-2 and Jal-11), 79.2 (Qui-2), 79.1 (Rha-4), 78.6 (Qui-3), 77.4 (Qui-4 and Glu-3), 77.1 (Qui′-4), 75.6 (Qui′-3), 75.3 (Qui′-2), 75.2 (Glu-5), 72.7 (Qui-5), 72.5 (Glu-4), 71.5 (Rha-3), 70.7 (Qui′-5), 70.0 (Rha-2), 69.3 (nla-3), 68.8 (Rha-5), 65.0 (Glu-6), 48.7 (nla-2), 41.3 (mba-2), 34.7 (Jal-2), 21.1 (nla-4), 18.8 (Qui-6), 18.7 (Rha-6), 18.2 (Qui′-6), 17.2 (mba-5), 14.5 (Jal-16), 14.4 (tga-4), 13.6 (nla-5), 12.4 (tga-5), 12.0 (mba-4); negative FAB-MS $m/z$ 1119 $[M-H]^-$, 1019 $[M-H-C_5H_8O_2]^-$, 973 $[1019-C_2H_4-H_2O]^-$, 789, 680, 661, 617, 561, 417, 271; HRFAB-MS $m/z$ 1119.6008 ($[M-H]^-$, calcd for $C_{55}H_{91}O_{23}$ +5.1 ppm error). $ED_{50}$ (μg/mL): 2.3 (KB), 16.6 (SQC-1), 10.0 (HCT-15), 14.5 (OVCAR).

**2.2.7. Orizabin XV (7).** Obtained as a white amorphous powder; mp 110–112°C; $[\alpha]_D$=−34.7 ($c$ 0.2, MeOH); $^1$H NMR (500 MHz, $C_5D_5N$): 6.98 (1H, qq, $J$=6.5, 1.5 Hz, tga-3), 6.41 (1H, dd, $J$=10.0, 3.0 Hz, Rha-3), 6.27 (1H, d, $J$=1.5 Hz, Rha-1), 6.22 (1H, dd, $J$=3.0, 1.5 Hz, Rha-2), 5.91 (1H, d, $J$=7.5 Hz, Glu-1), 5.35 (1H, dd, $J$=9.5, 9.5 Hz, Qui′-4), 5.02 (1H, d, $J$=8.0 Hz, Qui′-1), 4.97 (1H, dd, $J$=11.5, 2.5 Hz, Glu-6), 5.00 (1H, d, $J$=8.5 Hz, Qui-1), 4.82 (1H, dq, $J$=10.0, 6.5 Hz, Rha-5), 4.65 (1H, dd, $J$=11.5, 6.5 Hz, Glu-6′), 4.37 (1H, dq, $J$=7.0, 6.0 Hz, nla-3), 4.34 (1H, dd, $J$=10.0, 10.0 Hz, Rha-4), 4.31 (1H, dd, $J$=9.5, 8.5 Hz, Qui-2), 4.24 (1H, dd, $J$=8.5, 8.0 Hz, Glu-3), 4.17 (1H, dd, $J$=9.5, 9.5 Hz, Qui-3), 4.20 (1H, dd, $J$=8.5, 7.5 Hz, Glu-2), 4.18 (1H, dd, $J$=9.5, 8.0 Hz, Qui′-3), 3.99 (1H, ddd, $J$=8.0, 6.5, 2.5 Hz, Glu-5), 3.95 (1H, m, Jal-11), 3.91 (1H, dd, $J$=8.0, 8.0 Hz, Glu-4), 3.89 (1H, dd, $J$=8.0, 8.0 Hz, Qui′-2), 3.78–3.71 (2H, m, Qui-5 and Qui′-5), 3.61 (1H, dd, $J$=9.5, 8.0 Hz, Qui-4), 2.84 (1H, ddd, $J$=15.5, 8.5, 1.5 Hz, Jal-2), 2.84 (1H, dq, $J$=7.0, 7.0 Hz, nla-2), 2.62 (1H, ddd, $J$=15.5, 10.0, 3.0 Hz, Jal-2′), 2.39 (1H, tq, $J$=7.5, 7.5 Hz, mba-2), 1.84 (3H, quintet, $J$=1.0 Hz, tga-5), 1.62–1.56 (9H, m, Qui-6, Rha-6, tga-4), 1.37 (3H, d, $J$=6.5 Hz, nla-4), 1.33 (3H, d, $J$=6.5 Hz, Qui′-6), 1.29 (3H, d, $J$=7.5 Hz, nla-5), 1.16 (3H, d, $J$=7.5 Hz, mba-5), 0.99 (3H, t, $J$=7.5 Hz, mba-4), 0.87 (3H, t, $J$=7.0 Hz, Jal-16); $^{13}$C NMR: 175.5 (nla-1), 175.4 (mba-1), 173.3 (Jal-1), 167.5 (tga-1), 137.5 (tga-3), 129.1 (tga-2), 105.2 (Qui′-1), 101.2 (Glu-1), 100.9 (Qui-1), 96.4 (Rha-1), 79.2 (Glu-2 and Jal-11), 79.0 (Qui-2), 78.9 (Rha-4), 78.5 (Qui-3), 77.3 (Qui-4 and Glu-3), 76.9 (Qui′-4), 75.4 (Qui′-3), 75.2 (Qui′-2), 75.0 (Glu-5), 72.5 (Qui-5), 72.3 (Glu-4), 71.3 (Rha-3), 70.5 (Qui′-5), 69.8 (Rha-2), 69.1 (nla-3), 68.6 (Rha-5), 64.8 (Glu-6), 48.5 (nla-2), 41.1 (mba-2), 34.5 (Jal-2), 20.9 (nla-4), 18.5 (Qui-6 and Rha-6), 18.2 (Qui′-6), 17.0 (mba-5), 14.3 (Jal-16), 14.1 (tga-4), 13.4 (nla-5), 12.2 (tga-5), 11.7 (mba-4); negative FAB-MS $m/z$ 1119 $[M-H]^-$, 1019 $[M-H-C_5H_8O_2]^-$, 973 $[1019-C_2H_4-H_2O]^-$, 661, 617, 561, 417, 271; HRFAB-MS $m/z$ 1119.6001 ($[M-H]^-$, calcd for $C_{55}H_{91}O_{23}$ +4.4 ppm error). $ED_{50}$ (μg/mL): 1.1 (KB), >20 (SQC-1, HCT-15, OVCAR).

**2.2.8. Orizabin XVI (8).** Obtained as a white amorphous powder; mp 124−127°C; $[\alpha]_D$=−26 ($c$ 0.2, MeOH); $^1$H NMR (500 MHz, $C_5D_5N$): 6.98 (1H, qq, $J$=7.0, 1.0 Hz, tga-3), 6.41 (1H, dd, $J$=10.0, 2.0 Hz, Rha-3), 6.25 (1H, d, $J$=1.0 Hz, Rha-1), 6.19 (1H, dd, $J$=3.0, 1.0 Hz, Rha-2), 5.92 (1H, d, $J$=7.5 Hz, Glu-1), 5.35 (1H, dd, $J$=9.5, 9.0 Hz, Qui′-4), 5.02 (2H, br d, $J$=8.0 Hz, Qui-1 and Qui′-1), 5.01 (1H, dd, $J$=11.5, 2.0 Hz, Glu-6), 4.83 (1H, dq, $J$=9.5, 6.0 Hz, Rha-5), 4.60 (1H, dd, $J$=11.5, 7.0 Hz, Glu-H-6′), 4.37 (1H, dq, $J$=7.5, 6.5 Hz, nla-3), 4.34 (1H, dd, $J$=10.0, 9.5 Hz, Rha-4), 4.32 (1H, dd, $J$=9.0, 8.0 Hz, Qui-2), 4.24 (1H, dd, $J$=9.0, 9.0 Hz, Glu-3), 4.19 (2H, dd, $J$=9.0, 9.0 Hz, Qui-3 and Qui′-3), 4.19 (1H, dd, $J$=9.0, 7.0 Hz, Glu-2), 3.98 (1H, ddd, $J$=8.5, 7.0, 2.0 Hz, Glu-5),

3.97–3.94 (1H, m, Jal-11), 3.94 (1H, dd, *J*=9.0, 8.5 Hz, Glu-4), 3.90 (1H, dd, *J*=9.0, 8.0 Hz, Qui′-2), 3.78–3.71 (2H, m, Qui-5 and Qui′-5), 3.62 (1H, dd, *J*=9.0, 9.0 Hz, Qui-4), 2.87–2.82 (1H, m, Jal-2), 2.85 (1H, dq, *J*=7.5, 6.5 Hz, nla-2), 2.62 (1H, ddd, *J*=15.0, 10.0, 3.0 Hz, Jal-2′), 2.39 (1H, tq, *J*=7.0, 7.0 Hz, mba-2), 1.84 (3H, quintet, *J*=1.0 Hz, tga-5), 1.62–1.58 (9H, m, Qui-6, Rham-6 and tga-4), 1.36 (3H, d, *J*=6.5 Hz, nla-4), 1.33 (3H, d, *J*=6.0 Hz, Qui′-6), 1.31 (3H, d, *J*=6.5 Hz, nla-5), 1.16 (3H, d, *J*=7.0 Hz, mba-5), 0.99 (3H, t, *J*=7.0 Hz, mba-4), 0.87 (3H, t, *J*=7.0 Hz, Jal-16); $^{13}$C NMR: 175.4 (nla-1 and mba-1), 173.3 (Jal-1), 167.5 (tga-1), 137.5 (tga-3), 129.1 (tga-2), 105.2 (Qui′-1), 100.9 (Qui-1, Glu-1), 96.3 (Rha-1), 79.3 (Jal-11), 79.2 (Glu-2), 78.8 (Qui-2 and Rha-4), 78.4 (Qui-3), 77.3 (Qui-4), 77.2 (Glu-3), 76.9 (Qui′-4), 75.4 (Qui′-3), 75.1 (Qui′-2), 74.9 (Glu-5), 72.5 (Qui-5), 72.3 (Glu-4), 71.3 (Rha-3), 70.5 (Qui′-5), 69.9 (Rha-2), 69.0 (nla-3), 68.7 (Rha-5), 64.6 (Glu-6), 48.7 (nla-2), 41.1 (mba-2), 34.5 (Jal-2), 20.9 (nla-4), 18.5 (Qui-6 and Rha-6), 18.0 (Qui′-6), 17.0 (mba-5), 14.3 (Jal-16), 14.1 (tga-4), 13.3 (nla-5), 12.2 (tga-5), 11.7 (mba-4); negative FAB-MS *m/z* 1119 $[M-H]^-$, 1075 $[M-H-C_2H_4O]^-$, 1019 $[M-H-C_5H_8O_2]^-$, 973 $[1019-C_2H_4-H_2O]^-$, 661, 617, 417, 271; HRFAB-MS *m/z* 1119.6002 ($[M-H]^-$, calcd for $C_{55}H_{91}O_{23}$ +4.5 ppm error). $ED_{50}$ (μg/mL): 1.7 (KB), 12.0 (SQC-1), >20 (HCT-15), 15.8 (OVCAR).

**2.2.9. Orizabin XVII (9).** Obtained as a white amorphous powder; mp 120–122°C; $[\alpha]_D=-20.7$ (*c* 0.2, MeOH); $^1$H NMR (500 MHz, $C_5D_5N$): 6.96 (1H, qq, *J*=6.5, 1.5 Hz, tga-3), 6.39 (1H, dd, *J*=10.0, 3.0 Hz, Rha-3), 6.23 (1H, d, *J*=1.5 Hz, Rha-1), 6.17 (1H, dd, *J*=3.0, 1.5 Hz, Rha-2), 5.89 (1H, d, *J*=7.5 Hz, Glu-1), 5.33 (1H, dd, *J*=10.0, 9.5 Hz, Qui′-4), 4.99 (2H, d, *J*=8.0 Hz, Qui-1 and Qui′-1), 4.98 (1H, dd, *J*=11.5, 2.0 Hz, Glu-6), 4.81 (1H, dq, *J*=10.0, 6.0 Hz, Rha-5), 4.58 (1H, dd, *J*=11.5, 6.5 Hz, Glu-6′), 4.35 (1H, m, nla-3), 4.32 (1H, dd, *J*=10.0, 10.0 Hz, Rha-4), 4.30 (1H, dd, *J*=8.0, 7.5 Hz, Qui-2), 4.22 (1H, dd, *J*=9.0, 8.5 Hz, Glu-3), 4.19–4.15 (3H, m, Qui-3, Glu-2 and Qui′-3), 3.97 (1H, ddd, *J*=8.5, 6.5, 2.0 Hz, Glu-5), 3.95–3.91 (2H, m, Jal-11 and Glu-4), 3.88 (1H, dd, *J*=8.5, 8.0 Hz, Qui′-2), 3.76–3.69 (2H, m, Qui-5 and Qui′-5), 3.60 (1H, dd, *J*=9.0, 8.5 Hz, Qui-4), 2.84 (1H, dq, *J*=7.0, 7.0 Hz, nla-2), 2.82 (1H, ddd, *J*=15.5, 8.0, 3.5 Hz, Jal-2), 2.62 (1H, ddd, *J*=15.0, 10.0, 3.0 Hz, Jal-2′), 2.38 (1H, tq, *J*=7.0, 7.0 Hz, mba-2), 1.83 (3H, quintet, *J*=1.0 Hz, tga-5), 1.60 (3H, dq, *J*=6.5, 1.5 Hz, tga-4), 1.59 (3H, d, *J*=6.5 Hz, Qui-6), 1.58 (3H, d, *J*=6.5 Hz, Rha-6), 1.35 (3H, d, *J*=6.5 Hz, nla-4), 1.32 (3H, d, *J*=6.5 Hz, Qui′-6), 1.30 (3H, d, *J*=7.0 Hz, nla-5), 1.15 (3H, d, *J*=7.0 Hz, mba-5), 0.99 (3H, t, *J*=7.0 Hz, mba-4), 0.87 (3H, t, *J*=7.0 Hz, Jal-16); $^{13}$C NMR: 175.6 (nla-1 and mba-1), 173.5 (Jal-1), 167.7 (tga-1), 137.7 (tga-3), 129.3 (tga-2), 105.4 (Qui′-1), 101.1 (Qui-1, Glu-1), 96.5 (Rha-1), 79.5 (Jal-11), 79.4 (Glu-2), 79.1 (Qui-2), 79.0 (Rha-4), 78.6 (Qui-3), 77.5 (Qui-4), 77.4 (Glu-3), 77.1 (Qui′-4), 75.6 (Qui′-3), 75.3 (Qui′-2), 75.1 (Glu-5), 72.7 (Qui-5), 72.5 (Glu-4), 71.5 (Rha-3), 70.7 (Qui′-5), 70.2 (Rha-2), 69.3 (nla-3), 68.9 (Rha-5), 64.8 (Glu-6), 48.9 (nla-2), 41.3 (mba-2), 34.7 (Jal-2), 21.1 (nla-4), 18.8 (Rha-6),18.7 (Qui-6), 18.2 (Qui′-6), 17.2 (mba-5), 14.5 (Jal-16), 14.4 (tga-4), 13.5 (nla-5), 12.5 (tga-5), 11.9 (mba-4); negative FAB-MS *m/z* 1119 $[M-H]^-$, 1019 $[1119-C_5H_8O_2]^-$, 973 $[1019-C_2H_4-H_2O]^-$, 789, 679, 661, 561, 417, 271; HRFAB-MS *m/z* 1119.5968 ($[M-H]^-$, calcd for $C_{55}H_{91}O_{23}$ +1.5 ppm error). $ED_{50}$ (μg/mL): 1.9 (KB), 15.8 (SQC-1), 13.2 (HCT-15), 15.8 (OVCAR).

**2.2.10. Orizabin XVIII (10).** Obtained as a white amorphous powder; mp 112–115°C; $[\alpha]_D=-26.5$ (*c* 0.2, MeOH); $^1$H NMR (500 MHz, $C_5D_5N$): 6.40 (1H, dd, *J*=10.0, 3.0 Hz, Rha-3), 6.26 (1H, d, *J*=1.5 Hz, Rha-1), 6.22 (1H, dd, *J*=3.0, 1.5 Hz, Rha-2), 5.91 (1H, d, *J*=7.0 Hz, Glu-1), 5.25 (1H, dd, *J*=10.0, 9.5 Hz, Qui′-4), 5.00 (1H, d, *J*=8.0 Hz, Qui-1), 4.98 (1H, d, *J*=8.0 Hz, Qui′-1), 4.98 (1H, dd, *J*=11.5, 2.0 Hz, Glu-6), 4.82 (1H, dq, *J*=9.5, 6.0 Hz, Rha-5), 4.65 (1H, dd, *J*=11.5, 7.0 Hz, Glu-6′), 4.37 (1H, dq, *J*=7.0, 6.0 Hz, nla-3), 4.33 (1H, dd, *J*=10.0, 9.5 Hz, Rha-4), 4.31 (1H, dd, *J*=8.0, 8.0 Hz, Qui-2), 4.23 (1H, dd, *J*=9.0, 9.0 Hz, Glu-3), 4.19 (1H, dd, *J*=9.0, 7.0 Hz, Glu-2), 4.17 (1H, dd, *J*=9.0, 8.0 Hz, Qui-3), 4.10 (1H, dd, *J*=9.5, 9.5 Hz, Qui′-3), 3.98 (1H, ddd, *J*=9.0, 7.0, 2.0 Hz, Glu-5), 3.95 (1H, m, Jal-11), 3.91 (1H, dd, *J*=9.0, 9.0 Hz, Glu-4), 3.85 (1H, dd, *J*=9.0, 8.0 Hz, Qui′-2), 3.74 (1H, dq, *J*=9.0, 6.5 Hz, Qui-5), 3.70 (1H, dq, *J*=10.0, 6.5 Hz, Qui′-5), 3.61 (1H, dd, *J*=9.0, 9.0 Hz, Qui-4), 2.84 (1H, dq, *J*=7.5, 7.0 Hz, nla-2), 2.81 (1H, ddd, *J*=15.0, 7.5, 3.5 Hz, Jal-2), 2.60 (1H, ddd, *J*=15.0, 9.5, 2.5 Hz, Jal-2′), 2.49 (1H, tq, *J*=7.5, 7.5 Hz, mba′-2), 2.38 (1H, tq, *J*=7.0, 7.0 Hz, mba-2), 1.59 (6H, d, *J*=6.5 Hz, Qui-6 and Rha-6), 1.36 (3H, d, *J*=6.5 Hz, Qui′-6), 1.35 (3H, d, *J*=6.0 Hz, nla-4), 1.29 (3H, d, *J*=7.5 Hz, nla-5), 1.17 (3H, d, *J*=7.5 Hz, mba′-5), 1.13 (3H, d, *J*=7.0 Hz, mba-5), 0.99 (3H, t, *J*=7.0 Hz, mba-4), 0.96 (3H, t, *J*=7.5 Hz, mba′-4), 0.87 (3H, t, *J*=7.0 Hz, Jal-16); $^{13}$C NMR: 175.4 (mba′-1), 175.1 (nla-1), 174.9 (mba-1), 172.8 (Jal-1), 104.6 (Qui′-1), 100.6 (Glu-1), 100.4 (Qui-1), 95.8 (Rha-1), 78.7 (Glu-2 and Jal-11), 78.5 (Qui-2), 78.4 (Rha-4), 77.9 (Qui-3), 76.7 (Qui-4 and Glu-3), 75.8 (Qui′-4), 74.7 (Qui′-2 and Qui′-3), 74.5 (Glu-5), 71.9 (Qui-5), 71.8 (Glu-4), 70.8 (Rha-3), 69.8 (Qui′-5), 69.3 (Rha-2), 68.5 (nla-3), 68.1 (Rha-5), 64.3 (Glu-6), 48.0 (nla-2), 41.2 (mba′-2), 40.6 (mba-2), 33.9 (Jal-2), 20.4 (nla-4), 18.0 (Rha-6),17.9 (Qui-6), 17.5 (Qui′-6), 16.5 (mba-5 and mba′-5), 13.8 (Jal-16), 12.9 (nla-5), 11.3 (mba′-4), 11.2 (mba-4); negative FAB-MS *m/z* 1121 $[M-H]^-$, 1021 $[M-H-C_5H_8O_2]^-$, 975 $[1021-C_2H_4-H_2O]^-$, 789, 679, 661, 561, 417, 271; HRFAB-MS *m/z* 1121.6088 ($[M-H]^-$, calcd for $C_{55}H_{93}O_{23}$ −1.7 ppm error). $ED_{50}$ (μg/mL): 2.1 (KB), 19.5 (SQC-1), 20.0 (HCT-15), 11.5 (OVCAR).

**2.2.11. Orizabin XIX (11).** Obtained as a white amorphous powder; mp 114–116°C; $[\alpha]_D=-30.6$ (*c* 0.2, MeOH); $^1$H NMR (500 MHz, $C_5D_5N$): 6.37 (1H, dd, *J*=10.0, 3.0 Hz, Rha-3), 6.24 (1H, d, *J*=1.5 Hz, Rha-1), 6.19 (1H, dd, *J*=3.0, 1.5 Hz, Rha-2), 5.87 (1H, d, *J*=7.5 Hz, Glu-1), 5.22 (1H, dd, *J*=9.5, 9.0 Hz, Qui′-4), 4.97 (1H, d, *J*=8.0 Hz, Qui-1), 4.96 (1H, d, *J*=7.5 Hz, Qui′-1), 4.95 (1H, dd, *J*=12.0, 2.0 Hz, Glu-6), 4.79 (1H, dq, *J*=9.5, 6.5 Hz, Rha-5), 4.62 (1H, dd, *J*=12.0, 6.5 Hz, Glu-6′), 4.34 (1H, dq, *J*=7.0, 6.5 Hz, nla-3), 4.30 (1H, dd, *J*=10.0, 9.5 Hz, Rha-4), 4.28 (1H, dd, *J*=8.0, 8.0 Hz, Qui-2), 4.20 (1H, dd, *J*=9.0, 9.0 Hz, Glu-3), 4.17 (1H, dd, *J*=9.0, 7.0 Hz, Glu-2), 4.14 (1H, dd, *J*=9.0, 9.0 Hz, Qui-3), 4.07 (1H, dd, *J*=9.5, 9.5 Hz, Qui′-3), 3.96 (1H, ddd, *J*=8.0, 6.5, 2.0 Hz, Glu-5),

3.92 (1H, m, Jal-11), 3.88 (1H, dd, *J*=9.0, 8.0 Hz, Glu-4), 3.82 (1H, dd, *J*=9.5, 7.5 Hz, Qui′-2), 3.71 (1H, dq, *J*=9.0, 6.0 Hz, Qui-5), 3.67 (1H, dq, *J*=9.0, 6.5 Hz, Qui′-5), 3.58 (1H, dd, *J*=9.0, 9.0 Hz, Qui-4), 2.81 (1H, dq, *J*=7.0, 6.5 Hz, nla-2), 2.78 (1H, ddd, *J*=15.5, 8.0, 3.0 Hz, Jal-2), 2.57 (1H, ddd, *J*=15.5, 10.0, 2.5 Hz, Jal-2′), 2.47 (1H, tq, *J*=7.0, 7.0 Hz, mba′-2), 2.36 (1H, tq, *J*=7.0, 7.0 Hz, mba-2), 1.56 (6H, d, *J*=6.5 Hz, Qui-6 and Rha-6), 1.34 (3H, d, *J*=6.0 Hz, nla-4), 1.32 (3H, d, *J*=6.5 Hz, Qui′-6), 1.26 (3H, d, *J*=7.0 Hz, nla-5), 1.17 (3H, d, *J*=7.0 Hz, mba′-5), 1.13 (3H, d, *J*=7.0 Hz, mba-5), 0.96 (3H, t, *J*=7.0 Hz, mba-4), 0.93 (3H, t, *J*=7.0 Hz, mba′-4), 0.84 (3H, t, *J*=7.0 Hz, Jal-16); $^{13}$C NMR: 175.9 (mba′-1), 175.5 (nla-1), 175.4 (mba-1), 173.3 (Jal-1), 105.2 (Qui′-1), 101.1 (Glu-1), 100.9 (Qui-1), 96.3 (Rha-1), 79.2 (Glu-2 and Jal-11), 79.0 (Rha-4), 78.9 (Qui-2), 78.4 (Qui-3), 77.2 (Qui-4 and Glu-3), 76.3 (Qui′-4), 75.2 (Qui′-2 and Qui′-3), 75.0 (Glu-5), 72.4 (Qui-5), 72.3 (Glu-4), 71.3 (Rha-3), 70.3 (Qui′-5), 69.8 (Rha-2), 69.0 (nla-3), 68.6 (Rha-5), 64.8 (Glu-6), 48.5 (nla-2), 41.7 (mba′-2), 41.1 (mba-2), 34.4 (Jal-2), 20.9 (nla-4), 18.5 (Qui-6),18.4 (Rha-6), 18.0 (Qui′-6), 17.0 (mba-5 and mba′-5), 14.3 (Jal-16), 13.4 (nla-5), 11.8 (mba-4), 11.7 (mba′-4); negative FAB-MS *m/z* 1121 $[M-H]^-$, 1021 $[M-H-C_5H_8O_2]^-$, 975 $[1021-C_2H_4-H_2O]^-$, 789, 679, 661, 617, 561, 417, 271; HRFAB-MS *m/z* 1121.6194 ($[M-H]^-$, calcd for $C_{55}H_{93}O_{23}$ +7.6 ppm error). $ED_{50}$ (μg/mL): 2.3 (KB), 18.2 (SQC-1), 8.3 (HCT-15), 12.6 (OVCAR).

**2.2.12. Orizabin XX (12).** Obtained as a white amorphous power; mp 111–113°C; $[\alpha]_D=-21.5$ (*c* 0.2, MeOH); $^1$H NMR (500 MHz, $C_5D_5N$): 6.37 (1H, dd, *J*=10.0, 3.0 Hz, Rha-3), 6.22 (1H, d, *J*=1.5 Hz, Rha-1), 6.15 (1H, dd, *J*=3.0, 1.5 Hz, Rha-2), 5.88 (1H, d, *J*=7.5 Hz, Glu-1), 5.22 (1H, dd, *J*=9.0, 9.0 Hz, Qui′-4), 4.99 (1H, d, *J*=7.5 Hz, Qui-1), 4.98 (1H, dd, *J*=11.5, 2.0 Hz, Glu-6), 4.95 (1H, d, *J*=8.0 Hz, Qui′-1), 4.79 (1H, dq, *J*=9.5, 6.0 Hz, Rha-5), 4.57 (1H, dd, *J*=11.5, 6.5 Hz, Glu-6′), 4.34 (1H, dq, *J*=7.0, 6.0 Hz, nla-3), 4.30 (1H, dd, *J*=10.0, 9.5 Hz, Rha-4), 4.29 (1H, dd, *J*=9.5, 7.5 Hz, Qui-2), 4.21 (1H, dd, *J*=9.0, 9.0 Hz, Glu-3), 4.19–4.13 (2H, m, Qui-3 and Glu-2), 4.07 (1H, dd, *J*=9.0, 9.0 Hz, Qui′-3), 3.96 (1H, ddd, *J*=8.5, 6.5, 2.0 Hz, Glu-5), 3.93 (1H, m, Jal-11), 3.89 (1H, dd, *J*=9.0, 8.5 Hz, Glu-4), 3.82 (1H, dd, *J*=9.0, 8.0 Hz, Qui′-2), 3.72 (1H, dq, *J*=9.5, 6.5 Hz, Qui-5), 3.67 (1H, dq, *J*=9.0, 6.5 Hz, Qui′-5), 3.59 (1H, dd, *J*=9.5, 9.5 Hz, Qui-4), 2.82 (1H, dq, *J*=7.0, 7.0 Hz, nla-2), 2.79 (1H, ddd, *J*=15.0, 8.5, 3.0 Hz, Jal-2), 2.56 (1H, ddd, *J*=15.0, 9.5, 3.0 Hz, Jal-2′), 2.47 (1H, tq, *J*=7.0, 7.0 Hz, mba-2), 2.36 (1H, tq, *J*=7.0, 7.0 Hz, mba′-2), 1.56 (3H, d, *J*=6.0 Hz, Rha-6), 1.54 (3H, d, *J*=6.5 Hz, Qui-6), 1.33 (3H, d, *J*=6.0 Hz, nla-4), 1.31 (3H, d, *J*=6.5 Hz, Qui′-6), 1.28 (3H, d, *J*=7.5 Hz, nla-5), 1.18 (3H, d, *J*=7.0 Hz, mba-5), 1.13 (3H, d, *J*=7.0 Hz, mba′-5), 0.95 (3H, t, *J*=7.0 Hz, mba′-4), 0.93 (3H, t, *J*=7.5 Hz, mba-4), 0.84 (3H, t, *J*=7.0 Hz, Jal-16); $^{13}$C NMR: 175.9 (mba′-1), 175.4 (mba-1 and nla-1), 173.3 (Jal-1), 105.2 (Qui′-1), 100.9 (Qui-1 and Glu-1), 96.3 (Rha-1), 79.2 (Qui-2, and Jal-11), 78.9 (Rha-4), 78.4 (Qui-3), 77.3 (Qui-4 and Glu-3), 76.3 (Qui′-4), 75.2 (Qui′-2 and Qui′-3), 74.9 (Glu-5), 72.5 (Qui-5), 72.3 (Glu-4), 71.3 (Rha-3), 70.3 (Qui′-5), 70.0 (Rha-2), 69.0 (nla-3), 68.6 (Rha-5), 64.6 (Glu-6), 48.7 (nla-2), 41.7 (mba′-2), 41.1 (mba-2), 34.5 (Jal-2), 20.9 (nla-4), 18.5 (Qui-6 and Rha-6), 18.0 (Qui′-6), 17.0 (mba-5 and mba′-5), 14.3 (Jal-16), 13.3 (nla-5), 11.8 (mba-4), 11.7 (mba′-4); negative FAB-MS *m/z* (modo negativo) 1121 $[M-H]^-$, 1021 $[1121-C_5H_8O_2]^-$, 790, 679, 661, 618, 561, 417, 271; negativo HRFAB-MS *m/z* 1121.6130 ($[M-H]^-$, calcd for $C_{55}H_{93}O_{23}$ +1.9 ppm error). $ED_{50}$ (μg/mL): 1.8 (KB), 11.0 (SQC-1), >20 (HCT-15), 3.9 (OVCAR).

**2.2.13. Orizabin XXI (13).** Obtained as a white amorphous powder; mp 117–120°C; $[\alpha]_D=-26.5$ (*c* 0.2, MeOH); $^1$H NMR (500 MHz, $C_5D_5N$): 6.39 (1H, dd, *J*=10.0, 3.0 Hz, Rha-3), 6.23 (1H, d, *J*=1.5 Hz, Rha-1), 6.17 (1H, dd, *J*=3.0, 1.5 Hz, Rha-2), 5.90 (1H, d, *J*=7.5 Hz, Glu-1), 5.24 (1H, dd, *J*=9.5, 9.5 Hz, Qui′-4), 5.0 (1H, d, *J*=8.0 Hz, Qui-1), 4.99 (1H, dd, *J*=11.5, 2.0 Hz, Glu-6), 4.97 (1H, d, *J*=8.0 Hz, Qui′-1), 4.81 (1H, dq, *J*=9.5, 6.0 Hz, Rha-5), 4.59 (1H, dd, *J*=11.5, 6.5 Hz, Glu-6′), 4.37 (1H, dq, *J*=7.0, 6.0 Hz, nla-3), 4.31 (1H, dd, *J*=10.0, 9.5 Hz, Rha-4), 4.30 (1H, dd, *J*=9.0, 8.0 Hz, Qui-2), 4.23 (1H, dd, *J*=9.0, 8.5 Hz, Glu-3), 4.18 (1H, dd, *J*=9.0, 8.0 Hz, Qui-3), 4.17 (1H, dd, *J*=9.0, 7.5 Hz, Glu-2), 4.08 (1H, dd, *J*=9.5, 9.5 Hz, Qui′-3), 3.99–3.95 (2H, m, Glu-5 and Jal-11), 3.91 (1H, dd, *J*=9.0, 9.0 Hz, Glu-4), 3.84 (1H, dd, *J*=9.0, 8.0 Hz, Qui′-2), 3.74 (1H, dq, *J*=9.5, 6.5 Hz, Qui-5), 3.68 (1H, dq, *J*=9.5, 6.5 Hz, Qui′-5), 3.61 (1H, dd, *J*=9.5, 8.0 Hz, Qui-4), 2.84 (1H, dq, *J*=7.0, 7.0 Hz, nla-2), 2.81 (1H, ddd, *J*=15.0, 8.0, 3.0 Hz, Jal-2), 2.58 (1H, ddd, *J*=15.0, 10.0, 3.0 Hz, Jal-2′), 2.49 (1H, tq, *J*=7.0, 7.0 Hz, mba′-2), 2.38 (1H, tq, *J*=7.0, 7.0 Hz, mba-2), 1.59 (3H, d, *J*=6.5 Hz, Qui-6), 1.58 (3H, d, *J*=6.5 Hz, Rha-6), 1.35 (3H, d, *J*=6.0 Hz, nla-4), 1.34 (3H, d, *J*=6.5 Hz, Qui′-6), 1.30 (3H, d, *J*=7.0 Hz, nla-5), 1.20 (3H, d, *J*=7.0 Hz, mba′-5), 1.15 (3H, d, *J*=7.0 Hz, mba-5), 0.98 (3H, t, *J*=7.0 Hz, mba-4), 0.95 (3H, t, *J*=7.0 Hz, mba′-4), 0.87 (3H, t, *J*=7.0 Hz, Jal-16); $^{13}$C NMR: 176.1 (mba′-1), 175.6 (mba-1 and nla-1), 173.5 (Jal-1), 105.4 (Qui′-1), 101.1 (Qui-1 and Glu-1), 96.5 (Rha-1), 79.5 (Jal-11), 79.4 (Glu-2), 79.1 (Rha-4), 78.6 (Qui-3), 77.5 (Qui-4 and Glu-3), 76.5 (Qui′-4), 75.4 (Qui′-2 and Qui′-3), 75.1 (Glu-5), 72.7 (Qui-5), 72.5 (Glu-4), 71.5 (Rha-3), 70.5 (Qui′-5), 70.2 (Rha-2), 69.3 (nla-3), 68.8 (Rha-5), 64.8 (Glu-6), 48.9 (nla-2), 41.9 (mba′-2), 41.3 (mba-2), 34.7 (Jal-2), 21.1 (nla-4), 18.8 (Rha-6), 18.7 (Qui′-6),18.2 (Qui-6), 17.2 (mba-5 and mba′-5), 14.5 (Jal-16), 13.5 (nla-5), 12.0 (mba′-4), 11.9 (mba-4); negative FAB-MS *m/z* 1121 $[M-H]^-$, 1021 $[M-H-C_5H_8O_2]^-$, 975 $[1021-C_2H_4-H_2O]^-$, 789, 679, 661, 561, 417, 271; HRFAB-MS *m/z* 1121.6085 ($[M-H]^-$, calcd for $C_{55}H_{93}O_{23}$ −2.0 ppm error). $ED_{50}$ (μg/mL): 1.7 (KB), 15.8 (SQC-1), 17.4 (HCT-15), 6.3 (OVCAR).

### 2.3. Alkaline hydrolysis and identification of organic acids

The crude resin glycoside (200 mg) in 5% KOH–$H_2O$ (5 mL) was refluxed at 95°C for 1 h. The reaction mixture was acidified to pH 4.0 and extracted with $CHCl_3$ (10 mL). The organic layer was washed with $H_2O$, dried over anhydrous $Na_2SO_4$, and evaporated under reduced pressure. The residue was treated with $CH_2N_2$ and directly analyzed by GC-EIMS[28] (30 m×0.25 mm DB-1 column; He, 2 mL/min; 120–280°C, Δ 4°C/min) with three major peaks

detected which were identified as methyl 2-methylbutyrate: $t_R$ 15.5 min; *m/z* $[M]^+$ 116 (2), $[M-CH_3]^+$ 101 (8), 88 (98), 85 (34), 74 (18), 59 (23), 57 (100), 41 (55), 39 (14), 29 (45); methyl tiglate: $t_R$ 18.0 min; *m/z* $[M]^+$ 114 (2), $[M-CH_3]^+$ 99 (15), 83 (25), 59 (8), 57 (100); and methyl 3-hydroxy-2-methylbutyrate: $t_R$ 21.3 min; *m/z* $[M]^+$ 132 (4), $[M-CH_3]^+$ 117 (10), 101 (20), 88 (100), 59 (18), 57 (80); and additional minor peak was identified as methyl 2-methylpropanoate: $t_R$ 12.0 min; *m/z* $[M]^+$ 102 (15), $[M-CH_3]^+$ 87 (21), 71 (45), 59 (24), 43 (100), 27 (18). The aqueous phase was extracted with *n*-BuOH (10 mL). Then the organic layer was concentrated to give a colorless solid (102.5 mg). The physical and spectroscopic constants registered for this derivative were identical in all aspects to those previously reported[15,24] for scammonic acid A: mp 154–157°C; $[\alpha]_D=-51.1$ (MeOH, *c* 1.0).

The identification from **1**–**13** of (−)-2*R*,3*R*- or (+)-2*S*,3*S*-methyl 3-hydroxy-2-methylbutyric and (+)-2*S*-methylbutyric acids was performed as follows. Independent solutions of each orizabin (15–80 mg) were saponified according to the above mentioned procedure. After evaporation, each of the organic layer residues (1.6–14.0 mg) was treated with triethylamine (two drops) and 4-bromophenacyl bromide (10–30 mg) in dried acetone (5 mL) for 2 h at room temperature. The reaction mixture was evaporated to dryness, resuspended in $H_2O$ (10 mL) and extracted with $Et_2O$ (20 mL). The resulting organic phases were independently concentrated and their residues were fractionated by normal phase HPLC on an ISCO column (150×19 mm, μporasil, 10 μm), using *n*-hexane–AcOEt (7:3; flow rate=2.0 mL/min) to afford 4-bromophenacyl nilate ($t_R$ 12 min; 1.2–3.4 mg). A mixture of the 4-bromophenacyl ester derivatives of (2*S*)-2-methylbutyric and tiglic (from compounds **1** and **6**–**9**) or isobutyric (from compounds **2**–**5**) acids were also collected ($t_R$=6 min; 3–14 mg). Total separation of these two mixtures was achieved on the same normal phase column, but a different elution system was used, *n*-hexane–AcOEt (92:8); flow rate=2.0 mL/min: $t_R$=13.4 min, 4-bromophenacyl (2*S*)-2-methylbutyrate, mp 40–42°C; $[\alpha]_D=+18.6$ (*c* 1.0, MeOH); $t_R$=15.2 min, 4-bromophenacyl isobutyrate; $t_R$=16.1 min, 4-bromophenacyl tiglate. The same analysis allowed the detection of nilic and (2*S*)-2-methylbutyric acids in the liberated fraction produced by the saponification of compounds **10**–**13**. All the organic acids were identified by comparison of physical and spectral data with published values.[20,21] Orizabins **IX**, **X**, **XII**, **XIV**, **XVI**, **XVIII** and **XX** afforded 4-bromophenacyl (2*R*,3*R*)-nilate, $[\alpha]_D=-13.8$ (*c* 1.0, $CHCl_3$).[29] The enantiomeric (2*S*,2*S*)-nilate was liberated and derivatized from the remaing natural products, $[\alpha]_D=+14.0$ (*c* 1.0, $CHCl_3$).

### 2.4. Determination of the absolute configuration of nilic acid

Each individual solution of 4-bromophenacyl nilate (0.5–1.6 mg) was treated with DMAP (2 mg), DCC (2 mg) and (*R*)-(−)-α-methoxy-α-(trifluoromethyl)-phenylacetyl chloride (4–8 mg) in dry $CH_2Cl_2$ (1 mL). The reactions were allowed to stand overnight under $N_2$. Saturated aqueous $NaHCO_3$ and $Et_2O$ were added to the mixtures and stirred vigorously for 5 min to allow efficient hydrolysis of the excess MTPA-Cl.[30] The organic phases were washed with 0.5N HCl, dried with anhydrous $Na_2SO_4$ and concentrated. Each crude residue was purified by semipreparative HPLC on a normal phase column (150×19 mm, 10 μm) using *n*-hexane–AcOEt (95:5, flow rate=2.5 mL/min) to give the (*S*)-MTPA ester of 4-bromophenacyl nilate (0.3–1.4 mg). Treatment of each nilate derivative with (*S*)-(+)-MTPA-Cl as described above yielded the (*R*)-MTPA ester.

**2.4.1. (*R*)-MTPA ester of (2*R*,3*R*) *p*-bromophenacyl nilate.** Colorless needles; $[\alpha]_D=+7.0$ (*c* 1.0, $CHCl_3$); $^1H$ NMR ($CDCl_3$, 300 MHz) δ 7.75, 7.64 (each 2H, d, *J*=8.7 Hz, Ar*H*), 7.52 (2H, m, Ar*H*), 7.40 (3H, m, Ar*H*), 5.47 (1H, dq, *J*=7.2, 6.6 Hz, H-3), 5.28, 5.18 (each 1H, d, *J*=16.2 Hz, C$H_2$COAr), 3.51 (3H, q, *J*=1.2 Hz, OMe), 2.96 (1H, dq, *J*=7.2, 7.2 Hz, H-2), 1.36 (1H, d, *J*=6.6 Hz, Me-4), 1.31 (1H, d, *J*=7.2 Hz, Me-5).

**2.4.2. (*S*)-MTPA ester of (2*R*,3*R*) *p*-bromophenacyl nilate.** Colorless needles; $[\alpha]_D=-8.6$ (*c* 2.2, $CHCl_3$); $^1H$ NMR ($CDCl_3$, 300 MHz) δ 7.72, 7.64 (each 2H, d, *J*=8.7 Hz, Ar*H*), 7.53 (2H, m, ArH), 7.40 (3H, m, ArH), 5.46 (1H, dq, *J*=7.2, 6.6 Hz, H-3), 5.13, 5.05 (each 1H, d, *J*=16.5 Hz, C$H_2$COAr), 3.56 (3H, q, *J*=1.2 Hz, OMe), 2.93 (1H, dq, *J*=7.2, 7.2 Hz, H-2), 1.44 (3H, d, *J*=6.6 Hz, Me-4), 1.24 (3H, d, *J*=7.2 Hz, Me-5).

**2.4.3. (*R*)-MTPA ester of (2*S*,3*S*) *p*-bromophenacyl nilate.** Colorless needles; $[\alpha]_D=+8.8$ (*c* 1.1, $CHCl_3$); $^1H$ NMR data were identical to those of the (*S*)-MTPA ester of (2*R*,3*R*) *p*-bromophenacyl nilate.

**2.4.4. (*S*)-MTPA ester of (2*S*,3*S*) *p*-bromophenacyl nilate.** Colorless needles; $[\alpha]_D=-8.8$ (*c* 1.2, $CHCl_3$); $^1H$ NMR data were identical to those of the (*R*)-MTPA ester of (2*R*,2*R*) *p*-bromophenacyl nilate.

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Pursh, *I. purga* (Wender) Hayne, *I. quinquefolia* L., and *I. stans* Cav. along with other species from the same Convolvulaceae family. The Aztec medicinal approach was centered in the concept of discharging an illness from the body by means of various convolvulaceous species with cathartic action, as well as with other medicinal plants of purgative, diaphoretic, diuretic and emetic activities. For Pre-Hispanic concepts about illness (phlegm-based etiology) see: Ortiz de Montellano, B. R. *Aztec Medicine, Health and Nutrition*; Rutgers University: New Brunswick, 1990; pp 129–192.

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