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Synthesis of water soluble pentacyclic dihydroxyterpene carboxylic acid derivatives coupled amino acids and their inhibition activities on α-glucosidase

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ABSTRACT

Twenty maslinic acid and corosolic acid derivatives were obtained by coupling with L-amino acids at C-28 position. The α-glucosidase inhibitory activities of the present compounds were evaluated in vitro. Results reveal that some of the derivatives exhibit a better α-glucosidase inhibitory activity than that of acarbose in the test conditions of ethanol-water solution and DMSO. It is worth noting that maslinic acid and corosolic acid derivatives coupled aspartic acid (**9f**: IC_{50} = 382 μm and **10f**: IC_{50} = 364 μm, respectively) have the best water solubility and thus presented higher inhibitory activity than that of acarbose (IC_{50} = 484 μm). Unfortunately, all of the derivatives possess lower inhibitory properties of α-glucosidase than those of the parent compounds in the measurement system of DMSO solution, even if the derivatives exhibit better water solubility than that of the parent compounds.

1. Introduction

Diabetes mellitus (DM) is a chronic metabolic disease characterized by hyperglycemia resulting from defects in insulin secretion, insulin action or both [1–2]. More than 90% of diabetic patient suffers from type II DM, which is non-insulin dependent diabetes mellitus [3]. The treatment of type II DM is mainly by inhibiting the activity of α-glucosidase [4–5], protein tyrosine phosphatase-1B [6–7] and glycogen phosphorylase [8–10]. α-Glucosidase interferes with enzymatic action at the epithelium of the small intestine thus delays the hydrolysis and absorption of carbohydrates, leading to a lowering effect on postprandial blood glucose and insulin levels [11]. Therefore, inhibitors of α-glucosidase are employed to control the postprandial hyperglycemia in the treatment of type II DM. Besides, inhibition of α-glucosidase has attracted a great deal of interest by the pharmaceutical researchers for the treatment of carbohydrate mediated diseases such as cancer, viral infections, and hepatitis [12–13] due to the α-glucosidase enables monosaccharide removal from the related glycoproteins. Hence, the inhibitors of α-glucosidase are known to possess antioxidant [14–15], antiobesity [16–17], antimicrobial [18], antitumor [19], and antiviral [20] activities.

At present, some clinical hypoglycemic drugs, such as acarbose, miglitol, voglibose and so on are utilized as the inhibitor for the treatment of type II DM. However, it is not satisfactory and some consequent side effects including diarrhea, pain, intestinal flatulence were often caused due to prolonging use of the drugs mentioned above [20–22]. Recently, some non-glucosidic based inhibitors exhibiting potential inhibitory activities towards α-glucosidase in vitro such as thiadiazole analogs [2], coumarincarbohydrazones [3], oxindole based oxadiazole derivatives [12], indolcarbohydrazones [13], imidazole derivatives [19], flavone hydrazones [21] and piperidine derivatives [20] has been reported. These inhibitors are hopeful of being the potential therapeutic agents for the regulation of glucose absorption in type II DM. Consequently, it is necessarily to design and develop safe, low toxicity, and effective non-glycosidic based inhibitors from common natural products.

Maslinic acid (MA) and corosolic acid (CA) are natural pentacyclic triterpenoid carboxylic acids existed in great variety of plants such as *Olea europaea* [23] and *Lagerstroemia speciosa* L [24]. Some literatures have demonstrated that MA [25–26] and CA [27–28] (Fig. 1) showed a positive effect on lowing blood glucose levels, but they are provided with a low bioavailability due to the poor water solubility. Structure modification based on the skeleton of MA or CA is considered to be an effective method to enhance their water solubility. Introduction of polar groups or active groups (e.g. amino acid [17,29–30], glycosidic [31], N-heterocyclic residues [32]) into the parent compounds can significantly

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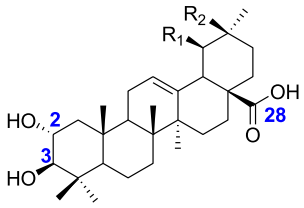


MA (R_1 = H, R_2 = CH_3)
CA (R_1 = CH_3, R_2 = H)

Fig. 1. The structures of maslinic acid and corosolic acid.

improve the water solubility of the pentacyclic triterpenoid carboxylic acids. Currently, a large number of pentacyclic triterpene acid derivatives coupling amino acids exhibited anti-HIV activity [33], cytotoxic activity [34–36] and inhibitory activity of glycogen phosphorylase [37], however, the inhibitory activity of α-glucosidase is rarely reported. In addition, many literatures show that amino acids can regulate insulin secretion from pancreatic β-cells in vivo and in vitro [38–39]. Based on the facts that introduction of amino acids into the parent compounds can improve their water solubility as well as achieve synergistic hypoglycemic effect resulting from superposition of the two actions. Meanwhile, it will be still completely apart into the desired natural products (MA or CA) and the non-hazardous amino acids even if the compounds are hydrolyzed by enzymes in vivo. Here, a series of derivatives of MA and CA as non-glucosidic based α-glucosidase inhibitors have been synthesized via introduction of amino acid groups at C-28 position and their α-glucosidase inhibitory activities in vitro were evaluated, the results of the preliminary structure-activity relationship (SAR) have also been discussed as well.

2. Results and discussion

2.1. Chemistry

The derivatives of pentacyclic dihydroxyterpene carboxylic acid coupling with L-amino acids were prepared as previously described [35,40–41] (Scheme 1). The precursor **MA** (or **CA**) was converted into the 2α, 3β-O-diacetate **3** (or **4**) by treatment with acetic anhydride, which then treated with oxalyl condensed with the appropriate amino acid methyl/ethyl ester hydrochloride (glycine, γ-aminobutyric acid, L-valine acid et.al) in the presence of Et_3N to give the derivatives **7a**−**7n** (or **8a**−**8f**). Hydrolysis of derivatives **7a**−**7n** (or **8a**−**8f**) gave the corresponding derivatives **9a**−**9n** (or **10a**−**10f**), respectively. The structures of the intermediate and the final derivatives were confirmed by ^{1}H NMR, ^{13}C NMR and HRMS.

The ^{1}H NMR spectra of the derivatives **7a**−**7n** (or **8a**−**8f**) showed the signal of the proton of the amide group appears at δ 6.86–6.00 ppm as a double or triple peak, depending on the substituent on the adjacent carbon atom. The presence of an olefinic proton resonating at δ 5.45–5.25 ppm, and a carbinylic proton at 5.11–5.06 or 4.75 ppm were assigned to H-12 and H-2α or H-3β, respectively. Due to the hydrolysis of the derivatives **7a**−**7n** (or **8a**−**8f**), the shift value of the amide proton derivatives (**9a**−**9n** or **10a**−**10f**) moves to a lower field shift of 6.99–7.41 ppm. The ^{1}H NMR data were generally consistent with those reported in the literature [33].

2.2. Water solubility

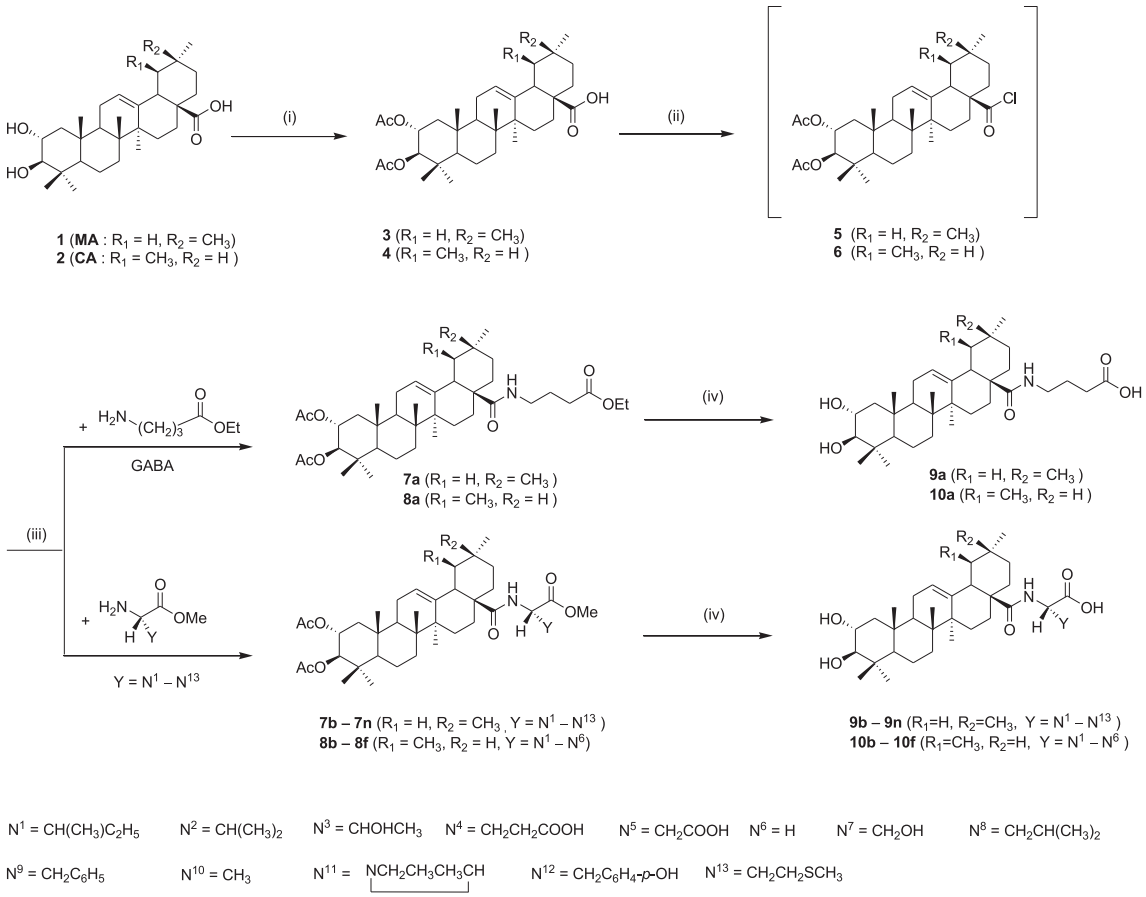
With the introduction of an amino acid molecule into the skeleton of pentacyclic triterpenoid carboxylic acids, the water solubility of the parent compounds is improved remarkably. The results are shown in Table 1. Among all of the derivatives, the glutamic acid derivative of maslinic acid (**9e**) and the aspartic acid derivatives of maslinic acid or corosolic acid (**9f or 10f**) exhibit excellent solubility in the ultrapure water. It's puzzling that the glutamic acid derivative of corosolic acid (**10e**) is poorly soluble in the ultrapure water, which probably due to its structure. The derivatives with a slightly longer hydrophobic side chain (e.g. **9a**, **9b**, **9c**, **9i**, **9n** or **10a**, **10b**, **10c**) are more difficult to dissolve than those of the derivatives with a short hydrophobic side chain (**9g**, **9k**). The derivatives with free hydroxyl group on the C-28 amide side chain (**9d**, **9 h**) can be completely dissolved in 20% ethanol solution, while the derivative **9m** with free hydroxyl group on the benzene ring side chain is slightly poor water-soluble. Moreover, the derivatives with free hydroxyl group on the C-28 amide side chain (**9d**, **9h**, **9m**) have better water solubility than those of the derivatives with hydrophobic side chains (e.g. **9a**, **9b**, **9c**, **9g**, **9i**, **9j**, **9k**, **9n** or **10a**, **10b**, **10c**). It is worth mentioning that the solubility of all derivatives is still significantly better than their parent compounds. When the concentration is below 1 mg/mL, all the derivatives can be completely dissolved in ultrapure water under heating conditions.

2.3. Inhibitory activity of α-glucosidase

In order to find the potent and low-toxic hypoglycemic compounds, the low toxicity ethanol-water system instead of the common solvent (DMSO) was used to dissolve the samples. Then, all of the derivatives and acarbose (as a positive control) were dissolved and their inhibitory activities against α-glucosidase were evaluated in vitro. The assay results are summarized in Table 2.

As shown in Table 2, most of the tested derivatives exhibit some certain inhibitory activity. The C-28 amide side chain of compounds with two free carboxyl groups (**9f**: IC_{50} = 382 μm and **10f**: IC_{50} = 364 μm, respectively) present higher inhibitory activity than that of acarbose (IC_{50} = 484 μm). Compared to the inhibitory activity of acarbose, the inhibitory activity of compounds (**9e** for IC_{50} = 495 μm, **9i** for IC_{50} = 608 μm, **9j** for IC_{50} = 798 μm, **9m** for IC_{50} = 684 μm and **9n** for IC_{50} = 598 μm, respectively) is only slightly weaker. Compounds with free hydroxyl group at the C-28 amide side chain (**9d**: IC_{50} = 998 μm and **9h**: IC_{50} = 1321 μm) show similar inhibitory activity as compounds without the hydroxyl group on the amide side chain (e.g. **9a** for IC_{50} = 987 μm, **9g** for IC_{50} = 1000 μm and **9k** for IC_{50} = 993 μm), indicating that a hydroxyl group at the C-28 amide side chain does not enhance the inhibition of α-glucosidase in despite of the superior water solubility of the former. It is remarkable that the compounds with two free carboxyl groups on the C-28 amide side chain (**9f**: IC_{50} = 382 μm, **9e**: IC_{50} = 495 μm and **10f**: IC_{50} = 364 μm) possess better inhibitory activities than those of the compounds with only one free carboxyl group on the C-28 amide side chain (e.g. **9i**: IC_{50} = 608 μm, **9j**: IC_{50} = 798 μm). Besides, the inhibitory activities of **CA** derivatives are generally lower than those of **MA** derivatives (e.g.**9a** vs **10a**, **9d** vs **10d**). Compounds **9c, 9l**, **10b** and **10e** cannot be dissolved well in ethanol-water at high concentrations for determining their inhibitory activity because of the poor water solubility.

Due to the parent compounds (**MA** and **CA**) cannot dissolve in the solvent system of ethanol-water, it is impossible to measure their bioactivity assay and evaluate the inhibitory activities of α-glucosidase with their derivatives. To further explore the inhibitory activities of α-glucosidase of the parent compounds and their derivatives under identical conditions, the parent compounds (**MA**, **CA**) and their derivatives with the high α-glucosidase inhibitory activity (**9e**, **9f**, **10f**) or with the poor water solubility (**9c**, **9l**, **10b**, **10e**) were dissolved in DMSO. The α-glucosidase inhibitory activities were tested using the method described above in ethanol-water system, and the assay results are presented in Table 3. As shown in Table 3, the derivatives show superior or inferior inhibitory activity than that of acarbose, but the inhibitory activity of all the derivatives and acarbose is lower than those of the parent compounds (**MA** for IC_{50} = 283 μm and **CA** for IC_{50} = 150 μm). The results reveal that coupling a series of amino acids



Scheme 1. Synthesis of pentacyclic dihydroxyterpene carboxylic acid derivatives. (i) $(CH_3CO)_2O$, Py, rt, 18 h; (ii) $(COCl)_2$, Et_3N, CH_2Cl_2, rt, 24 h; (iii) amino acid methyl (ethyl) ester hydrochloride, Et_3N, CH_2Cl_2, 4 h; (iv) 4 N NaOH, CH_3OH, THF, rt, 1 h.

Table 1
Water solubility of the derivatives in the ethanol-water system.

Compounds number	Water solubility	Compounds number	Water solubility
9a	40% ethanol	**9k**	35% ethanol
9b	40% ethanol	**9l**	ethanol
9c	ethanol	**9m**	35% ethanol
9d	20% ethanol	**9n**	40% ethanol
9e	ultrapure water	**10a**	40% ethanol
9f	ultrapure water	**10b**	40% ethanol
9g	35% ethanol	**10c**	40% ethanol
9h	20% ethanol	**10d**	40% ethanol
9i	40% ethanol	**10e**	ethanol
9j	40% ethanol	**10f**	ultrapure water
MA	insoluble	**CA**	insoluble

Table 2
IC_{50} values (μm) of the derivatives for the inhibition of α-glucosidase.

Compounds number	IC_{50}[a]	Compounds number	IC_{50}[a]
9a	987 ± 19	**9k**	993 ± 6
9b	847 ± 15	**9l**	[b]NI
9c	[b]NI	**9m**	684 ± 14
9d	998 ± 8	**9n**	598 ± 5
9e	495 ± 18	**10a**	1112 ± 26
9f	382 ± 5	**10b**	[c]/
9g	1000 ± 14	**10c**	1261 ± 11
9h	1321 ± 20	**10d**	1054 ± 6
9i	608 ± 2	**10e**	[c]/
9j	798 ± 11	**10f**	364 ± 7
Acarbose	484 ± 6		

[a] Values are the mean of four experiments.
[b] NI = no inhibition.
[c] The derivatives cannot dissolved in the solvent system of measurement.

to the C-28 carboxyl site of the parent compounds cannot enhance their inhibitory activities of α-glucosidase, even though the water solubility of the derivatives is improved greatly.

3. Conclusion

In summary, a series of maslinic acid (or corosolic acid) coupled amino acid derivatives have been synthesized and their inhibitory activity against α-glucosidase were performed in vitro. All derivatives possess significantly better water solubility than that of the parent compounds and exhibit superior or inferior α-glucosidase inhibitory activity than that of acarbose utilizing ethanol-water or DMSO as solvent. The inhibitory activities of all the derivatives and acarbose are inferior to those of the parent compounds using DMSO as solvent. Among all the presented derivatives, except for **9e**, the C-28 amide side

Table 3
IC_{50} values (μm) of the derivatives for the inhibition of α-glucosidase.

Compounds number	IC_{50}[a]	Compounds number	IC_{50}[a]
9e	486 ± 2	**9l**	[b]NI
9f	438 ± 6	**10b**	742 ± 8
9c	[b]NI	**10e**	678 ± 9
Acarbose	447 ± 7	**10f**	430 ± 3
MA	283 ± 6	**CA**	150 ± 3

[a] Values are the mean of four experiments.
[b] NI = no inhibition.

chain of derivatives with two free carboxyl groups (**9f**: IC_{50} = 382 and **10f**: IC_{50} = 364 μm, respectively) show a higher inhibitory activities than that of acarbose (IC_{50} = 484 μm) in the ethanol or DMSO system. Most of derivatives reveal slightly lower inhibitory activities than that of acarbose in the ethanol-water system. It is remarkable that the compounds with two free carboxyl groups on the C-28 amide side chain possess better inhibitory activities than those of the compounds with only one free carboxyl group on the C-28 amide side chain. However, compounds which the free hydroxyl at the C-28 amide side chain cannot influence the inhibitory activities. For a fair comparison, the inhibitory activities of **CA** derivatives are generally lower than those of **MA** derivatives.

4. Experimental section

4.1. Chemicals and general methods

All chemicals and solvents we used were bought from commercial suppliers in an analytic grade. Analytical thin layer chromatography was performed on Merck silica-gel 60 F_{254} plates and spots were rendered visible by spraying with a solution of H_2SO_4 − ethanol and heating. Crude products were purified via column chromatography with chemically pure silica gel (200–300 mesh). The melting points were determined using Tektronix X4 microscopic melting point apparatus and uncorrected. NMR spectra were measured in $CDCl_3$ or DMSO-d_6 on a Bruker 400 or 600 MHz spectrometer with the tetramethylsilane (TMS) as an internal standard. Chemical shifts (δ) are presented in ppm and coupling constants (J values) are expressed with Hz. High resolution mass spectral analysis (HRMS) was taken on a Finnigan MAT 95 XP mass spectrometer (Thermo Electron Corporation) via a direct insertion probe. 4-Nitrophenyl-α-D-glucopyranoside (PNPG) and acarbose were purchased from Aladdin. α-Glucosidase (EC 3.2.1.20) from saccharomyces cerevisiae (product number: G5003, CAS: 9001-42-7) was purchased from Sigma.

4.2. Synthesis

4.2.1. Preparation of parent compounds

Maslinic acid and corosolic acid were synthesized by using the reported methods [42–43].

4.2.2. 2α, 3β-Diacetoxy-ole-12-en-28-oic acid (3)

MA (0.63 mmol) was dissolved in pyridine (15 mL), then acetic anhydride (10 mL) was added dropwise over 10 min to mixture under an ice-water bath which was stirred at temperature for 18 h after evacuating ice-water bath. The reaction liquid was washed with 1 N hydrochloric acid (50 mL × 3). After 10 min, the organic layer was separated, washed with saturated sodium bicarbonate solution and distilled water to pH 7, and then dried over anhydrous sodium sulfate, filtered and evaporated in vacuum. The residue was purified by chromatography column on silica gel, eluted by petroleum ether/ethyl acetate (v : v) = 4 : 1 to give **3** as a white powder (yield: 93.2%). Mp: 228–233 °C. ^{1}H NMR (600 MHz, $CDCl_3$): δ 5.26 (t, *J* = 12.0 Hz, 1H), 5.11–5.07 (m, 1H), 4.75 (d, *J* = 12.0 Hz, 1H), 2.82 (dd, *J* = 6.0, 6.0 Hz, 1H), 2.06 (d, *J* = 12.0 Hz, 3H), 2.02–1.99 (m, 1H), 1.98 (s, 3H), 1.95–1.90 (m, 1H), 1.87–1.82 (m, 1H), 1.79–1.73 (m, 1H), 1.71–1.67 (m, 1H), 1.63–1.61 (m, 1H), 1.60 (s, 1H), 1.58–1.57 (d, *J* = 12.0 Hz, 1H), 1.56–1.54 (m, 1H), 1.48–1.45 (m, 1H), 1.44–1.41 (m, 1H), 1.39–1.36 (m, 1H), 1.34–1.29 (m, 2H), 1.26–1.25 (m, 1H), 1.22–1.20 (m, 1H), 1.15 (d, *J* = 12.0 Hz, 1H), 1.12 (s, 3H), 1.08–1.06 (m, 1H), 1.05 (s, 3H), 1.00–0.96 (m, 2H), 0.92 (s, 3H), 0.90 (d, *J* = 6.0 Hz, 9H), 0.88 (d, *J* = 6.0 Hz, 1H), 0.73 (s, 3H). ^{13}C NMR (125 MHz, $CDCl_3$): δ 184.1, 170.8, 170.6, 143.6, 122.2, 80.6, 70.0, 54.8, 47.5, 46.5, 45.8, 41.5, 40.8, 39.4, 39.3, 39.3, 38.2, 33.7, 33.0, 32.4, 32.3, 31.6, 30.6, 28.4, 27.6, 25.9, 23.5, 23.4, 22.8, 22.6, 21.3, 21.1, 21.0, 20.9, 18.1, 18.0, 17.6, 17.1, 17.0, 16.4, 14.1. HRMS (ESI, *m/z*): $[M]^+$ Calcd for $C_{34}H_{52}O_6$: 556.3758; found: 556.3685.

4.2.3. N-[2α, 3β-Diacetoxy-ole-12-en-28-oyl]-γ-aminobutyric acid ethyl ester [33] (7a)

To a solution of **3** (0.089 mmol) in dry dichloromethane (10 mL) with a drop of triethylamine added oxalyl chloride (50 μl) slowly and the reaction mixture was stirred at an ice-water bath for 15 min, then further stirred at room temperature overnight. The solvent was removed to dryness under reduced pressure using a rotary evaporator (40 °C). Dry dichloromethane (15 mL × 3) was added to the residue for the sake of distilling off the remaining oxalyl chloride, then the concentrated to dryness to yield crude 28-O-acetylursolyl chloride **5**. Next in an ice-water bath, a solution of **5** (0.84 mmol) in dry dichloromethane (20 mL), were successively treated with the corresponding γ-aminobutyric acid ethyl ester hydrochloride (1.6 mmol) in the presence of triethylamine (0.20 mL). After stirring in the ice-water bath for 15 min, the reaction mixture was stirred at room temperature for 4 h. The combined organic layer was washed in turn with distilled water and saturated sodium bicarbonate solution after acidified using 1 N aqueous hydrochloric acid solution. The reaction mixture was dried over anhydrous sodium sulfate, filtered and evaporated in vacuum. Then the residue was purified by chromatography column on silica gel, eluted by petroleum ether/ethyl acetate (v : v) = 5 : 1 to give **7a** as a white foamy solid (yield: 80.7%). Mp: 96–107 °C. ^{1}H NMR (600 MHz, $CDCl_3$): δ 6.11 (s, 1H), 5.38 (s, 1H), 5.11–5.07 (m, 1H), 4.75 (d, *J* = 6.0 Hz, 1H), 4.15–4.12 (m, 2H), 3.41 (br, 1H), 3.04 (br, 1H), 2.54 (d, *J* = 18.0 Hz, 1H), 2.35–2.32 (m, 2H), 2.06 (s, 3H), 2.04–2.00 (m, 2H), 1.98 (s, 3H), 1.97–1.94 (m, 2H), 1.82–1.80 (m, 2H), 1.77–1.72 (m, 2H), 1.67 (s, 6H), 1.65–1.62 (m, 2H), 1.27 (s, 1H), 1.26 (d, *J* = 6.0 Hz, 4H), 1.21–1.18 (m, 2H), 1.14 (s, 3H), 1.05 (s, 3H), 1.02 (s, 1H), 0.98 (d, *J* = 18.0 Hz, 1H), 0.91 (d, *J* = 12.0 Hz, 12H), 0.74 (s, 3H).

4.2.4. N-[2α, 3β-Diacetoxy-ole-12-en-28-oyl]-L-isoleucine acid methyl ester (7b)

Compound **7b** was prepared as a white foamy solid (yield: 82.4%) from L-isoleucine acid methyl ester hydrochloride by using the same method established for **7a**. Mp: 117–136 °C. ^{1}H NMR (600 MHz, $CDCl_3$): δ 6.44 (d, *J* = 6.0 Hz, 1H), 5.42 (t, *J* = 6.0 Hz, 1H), 5.11–5.06 (m, 1H), 4.75 (d, *J* = 12.0 Hz, 1H), 4.50–4.48 (m, 1H), 3.69 (s, 3H), 2.65 (dd, *J* = 6.0, 3.6 Hz, 1H), 2.04 (s, 3H), 2.03–1.98 (m, 2H), 1.97 (s, 3H), 1.96–1.93 (m, 1H), 1.89–1.84 (m, 2H), 1.77 (t, *J* = 24.0 Hz, 1H), 1.65–1.52 (m, 8H), 1.48–1.44 (m, 2H), 1.41–1.34 (m, 2H), 1.29–1.27 (m, 1H), 1.21–1.17 (m, 3H), 1.14 (s, 3H), 1.04 (s, 3H), 0.94 (t, *J* = 12.0 Hz, 3H), 0.91 (s, 6H), 0.89 (d, *J* = 3.6 Hz, 6H), 0.87 (d, *J* = 12.0 Hz, 3H), 0.68 (s, 3H). ^{13}C NMR (125 MHz, $CDCl_3$): δ 177.4, 172.2, 170.8, 170.5, 143.8, 122.8, 80.5, 70.0, 56.3, 54.8, 51.8, 47.5, 46.5, 46.4, 43.9, 42.1, 42.0, 39.4, 39.3, 38.2, 38.1, 34.1, 33.1, 32.9, 32.4, 31.5, 30.7, 28.4, 27.3, 25.6, 25.6, 23.5, 23.5, 22.6, 22.1, 20.8, 18.1, 17.6, 16.6, 16.4, 15.2, 14.1, 11.5. HRMS (ESI, *m/z*): $[M+H]^+$ Calcd for $C_{41}H_{65}NO_7$: 684.4833; found: 684.4807.

4.2.5. N-[2α, 3β-Diacetoxy-ole-12-en-28-oyl]-L-valine acid methyl ester (7c)

Compound **7c** was prepared as a white foamy solid (yield: 85.6%) from L-valine acid methyl ester hydrochloride by using the same method established for **7a**. Mp: 168–180 °C. ^{1}H NMR (600 MHz, $CDCl_3$): δ 6.40 (d, *J* = 6.0 Hz, 1H), 5.43 (t, *J* = 6.0 Hz, 1H), 5.11–5.06 (m, 1H), 4.75 (d, *J* = 12.0 Hz, 1H), 4.45–4.43 (m, 1H), 3.70 (s, 3H), 2.66 (dd, *J* = 6.0, 3.6 Hz, 1H), 2.13–2.06 (m, 1H), 2.05 (s, 3H), 2.03–2.00 (m, 2H), 1.98 (s, 3H), 1.97–1.93 (m, 1H), 1.90–1.85 (m, 1H), 1.77–1.73 (m, *J* = 24.0 Hz, 1H), 1.70 (s, 1H), 1.66–1.63 (m, 2H), 1.61–1.58 (m, 2H), 1.56–1.52 (m, 1H), 1.49–1.44 (m, 1H), 1.42–1.33 (m, 2H), 1.29–1.27 (m, 1H), 1.25 (s, 3H), 1.21–1.18 (m, 2H), 1.14 (s, 3H), 1.08 (d, *J* = 12.0 Hz, 1H), 1.04 (s, 3H), 0.97–0.95 (m, 1H), 0.93–0.89 (m, 15H), 0.67 (s, 3H). ^{13}C NMR (125 MHz, $CDCl_3$): δ 177.6, 172.3, 170.7, 170.4, 143.8, 122.8, 80.6, 70.0, 57.2, 54.8, 51.9, 47.5, 46.6, 46.4, 43.9, 42.1,

42.0, 39.5, 39.3, 38.1, 34.1, 33.2, 32.9, 32.4, 31.7, 30.7, 28.4, 27.3, 25.6, 23.5, 21.1, 20.8, 18.7, 18.2, 18.2, 17.6, 16.6, 16.4. HRMS (ESI, *m/z*): $[M]^+$ Calcd for $C_{40}H_{63}NO_7$: 669.4599; found 669.4605.

4.2.6. N-[2α, 3β-Diacetoxy-ole-12-en-28-oyl]-L-threonine acid methyl ester (7d)

Compound **7d** was prepared as a white foamy solid (yield: 78.4%) from L-threonine acid methyl ester hydrochloride by using the same method established for **7a** Mp: 118–136 °C. ^{1}H NMR (600 MHz, $CDCl_3$): δ 6.59 (d, J = 6.0 Hz, 1H), 5.43 (t, J = 6.0 Hz, 1H), 5.11–5.06 (m, 1H), 4.75 (d, J = 12.0 Hz, 1H), 4.52–4.51 (m, 1H), 4.25–4.21 (m, 1H), 3.74 (s, 3H), 2.71 (dd, J = 6.0, 3.6 Hz, 1H), 2.05 (s, 3H), 2.04–2.00 (m, 2H), 1.98 (s, 3H), 1.95–1.92 (m, 1H), 1.89–1.84 (m, 3H), 1.77 (t, J = 24.0 Hz, 1H), 1.67–1.64 (m, 2H), 1.62 (d, J = 6.0 Hz, 1H), 1.60–1.58 (m, 1H), 1.55–1.53 (m, 1H), 1.50–1.46 (m, 1H), 1.42–1.40 (m, 1H), 1.39–1.33 (m, 1H), 1.30–1.27 (m, 1H), 1.25 (s, 3H), 1.20 (d, J = 6.0 Hz, 3H), 1.15 (s, 3H), 1.08–1.06 (m, 1H), 1.04 (s, 3H), 0.97–0.95 (m, 1H), 0.92 (dd, J = 6.0, 6.0 Hz, 12H), 0.70 (s, 3H). ^{13}C NMR (125 MHz, $CDCl_3$): δ 178.7, 171.5, 170.8, 170.5, 143.6, 122.8, 80.5, 69.9, 68.4, 57.5, 52.4, 47.4, 46.7, 42.0, 41.9, 39.4, 39.3, 38.1, 33.2, 33.9, 32.4, 30.7, 28.4, 27.3, 25.6, 25.5, 23.7, 23.5, 21.1, 21.0, 20.9, 20.8, 19.9, 19.8, 18.1, 17.6, 16.8. HRMS (ESI, *m/z*): $[M+Na]^+$ Calcd for $C_{39}H_{61}NO_8$: 694.4289; found: 694.4257.

4.2.7. N-[2α, 3β-Diacetoxy-ole-12-en-28-oyl]-L-glutamic acid diethyl ester (7e)

Compound **7e** was prepared as a white foamy solid (yield: 78.7%) from L-glutamic acid diethyl ester hydrochloride by using the same method established for **7a**. Mp: 94–106 °C. ^{1}H NMR (600 MHz, $CDCl_3$): δ 6.60 (d, J = 6.0 Hz, 1H), 5.42 (t, J = 6.0 Hz, 1H), 5.10–5.06 (m, 1H), 4.74 (d, J = 6.0 Hz, 1H), 4.48–4.45 (m, 1H), 4.23–4.19 (m, 1H), 4.18–4.10 (m, 4H), 2.67 (dd, J = 6.0, 3.6 Hz, 1H), 2.42–2.36 (m, 1H), 2.32–2.27 (m, 1H), 2.20–2.14 (m, 1H), 2.05 (s, 3H), 2.02 (t, J = 12.0 Hz, 1H), 2.00–1.99 (m, 1H), 1.97 (s, 3H), 1.96–1.93 (m, 1H), 1.89–1.84 (m, 1H), 1.76 (t, J = 24.0 Hz, 1H), 1.61–1.52 (m, 6H), 1.48–1.44 (m, 1H), 1.39–1.33 (m, 2H), 1.28 (d, J = 6.0 Hz, 3H), 1.26 (d, J = 6.0 Hz, 2H), 1.24 (d, J = 6.0 Hz, 3H), 1.21–1.18 (m, 2H), 1.14 (s, 3H), 1.08 (d, J = 12.0 Hz, 1H), 1.03 (s, 3H), 0.96–0.94 (m, 1H), 0.92 (s, 3H), 0.91 (s, 3H), 0.89 (d, J = 3.0 Hz, 6H), 0.66 (s, 3H). ^{13}C NMR (125 MHz, $CDCl_3$): δ 177.8, 172.8, 172.0, 170.8, 170.5, 143.7, 122.9, 80.5, 69.9, 61.5, 60.6, 54.8, 51.9, 47.5, 46.4, 46.3, 43.9, 41.9, 41.9, 39.4, 39.3, 38.1, 34.0, 33.0, 33.0, 32.3, 30.7, 30.1, 28.4, 27.6, 27.3, 25.7, 23.6, 23.5, 23.4, 21.1, 20.9, 18.1, 17.6, 16.5, 16.4, 14.1. HRMS (ESI, *m/z*): $[M+K]^+$ Calcd for $C_{43}H_{67}NO_9$: 780.4447; found: 780.4401.

4.2.8. N-[2α, 3β-Diacetoxy-ole-12-en-28-oyl]-L-aspartic acid dimethyl ester (7f)

Compound **7f** was prepared as a white foamy solid (yield: 77.8%) from L-aspartic acid dimethyl ester hydrochloride by using the same method established for **7a**. Mp: 94–106 °C. ^{1}H NMR (600 MHz, $CDCl_3$): δ 6.80 (d, J = 6.0 Hz, 1H), 5.42 (t, J = 6.0 Hz, 1H), 5.11–5.07 (m, 1H), 4.75 (d, J = 12.0 Hz, 1H), 4.69–4.67 (m, 1H), 3.67 (s, 3H), 3.41 (s, 3H), 2.94 (d, J = 6.0 Hz, 2H), 2.68 (dd, J = 3.6, 6.0 Hz, 1H), 2.05 (s, 3H), 2.04–2.01 (m, 2H), 1.98 (s, 3H), 1.90–1.85 (m, 1H), 1.72 (t, J = 24.0 Hz, 1H), 1.61 (t, J = 12.0 Hz, 1H), 1.59–1.56 (m, 4H), 1.54–1.53 (m, 1H), 1.50–1.45 (m, 1H), 1.42–1.39 (m, 1H), 1.38–1.32 (m, 2H), 1.28–1.25 (m, 2H), 1.20–1.18 (m, 2H), 1.14 (s, 3H), 1.08 (d, J = 12.0 Hz, 1H), 1.05 (s, 3H), 0.97–0.95 (m, 1H), 0.90–0.89 (m, 12H), 0.71 (s, 3H). ^{13}C NMR (125 MHz, $CDCl_3$): δ 177.5, 171.3, 171.2, 170.8, 170.5, 143.5, 122.8, 80.5, 70.0, 54.8, 52.7, 51.8, 49.0, 47.5, 46.4, 46.3, 43.9, 41.9, 41.8, 39.4, 39.3, 38.1, 36.0, 34.0, 33.0, 33.0, 32.5, 31.5, 30.7, 28.4, 27.2, 25.7, 23.5, 23.5, 23.4, 21.1, 20.9, 18.2, 17.6, 16.6, 16.5, 14.1. HRMS (ESI, *m/z*): $[M+K]^+$ Calcd for $C_{40}H_{61}NO_9$: 738.3977; found: 738.3983.

4.2.9. N-[2α, 3β-Diacetoxy-ole-12-en-28-oyl]-L-glycine acid methyl ester [33] (7g)

Compound **7g** was prepared as a white foamy solid (yield: 77.5%) from L-glycine acid methyl ester hydrochloride by using the same method established for **7a**. Mp: 123–132 °C. ^{1}H NMR (600 MHz, $CDCl_3$): δ 6.49 (t, J = 4.0 Hz, 1H), 5.44 (d, J = 4.0 Hz, 1H), 5.12–5.06 (m, 1H), 4.75 (d, J = 8.0 Hz, 1H), 4.16–4.10 (m, 1H), 3.84–3.79 (m, 1H), 3.76 (s, 3H), 2.63 (t, J = 16.0 Hz, 1H), 2.05 (s, 3H), 2.03–2.00 (m, 1H), 1.98 (s, 3H), 1.95–1.85 (m, 2H), 1.79–1.70 (m, 2H), 1.66–1.60 (m, 3H), 1.58–1.52 (m, 2H), 1.48–1.45 (m, 1H), 1.42–1.35 (m, 2H), 1.33–1.25 (m, 2H), 1.22–1.19 (m, 2H), 1.15 (s, 3H), 1.12 (t, J = 24.0 Hz, 1H), 1.04 (s, 3H), 1.02–0.95 (m, 2H), 0.91 (t, J = 8.0, 12H), 0.70 (s, 3H).

4.2.10. N-[2α, 3β-Diacetoxy-ole-12-en-28-oyl]-L-serine acid methyl ester (7h)

Compound **7h** was prepared as a white foamy solid (yield: 84.6%) from L-serine acid methyl ester hydrochloride by using the same method established for **7a**. Mp: 186–190 °C. ^{1}H NMR (600 MHz, $CDCl_3$): δ 6.86 (d, J = 6.0 Hz, 1H), 5.45 (t, J = 6.0 Hz, 1H), 5.11–5.06 (m, 1H), 4.75 (d, J = 12.0 Hz, 1H), 4.54–4.51 (m, 1H), 3.97 (dd, J = 3.0, 3.6 Hz, 1H), 3.86–3.83 (m, 1H), 3.78 (s, 3H), 2.69 (dd, J = 6.0, 3.6 Hz, 1H), 2.05 (s, 3H), 2.04–2.00 (m, 2H), 1.97 (s, 3H), 1.96–1.93 (m, 1H), 1.91–1.86 (m, 1H), 1.75 (t, J = 30.0 Hz, 1H), 1.65–1.60 (m, 4H), 1.59–1.53 (m, 2H), 1.48–1.45 (m, 1H), 1.41–1.34 (m, 3H), 1.30–1.25 (m, 2H), 1.22–1.19 (m, 2H), 1.15 (s, 3H), 1.08–1.06 (m, 1H), 1.04 (s, 3H), 0.97–0.95 (m, 1H), 0.92 (d, J = 3.6 Hz, 6H), 0.90 (d, J = 6.0 Hz, 6H), 0.68 (s, 3H). ^{13}C NMR (125 MHz, $CDCl_3$): δ 179.1, 170.8, 170.5, 143.6, 123.1, 80.5, 69.9, 64.3, 55.7, 54.8, 52.7, 47.4, 46.5, 46.3, 43.9, 42.0, 41.9, 39.5, 39.3, 38.0, 34.0, 33.0, 33.0, 32.3, 31.5, 30.7, 28.4, 27.3, 25.7, 23.6, 23.5, 23.5, 22.6, 21.1, 20.9, 18.1, 17.6, 16.4, 16.4, 14.1. HRMS (ESI, *m/z*): $[M+H]^+$ Calcd for $C_{38}H_{59}NO_8$: 658.4313; found: 658.4302.

4.2.11. N-[2α, 3β-Diacetoxy-ole-12-en-28-oyl]-L-leucine acid methyl ester (7i)

Compound **7i** was prepared as a white foamy solid (yield: 82.3%) from L-leucine acid methyl ester hydrochloride by using the same method established for **7a**. Mp: 98–115 °C. ^{1}H NMR (600 MHz, $CDCl_3$): δ 6.32 (d, J = 12.0 Hz, 1H), 5.40 (t, J = 6.0 Hz, 1H), 5.11–5.06 (m, 1H), 4.75 (d, J = 12.0 Hz, 1H), 4.54–4.51 (m, 1H), 3.70 (s, 3H), 2.65 (dd, J = 6.0, 3.6 Hz, 1H), 2.05 (s, 3H), 2.03 (d, J = 6.0 Hz, 1H), 2.00 (t, J = 6.0 Hz, 1H), 1.97 (s, 3H), 1.90–1.85 (m, 1H), 1.74 (t, J = 24.0 Hz, 1H), 1.65–1.59 (m, 8H), 1.56–1.52 (m, 2H), 1.50–1.45 (m, 1H), 1.43–1.36 (m, 1H), 1.35–1.32 (m, 1H), 1.31–1.25 (m, 2H), 1.21–1.16 (m, 2H), 1.14 (s, 3H), 1.08 (d, J = 12.0 Hz, 1H), 1.05 (s, 3H), 1.02–0.95 (m, 2H), 0.94–0.92 (m, 6H), 0.90 (t, J = 6.0 Hz, 12H), 0.71 (s, 3H). ^{13}C NMR (125 MHz, $CDCl_3$): δ 177.5, 173.4, 170.8, 170.5, 143.9, 122.7, 80.6, 70.0, 54.8, 52.1, 51.0, 47.5, 46.5, 46.3, 43.9, 42.0, 42.0, 41.9, 39.4, 39.3, 38.1, 34.1, 33.0, 32.9, 32.4, 30.7, 28.4, 27.2, 25.6, 25.0, 23.7, 23.6, 23.5, 22.7, 22.4, 22.1, 20.9, 18.2, 17.6, 16.6, 16.4. HRMS (ESI, *m/z*): $[M+Na]^+$ Calcd for $C_{41}H_{66}NO_7$:706.4653; found: 706.4664.

4.2.12. N-[2α, 3β-Diacetoxy-ole-12-en-28-oyl]-L-phenylalanine acid methyl ester (7j)

Compound **7j** was prepared as a white foamy solid (yield: 81.2%) from L-phenylalanine acid methyl ester hydrochloride by using the same method established for **7a**. Mp: 101–115 °C. ^{1}H NMR (600 MHz, $CDCl_3$): δ 7.28–7.26 (m, 1H), 7.25 (s, 1H), 7.24–7.21 (m, 1H), 7.10 (t, J = 12.0 Hz, 3H), 6.35 (d, J = 6.0 Hz, 1H), 5.27 (t, J = 6.0 Hz, 1H), 5.09–5.05 (m, 1H), 4.75–4.72 (m, 2H), 3.68 (s, 3H), 3.19–3.16 (m, 1H), 3.05–3.02 (m, 1H), 2.04 (s, 3H), 1.97 (s, 3H), 1.95–1.86 (m, 2H), 1.84–1.79 (m, 1H), 1.72–1.68 (m, 1H), 1.64–1.60 (m, 2H), 1.58–1.56 (m, 1H), 1.55–1.50 (m, 2H), 1.48–1.41 (m, 2H), 1.37–1.30 (m, 2H), 1.25 (s, 2H), 1.23–1.17 (m, 2H), 1.10 (s, 3H), 1.01 (s, 3H), 0.98–0.93 (m, 2H), 0.89 (d, J = 6.0 Hz, 9H), 0.85 (s, 3H), 0.61 (s, 3H). ^{13}C NMR

(125 MHz, $CDCl_3$): δ 177.4, 171.9, 170.8, 170.5, 143.7, 136.1, 129.3, 128.4, 127.0, 122.9, 80.5, 70.0, 54.8, 53.4, 52.1, 47.4, 46.5, 46.3, 43.9, 41.9, 41.8, 39.4, 39.3, 38.0, 37.8, 34.1, 32.9, 32.6, 32.3, 30.7, 29.7, 28.4, 27.2, 25.6, 23.7, 23.5, 23.4, 21.1, 20.9, 18.1, 17.6, 16.4, 16.4. HRMS (ESI, *m/z*): $[M+K]^+$ Calcd for $C_{44}H_{63}NO_7$: 756.4236; found: 756.4229.

4.2.13. N-[2α, 3β-Diacetoxy-ole-12-en-28-oyl]-L-alanine acid methyl ester (7k)

Compound **7k** was prepared as a white foamy solid (yield: 80.1%) from L-alanine acid methyl ester hydrochloride by using the same method established for **7a**. Mp: 117–129 °C. ^1H NMR (600 MHz, $CDCl_3$): δ 6.55 (d, *J* = 6.0 Hz, 1H), 5.42 (t, *J* = 12.0 Hz, 1H), 5.10–5.05 (m, 1H), 4.74 (d, *J* = 12.0 Hz, 1H), 4.46–4.42 (m, 1H), 3.72 (s, 3H), 2.65 (dd, *J* = 6.0, 3.6 Hz, 1H), 2.04 (s, 3H), 2.02–1.99 (m, 2H), 1.96 (s, 3H), 1.95–1.93 (m, 1H), 1.89–1.84 (m, 1H), 1.75 (t, *J* = 24.0 Hz, 1H), 1.60–1.57 (m, 4H), 1.55–1.52 (m, 1H), 1.48–1.39 (m, 2H), 1.37 (d, *J* = 6.0 Hz, 3H), 1.34–1.24 (m, 3H), 1.19–1.17 (m, 2H), 1.13 (s, 3H), 1.07 (d, *J* = 12.0 Hz, 1H), 1.03 (s, 3H), 0.96–0.94 (m, 1H), 0.90 (d, *J* = 3.6 Hz, 6H), 0.89 (d, *J* = 6.0 Hz, 6H), 0.69 (s, 3H). ^{13}C NMR (125 MHz, $CDCl_3$): δ 177.3, 173.6, 170.8, 170.5, 143.9, 122.8, 80.5, 69.9, 54.8, 52.3, 48.3, 47.5, 46.4, 46.2, 43.9, 41.9, 41.9, 39.4, 39.3, 38.1, 34.1, 33.0, 32.8, 32.3, 30.7, 28.4, 27.2, 25.7, 23.6, 23.6, 21.1, 20.8, 18.6, 18.1, 17.6, 16.5, 16.4. HRMS (ESI, *m/z*): $[M+K]^+$ Calcd for $C_{38}H_{59}NO_7$: 642.4364; found: 642.4413.

4.2.14. N-[2α, 3β-Diacetoxy-ole-12-en-28-oyl]-L-proline acid methyl ester (7l)

Compound **7l** was prepared as a white foamy solid (yield: 86.2%) from L-proline acid methyl ester hydrochloride by using the same method established for **7a**. Mp: 116–138 °C. ^1H NMR (600 MHz, $CDCl_3$): δ 5.25 (t, *J* = 12.0 Hz, 1H), 5.11–5.07 (m, 1H), 4.74 (d, *J* = 12.0 Hz, 1H), 4.44 (br.s, 1H), 3.89 (t, *J* = 18.0 Hz, 1H), 3.67 (s, 3H), 3.49 (t, *J* = 12.0 Hz, 1H), 3.11 (d, *J* = 12.0 Hz, 1H), 2.10–2.05 (m, 2H), 2.04 (s, 3H), 2.01–1.98 (m, 2H), 1.96 (s, 3H), 1.95–1.89 (m, 2H), 1.83–1.74 (m, 2H), 1.68–1.59 (m, 4H), 1.57–1.51 (m, 2H), 1.47–1.38 (m, 2H), 1.34–1.26 (m, 3H), 1.21–1.13 (m, 2H), 1.12 (s, 3H), 1.06 (d, *J* = 12.0 Hz, 1H), 1.03 (s, 3H), 1.01–0.95 (m, 1H), 0.92 (s, 3H), 0.89 (t, *J* = 6.0 Hz, 9H), 0.86–0.77 (m, 1H), 0.70 (s, 3H). ^{13}C NMR (125 MHz, $CDCl_3$): δ 175.2, 173.3, 170.8, 170.4, 144.4, 132.8, 129.5, 128.3, 121.6, 80.7, 70.0, 54.9, 51.9, 47.7, 47.0, 45.9, 43.8, 42.4, 41.9, 39.3, 39.2, 38.2, 33.8, 33.2, 32.5, 31.5, 30.5, 29.2, 28.4, 27.5, 27.4, 26.0, 25.6, 24.0, 23.4, 22.6, 22.1, 20.9, 18.2, 17.6, 16.8, 16.4, 14.1. HRMS (ESI, *m/z*): $[M+H]^+$ Calcd for $C_{40}H_{61}NO_7$: 668.4520; found: 668.4520.

4.2.15. N-[2α, 3β-Diacetoxy-ole-12-en-28-oyl]-L-tyrosine acid methyl ester (7m)

Compound **7m** was prep ared as a white foamy solid (yield: 84.8%) from L-tyrosine acid methyl ester hydrochloride by using the same method established for **7a**. Mp: 130–155 °C. ^1H NMR (600 MHz, $CDCl_3$): δ 6.96 (d, *J* = 12.0 Hz, 2H), 6.74 (d, *J* = 6.0 Hz, 2H), 6.42 (d, *J* = 6.0 Hz, 1H), 5.30 (t, *J* = 6.0 Hz, 1H), 5.10–5.06 (m, 1H), 4.74–4.70 (m, 2H), 3.68 (s, 3H), 3.11–3.08 (m, 1H), 2.97–2.94 (m, 1H), 2.47–2.45 (m, 1H), 2.05 (s, 3H), 1.97 (s, 3H), 1.96–1.92 (m, 1H), 1.91–1.87 (m, 1H), 1.85–1.80 (m, 1H), 1.74–1.69 (m, 1H), 1.64–1.59 (m, 2H), 1.58–1.55 (m, 1H), 1.53–1.51 (m, 1H), 1.49–1.42 (m, 2H), 1.40–1.32 (m, 2H), 1.31–1.29 (m, 1H), 1.26–1.24 (m, 1H), 1.20–1.15 (m, 2H), 1.11 (s, 3H), 1.06 (d, *J* = 12.0 Hz, 1H), 1.02 (s, 3H), 1.00 (d, *J* = 12.0 Hz, 1H), 0.95 (d, *J* = 12.0 Hz, 1H), 0.89 (t, *J* = 6.0 Hz, 9H), 0.87 (s, 3H), 0.62 (s, 3H). ^{13}C NMR (125 MHz, $CDCl_3$): δ 177.8, 172.0, 170.9, 170.6, 155.4, 143.6, 130.4, 127.4, 123.0, 115.4, 80.6, 70.0, 54.8, 53.6, 52.2, 47.4, 46.4, 46.4, 43.9, 41.9, 41.9, 39.4, 39.3, 38.0, 37.2, 34.1, 32.9, 32.6, 32.3, 30.7, 28.4, 27.2, 25.6, 23.6, 23.5, 23.4, 22.6, 21.1, 20.9, 18.1, 17.6, 16.4, 16.4, 14.1. HRMS (ESI, *m/z*): $[M+H]^+$ Calcd for $C_{44}H_{63}NO_8$: 734.4626; found: 734.4621.

4.2.16. N-[2α, 3β-Diacetoxy-ole-12-en-28-oyl]-L-methionine acid methyl ester (7n)

Compound **7n** was prepared as a white foamy solid (yield: 84.9%) from L-methionine acid methyl ester hydrochloride by using the same method established for **7a**. Mp: 123–138 °C. ^1H NMR (600 MHz, $CDCl_3$): δ 6.61 (d, *J* = 6.0 Hz, 1H), 5.43 (t, *J* = 6.0 Hz, 1H), 5.11–5.06 (m, 1H), 4.75 (d, *J* = 12.0 Hz, 1H), 4.60–4.57 (m, 1H), 3.73 (s, 3H), 2.65 (dd, *J* = 6.0, 6.0 Hz, 1H), 2.51–2.44 (m, 2H), 2.19–2.13 (m, 1H), 2.08 (s, 3H), 2.05 (s, 3H), 2.03 (d, *J* = 6.0 Hz, 1H), 2.01–2.00 (m, 1H), 1.99 (d, *J* = 6.0 Hz, 1H), 1.98 (s, 3H), 1.96–1.94 (m, 1H), 1.90–1.86 (m, 1H), 1.75 (t, *J* = 24.0 Hz, 1H), 1.63–1.53 (m, 6H), 1.49–1.44 (m, 1H), 1.42–1.33 (m, 2H), 1.29–1.25 (m, 1H), 1.21–1.18 (m, 2H), 1.14 (s, 3H), 1.08 (d, *J* = 12.0 Hz, 1H), 1.04 (s, 3H), 1.03–0.98 (m, 1H), 0.97–0.95 (m, 1H), 0.91 (s, 6H), 0.90 (d, *J* = 6.0 Hz, 6H), 0.68 (s, 3H). ^{13}C NMR (125 MHz, $CDCl_3$): δ 177.7, 172.4, 170.8, 170.5, 143.7, 122.9, 80.5, 70.0, 54.8, 52.4, 51.7, 47.5, 46.4, 46.4, 43.9, 42.0, 41.9, 39.5, 39.3, 38.1, 34.1, 33.0, 33.0, 32.4, 31.8, 30.7, 29.9, 28.4, 27.3, 25.7, 23.6, 23.6, 23.5, 21.1, 20.9, 18.1, 17.6, 16.5, 16.4, 15.5. HRMS (ESI, *m/z*): $[M+K]^+$ Calcd for $C_{40}H_{63}NO_7S$: 740.3956; found: 740.3996.

4.2.17. N-[2α, 3β-Diacetoxy-urs-12-en-28-oyl]-γ-aminobutyric acid ethyl ester (8a)

Compound **8a** was prepared as a white foamy solid (yield: 80.7%) from γ-aminobutyric acid ethyl ester hydrochloride by using the same method established for **7a**. Mp: 98–110 °C. ^1H NMR (600 MHz, $CDCl_3$): δ 6.00 (t, *J* = 12.0 Hz, 1H), 5.30 (t, *J* = 4.0 Hz, 1H), 5.12–5.06 (m, 1H), 4.76 (d, *J* = 12.0 Hz, 1H), 4.15–4.10 (m, 2H), 3.41–3.33 (m, 1H), 3.06–2.98 (m, 1H), 2.33 (t, *J* = 16.0 Hz, 1H), 2.05 (s, 3H), 1.97 (s, 3H), 1.96–1.92 (m, 2H), 1.87–1.77 (m, 5H), 1.71–1.55 (m, 5H), 1.52–1.29 (m, 8H), 1.25 (t, *J* = 16.0 Hz, 4H), 1.14–1.09 (m, 1H), 1.07 (s, 3H), 1.06 (s, 3H), 1.01–0.97 (m, 1H), 0.94 (s, 3H), 0.90 (s, 3H), 0.89 (s, 3H), 0.86 (d, *J* = 16.0 Hz, 1H), 0.75 (s, 3H). ^{13}C NMR (125 MHz, $CDCl_3$): δ 178.0, 173.3, 170.8, 170.5, 139.9, 125.2, 80.6, 70.0, 60.5, 54.8, 53.7, 47.7, 47.4, 44.1, 42.5, 39.7, 39.6, 39.3, 39.1, 38.9, 38.0, 37.3, 32.5, 31.9, 30.8, 28.4, 27.8, 24.7, 24.5, 23.4, 23.2, 21.2, 21.1, 20.8, 18.1, 17.6, 17.2, 16.9, 16.5, 14.2. HRMS (ESI, *m/z*): $[M]^+$ Calcd for $C_{44}H_{63}NO_8$: 669.4599; found: 669.4594.

4.2.18. N-[2α, 3β-Diacetoxy-urs-12-en-28-oyl]-L-isoleucine acid ethyl ester (8b)

Compound **8b** was prepared as a white foamy solid (yield: 84.3%) from L-isoleucine acid methyl ester hydrochloride by using the same method established for **7a**. Mp: 123–138 °C. ^1H NMR (600 MHz, $CDCl_3$): δ 6.44 (d, *J* = 12.0 Hz, 1H), 5.37 (t, *J* = 6.0 Hz, 1H), 5.11–5.06 (m, 1H), 4.75 (d, *J* = 6.0 Hz, 1H), 4.50–4.48 (m, 1H), 3.69 (s, 3H), 2.07 (d, *J* = 6.0 Hz, 1H), 2.04 (s, 3H), 2.01–1.98 (m, 1H), 1.97 (s, 3H), 1.96–1.93 (m, 4H), 1.84–1.79 (m, 2H), 1.75–1.63 (m, 3H), 1.59–1.56 (m, 1H), 1.53–1.48 (m, 3H), 1.46–1.45 (m, 1H), 1.45–1.40 (m, 2H), 1.38–1.34 (m, 1H), 1.32–1.24 (m, 2H), 1.21–1.13 (m, 1H), 1.11 (d, *J* = 12.0 Hz, 1H), 1.08 (s, 3H), 1.05 (s, 3H), 1.02–1.00 (m, 1H), 0.95 (s, 3H), 0.92 (t, *J* = 12.0 Hz, 3H), 0.89–0.87 (m, 9H), 0.85 (d, *J* = 6.0 Hz, 3H), 0.66 (s, 3H). ^{13}C NMR (125 MHz, $CDCl_3$): δ 177.3, 172.3, 170.8, 170.5, 138.4, 126.0, 80.5, 70.0, 56.2, 54.8, 53.9, 51.8, 47.9, 47.5, 44.1, 42.3, 39.6, 39.6, 39.3, 39.1, 38.4, 38.0, 37.6, 32.7, 30.8, 28.4, 27.8, 25.5, 24.6, 23.4, 23.3, 21.1, 21.1, 20.9, 18.1, 17.6, 17.1, 16.5, 16.5, 15.1, 11.5. HRMS (ESI, *m/z*): $[M+Na]^+$ Calcd for $C_{41}H_{65}NO_7$: 706.4653; found: 706.4704.

4.2.19. N-[2α, 3β-Diacetoxy-urs-12-en-28-oyl]-L-valine acid ethyl ester (8c)

Compound **8c** was prepared as a white foamy solid (yield: 84.5%) from L-valine acid methyl ester hydrochloride by using the same method established for **7a**. Mp: 123–138 °C. ^1H NMR (600 MHz, $CDCl_3$): δ 6.40 (d, *J* = 8.0 Hz, 1H), 5.37 (t, *J* = 8.0 Hz, 1H), 5.12–5.05 (m, 1H), 4.76 (d, *J* = 12.0 Hz, 1H), 4.43 (t, *J* = 12. 0 Hz, 1H), 3.69 (s, 3H), 2.20–2.06 (m, 2H), 2.05 (s, 3H), 1.97 (s, 3H), 1.95–1.93 (m, 2H),

1.84–1.65 (m, 7H),1.59–1.45 (m, 7H), 1.38–1.20 (m, 7H), 1.12–1.11 (m, 1H), 1.07 (s, 3H), 1.04 (s, 3H), 0.95 (s, 3H), 0.92 (s, 2H), 0.90 (d, J = 4.0 Hz, 9H), 0.65 (s, 2H). ^{13}C NMR (125 MHz, $CDCl_3$): δ 177.5, 172.4, 170.8, 170.5, 138.4, 126.0, 80.6, 70.0, 57.1, 54.9, 54.0, 51.9, 48.0, 47.5, 44.1, 42.4, 39.7, 39.3, 39.1, 38.1, 37.8, 32.7, 32.0, 30.9, 29.7, 28.4, 27.8, 24.6, 23.4, 23.4, 21.1, 21.1, 20.9, 18.7, 18.3, 18.1, 17.6, 17.1, 16.6, 16.5. HRMS (ESI, m/z): $[M]^+$ Calcd for $C_{40}H_{63}NO_7$: 669.4599; found: 669.4559.

4.2.20. N-[2α, 3β-Diacetoxy-urs-12-en-28-oyl]-L-threonine acid ethyl ester (8d)

Compound **8d** was prepared as a white foamy solid (yield: 83.2%) from L-threonine acid methyl ester hydrochloride by using the same method established for **7a**. Mp: 136–142 °C. ^{1}H NMR (600 MHz, $CDCl_3$): δ 6.58 (d, J = 6.0 Hz, 1H), 5.38 (t, J = 6.0 Hz, 1H), 5.11–5.06 (m, 1H), 4.75 (d, J = 6.0 Hz, 1H), 4.53–4.51 (m, 1H), 4.19–4.15 (m, 1H), 3.73 (s, 3H), 2.07 (d, J = 6.0 Hz, 1H), 2.04 (s, 3H), 2.02–2.00 (m, 2H), 1.97 (s, 3H), 1.96–1.94 (m, 1H), 1.85–1.81 (m, 1H), 1.74–1.67 (m, 2H), 1.58–1.40 (m, 8H), 1.38–1.25 (m, 5H), 1.18 (d, J = 6.0 Hz, 3H), 1.13 (t, J = 12.0 Hz, 1H), 1.09 (s, 3H), 1.07 (s, 1H), 1.05 (s, 3H), 0.96 (d, J = 6.0 Hz, 3H), 0.89 (d, J = 2.4 Hz, 6H), 0.88 (d, J = 6.0 Hz, 3H), 0.68 (s, 3H). ^{13}C NMR (125 MHz, $CDCl_3$): δ 178.7, 171.5, 170.8, 170.5, 138.3, 126.1, 80.5, 70.0, 68.8, 57.4, 54.9, 53.7, 52.4, 48.2, 47.5, 44.1, 42.4, 39.7, 39.6, 39.3, 39.0, 38.0, 37.8, 32.8, 31.5, 30.8, 28.4, 27.8, 24.6, 23.4, 23.3, 21.1, 21.1, 20.8, 19.9, 18.1, 17.6, 17.1, 16.7, 16.6. HRMS (ESI, m/z): $[M+H]^+$ Calcd for $C_{39}H_{61}NO_8$: 672.4469; found: 672.4462.

4.2.21. N-[2α, 3β-Diacetoxy-urs-12-en-28-oyl]-L-glutamic acid diethyl ester (8e)

Compound **8e** was prepared as a white foamy solid (yield: 79.1%) from L-glutamic acid diethyl ester hydrochloride by using the same method established for **7a**. Mp: 116–122 °C. ^{1}H NMR (600 MHz, $CDCl_3$): δ 6.60 (d, J = 6.0 Hz, 1H), 5.37 (t, J = 6.0 Hz, 1H), 5.10–5.05 (m, 1H), 4.74 (d, J = 6.0 Hz, 1H), 4.49–4.46 (m, 1H), 4.23–4.18 (m, 1H), 4.15–4.09 (m, 3H), 2.40–2.35 (m, 1H), 2.29–2.23 (m, 1H), 2.18–2.12 (m, 1H), 2.04 (s, 3H), 2.00–1.98 (m, 1H), 1.96 (s, 3H), 1.95–1.93 (m, 3H), 1.81–1.79 (m, 1H), 1.75–1.70 (m, 2H), 1.67–1.61 (m, 1H), 1.58–1.52 (m, 1H), 1.50–1.29 (m, 8H), 1.28 (d, J = 6.0 Hz, 3H), 1.25 (d, J = 2.4 Hz, 2H), 1.24 (d, J = 12.0 Hz, 3H), 1.10 (d, J = 12.0 Hz, 1H), 1.07 (s, 3H), 1.02 (s, 3H), 1.01–0.98 (m, 1H), 0.95 (s, 3H), 0.94 (s, 1H), 0.88–0.86 (m, 9H), 0.64 (s, 3H). ^{13}C NMR (125 MHz, $CDCl_3$): δ 177.7, 172.8, 172.0, 170.7, 170.4, 138.3, 126.1, 80.5, 70.0, 61.5, 60.5, 54.8, 53.6, 51.6, 47.7, 47.4, 44.0, 42.2, 39.6, 39.5, 39.3, 39.0, 38.0, 37.5, 32.6, 31.5, 30.8, 30.1, 28.4, 27.8, 27.7, 24.4, 27.8, 27.7, 24.4, 23.4, 23.3, 22.6, 21.1, 21.1, 20.8, 18.1, 17.6, 17.1, 16.5, 16.4, 14.1, 14.1. HRMS (ESI, m/z): $[M+K]^+$ Calcd for $C_{43}H_{67}NO_9$: 780.4447; found: 780.4514.

4.2.22. N-[2α, 3β-Diacetoxy-urs-12-en-28-oyl]-L-aspartic acid dimethyl ester (8f)

Compound **8f** was prepared as a white foamy solid (yield: 79.3%) from L-aspartic acid dimethyl ester hydrochloride by using the same method established for **7a**. Mp: 119–126 °C. ^{1}H NMR (600 MHz, $CDCl_3$): δ 6.78 (d, J = 6.0 Hz, 1H), 5.39 (t, J = 6.0 Hz, 1H), 5.11–5.06 (m, 1H), 4.75 (d, J = 12.0 Hz, 1H), 4.65–4.62 (m, 1H), 3.75 (s, 3H), 3.66 (s, 3H), 2.92 (d, J = 4.8 Hz, 2H), 2.07–2.05 (m, 1H), 2.04 (s, 3H), 2.03–1.99 (m, 2H), 1.97 (s, 3H), 1.96–1.92 (m, 1H), 1.77–1.74 (m, 1H), 1.69–1.64 (m, 3H), 1.59–1.56 (m, 1H), 1.53–1.47 (m, 3H), 1.44–1.41 (m, 1H), 1.40–1.37 (m, 1H), 1.36–1.32 (m, 1H), 1.31–1.20 (m, 3H), 1.13–1.09 (m, 1H), 1.08 (s, 3H), 1.05 (s, 3H), 1.03–1.00 (m, 1H), 0.96 (s, 1H), 0.94 (s, 3H), 0.89–0.86 (m, 9H), 0.70 (s, 3H). ^{13}C NMR (125 MHz, $CDCl_3$): δ 177.5, 171.3, 171.3, 170.8, 170.5, 138.2, 126.0, 80.5, 70.0, 54.8, 53.4, 52.7, 51.8, 49.0, 47.8, 47.4, 44.1, 42.3, 39.6, 39.5, 39.3, 38.9, 38.0, 37.4, 36.0, 32.7, 30.8, 28.4, 27.7, 24.5, 23.4, 23.3, 21.1, 21.1, 20.9, 18.1, 17.6, 17.0, 16.6, 16.6. HRMS (ESI, m/z): $[M+Na]^+$ Calcd for $C_{40}H_{61}NO_9$: 722.4238; found: 722.4209.

4.2.23. N-[2α, 3β-Dihydroxy-ole-12-en-28-oyl]-γ-aminobutyric acid acid [33] (9a)

An aqueous solution of 4 N NaOH (0.50 mL) was added to a solution of the ethyl ester derivative (**7a**) (0.21 mmol) in MeOH/THF (v : v = 2 : 3, 10 mL) and was stirred for 1 h at room temperature. The hydrolysis product was neutralized to pH 7 with 1 N hydrochloric acid. After that, the organic solvent was dried over anhydrous sodium sulfate, filtered and evaporated in vacuum. The residue was purified by chromatography column on silica gel, eluted by dichloromethane/methanol (v : v) = 10 : 1 to give **9a** as a white powder (yield: 70.4%). Mp: 131–156 °C. ^{1}H NMR (600 MHz, DMSO-d_6): δ 12.0 (s, 1H), 7.31 (t, J = 12.0 Hz, 1H), 5.21 (t, J = 6.0 Hz, 1H), 4.38–4.37 (dd, J = 6.0, 3.0 Hz, 2H), 3.04–2.96 (m, 2H), 2.79–2.76 (m, 1H), 2.73–2.71 (m, 1H), 2.17–2.15 (t, J = 12.0 Hz, 2H), 1.91–1.79 (m, 3H), 1.75–1.72 (m, 1H), 1.66 (t, J = 24.0 Hz, 1H), 1.60–1.55 (m, 4H), 1.50–1.42 (m, 3H), 1.38–1.36 (m, 2H), 1.31–1.28 (m, 2H), 1.26–1.17 (m, 2H), 1.07 (s, 3H), 0.90 (s, 3H), 0.87 (s, 6H), 0.85 (s, 3H), 0.76 (t, J = 24.0 Hz, 2H), 0.68 (s, 3H), 0.63 (s, 3H).

4.2.24. N-[2α, 3β-Dihydroxy-ole-12-en-28-oyl]-L-isoleucine acid (9b)

Compound **9b** was prepared as a white powder (yield: 65.4%) from the hydrolysis of **7b** by using the same method established for **9a**. Mp: 147–184 °C. ^{1}H NMR (600 MHz, DMSO-d_6): δ 6.99 (d, J = 6.0 Hz, 1H), 5.23 (t, J = 12.0 Hz, 1H), 4.08 (t, J = 12.0 Hz, 1H), 3.43–3.38 (m, 1H), 2.73–2.70 (m, 2H), 1.94–1.78 (m, 4H), 1.76–1.73 (m, 1H), 1.69–1.64 (m, 2H), 1.55–1.47 (m, 3H), 1.45–1.37 (m, 4H), 1.34–1.28 (m, 2H), 1.26–1.21 (m, 1H), 1.20–1.12 (m, 3H), 1.08 (s, 3H), 0.94 (t, J = 6.0 Hz, 1H), 0.91 (s, 3H), 0.87 (t, J = 12.0 Hz, 9H), 0.82 (t, J = 18.0 Hz, 6H), 0.77–0.72 (m, 2H), 0.69 (s, 3H), 0.62 (s, 3H). ^{13}C NMR (125 MHz, DMSO-d_6): δ 173.7, 170.5, 141.2, 119.3, 79.7, 64.6, 53.7, 52.2, 44.6, 44.3, 43.4, 43.0, 38.9, 38.3, 36.4, 35.1, 33.7, 31.1, 30.3, 29.9, 29.8, 27.8, 26.3, 24.3, 23.0, 22.5, 20.9, 20.5, 19.9, 15.5, 14.6, 14.2, 13.8, 12.8. HRMS (ESI, m/z): $[M]^+$ Calcd for $C_{36}H_{59}NO_5$: 585.4387; found: 585.4387.

4.2.25. N-[2α, 3β-Dihydroxy-ole-12-en-28-oyl]-L-valine acid (9c)

Compound **9c** was prepared as a white powder (yield: 68.4%) from the hydrolysis of **7c** by using the same method established for **9a**. Mp: 112–128 °C. ^{1}H NMR (600 MHz, DMSO-d_6): δ 7.23 (d, J = 6.0 Hz, 1H), 5.20 (t, J = 6.0 Hz, 1H), 4.34 (d, J = 6.0 Hz, 1H), 4.24 (d, J = 4.2 Hz, 1H), 4.01 (t, J = 12.0 Hz, 1H), 3.58 (s, 3H), 3.43–3.38 (m, 1H), 3.30 (s, 3H), 2.78 (dd, 1H, J = 6.0, 6.0 Hz), 2.73–2.71 (m, 1H), 2.06–2.01 (m, 1H), 1.94–1.89 (m, 1H), 1.82–1.80 (m, 2H), 1.75–1.73 (m, 1H), 1.71–1.68 (m, 1H), 1.66 (d, J = 12.0 Hz, 1H), 1.57–1.52 (m, 2H), 1.50 (t, J = 18.0 Hz, 1H), 1.45–1.41 (m, 2H), 1.39–1.37 (m, 1H), 1.34–1.28 (m, 2H), 1.26–1.23 (m, 1H), 1.18–1.15 (m, 1H), 1.08 (s, 3H), 0.90 (s, 3H), 0.88 (s, 3H), 0.86 (s, 6H), 0.84 (s, 1H), 0.83 (s, 1H), 0.69 (s, 3H), 0.60 (s, 3H). ^{13}C NMR (125 MHz, DMSO-d_6): δ 176.7, 172.2, 144.0, 121.5, 82.3, 67.2, 58.1, 54.8, 51.5, 47.1, 46.9, 45.9, 45.5, 41.6, 40.6, 39.0, 38.9, 37.7, 33.6, 32.9, 32.5, 32.2, 30.4, 29.7, 28.8, 26.9, 25.6, 23.6, 23.1, 22.1, 22.1, 19.2, 19.1, 18.1, 17.2, 16.8, 16.3. HRMS (ESI, m/z): $[M+H]^+$ Calcd for $C_{35}H_{57}NO_5$: 572.4309; found: 572.4309.

4.2.26. N-[2α, 3β-Dihydroxy-ole-12-en-28-oyl]-L-threonine acid (9d)

Compound **9d** was prepared as a white powder (yield: 65.7%) from the hydrolysis of **7d** by using the same method established for **9a**. Mp: 156–168 °C. ^{1}H NMR (600 MHz, DMSO-d_6): δ 7.02 (d, J = 6.0 Hz, 1H), 5.25 (s, 1H), 4.39 (br, 2H), 3.99 (t, J = 6.0 Hz, 1H), 3.89 (s, 1H), 2.73 (d, J = 6.0 Hz, 1H), 2.68 (d, J = 12.0 Hz, 1H), 1.97–1.88 (m, 2H), 1.85–1.79 (m, 2H), 1.76–1.66 (m, 3H), 1.56–1.42 (m, 8H), 1.39–1.28 (m, 4H), 1.22–1.16 (m, 2H), 1.08 (s, 3H), 0.92 (d, J = 12.0 Hz, 6H), 0.87 (t, J = 12.0 Hz, 9H), 0.75–0.72 (m, 2H), 0.68 (s, 3H), 0.63 (s, 3H). ^{13}C NMR (125 MHz, DMSO-d_6): δ 175.9, 172.6, 143.2, 122.2, 82.2, 67.1, 65.6, 57.1, 54.7, 47.1, 46.9, 45.9, 45.4, 41.4, 41.0, 37.6, 33.6,

32.8, 32.4, 30.4, 28.8, 26.9, 25.6, 23.5, 23.4, 23.0, 22.8, 18.0, 17.1, 16.6, 16.4. HRMS (ESI, *m/z*): $[M+H]^+$ Calcd for $C_{34}H_{55}NO_6$: 574.4102; found: 574.4101.

*4.2.27. N-[2α, 3β-Dihydroxy-ole-12-en-28-oyl]-L-glutamic acid (**9e**)*

Compound **9e** was prepared as a white powder (yield: 62.8%) from the hydrolysis of **7e** by using the same method established for **9a**. Mp: 212–235 °C. ^{1}H NMR (600 MHz, DMSO-d_6): δ 7.18 (s, 1H), 5.23 (s, 1H), 3.90 (d, J = 6.0 Hz, 1H), 3.43–3.39 (m, 2H), 2.73 (d, J = 12.0 Hz, 1H), 2.68 (d, J = 12.0 Hz, 1H), 2.21–2.05 (m, 3H), 1.92–1.64 (m, 8H), 1.52–1.47 (m, 2H), 1.46–1.44 (m, 2H), 1.38–1.10 (m, 6H), 1.07 (s, 3H), 0.90 (s, 3H), 0.86 (t, J = 6.0 Hz, 9H), 0.76–0.71 (m, 2H), 0.68 (s, 3H), 0.61 (s, 3H). ^{13}C NMR (125 MHz, DMSO-d_6): δ 173.1, 140.9, 119.5, 99.0, 79.7, 64.6, 52.2, 44.6, 43.5, 42.7, 38.8, 36.4, 36.4, 35.1, 31.1, 30.4, 30.2, 29.9, 27.9, 26.3, 24.4, 23.1, 20.9, 20.5, 20.2, 15.5, 14.6, 14.1, 13.9. HRMS (ESI, *m/z*): $[M+H]^+$ Calcd for $C_{35}H_{55}NO_7$: 602.4051; found: 602.4050.

*4.2.28. N-[2α, 3β-Dihydroxy-ole-12-en-28-oyl]-L-aspartic acid (**9f**)*

Compound **9f** was prepared as a white powder (yield: 64.2%) from the hydrolysis of **7f** by using the same method established for **9a**. Mp: 112–135 °C. ^{1}H NMR (600 MHz, DMSO-d_6): δ 7.17 (d, J = 3.0 Hz, 1H), 5.28 (t, J = 6.0 Hz, 1H), 4.35 (d, J = 36.0 Hz, 2H), 3.91 (s, 1H), 3.44–3.39 (m, 3H), 2.73 (d, J = 6.0 Hz, 1H), 1.11 (d, J = 12.0 Hz, 1H), 2.46 (d, J = 12.0 Hz, 1H), 2.34–2.29 (m, 1H), 1.95–1.89 (m, 1H), 1.86–1.80 (m, 2H), 1.77–1.74 (m, 1H), 1.69 (t, J = 24.0 Hz, 1H), 1.50–1.48 (m, 2H), 1.45–1.43 (m, 3H), 1.39–1.36 (m, 1H), 1.34–1.29 (m, 2H), 1.19 (d, J = 12.0 Hz, 1H), 1.09 (s, 3H), 1.04 (d, J = 12.0 Hz, 1H), 0.90 (s, 3H), 0.87 (d, J = 6.0 Hz, 9H), 0.77–0.72 (m, 2H), 0.68 (s, 3H), 0.62 (s, 3H). ^{13}C NMR (125 MHz, DMSO-d_6): δ 140.6, 119.9, 79.7, 64.6, 53.5, 52.2, 44.6, 44.4, 43.4, 42.8, 38.6, 38.6, 35.0, 31.0, 30.3, 30.2, 29.8, 27.9, 26.3, 24.4, 23.1, 20.9, 20.6, 20.2, 16.0, 15.5, 14.6, 13.9. HRMS (ESI, *m/z*): $[M+Na]^+$ Calcd for $C_{34}H_{53}NO_7$: 610.3714; found: 610.3714.

*4.2.29. N-[2α, 3β-Dihydroxy-ole-12-en-28-oyl]-L-glycine acid [33] (**9g**)*

Compound **9g** was prepared as a white powder (yield: 75.9%) from the hydrolysis of **7g** by using the same method established for **9a**. Mp: 179–195 °C. ^{1}H NMR (600 MHz, DMSO-d_6): δ 7.34 (s, 1H), 5.22 (s, 1H), 3.64 (dd, J = 8.0, 4.0 Hz, 1H), 3.46–3.38 (m, 2H), 2.73–2.68 (t, J = 20.0 Hz, 2H), 1.94–1.81 (m, 3H), 1.76–1.66 (m, 2H), 1.63–1.55 (m, 2H), 1.51–1.50 (m, 2H), 1.45–1.28 (m, 6H), 1.22–1.11 (m, 2H), 1.08 (s, 3H), 0.90 (s, 3H), 0.86 (s, 9H), 0.80 (d, J = 8.0 Hz, 1H), 0.74 (d, J = 12.0 Hz, 2H), 0.68 (s, 3H), 0.62 (s, 3H).

*4.2.30. N-[2α, 3β-Dihydroxy-ole-12-en-28-oyl]-L-serine acid (**9h**)*

Compound **9h** was prepared as a white powder (yield: 68.6%) from the hydrolysis of **7h** by using the same method established for **9a**. Mp: 109–125 °C. ^{1}H NMR (600 MHz, DMSO-d_6): δ 7.14 (s, 1H), 5.25 (d, J = 12.0 Hz, 1H), 4.42 (br.s 1H), 3.77 (s, 1H), 3.64–3.62 (m, 1H), 3.43–3.39 (m, 2H), 3.35–3.33 (m, 1H), 2.73 (d, J = 6.0 Hz, 1H), 2.68–2.66 (m, 1H), 1.95–1.91 (m, 1H), 1.88–1.80 (m, 2H), 1.76–1.73 (m, 1H), 1.70–1.65 (m, 1H), 1.59–1.52 (m, 1H), 1.50–1.42 (m, 5H), 1.38–1.37 (m, 1H), 1.33–1.28 (m, 2H), 1.23–1.17 (m, 2H), 1.08 (s, 3H), 0.90 (s, 3H), 0.87 (s, 3H), 0.86 (s, 3H), 0.85 (s, 3H), 0.77–0.71 (m, 2H), 0.68 (s, 3H), 0.66 (s, 1H), 0.64 (s, 3H). ^{13}C NMR (125 MHz, DMSO-d_6): δ 173.6, 140.7, 119.7, 79.7, 64.6, 59.6, 52.2, 44.6, 44.4, 43.4, 42.8, 38.8, 38.5, 35.1, 31.1, 30.4, 29.8, 28.4, 27.9, 26.3, 24.4, 23.1, 20.9, 20.6, 20.3, 19.5, 15.5, 14.6, 13.9, 13.9, 11.4. HRMS (ESI, *m/z*): $[M+K]^+$ Calcd for $C_{33}H_{53}NO_6$: 598.3504; found: 598.3505.

*4.2.31. N-[2α, 3β-Dihydroxy-ole-12-en-28-oyl]-L-leucine acid (**9i**)*

Compound **9i** was prepared as a white powder (yield: 66.3%) from the hydrolysis of **7i** by using the same method established for **9a**. Mp: 166–179 °C. ^{1}H NMR (600 MHz, DMSO-d_6): δ 7.33 (d, J = 6.0 Hz, 1H), 5.16 (t, J = 12.0 Hz, 1H), 4.16–4.12 (m, 1H), 3.43–3.38 (m, 2H), 2.79 (dd, J = 6.0, 3.6 Hz, 1H), 2.73 (d, J = 12.0 Hz, 1H), 1.95–1.90 (m, 1H), 1.85–1.79 (m, 1H), 1.77–1.73 (m, 1H), 1.67–1.61 (m, 4H), 1.56–1.52 (m, 2H), 1.50–1.46 (m, 2H), 1.44–1.37 (m, 3H), 1.33–1.22 (m, 3H), 1.17–1.09 (m, 2H), 1.07 (s, 3H), 0.98–0.93 (m, 1H), 0.91 (s, 3H), 0.88–0.84 (m, 12H), 0.81 (d, J = 6.0 Hz, 3H), 0.77–0.72 (m, 2H), 0.69 (s, 3H), 0.63 (s, 3H). ^{13}C NMR (125 MHz, DMSO-d_6): δ 173.9, 171.9, 141.5, 126.7, 125.9, 118.7, 79.7, 64.6, 52.3, 47.9, 44.6, 44.3, 43.5, 42.7, 38.9, 37.9, 36.4, 36.3, 35.1, 31.1, 30.4, 30.1, 29.6, 27.8, 26.3, 24.3, 23.0, 21.8, 20.9, 20.6, 20.5, 19.7, 18.7, 15.5, 14.6, 14.4, 13.8. HRMS (ESI, *m/z*): $[M+K]^+$ Calcd for $C_{36}H_{59}NO_5$: 624.4024; found: 624.4024.

*4.2.32. N-[2α, 3β-Dihydroxy-ole-12-en-28-oyl]-L-phenylalanine acid (**9j**)*

Compound **9j** was prepared as a white powder (yield: 63.2%) from the hydrolysis of **7j** by using the same method established for **9a**. Mp: 167–186 °C. ^{1}H NMR (600 MHz, DMSO-d_6): δ 7.30 (d, J = 6.0 Hz, 1H), 7.21 (d, J = 6.0 Hz, 3H), 7.16–7.14 (m, 1H), 5.06 (s, 1H), 4.25 (t, J = 18.0 Hz, 1H), 3.44–3.36 (m, 5H), 3.07–3.04 (m, 1H), 2.95–2.91 (m, 1H), 2.71 (d, J = 6.0 Hz, 1H), 2.59 (dd, J = 4.2, 6.0 Hz, 1H), 1.86–1.82 (m, 1H), 1.73–1.68 (m, 3H), 1.60–1.53 (m, 3H), 1.41–1.36 (m, 2H), 1.30–1.20 (m, 4H), 1.09–1.01 (m, 3H), 0.99 (s, 3H), 0.90 (s, 3H), 0.84 (s, 3H), 0.81 (d, J = 6.0 Hz, 6H), 0.72 (d, J = 12.0 Hz, 1H), 0.68 (s, 3H), 0.23 (s, 3H). ^{13}C NMR (125 MHz, DMSO-d_6): δ 176.2, 173.4, 143.9, 138.2, 129.4, 128.0, 126.2, 121.4, 82.3, 67.2, 54.9, 54.2, 47.2, 46.9, 46.0, 45.2, 41.3, 40.5, 39.0, 38.8, 37.6, 36.3, 33.7, 32.9, 32.0, 30.4, 28.9, 26.8, 25.6, 23.4, 23.0, 22.4, 17.9, 17.2, 16.4, 16.0. HRMS (ESI, *m/z*): $[M+Na]^+$ Calcd for $C_{39}H_{57}NO_5$: 596.3921; found: 596.3922.

*4.2.33. N-[2α, 3β-Dihydroxy-ole-12-en-28-oyl]-L-alanine acid (**9k**)*

Compound **9k** was prepared as a white powder (yield: 69.3%) from the hydrolysis of **7k** by using the same method established for **9a**. Mp: 153–165 °C. ^{1}H NMR (600 MHz, DMSO-d_6): δ 7.35 (d, J = 6.0 Hz, 1H), 5.20 (t, J = 6.0 Hz, 1H), 4.09–4.05 (m, 1H), 3.43–3.38 (m, 2H), 2.75–2.71 (m, 2H), 1.95–1.90 (m, 1H), 1.83–1.79 (m, 2H), 1.75–1.72 (m, 1H), 1.68–1.63 (m, 1H), 1.59–1.54 (m, 2H), 1.52–1.49 (m, 1H), 1.47–1.45 (m, 2H), 1.43–1.37 (m, 2H), 1.33–1.25 (m, 3H), 1.23 (d, J = 12.0 Hz, 3H), 1.20–1.18 (m, 1H), 1.11 (d, J = 2.4 Hz, 1H), 1.08 (s, 3H), 0.90 (s, 3H), 0.87 (s, 3H), 0.86 (d, J = 1.8 Hz, 6H), 0.75–0.72 (m, 2H), 0.68 (s, 3H), 0.63 (s, 3H). ^{13}C NMR (125 MHz, DMSO-d_6): δ 175.9, 174.5, 144.0, 121.8, 82.4, 67.3, 54.9, 48.0, 47.3, 47.0, 46.1, 45.2, 41.5, 40.9, 39.1, 39.1, 37.8, 33.7, 33.1, 32.5, 31.1, 30.6, 29.0, 27.0, 25.8, 23.6, 23.2, 22.4, 22.2, 18.2, 17.8, 17.3, 16.8, 16.5, 14.1. HRMS (ESI, *m/z*): $[M+H]^+$ Calcd for $C_{33}H_{53}NO_5$: 544.3996; found: 544.3999.

*4.2.34. N-[2α, 3β-Dihydroxy-ole-12-en-28-oyl]-L-proline acid (**9l**)*

Compound **9l** was prepared as a white powder (yield: 71.9%) from the hydrolysis of **7l** by using the same method established for **9a**. Mp: 127–135 °C. ^{1}H NMR (600 MHz, DMSO-d_6): δ 5.07 (d, J = 6.0 Hz, 1H), 4.34 (d, J = 6.0 Hz, 1H), 4.25 (d, J = 6.0 Hz, 1H), 4.15–4.11 (m, 1H), 3.8 (br.s, 1H), 3.56 (s, 3H), 3.45–3.38 (m, 2H), 2.94 (d, J = 12.0 Hz, 1H), 2.73–2.71 (m, 1H), 2.02–1.99 (m, 1H), 1.97–1.95 (m, 1H), 1.93–1.91 (m, 1H), 1.80–1.78 (m, 1H), 1.75–1.72 (m, 2H), 1.68–1.63 (m, 2H), 1.52–1.48 (m, 2H), 1.45 (d, J = 6.0 Hz, 1H), 1.40–1.37 (m, 1H), 1.36–1.33 (m, 1H), 1.32–1.29 (m, 1H), 1.23–1.22 (m, 1H), 1.21–1.18 (m, 1H), 1.14–1.12 (m, 1H), 1.08 (s, 3H), 0.91 (s, 3H), 0.88 (s, 3H), 0.87 (s, 3H), 0.85 (s, 3H), 0.77–0.73 (m, 2H), 0.69 (s, 3H), 0.62 (s, 3H). ^{13}C NMR (125 MHz, DMSO-d_6): δ 171.6, 170.1, 141.9, 118.5, 79.7, 64.6, 52.3, 48.9, 44.8, 44.6, 44.3, 43.7, 43.0, 39.6, 39.0, 36.4, 36.2, 35.1, 30.7, 30.3, 29.8, 28.4, 27.6, 26.3, 26.2, 24.5, 23.2, 22.9, 21.2, 20.4, 19.5, 15.4, 14.6, 13.9, 13.8. HRMS (ESI, *m/z*): $[M+Na]^+$ Calcd for $C_{35}H_{55}NO_5$: 592.3972; found: 592.3956.

*4.2.35. N-[2α, 3β-Dihydroxy-ole-12-en-28-oyl]-L-tyrosine acid (**9m**)*

Compound **9m** was prepared as a white powder (yield: 73.6%) from the hydrolysis of **7m** by using the same method established for **9a**. Mp:

185–219 °C. ^{1}H NMR (600 MHz, DMSO-d_6): δ 7.19 (d, J = 6.0 Hz, 1H), 6.98 (d, J = 12.0 Hz, 2H), 6.60 (d, J = 12.0 Hz, 2H), 5.07 (t, J = 6.0 Hz, 1H), 4.19–4.16 (m, 1H), 3.41–3.36 (m, 2H), 2.94–2.91 (m, 1H), 2.83–2.79 (m, 1H), 2.71 (d, J = 6.0 Hz, 1H), 2.59 (dd, J = 3.6, 3.0 Hz, 1H), 1.87–1.82 (m, 1H), 1.73–1.68 (m, 3H), 1.61–1.53 (m, 3H), 1.42–1.38 (m, 3H), 1.31–1.22 (m, 5H), 1.16 (d, J = 24.0 Hz, 1H), 1.10–1.07 (m, 2H), 1.00 (s, 3H), 0.90 (s, 3H), 0.84 (s, 3H), 0.82 (d, J = 6.0 Hz, 6H), 0.73 (d, J = 12.0 Hz, 1H), 0.69 (s, 3H), 0.28 (s, 3H). ^{13}C NMR (125 MHz, DMSO-d_6): δ 173.6, 153.5, 141.4, 127.6, 125.4, 118.9, 112.3, 79.7, 64.6, 52.3, 51.9, 44.6, 44.3, 43.5, 42.6, 38.7, 38.0, 36.4, 36.2, 35.0, 32.9, 31.1, 30.3, 29.5, 27.8, 26.3, 24.2, 23.1, 20.8, 20.4, 19.9, 15.4, 14.6, 13.8, 13.5. HRMS (ESI, m/z): $[M+Na]^+$ Calcd for $C_{39}H_{57}NO_6$: 658.4078; found: 658.4078.

4.2.36. N-[2α, 3β-Dihydroxy-ole-12-en-28-oyl]-L-methionine acid (9n)

Compound **9n** was prepared as a white powder (yield: 62.4%) from the hydrolysis of **7n** by using the same method established for **9a**. Mp: 108–135 °C. ^{1}H NMR (600 MHz, DMSO-d_6): δ 7.41 (d, J = 8.0 Hz, 1H), 5.17 (s, 1H), 4.22 (d, J = 8.0 Hz, 1H), 2.77–2.71 (m, 2H), 2.46–2.34 (m, 2H), 2.00 (s, 3H), 1.96–1.88 (m, 3H), 1.79–1.71 (m, 3H), 1.67–1.60 (m, 2H), 1.55–1.42 (m, 6H), 1.39–1.27 (m, 4H), 1.22–1.12 (m, 3H), 1.07 (s, 3H), 0.90 (s, 3H), 0.87 (d, J = 8.0 Hz, 9H), 0.80–0.71 (m, 3H), 0.68 (s, 3H), 0.63 (s, 3H). ^{13}C NMR (125 MHz, DMSO-d_6): δ 175.9, 173.0, 143.4, 120.9, 81.7, 66.6, 54.3, 50.7, 46.6, 46.4, 45.4, 44.8, 41.0, 40.9, 40.0, 37.1, 33.1, 32.4, 32.0, 31.8, 29.8, 29.6, 28.3, 26.4, 25.1, 22.9, 22.5, 22.0, 21.7, 21.5, 17.8, 17.5, 16.6, 16.4, 16.3, 15.8, 14.0. HRMS (ESI, m/z): $[M+H]^+$ Calcd for $C_{35}H_{57}NO_5S$: 604.4030; found: 604.4030.

4.2.37. N-[2α, 3β-Dihydroxy-urs-12-en-28-oyl]-γ-aminobutyric acid (10a)

Compound **10a** was prepared as a white powder (yield: 71.8%) from the hydrolysis of **8a** by using the same method established for **9a**. Mp: 175–180 °C. ^{1}H NMR (600 MHz, DMSO-d_6): δ 7.18 (t, J = 12.0 Hz, 1H), 5.20 (t, J = 6.0 Hz, 1H), 3.43–3.39 (m, 1H), 3.01–2.94 (m, 2H), 2.73 (d, J = 6.0 Hz, 1H), 2.16 (t, J = 12.0 Hz, 3H), 1.91–1.84 (m, 3H), 1.79–1.70 (m, 2H), 1.61–1.54 (m, 3H), 1.52–1.37 (m, 7H), 1.34–1.21 (m, 5H), 1.02 (s, 3H), 0.91 (d, J = 6.0 Hz, 9H), 0.82 (d, J = 6.0 Hz, 3H), 0.79–0.71 (m, 2H), 0.69 (s, 3H), 0.66 (s, 3H). ^{13}C NMR (125 MHz, DMSO-d_6): δ 176.4, 174.6, 138.7, 124.7, 82.4, 67.3, 54.9, 52.0, 47.2, 47.2, 46.7, 41.8, 39.3, 39.1, 39.0, 38.6, 38.4, 37.7, 37.3, 32.9, 31.5, 30.6, 29.0, 27.5, 24.6, 23.6, 23.5, 23.1, 21.3, 18.2, 17.4, 17.3, 17.0, 16.6. HRMS (ESI, m/z): $[M]^+$ Calcd for $C_{34}H_{55}NO_5$: 557.4074; found: 557.4068.

4.2.38. N-[2α, 3β-Dihydroxy-urs-12-en-28-oyl]-L-isoleucine acid (10b)

Compound **10b** was prepared as a white powder (yield: 69.7%) from the hydrolysis of **8b** by using the same method established for **9a**. Mp: 174–192 °C. ^{1}H NMR (600 MHz, DMSO-d_6): δ 6.87 (d, J = 4.0 Hz, 1H), 5.23 (s, 1H), 4.09 (t, J = 12.0 Hz, 1H), 2.73 (d, J = 8.0 Hz, 1H), 2.49 (s, 1H), 2.09 (d, J = 12.0 Hz, 1H), 1.93–1.84 (m, 3H), 1.79–1.73 (m, 2H), 1.68–1.61 (m, 3H), 1.47–1.31 (m, 8H), 1.29–1.11 (m, 6H), 1.02 (s, 3H), 0.90 (d, J = 8.0 Hz, 9H), 0.83–0.79 (m, 9H), 0.77–0.73 (m, 1H), 0.68 (s, 3H), 0.61 (s, 3H). ^{13}C NMR (125 MHz, DMSO-d_6): δ 175.6, 172.4, 137.4, 124.7, 81.8, 66.6, 55.5, 54.2, 51.9, 46.6, 46.6, 46.4, 41.2, 37.8, 37.0, 36.3, 36.1, 32.2, 29.9, 28.3, 26.8, 24.5, 23.1, 22.7, 22.4, 20.5, 17.5, 16.6, 16.5, 16.1, 15.9, 14.7. HRMS (ESI, m/z): $[M+H]^+$ Calcd for $C_{36}H_{59}NO_5$: 586.4466; found: 586.4464

4.2.39. N-[2α, 3β-Dihydroxy-urs-12-en-28-oyl]-L-valine acid (10c)

Compound **10c** was prepared as a white powder (yield: 76.8%) from the hydrolysis of **8c** by using the same method established for **9a**. Mp: 187–197 °C. ^{1}H NMR (600 MHz, DMSO-d_6): δ 6.87 (d, J = 6.0 Hz, 1H), 5.23 (t, J = 6.0 Hz, 1H), 4.36 (s, 1H), 4.24 (s, 1H), 4.02 (t, J = 18.0 Hz, 1H), 3.43–3.39 (m, 1H), 2.73 (d, J = 6.0 Hz, 1H), 2.10 (d, J = 6.0 Hz, 1H), 2.02–1.96 (m, 1H), 1.93–1.88 (m, 1H), 1.86–1.84 (m, 1H), 1.79–1.77 (m, 1H), 1.69–1.63 (m, 3H), 1.47–1.42 (m, 3H), 1.40–1.34 (m, 3H), 1.31–1.19 (m, 3H), 1.03 (s, 3H), 0.90 (d, J = 5.4 Hz, 6H), 0.88 (s, 3H), 0.86–0.83 (m, 9H), 0.81–0.71 (m, 3H), 0.69 (s, 3H), 0.62 (s, 3H). ^{13}C NMR (125 MHz, DMSO-d_6): δ 176.5, 173.1, 138.0, 125.4, 82.4, 67.3, 57.5, 54.9, 52.6, 47.3, 47.2, 47.1, 41.9, 39.1, 39.0, 38.5, 37.7, 37.0, 32.9, 30.6, 29.0, 27.5, 23.8, 23.4, 23.1, 21.2, 19.2, 18.8, 18.2, 17.3, 17.2, 16.8, 16.6. HRMS (ESI, m/z): $[M]^+$ Calcd for $C_{35}H_{57}NO_5$: 571.4231; found: 571.4252.

4.2.40. N-[2α, 3β-Dihydroxy-urs-12-en-28-oyl]-L-threonine acid (10d)

Compound **10d** was prepared as a white powder (yield: 73.4%) from the hydrolysis of **8d** by using the same method established for **9a**. Mp: 178–189 °C. ^{1}H NMR (600 MHz, DMSO-d_6): δ 6.99 (s, 1H), 5.27 (s, 1H), 4.41 (s, 1H), 4.33 (s, 1H), 3.96 (s, 1H), 3.79 (s, 1H), 2.73 (d, J = 6.0 Hz, 1H), 1.96–1.90 (m, 1H), 1.88–1.78 (m, 3H), 1.62–1.53 (m, 3H), 1.46–1.35 (m, 6H), 1.33–1.19 (m, 6H), 1.03 (s, 3H), 0.90 (s, 6H), 0.88–0.83 (m, 9H), 0.77–0.71 (m, 2H), 0.68 (s, 3H), 0.63 (s, 3H). ^{13}C NMR (125 MHz, DMSO-d_6): δ 175.9, 172.9, 137.5, 125.9, 82.5, 67.3, 65.5, 57.1, 54.9, 53.1, 47.4, 47.3, 47.0, 41.9, 39.1, 39.1, 38.8, 37.7, 37.5, 32.8, 30.6, 29.0, 27.5, 24.3, 23.5, 23.2, 21.3, 18.5, 18.2, 17.3, 17.3, 16.7, 16.6. HRMS (ESI, m/z): $[M]^+$ Calcd for $C_{34}H_{55}NO_6$: 573.4023; found: 573.4009.

4.2.41. N-[2α, 3β-Dihydroxy-urs-12-en-28-oyl]-L-glutamic acid (10e)

Compound **10e** was prepared as a white powder (yield: 62.8%) from the hydrolysis of **8e** by using the same method established for **9a**. Mp: 223–237 °C. ^{1}H NMR (600 MHz, DMSO-d_6): δ 7.07 (s, 1H), 5.24 (s, 1H), 4.41 (br.s, 1H), 3.83 (s, 1H), 2.72 (s, 1H), 2.11–1.22 (m, 25H), 1.02 (s, 3H), 0.90 (d, J = 6.0 Hz, 9H), 0.83 (s, 3H), 0.68 (s, 3H), 0.63 (s, 3H). ^{13}C NMR (125 MHz, DMSO-d_6): δ 174.9, 137.0, 125.0, 81.8, 66.7, 54.2, 52.5, 52.3, 46.6, 46.2, 41.2, 38.1, 37.0, 32.1, 30.0, 28.3, 26.9, 23.5, 22.7, 22.6, 20.6, 17.5, 16.6, 16.0. HRMS (ESI, m/z): $[M+H]^+$ Calcd for $C_{35}H_{55}NO_7$: 602.4051; found: 602.4051.

4.2.42. N-[2α, 3β-Dihydroxy-urs-12-en-28-oyl]-L-aspartic acid (10f)

Compound **10f** was prepared as a white powder (yield: 65.3%) from the hydrolysis of **8f** by using the same method established for **9a**. Mp: 211–225 °C. ^{1}H NMR (600 MHz, DMSO-d_6): δ 7.23 (d, J = 5.4 Hz, 1H), 5.24 (t, J = 6.0 Hz, 1H), 4.39 (s, 1H), 4.31 (s, 1H), 4.08 (s, 1H), 3.78–3.63 (m, 1H), 3.42–3.39 (m, 2H), 3.03 (s, 1H), 2.73 (d, J = 6.0 Hz, 1H), 2.00 (d, J = 6.0 Hz, 1H), 1.93–1.86 (m, 4H), 1.80–1.77 (m, 1H), 1.64–1.57 (m, 2H), 1.47–1.41 (m, 3H), 1.38–1.33 (m, 3H), 1.29–1.19 (m, 3H), 1.02 (s, 3H), 0.90 (d, J = 6.0 Hz, 9H), 0.83 (d, J = 6.0 Hz, 3H), 0.79–0.71 (m, 2H), 0.68 (s, 3H), 0.64 (s, 3H). ^{13}C NMR (125 MHz, DMSO-d_6): δ 176.2, 173.3, 172.5, 172.2, 137.8, 125.7, 82.5, 67.4, 63.4, 54.9, 52.7, 49.0, 47.3, 47.3, 46.9, 42.7, 41.9, 39.1, 39.0, 38.6, 37.7, 37.1, 32.8, 30.6, 29.0, 27.4, 24.0, 23.4, 23.2, 21.3, 18.2, 17.4, 17.3, 16.7, 16.6. HRMS (ESI, m/z): $[M+H]^+$ Calcd for $C_{34}H_{53}NO_7$: 588.3894; found: 588.3896.

4.3. Enzyme inhibition assay

4.3.1. α-Glucosidase inhibition assay

The inhibitory activity of the compounds on α-glucosidase was carried out according to the literature method with a slight modification [44–45]. In brief, the reaction system contained 67 mM phosphate buffer ($KH_2PO_4-K_2HPO_4$, pH 6.80), 10 μl, 0.8 U/ml α-glucosidase and a certain volume of sample solution (dissolved in the different ratios of ethanol–water solution), which were well mixed to a final volume of 170 μl and allowed to pre-incubated for 20 min at 37.5 °C in the 96-well plates. Afterwards, 20 μl of 10 mM PNPG (4-nitrophenyl-α-D-glucopyranoside) was added and the reaction mixture was further incubated for 6 min at 37.5 °C. The absorbance was recorded at 405 nm using a microplate reader. Acarbose was used as a positive control. Moreover, due to the low water solubility of the parent compounds and their derivatives (**9c**, **9l**, **10b**, **10e**), it is necessary to further compare the inhibitory

of activity of the parent compounds and their derivatives on α-glucosidase replacing the ethanol–water solution with DMSO in the above method. The inhibition percentage was calculated as follows:

$$\alpha - \text{glucosidase inhibition } (\%) = [A-(A_1 - A_2)/A] \times 100$$

Where A was the absorbance without the sample solution; A_1 was the absorbance with the sample solution; A_2 was background reference of the sample solution. The IC_{50} value is defined as the concentration of the sample solution at an inhibition rate of 50%.

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Appendix A. Supplementary material

Supplementary data to this article can be found online at https://doi.org/10.1016/j.bioorg.2019.02.001.

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