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ORIGINAL PAPER

An unexpected reaction of cinobufagin analogues in the presence of PPh_3/I_2

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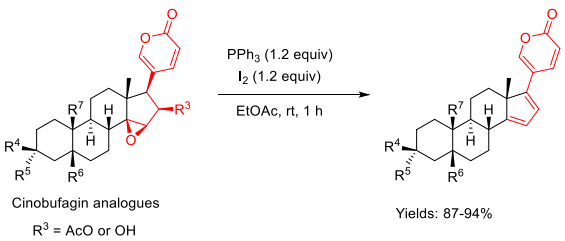
Abstract

An unexpected reaction of cinobufagin analogues in the presence of PPh_3/I_2 in EtOAc at room temperature to obtain diene compounds has been described. The whole process is carried out under mild conditions and the desired products are formed in good yields. It provides a simple, effective, and novel reaction method for the synthesis of diene compounds from corresponding substrates.

Graphic abstract



Keywords Cinobufagin · Bufalene · PPh_3/I_2 · Cyclopenta-1 · 3-Diene · Natural product

Introduction

Chansu is a Traditional Chinese Medicine made from the dried skin and parotid venom glands of *Bufo bufo gargarizans* Cantor or *Bufo melanostictus* Schneider [1, 2]. Bufalenes are the main active ingredients which show the significant biological activity [3–5]. Cinobufagin is one of bufalenes, and the structure is shown in Fig. 1. The structure of cinobufagin includes the following functional groups: hydroxy on C3, epoxy on C14–15, acetoxyl on C16, and 2*H*-pyran-2-one-5-yl on C17. To study structure–activity relationships, a series of cinobufagin derivatives, especially modification on C3, are synthesized.

According to the literature [6], the hydroxyl group could be replaced by iodine atom in the presence of PPh_3/I_2/DMAP. Therefore, we carried out the reaction of cinobufagin and PPh_3/I_2/DMAP under the reported conditions to synthesis of 3-iodide compound. After the completion of the reaction, the main product was isolated by column

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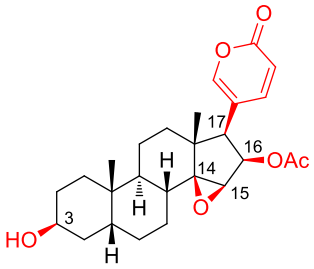


Fig. 1 Structure of cinobufagin

chromatography on silica gel. The product was light yellow, and the mass spectrometry data did not agree with the expected compounds. This phenomenon attracted our attention, so we used NMR to confirm the structure of the compound. The NMR results clearly showed that the compound was not the desired iodide product, but the elimination product (Scheme 1). This structure was also consistent with previous mass spectrometry data. We repeated the experiment three times under the same conditions and got the same results.

The results were completely unexpected, and we were excited by the results, even though the products were not what we expected. This is a novel reaction and has not been reported in the literature so far, so it is meaningful and valuable to further study this reaction.

Results and discussion

The following experiment showed that the base played no role in the reaction, and the same results were obtained with or without it. Then, to further improve the yields of the reaction, we carried out condition optimization experiments (Table 1).

Initially, the amount of PPh_3 was fixed at 1.5 equiv to examine the effect of different amounts of I_2 on the reaction. As shown in Table 1, with the increase of the amount of I_2, the yields of **3a** gradually increased. When the amount of I_2 was higher than 1.2 equiv, the yields were no longer increased, which indicates that 1.2 equiv I_2 was enough for this reaction. Then, the amount of PPh_3 was also optimized and 1.2 equiv PPh_3 showed best experimental result with the desired product **3a** in 89% yield (Table 1, entry 6). Therefore, PPh_3 (1.2 equiv) and I_2 (1.2 equiv) were selected for all further reaction.

Then, to investigation the solvent effect, various solvents such as DCM, THF, 1,4-dioxane, toluene, EtOAc, DMF, and acetone were applied to promote this elimination reaction (Table 2). All the solvents studied on this reaction showed good effect in terms of the yields of **3a** (80–92%) but THF (17%) and DMF (15%). Considering that hydroxyl group may react with PPh_3/I_2, we did not choose alcohol solvents as the reaction medium. The

Scheme 1

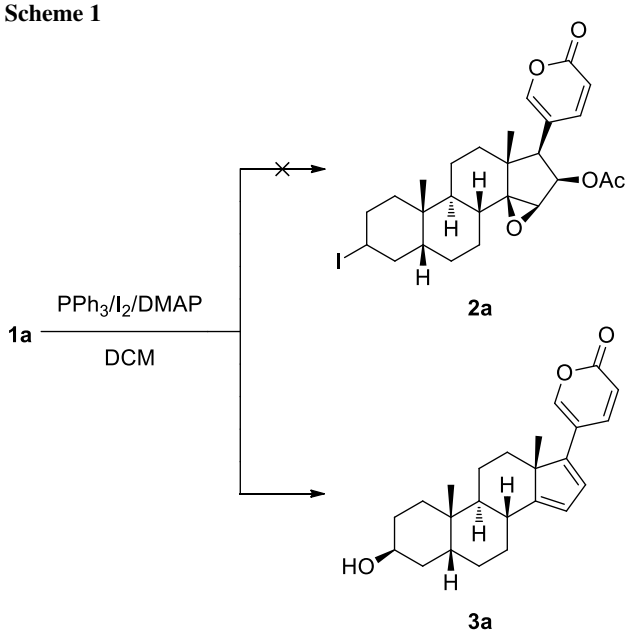
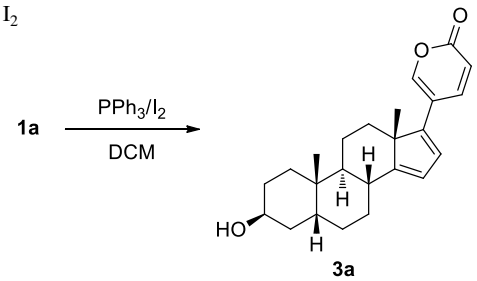


Table 1 Optimization of the amount of PPh_3 and I_2



Entry	PPh_3/equiv	I_2/equiv	Yield of **3a**/%[a]
1	1.5	0.5	32
2	1.5	0.8	54
3	1.5	1.0	75
4	1.5	1.2	86
5	1.5	1.5	88
6	1.2	1.2	89
7	1.0	1.2	72
8	0.8	1.2	55
9	0.5	1.2	35

Reaction conditions: 443 mg **1a** (1.0 mmol), PPh_3, I_2, 10 cm^3 DCM, rt for 1 h

[a]Isolated yields

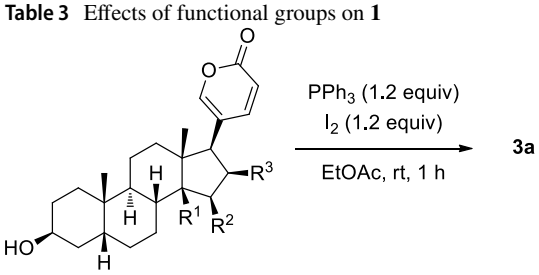
Table 2 Solvent screen

Entry	Solvent	Yield of **3a**/%[a]
1	DCM	89
2	THF	17
3	1,4-Dioxane	90
4	Toluene	90
5	EtOAc	92
6	DMF	15
7	Acetone	80

Reaction conditions: 443 mg **1a** (1.0 mmol), 315 mg PPh_3 (1.2 mmol, 1.2 equiv), 305 mg I_2 (1.2 mmol, 1.2 equiv), 10 cm^3 solvent, and rt for 1 h

[a]Isolated yields

Table 3 Effects of functional groups on **1**



1

Entry	Substrate **1**	Yield of **3a**/%[a]
1	**1a**: R^1, R^2=O, R^3=AcO	92
2	**1b**: R^1, R^2=O, R^3=OH	90
3	**1c**: R^1, R^2=O, R^3=H	0
4	**1d**: R^1=OH, R^2=H, R^3=AcO	0
5	**1e**: R^1=OH, R^2=H, R^3=OH	0
6	**1f**: R^1=OH, R^2=H, R^3=H	0

Reaction conditions: **1** (1.0 mmol), 315 mg PPh_3 (1.2 mmol, 1.2 equiv), 305 mg I_2 (1.2 mmol, 1.2 equiv), 10 cm^3 EtOAc, and rt for 1 h

[a]Isolated yields

results showed that amongst these solvents, EtOAc was the solvent of choice in terms of yields.

In Chansu, a number of bufalenes was found and isolated [7–10]. Therefore, six compounds, namely cinobufagin (**1a**), desacetylcinobufagin (**1b**), desibufogenin (**1c**), bufotaline (**1d**), desacetylbufatalin (**1e**), and bufalin (**1f**), were selected to study the effect of functional groups on the reaction (Table 3). The experimental results indicate that the epoxy structure on C14 and C15 was necessary (Table 3, entries 1, 2); otherwise, the reaction could not proceed smoothly to obtain corresponding product **3a** (Table 3, entries 4–6). While retaining the epoxy structure, the reaction of acetoxyl or hydroxyl substituted substrates on C16 could proceed smoothly. If it was a methylene without substituted by acetoxyl or hydroxyl on C16, the desired product **3a** could not form even if the epoxy structure was still existed (Table 3, entry 3). Therefore, to summarize, both epoxy structure on C14 and C15 and acetoxyl or hydroxyl on C16 was necessary for this elimination reaction.

Having established the optimized reaction conditions, we then successfully synthesized a variety of compounds **3** and the results are summarized in Table 4. Several cinobufagin analogues isolated from Chansu were chosen as subtrates [11]. All the substrates were carried out under standard conditions, and the desired products **3a**–**3h** were obtained in 87–94% yields (Table 4). These results clearly indicated that this method had good substrate applicability.

A tentative mechanism to rationalize the products **3** formation is shown in Scheme 2 [12–14]. Initially, PPh_3 reacts with I_2 to form **A**, and **A** reacts with trace amount of water in solvent to form triphenylphosphine oxide and hydrogen iodide. This is an important reason why the reaction liquid is acidic. Intermediate **A** also reacts with **1** to form intermediate **B**. Ring-opening reaction of epoxy of **B** takes place to form **C**, which is decomposed to intermediate **D**. Intermediate **D** removes a molecule of acetic acid or water to form desired product **3**.

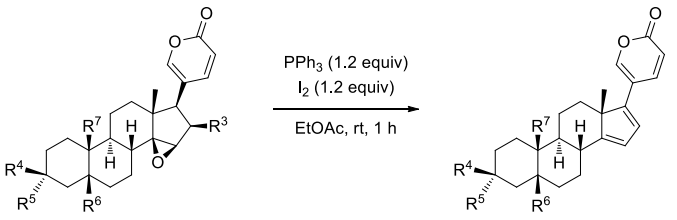
Conclusion

In conclusion, we have developed an efficient and convenient method for the synthesis of diene compounds via the reaction of cinobufagin analogues in the presence of PPh_3/I_2 in EtOAc at room temperature. The simple experimental procedure combined with the easy workup and excellent yields of products are salient features of the presented method. Furthermore, cinobufagin analogues have many biological activities and this method provides a novel strategy for the modification of these compounds.

Experimental

Melting points were measured by a WRS-1B micromelting point apparatus. NMR spectra were recorded on a Bruker AMX 500 instrument using solvent peaks as $CDCl_3$ solutions. Yields of condition screen experiment were determined by LC–MS (Waters LCMS/SQD + UPLC H-class spectrometer). HR-ESI–MS were determined on a Micromass Q-Tof Global mass spectrometer and ESI–MS were run on a Bruker Esquire 3000 Plus Spectrometer. TLC was performed on GF254 silica gel plates (Yantai Huiyou Inc., China).

Table 4 Synthesis of compounds **3**



1 **3**

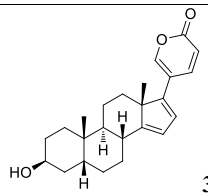
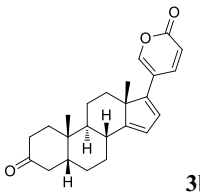
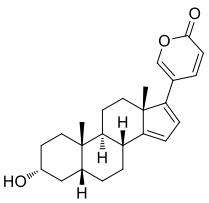
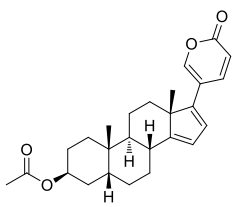
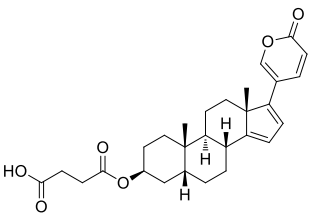
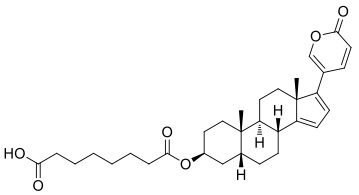
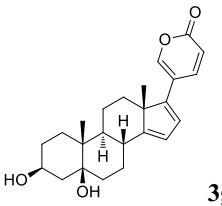
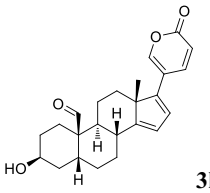
Entry	Substrate **1**	Product **3**	Yield / %[b]
1	Cinobufagin (**1a**): R^3 = AcO, R^4 = OH, R^5 = R^6 = H, R^7 = CH_3	 **3a**	92
2	Desacetylcinobufagin (**1b**): R^3 = R^4 = OH, R^5 = R^6 = H, R^7 = CH_3	**3a**	90
3	3-Oxocinobufagin (**1e**): R^3 = AcO, R^4, R^5 = O, R^6 = H, R^7 = CH_3	 **3b**	94
4	3-Oxodesacetylcinobufagin (**1f**): R^3 = OH, R^4, R^5 = O, R^6 = H, R^7 = CH_3	**3b**	87
5	3-*epi*-Cinobufagin (**1g**): R^3 = AcO, R^5 = OH, R^4 = R^6 = H, R^7 = CH_3	 **3c**	90
6	3β-Acetoxy-cinobufagin (**1h**): R^3 = R^4 = AcO, R^5 = R^6 = H, R^7 = CH_3	 **3d**	92
7	Cinobufagin-3-hemisuccinate (**1i**): R^3 = AcO, R^4 = $HOOC(CH_2)_2COO$, R^5 = R^6 = H, R^7 = CH_3		89

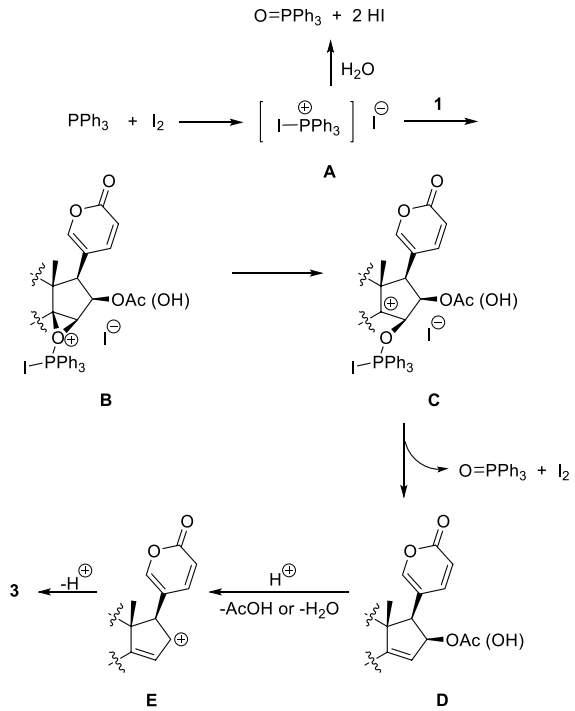
Table 4 (continued)

Entry	Substrate **1**	Product **3**	Yield / %[b]
8	Cinobufagin-3-hemisuberate (**1j**): R^3 = AcO, R^4 = $HOOC(CH_2)_6COO$, R^5 = R^6 = H, R^7 = CH_3 cinobufotalin	 **3e** **3f**	87
9	Cinobufotalin (**1k**): R^3 = AcO, R^4 = R^6 = OH, R^5 = H, R^7 = CH_3	 **3g**	87
10	19-Oxocinobufagin (**1l**): R^3 = AcO, R^4 = OH, R^5 = R^6 = H, R^7 = CHO	 **3h**	88

Reaction conditions: **1** (1.0 mmol), 315 mg PPh_3 (1.2 mmol, 1.2 equiv), 305 mg I_2 (1.2 mmol, 1.2 equiv), 10 cm^3 EtOAc, and rt for 1 h

[a]Isolated yields

Scheme 2



General procedure for the synthesis of compounds 3

A mixture of 315 mg PPh_3 (1.2 mmol, 1.2 equiv), 305 mg I_2 (1.2 mmol, 1.2 equiv), and 10 cm^3 EtOAc was stirred at rt for 30 min. Then, compound **1** (1.0 mmol) was added to the mixture, and stirred at rt for another 1 h. After completion of the reaction (TLC), 10 cm^3 EtOAc was added and the solution was washed with 20 cm^3 NaS_2O_3 (10%). Then, organic layer was washed with water (2×15 cm^3) and brine (2×15 cm^3), and was dried over anhyd. $MgSO_4$. The organic layer was concentrated under reduced pressure and the crude product was purified by column chromatography to afford the corresponding product **3**.

5-[(3*S*,5*R*,8*R*,9*S*,10*S*,13*S*)-3-Hydroxy-10,13-dimethyl-2,3,4,5,6,7,8,9,10,11,12,13-dodecahydro-1*H*-cyclopenta[*a*]phenanthren-17-yl]-2*H*-pyran-2-one (3a, $C_{24}H_{30}O_3$) Yield: 92%; light yellow solid; m.p.: 212.1–217.4 °C; ^{1}H NMR (500 MHz, DMSO-d_6): $\delta=7.61$ (dd, $J=9.8$, 2.6 Hz, 1H), 7.50 (d, $J=2.3$ Hz, 1H), 6.53 (d, $J=2.2$ Hz, 1H), 6.36 (d, $J=9.7$ Hz, 1H), 5.94 (t, $J=1.9$ Hz, 1H), 4.09–4.08 (m, 1H), 2.40–2.35 (m, 1H), 2.09–2.00 (m, 2H), 1.94 (td, $J=14.0$, 2.9 Hz, 1H), 1.83–1.79 (s, 1H), 1.73–1.63 (m, 3H), 1.57–1.48 (m, 5H), 1.43 (dd, $J=12.6$, 3.1 Hz, 1H), 1.39–1.29 (m, 3H), 1.10 (s, 3H), 1.07 (s, 3H),

1.03 (dd, J = 12.8, 4.0 Hz, 1H) ppm; ^{13}C NMR (126 MHz, DMSO-d_6): δ = 165.3, 161.4, 147.0, 145.3, 143.9, 127.0, 118.2, 116.8, 115.6, 67.0, 53.3, 44.1, 37.5, 36.5, 35.9, 35.8, 33.4, 30.4, 27.9, 26.3, 23.9, 23.8, 21.3, 20.3 ppm; MS (ESI): m/z = 367.4 ([M + H]$^+$); HRMS (ESI): m/z calcd for $C_{24}H_{31}O_3$ ([M + H]$^+$) 367.2268, found 367.2261.

5-[(5*R*,8*R*,9*S*,10*S*,13*S*)-10,13-Dimethyl-3-oxo-2,3,4,5,6,7,8,9,10,11,12,13-dodecahydro-1*H*-cyclopenta[*a*]phenanthren-17-yl]-2*H*-pyran-2-one (3b, $C_{24}H_{28}O_3$) Yield: 94%; light yellow solid; m.p.: 161.2–176.4 °C; ^{1}H NMR (500 MHz, DMSO-d_6): δ = 7.59 (dd, J = 9.8, 2.7 Hz, 1H), 7.49 (d, J = 2.3 Hz, 1H), 6.53 (d, J = 2.3 Hz, 1H), 6.35 (dd, J = 9.8, 1.0 Hz, 1H), 5.97 (t, J = 2.0 Hz, 1H), 2. 65 (t, J = 14.5 Hz, 1H), 2.47–2.41 (m, 1H), 2.24 (td, J = 14.5, 5.4 Hz, 1H), 2.16–2.10 (m, 2H), 2.06–1.98 (m, 3H), 1.90–1.84 (m, 1H), 1.82–1.77 (m, 1H), 1.73–1.65 (m, 1H), 1.65–1.57 (m, 1H), 1.57–1.50 (m, 2H), 1.46 (td, J = 14.5, 4.5 Hz, 1H), 1.41–1.36 (m, 1H), 1.12 (s, 3H), 1.12 (s, 3H), 1.10–1.06 (m, 1H) ppm; ^{13}C NMR (126 MHz, DMSO-d_6): δ = 212.6, 164.0, 161.1, 147.1, 145.4, 143.7, 126.9, 118.7, 116.8, 115.5, 53.2, 44.8, 44.1, 42.2, 37.3, 37.2, 37.1, 35.6, 35.5, 26.3, 23.3, 22.7, 21.3, 20.2 ppm; MS (ESI): m/z = 365.3 ([M + H]$^+$); HRMS (ESI): m/z calcd for $C_{24}H_{29}O_3$ ([M + H]$^+$) 365.2111, found 365.2118.

5-[(3*R*,5*R*,8*R*,9*S*,10*S*,13*S*)-3-Hydroxy-10,13-dimethyl-2,3,4,5,6,7,8,9,10,11,12,13-dodecahydro-1*H*-cyclopenta[*a*]phenanthren-17-yl]-2*H*-pyran-2-one (3c, $C_{24}H_{30}O_3$) Yield: 90%; light yellow solid; m.p.: 101.1–104.9 °C; ^{1}H NMR (500 MHz, DMSO-d_6): δ = 7.60 (dd, J = 9.8, 2.7 Hz, 1H), 7.49 (d, J = 2.4 Hz, 1H), 6.53 (d, J = 2.3 Hz, 1H), 6.36 (dd, J = 9.8, 1.1 Hz, 1H), 5.94 (t, J = 2.1 Hz, 1H), 3.66–3.60 (m, 1H), 2.39–2.34 (m, 1H), 2.07 (dt, J = 12.7, 3.0 Hz, 1H), 2.03–1.96 (m, 1H), 1.80 (dt, J = 14.2, 3.2 Hz, 1H), 1.74–1.65 (m, 6H), 1.57–1.53 (m, 2H), 1.49–1.38 (m, 4H), 1.29–1.20 (m, 1H), 1.09 (s, 3H), 1.07–1.05 (m, 1H), 1.03 (s, 3H) ppm; ^{13}C NMR (126 MHz, DMSO-d_6): δ = 165.1, 161.5, 146.9, 145.3, 144.0, 127.1, 118.3, 116.8, 115.7, 71.7, 53.3, 44.7, 42.0, 37.5, 36.3, 36.0, 35.7, 35.2, 30.5, 26.8, 23.9, 23.4, 21.1, 20.2 ppm; MS (ESI): m/z = 367.4 ([M + H]$^+$); HRMS (ESI): m/z calcd for $C_{24}H_{31}O_3$ ([M + H]$^+$) 367.2268, found 367.2272.

(3*S*,5*R*,8*R*,9*S*,10*S*,13*S*)-10,13-Dimethyl-17-(2-oxo-2*H*-pyran-5-yl)-2,3,4,5,6,7,8,9,10,11,12,13-dodecahydro-1*H*-cyclopenta[*a*]phenanthren-3-yl acetate (3d, $C_{26}H_{32}O_4$) Yield: 92%; light yellow solid; m.p.: 174.0–180.6 °C; ^{1}H NMR (500 MHz, DMSO-d_6): δ = 7.56 (dd, J = 9.8, 2.6 Hz, 1H), 7.45 (d, J = 2.3 Hz, 1H), 6.48 (d, J = 2.3 Hz, 1H), 6.30 (d, J = 9.8 Hz, 1H), 5.88 (t, J = 1.9 Hz, 1H), 4.97 (s, 1H), 2.35–2.30 (m, 1H), 2.05–2.01 (m, 1H), 1.98 (s, 3H), 1.97–1.93 (m, 1H), 1.85 (td, J = 14.2, 2.9 Hz, 1H), 1.68–1.59 (m, 3H), 1.56–1.47 (m, 3H), 1.42–1.30 (m, 5H), 1.26–1.23 (m, 1H), 1.04 (s, 3H), 1.02 (s, 3H), 0.98 (dd, J = 12.8, 4.0 Hz, 1H) ppm; ^{13}C NMR (126 MHz, DMSO-d_6): δ = 170.5, 164.8, 161.0, 146.8, 145.1, 143.7, 126.8, 118.1, 116.6, 115.4, 70.3, 53.0, 44.0, 37.2, 37.1, 35.6, 35.4, 31.0, 30.4, 26.0, 24.9, 23.7, 23.5, 21.4, 21.1, 20.0 ppm; MS (ESI): m/z = 409.4 ([M + H]$^+$); HRMS (ESI): m/z calcd for $C_{26}H_{33}O_4$ ([M + H]$^+$) 409.2373, found 409.2370.

4-[[(3*S*,5*R*,8*R*,9*S*,10*S*,13*S*)-10,13-Dimethyl-17-(2-oxo-2*H*-pyran-5-yl)-2,3,4,5,6,7,8,9,10,11,12,13-dodecahydro-1*H*-cyclopenta[*a*]phenanthren-3-yl]oxy]-4-oxobutanoic acid (3e, $C_{28}H_{34}O_6$) Yield: 89%; light yellow solid; m.p.: 162.8–166.3 °C; ^{1}H NMR (500 MHz, DMSO-d_6): δ = 7.60 (dd, J = 9.8, 2.6 Hz, 1H), 7.50 (d, J = 2.2 Hz, 1H), 6.53 (d, J = 2.3 Hz, 1H), 6.38 (dd, J = 9.8, 1.0 Hz, 1H), 5.95 (t, J = 2.0 Hz, 1H), 5.08 (s, 1H), 2.70–2.62 (m, 4H), 2.40–2.36 (m, 1H), 2.08 (dt, J = 13.0, 2.9 Hz, 1H), 2.06–1.98 (m, 1H), 1.95–1.89 (m, 1H), 1.72–1.69 (m, 2H), 1.62–1.58 (m, 1H), 1.58–1.53 (m, 2H), 1.48–1.45 (m, 2H), 1.43–1.36 (m, 4H), 1.29–1.24 (m, 2H), 1.10 (s, 3H), 1.08 (s, 3H), 1.04 (dd, J = 12.9, 4.2 Hz, 1H) ppm; ^{13}C NMR (126 MHz, DMSO-d_6): δ = 177.9, 171.6, 165.1, 161.5, 147.0, 145.4, 143.9, 127.0, 118.3, 116.8, 115.7, 71.2, 53.3, 44.2, 37.4, 37.3, 35.9, 35.5, 31.2, 30.5, 29.5, 29.2, 26.2, 25.1, 23.9, 23.7, 21.3, 20.3 ppm; MS (ESI): m/z = 467.2 ([M + H]$^+$); HRMS (ESI): m/z calcd for $C_{28}H_{35}O_6$ ([M + H]$^+$) 467.2428, found 467.2436.

8-[[(3*S*,5*R*,8*R*,9*S*,10*S*,13*S*)-10,13-Dimethyl-17-(2-oxo-2*H*-pyran-5-yl)-2,3,4,5,6,7,8,9,10,11,12,13-dodecahydro-1*H*-cyclopenta[*a*]phenanthren-3-yl]oxy]-8-oxooctanoic acid (3f, $C_{32}H_{42}O_6$) Yield: 87%; light yellow solid; m.p.: 107.9-115.7 °C; ^{1}H NMR (500 MHz, DMSO-d_6): δ = 7.60 (dd, J = 9.8, 2.6 Hz, 1H), 7.48 (d, J = 2.2 Hz, 1H), 6.52 (d, J = 2.2 Hz, 1H), 6.36 (d, J = 9.8 Hz, 1H), 5.93 (t, J = 1.8 Hz, 1H), 5.03 (s, 1H), 2.39–2.35 (m, 1H), 2.34–2.27 (m, 6H), 2.07 (dt, J = 13.0, 3.0 Hz, 1H), 2.02–1.97 (m, 1H), 1.90 (td, J = 15.0, 3.0 Hz, 1H), 1.72–1.67 (m, 2H), 1.63–1.58 (m, 6H), 1.45–1.39 (m, 4H), 1.36–1.31 (m, 6H), 1.08 (s, 3H), 1.06 (s, 3H), 1.02 (dd, J = 12.8, 4.0 Hz, 1H), 0.95–0.92 (m, 1H) ppm; ^{13}C NMR (126 MHz, DMSO-d_6): δ = 179.4, 173.4, 165.0, 161.5, 146.9, 145.3, 143.9, 127.0, 118.3, 116.7, 115.7, 70.3, 53.2, 44.2, 37.3 (2C), 35.8, 35.5, 34.7, 34.0, 31.2, 30.6, 28.8, 28.8, 26.1, 25.1, 24.9, 24.6, 23.9, 23.6, 21.2, 20.2 ppm; MS (ESI): m/z = 523.2 ([M + H]$^+$); HRMS (ESI): m/z calcd for $C_{32}H_{43}O_6$ ([M + H]$^+$) 523.3054, found 523.3059.

5-[(3*S*,5*S*,8*R*,9*S*,10*R*,13*S*)-3,5-Dihydroxy-10,13-dimethyl-2,3,4,5,6,7,8,9,10,11,12,13-dodecahydro-1*H*-cyclopenta[*a*]phenanthren-17-yl]-2*H*-pyran-2-one (3 g, $C_{24}H_{30}O_6$) Yield: 87%; light yellow solid; m.p.: 198.5-200.8 °C; ^{1}H NMR (500 MHz, DMSO-d_6): δ = 7.94 (dd, J = 9.8, 2.5 Hz, 1H), 7.90–7.83 (m, 1H), 6.76 (d,

$J = 1.9$ Hz, 1H), 6.40 (d, $J = 9.8$ Hz, 1H), 5.95 (s, 1H), 5.21 (d, $J = 3.9$ Hz, 1H), 4.84 (s, 1H), 4.03–3.90 (m, 1H), 2.35 (t, $J = 11.0$ Hz, 1H), 2.21 (d, $J = 12.8$ Hz, 1H), 2.03 (dd, $J = 14.4$, 2.4 Hz, 1H), 1.77–1.71 (m, 3H), 1.57–1.50 (m, 2H), 1.44–1.33 (m, 4H), 1.31–1.20 (m, 3H), 1.06 (s, 3H), 0.97 (s, 3H), 0.85 (td, $J = 12.8$, 3.4 Hz, 1H) ppm; ^{13}C NMR (126 MHz, DMSO-d_6): $\delta = 164.3$, 160.3, 146.9, 145.9, 144.4, 126.7, 118.3, 116.2, 114.6, 73.6, 66.4, 52.4, 46.9, 40.7, 36.8, 36.0, 34.5, 34.3, 27.3, 25.5, 25.4, 21.3, 19.4, 17.0 ppm; MS (ESI): $m/z = 383.2$ ($[M+H]^+$); HRMS (ESI): m/z calcd for $C_{24}H_{31}O_6$ ($[M+H]^+$) 383.2217, found 383.2212.

(3*S*,5*R*,8*R*,9*S*,10*R*,13*S*)-3-Hydroxy-13-methyl-17-(2-oxo-2*H*-pyran-5-yl)-2,3,4,5,6,7,8,9,10,11,12,13-dodecahydro-1*H*-cyclopenta[*a*]phenanthrene-10-carbaldehyde (3 h, $C_{24}H_{28}O_4$) Yield: 88%; light yellow solid; m.p.: 186.9–193.1 °C; ^{1}H NMR (500 MHz, DMSO-d_6): $\delta = 9.59$ (d, $J = 2.0$ Hz, 1H), 7.60 (dd, $J = 9.8$, 2.7 Hz, 1H), 7.51 (d, $J = 2.2$ Hz, 1H), 6.53 (d, $J = 2.2$ Hz, 1H), 6.37 (dd, $J = 9.8$, 1.0 Hz, 1H), 5.95 (t, $J = 2.0$ Hz, 1H), 4.15–4.14 (m, 1H), 2.70–2.67 (m, 1H), 2.41–2.39 (m, 1H), 2.14–2.03 (m, 2H), 1.91 (td, $J = 14.2$, 2.9 Hz, 1H), 1.86 (td, $J = 14.2$, 3.9 Hz, 1H), 1.80–1.76 (m, 1H), 1.75–1.71 (m, 1H), 1.67–1.55 (m, 6H), 1.53–1.50 (m, 1H), 1.48–1.42 (m, 2H), 1.19 (s, 3H), 0.99 (td, $J = 13.1$, 3.8 Hz, 1H) ppm; ^{13}C NMR (126 MHz, DMSO-d_6): $\delta = 206.0$, 164.2, 161.3, 147.2, 145.5, 143.8, 126.9, 118.4, 116.9, 115.6, 65.7, 53.4, 51.4, 42.3, 37.5, 35.9, 32.5, 29.3, 27.9, 26.4, 23.7, 21.6, 21.0, 20.0 ppm; MS (ESI): $m/z = 381.2$ ($[M+H]^+$); HRMS (ESI): m/z calcd for $C_{24}H_{29}O_4$ ($[M+H]^+$) 381.2060, found 381.2055.

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