Synthesis of non-enolizable α- and β-peltatins

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The preparation of non-enolizable derivatives of both α- and β-peltatins via reaction of the lactone enolates of the corresponding TBDMS derivatives with CH_3I and hexachloroethane is described. These derivatives are considerably less toxic than the precursor peltatins but show, at best, only marginal activity in in vivo tests against P 388 leukemia.

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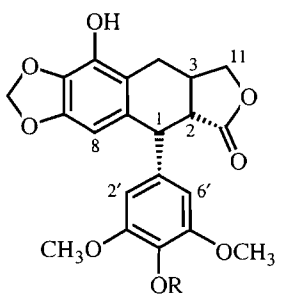
On décrit la préparation de dérivés non-énolisables des α- ainsi que β-peltatines par l'intermédiaire d'une réaction des énolates de la lactone des dérivés TBDMS correspondants avec le CH_3I et l'hexachloroéthane. Ces dérivés sont beaucoup moins toxiques que les peltatines qui leur donnent naissance; lors d'essais in vivo, ils ne présentent toutefois qu'une activité marginale contre la leucémie P388.

[Traduit par la rédaction]

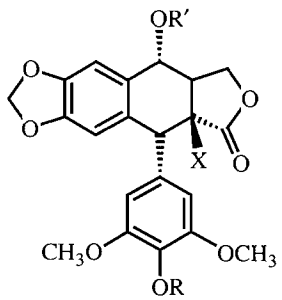
Introduction

α- and β-Peltatin **1** and **2** are members of the lignan family of compounds (1). These compounds were first isolated and characterized by Hartwell in 1956 from North American podophyllin (*podophyllum peltatum L*) (2). Like the closely related podophyllotoxin, **3**, both peltatins show considerable antineoplastic activity. Clinical use of these compounds was briefly investigated in the 1950's by Greenspan but abandoned due to their high toxicity, unfavourable therapeutic ratios, and their inability to effect more than short-term tumor remissions (3).

In the case of podophyllotoxin it was eventually possible to obtain clinically important derivatives such as Etoposide and Teniposide. No therapeutically useful derivatives have been prepared from either **1** or **2** (1).



1 R = H
2 R = CH_3



3 R = CH_3, R′ = H
X = H

In 1986 we reported the preparation of several non-enolizable podophyllotoxin derivatives, that is, compounds in which the hydrogen α to the lactone carbonyl group had been replaced by substituents such as CH_3, Cl, Br, and SCH_3 (4). In general, those compounds in which the podophyllotoxin geometry (*trans*-fused lactone, structure **3**) had been retained showed comparable or better in vivo activity against P 388 (T/C = 167 for the chloro derivative (X = Cl) at 40 mg/kg vs. 110 for podophyllotoxin) compared to podophyllotoxin itself. In addition, these derivatives were considerably less toxic. Not surprisingly, those compounds that had the picropodophyllotoxin geometry (*cis*-fused lactone) were inactive and nontoxic at the levels tested. We therefore decided to prepare the analogous derivatives of both α- and β-peltatins to see if a reduction in toxicity might allow one to observe useful anticancer activity.

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Results and discussion

The peltatins were isolated from commercially available podophyllin resin. We found that further separation of the crude chromatography fractions containing both peltatins was facilitated by conversion of the phenolic hydroxyl groups to TBDMS ethers. Using this approach, we were able to isolate the TBDMS derivatives **4** and **5**. The amounts obtained are equivalent to a recovery of 8.9 and 7.6% of **1** and **2** from the commercial resin. These compare favourably with the 6.4 and 6.0% yield of these compounds obtained by Hartwell and Detty (2*b*). Further details of the isolation procedure are given in the experimental section.

The protection of the phenolic groups in **1** and **2** was also useful for the subsequent enolate formation. This proceeded smoothly for both **4** and **5** with LDA in THF at −78°C. Protonation of the enolates of **4** and **5** with H_2O at −78°C resulted in their regeneration accompanied by the *cis*-fused isomers **6** and **7**, respectively, in roughly 1:3 ratios. These ratios can be compared with the 45:55 podo- to picropodophyllotoxin ratio obtained upon protonation of the enolate of the THP of podophyllotoxin with acetic acid (5).

Reaction of the enolate of **4** with CH_3I afforded the 2-methyl *trans*-lactone **8** and the isomeric *cis*-fused lactone **9** in 38 and 56% isolated yields, respectively. Similar reaction of the enolate derived from **5** gave the methylated products **10** and **11** in 52 and 46% yields. Chlorination of these enolates with hexachloroethane was considerably more selective. Thus, the enolate of **4** gave 92% of the *trans*-lactone **12** and only 4% of the *cis*-lactone **13**, while 83% of **14** and 6% of **15** were obtained from the enolate of **5**. Similar trends in the ratios of *cis*- to *trans*-fused lactone products were observed in the podophyllotoxin series (4).

The structure and stereochemistry of the 2-substituted products were readily assigned on the basis of their ^1H NMR spectra. The relevant data are summarized in Table 1 and compared with the unsubstituted and isomerized peltatins.

Substitution at C-2 was clearly indicated by the presence of a sharp singlet due to the remaining doubly benzylic hydrogen at C-1. This signal was found in the δ = 4.2 range

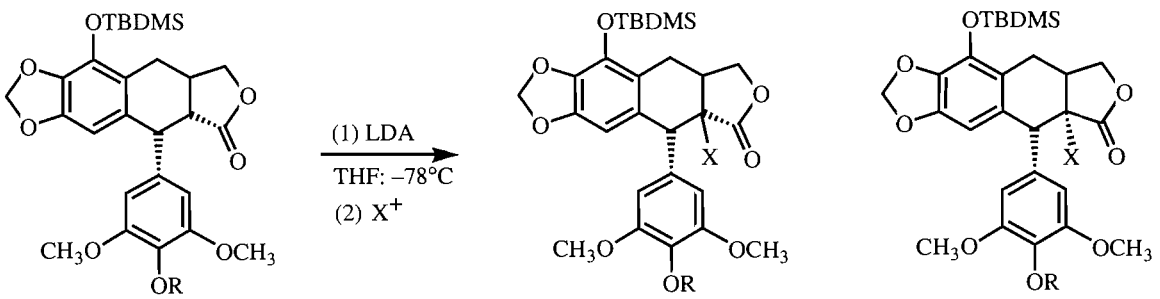
TABLE 1. Key chemical shifts and coupling constants in the TBDMS protected α- and β-peltatins

Compound	γ-Lactone fusion	Chemical shifts (δ) H-1	H-8	C-2′,6′	Coupling constant H3–H11 (Hz)
4	*trans*	4.57	6.27	108.4	10.1
6	*cis*	4.35	6.32	105.0	3.4
5	*trans*	4.59	6.26	108.2	10.2
7	*cis*	4.36	6.31	104.9	3.4
8	*trans*	4.22	6.23	109.1	11.0
9	*cis*	4.18	6.35	106.9	2.9
10	*trans*	4.24	6.23	108.8	11.3
11	*cis*	4.20	6.34	106.8	3.1
12	*trans*	4.73	6.26	109.1	9.7
13	*cis*	4.48	6.36	—	0
14	*trans*	4.75	6.25	108.9	9.7
15	*cis*	4.51	6.34	—	0

for the methylated compounds **8–11** and δ 4.5–4.7 for the chlorinated derivatives **12–15**.

The *cis*- or *trans*-fusion of the lactone ring was assigned on the basis of J_{H3-11}. As has been noted for the corresponding podophyllotoxins (4), *trans* ring fusion results in >9 Hz coupling constants while the *cis*-fused isomers have J_{H3-11} < 3. Several other consistent trends can be noted from Table 1. The C-1 proton of the isomer having the *trans*-fused lactone is always shifted further downfield than that of the corresponding *cis* isomer. The opposite effect is observed for H-8. In the latter case the aromatic ring at C-1 is essentially perpendicular to the other rings when the lactone fusion is *trans* and thus should have little anisotropy effect on H-8. In the case of the *cis*-fused lactones the half-chair conformation places H-8 in an aromatic ring deshielding zone. Finally, the ^{13}C chemical shifts for C-2′, C-6′ in the *trans*-fused isomers were typically found in the δ = 108–109 ppm range; these were consistently deshielded relative to the corresponding *cis* isomer by at least 2 ppm.

Desilylation of the compounds **8–15** was carried out with either 4 equivalents of tetrabutylammonium fluoride (TBAF) in THF at 0°C for 1 min followed by quenching with saturated NH_4Cl solution or, in the case of the more sensitive chloro compounds, with the same reagents in acetic acid. The proton spectra of the desilylated products were consistent with the structure assignments. See Table 1 and experimental section. Again the size of the J_{H3-11} was crucial in confirming the stereochemistry of the lactone ring junction.



4 R = CH_3
5 R = TBDMS

8 R = CH_3, X = CH_3
10 R = TBDMS, X = CH_3
12 R = CH_3, X = Cl
14 R = TBDMS, X = Cl

6 R = TBDMS, X = H
7 R = CH_3, X = H
9 R = CH_3, X = CH_3
11 R = TBDMS, X = CH_3
13 R = CH_3, X = Cl
15 R = TBDMS, X = Cl

Biological screening results

Testing against leukemia P 388 was performed on the peltatins, all four methylated isomers, and the two *trans*-fused chloro isomers at the Anti-Tumor Division of Bristol Laboratories using the following protocol (6). Ascitic fluid containing 10^6 cancer cells was implanted interperitoneally in female CDF1 mice (4 mice per test group). Treatment began 24 h after implant. The results are given in Table 2.

Under these conditions, α- and β-peltatins are toxic (the majority of the test animals died within 5 days of the administration of the compounds, compared to the 9-day life expectancy of the control group) at levels of 4 and 16 mg/kg. The *cis*-2-methyl derivatives of both α- and β-peltatins were inactive at dosages up to 64 mg/kg. All of the *trans*-fused derivatives were considerably less toxic than the parent peltatins, but only the *trans*-2-methyl-β-peltatin showed any marginal anti-cancer activity (T/C = 133 at 64 mg/kg) at the levels tested. The possibility that some of these com-

TABLE 2. In vivo effect of peltatins on P388 leukemia

Compound	Dosage (mg/kg)	T/C[a]
α-Peltatin **4**	4	toxic
	0.4	110
2-Methyl-α-peltatin (*trans*) **8**	4	100
	16	111
	64	128
2-Methyl-α-peltatin (*cis*) **9**	16	100
	64	106
2-Chloro-α-peltatin **12**	4	110
	8	100
β-Peltatin **5**	4	toxic
	1	125
2-Methyl-β-peltatin (*trans*)	4	100
	16	122
	64	133
2-Methyl-β-peltatin (*cis*)		
	16	100
	64	100
2-Chloro-β-peltatin	0.8	100
	1.6	90
	3.2	90
Etoposide[b]	30	270

[a]Lifespan, after implantation of cancer cells of treated (T) animals/that of the control (c) group × 100.

[b]Determined at Bristol Laboratories using the standard procedure (6).

pounds might show improved biological activity after glycosylation remains to be investigated.

Experimental section

General comments

^1H and ^{13}C NMR spectra were recorded on Varian XL200 and Varian FT-80 spectrometers, respectively, using $CDCl_3$ as solvent and TMS as internal standard. Melting points were determined on a Gallenkamp apparatus and are uncorrected. Optical rotations were recorded as $CHCl_3$ solution (c = 1.0) unless otherwise indicated, using a Perkin Elmer 241 polarimeter. Preparative thin-layer chromatography (PTLC) was carried out on glass plates coated with a 1.0 mm layer of Kieselgel 60GF 254. High-performance liquid chromatography was carried out using PrePak-500 silica gel cartridges on a Waters 500 instrument. Typical work-up refers to quenching the reaction mixture with saturated NH_4Cl solution, several extractions with either ethyl acetate or CH_2Cl_2, drying, and evaporating the organic solvents on a rotatory evaporator. All yields refer to isolated, chromatographically homogeneous products.

Preparation of the TBDMS derivatives of α- and β-peltatin from crude podophyllin resin

Podophyllin resin (10.2 g, U.S. Biochemicals) was stirred overnight with 50 mL of $CHCl_3$. The solution was filtered and the residue was similarly treated two more times. Evaporation of the $CHCl_3$ afforded 5.86 g of yellow-brown foam.

The chloroform soluble material (6.0 g) was further fractionated by placing it on 30 g of silica and eluting with 200 mL each of (1) hexanes; (2) 20% CH_2Cl_2 – 80% hexanes; (3) 40% CH_2Cl_2 – 60% hexanes; (4) 60% CH_2Cl_2 – 40% hexanes; (5) 80% CH_2Cl_2 – 20% hexanes; (6) CH_2Cl_2; (7)–(10) fractions contain 2, 4, 6, and 8% ethyl acetate in CH_2Cl_2. The last 5 fractions were combined and evaporated to yield 3.5 g of crude peltatin mixture.

The above mixture (2.38 g), imidazole (16.1 g), and *tert*-butyldimethylsilyl chloride (1.71 g) were combined in 11 mL of DMF, heated overnight at 50°C, and then worked up in the usual manner to yield 3.30 g of a yellow-brown foam. Purification by HPLC (1:9 ethyl acetate – hexanes) afforded 987 mg (14%) of the bis-TBDMS of α-peltatin and 630 mg (9%) of the TBDMS derivative of β-peltatin after recrystallization from hexanes.

*α-Peltatin-bis-TBDMS (**4**):* mp 172–174°C; $[\alpha]_D$ = −106; ^1H NMR, δ (ppm): 0.10 (s, 6H), 0.24 (s, 3H), 0.29 (s, 3H), 0.99 (s, 9H), 1.03 (s, 9H), 2.41 (dd, J = 11.2, 16.0 Hz, 1H), 2.68–2.72 (m, 2H), 3.10–3.24 (m, 1H), 3.68 (s, 6H), 3.92 (dd, J = 8.9, 10.1 Hz, 1H), 4.46 (dd, J = 6.2, 8.4 Hz, 1H), 4.56 (d, J = 3.4 Hz, 1H), 5.89 (d, J = 1.4 Hz, 1H), 5.91 (d, J = 1.4 Hz, 1H), 6.26 (s, 1H), 6.30 (s, 2H); ^{13}C NMR, δ (ppm): −4.56 (q), −4.04 (q), 18.67 (s), 18.7 (s), 25.8 (q), 25.9 (q), 28.2 (t), 32.3 (d), 43.8 (d), 47.4 (d), 55.9 (q), 72.4 (t), 100.8 (t), 104.3 (d), 108.4 (d), 122.17 (s), 132.0 (s), 133.2 (s), 133.4 (s), 135.5 (s), 136.5 (s), 147.5 (s), 150.8 (s), 175.1 (s). Anal. calcd. for $C_{33}H_{48}O_8Si_2$: C 63.02, H 7.69; found: C 62.93, H 8.05.

*β-Peltatin TBDMS (**5**):* mp 172–174°C; $[\alpha]_D$ = −106; ^1H NMR, δ (ppm): 0.24 (s, 3H), 0.29 (s, 3H), 1.03 (s, 9H), 2.42 (dd, J = 11.6, 15.8 Hz, 1H), 2.5–2.7 (m, 2H), 3.18 (dd, J = 4.1, 15.5 Hz, 1H), 3.75 (s, 6H), 3.80 (s, 3H), 3.948 (dd, J = 8.8, 10.2 Hz, 1H), 4.49 (dd, J = 6.3, 8.5 Hz, 1H), 4.596 (d, J = 3.8 Hz, 1H), 5.90 (d, J = 1.4 Hz, 1H), 5.91 (d, J = 1.4 Hz, 1H), 6.27 (s, 1H), 6.35 (s, 2H); ^{13}C NMR, δ (ppm): −4.04 (q), 18.6 (s), 25.9 (q), 28.1 (t), 32.4 (d), 43.9 (d), 47.3 (d), 56.2 (q), 60.7 (q), 72.4 (t), 100.9 (t), 104.2 (d), 108.2 (d), 122.1 (s), 131.6 (s), 135.6 (s), 136.4 (s), 136.5 (s), 137.0 (s), 147.6 (s), 152.5 (s), 175.0 (s). Anal. calcd. for $C_{28}H_{36}O_8Si$: C 63.61, H 6.86; found: C 63.47, H 7.00.

*Preparation of the lactone enolates derived from **4** and **5** and subsequent reaction with electrophiles*

Representative procedure

TBDMS derivative **4** (510 mg, 0.81 mmol) was dissolved in 5 mL of dry THF and added slowly to a −78°C solution of 0.85 mmol of LDA in 10 mL of dry THF. The solution (bluish color) was stirred for 15 min at −78°C and 0.5 mL of CH_3I was added. The reaction mixture was allowed to warm to room temperature and stirred for a further 10–20 h. Usual work-up afforded a crude product that was separated via preparative TLC (1:7 ethyl acetate – hexane) to afford 200 mg (38%) of the *trans*-fused, less polar isomer **8** and 292 mg (56%) of the methylated *cis*-fused lactone **9**.

*Isomer **8**:* mp 78–83°C; $[\alpha]_D$ = −102; ^1H NMR, δ (ppm): 0.10 (s, 6H), 0.24 (s, 3H), 0.27 (s, 3H), 0.98 (s, 9H), 1.03 (s, 9H), 1.22 (s, 3H), 2.36 (dd, J = 12.6, 16.2 Hz, 1H), 2.69–2.92 (m, 1H), 3.01 (dd, J = 5.3, 16.3 Hz, 1H), 3.68 (s, 6H), 3.98 (dd, J = 8.4, 11.0 Hz, 1H), 4.22 (s, 1H), 4.28–4.40 (m, 1H), 5.90 (s, 2H), 6.23 (s, 1H), 6.30 (s, 2H); ^{13}C NMR, δ (ppm): −4.55 (q), −4.06 (q), 16.4 (q), 18.7 (s), 23.1 (t), 25.8 (q), 25.9 (q), 34.3 (d), 46.1 (s), 52.3 (d), 56.0 (q), 70.1 (t), 100.8 (t), 105.2 (d), 109.1 (d), 120.9 (s), 131.3 (s), 133.4 (s), 135.4 (s), 136.3 (s), 147.8 (s), 150.6 (s), 178.6 (s). Anal. calcd. for $C_{34}H_{50}O_8Si_2$: C 63.52, H 7.84; found: C 63.46, H 7.81.

2-Methylated cis*-fused lactone (**9**):* mp 69–75°C; $[\alpha]_D$ = −106; ^1H NMR, δ (ppm): 0.11 (s, 6H), 0.21 (s, 3H), 0.24 (s, 3H), 0.99 (s, 9H), 1.03 (s, 9H), 1.19 (s, 3H), 2.63–2.75 (m, 1H), 2.99–3.09 (m, 2H), 3.71 (s, 6H), 3.99 (dd, J = 2.9, 9.1 Hz, 1H), 4.16 (s, 1H), 4.48 (dd, J = 6.9, 9.1 Hz, 1H), 5.85 (s, 2H), 6.30 (s, 2H), 6.35 (s, 1H); ^{13}C NMR, δ (ppm): −4.58 (q), −4.17 (q), 18.5 (s), 18.7 (s), 22.9 (q), 25.8 (q), 27.0 (t), 39.7 (q), 47.0 (s), 51.3 (d), 55.8 (q), 73.5 (t), 100.6 (t), 103.6 (d), 106.9 (d), 119.6 (s), 132.3 (s), 132.6 (s), 135.7 (s), 136.29 (s), 147.3 (s), 151.2 (s), 169.3 (s), 181.4 (s). Anal. calcd. for $C_{34}H_{50}O_8Si_2$: C 63.52, H 7.84; found: C 63.45, H 7.95.

*Preparation of the 2-methylated derivatives of β-peltatin (**10** and **11**)*

From 544 mg (1.03 mmol) of **5** was obtained 292 mg (52%) of the methylated *trans*-fused isomer **10** and 258 mg (46%) of the *cis*-fused product **11**.

*Compound **10**:* mp 76–82°C; $[\alpha]_D$ = −107; ^1H NMR, δ (ppm): 0.25 (s, 3H), 0.28 (s, 3H), 1.04 (s, 9H), 1.24 (s, 3H), 2.38 (dd, J = 12.4, 16.2 Hz, 1H), 2.70–2.92 (m, 1H), 3.04 (dd, J = 5.4, 16.4 Hz, 1H), 3.75 (s, 6H), 3.80 (s, 3H), 4.01 (dd, J = 8.6, 11.3

Hz, 1H), 4.24 (s, 1H), 4.37 (dd, J = 7.4, 8.2 Hz, 1H), 5.90 (s, 2H), 6.23 (s, 1H), 6.35 (s, 2H); ^{13}C NMR, δ (ppm): −4.1 (q), 16.4 (q), 18.6 (s), 23.1 (t), 25.9 (q), 34.4 (d), 45.9 (s), 52.4 (d), 56.6 (q), 60.7 (q), 70.1 (t), 100.9 (t), 105.1 (d), 108.8 (d), 120.9 (s), 130.9 (s), 135.5 (s), 136.4 (s), 136.5 (s), 136.9 (s), 147.8 (s), 152.2 (s), 178.5 (s). Anal. calcd. for $C_{25}H_{38}O_8Si$: C 64.18, H 7.06; found: C 59.19, H 6.47. The low carbon and hydrogen values appear to be due to occluded halocarbons, which were difficult to remove from the foamy solid even under high vacuum.

Compound 11: foam, mp 71–75°C; $[\alpha]_D$ = −107; ^{1}H NMR, δ (ppm): 0.21 (s, 3H), 0.24 (s, 3H), 1.03 (s, 9H), 1.23 (s, 3H), 2.64–2.77 (m, 1H), 3.02 (d, J = 5.4 Hz, 2H), 3.79 (s, 6H), 3.81 (s, 3H), 4.01 (dd, J = 3.1, 9.3 Hz, 4.21 (s, 1H), 4.51 (dd, J = 6.9, 9.3 Hz, 1H), 5.85 (s, 2H), 6.35 (s, 3H); ^{13}C NMR, δ (ppm): −4.17 (q), 18.5 (s), 22.8 (q), 25.9 (q), 26.9 (t), 39.6 (d), 47.0 (s), 51.4 (d), 56.1 (q), 60.8 (q), 73.4 (t), 100.7 (t), 103.4 (d), 106.8 (d), 119.6 (s), 132.2 (s), 135.5 (s), 135.8 (s), 136.3 (s), 137.0 (s), 147.4 (s), 152.9 (s), 181.2 (s). Anal. calcd. for $C_{29}H_{38}O_8Si$: C 64.18, H 7.06; found: C 61.19, H 6.57. The low C and H values appear to be due to occluded halocarbon solvents, which could not be removed from the foamy solid even after prolonged high vacuum.

*Reaction of the enolates of **4** and **5** with hexachloroethane*

The enolate prepared as above from 426 mg (0.68 mmol) of **4** was allowed to react with 1.29 (5.5 mmol) of hexachloroethane, first at −78°C, then at room temperature for 22 h. Usual work-up and purification by PTLC (1:6 ethyl acetate – hexanes) afforded the 2-chloro-*trans* derivative **12** (414 mg, 92%) and 2-chloro-*cis* product **13** (19 mg, 4%).

*2-Chloro-*trans *isomer **12**:* mp 79–86°C; ^{1}H NMR, δ (ppm): 0.10 (s, 6H), 0.26 (s, 3H), 0.29 (s, 3H), 0.98 (s, 9H), 1.04 (s, 9H), 2.65 (dd, J = 10.9, 15.7 Hz, 1H), 2.82–3.00 (m, 1H), 3.08 (dd, J = 5.6, 15.6 Hz, 1H), 3.68 (s, 6H), 4.16 (dd, J = 8.5, 9.7 Hz, 1H), 4.40 (dd, J = 6.7, 8.5 Hz, 1H), 4.73 (s, 1H), 5.90 (d, J = 1.4 Hz, 1H), 5.93 (d, J = 1.4 Hz, 1H), 6.26 (s, 1H), 6.34 (s, 2H); ^{13}C NMR, δ (ppm): −4.54 (q), −4.02 (q), 18.7 (s), 23.5 (t), 25.8 (q), 25.9 (q), 36.7 (d), 53.3 (d), 56.0 (q), 70.6 (t), 71.5 (s), 101.0 (t), 104.6 (d), 109.1 (d), 120.3 (s), 129.5 (s), 131.0 (s), 134.2 (s), 135.8 (s), 136.3 (s), 148.0 (s), 150.9 (s), 170.9 (s). Anal. calcd. for $C_{33}H_{47}O_8Si_2Cl$: C 59.75, H 7.14; found: C 59.21, H 7.38.

*2-Chloro-*cis *isomer **13**:* foam; ^{1}H NMR, δ (ppm): 0.115 (s, 6H), 0.207 (s, 3H), 0.235 (s, 3H), 0.994 (s, 9H), 1.014 (s, 9H), 2.95–3.40 (m, 3H), 3.725 (s, 6H), 4.171 (d, J = 9.0 Hz, 1H), 4.475 (s, 1H), 4.738 (dd, J = 4.9, 8.9 Hz, 1H), 5.881 (s, 2H), 6.358 (s, 1H), 6.421 (s, 2H). This material was not characterized further.

Preparation of the 2-chloro derivatives of TBDMS-protected β-peltatin

The lactone enolate derived from 465 mg (0.88 mmol) of **5** was reacted with excess (1.63 g) of hexachloroethane as above. Work-up and purification (PTLC, 1:5 ethyl acetate – hexanes) afforded 409 mg (83%) of the *trans*-2-chloro isomer **14** and 27 mg (60%) of the *cis* isomer **15**.

trans-*2-Chloro isomer **14**:* mp 212–215°C; $[\alpha]_D$ (c = 1.0) = −133; ^{1}H NMR, δ (ppm): 0.26 (s, 3H), 0.29 (s, 3H), 1.04 (s, 9H), 2.67 (dd, J = 11.0, 15.6 Hz, 1H), 2.80–3.04 (m, 1H), 3.10 (dd, J = 5.5, 15.7 Hz, 1H), 3.75 (s, 6H), 3.80 (s, 3H), 4.19 (dd, J = 8.7, 9.7 Hz, 1H), 4.43 (dd, J = 6.6, 8.4 Hz, 1H), 4.75 (s, 1H), 5.91 (d, J = 1.4 Hz, 1H), 5.93 (d, J = 1.4 Hz, 1H), 6.25 (s, 1H), 6.39 (s, 2H); ^{13}C NMR, δ (ppm): −4.03 (q), 18.6 (s), 23.4 (t), 25.9 (q), 36.8 (d), 53.4 (d), 56.3 (q), 60.7 (q), 70.6 (t), 71.2 (s), 101.0 (t), 104.5 (d), 108.9 (d), 120.3 (s), 129.1 (s), 134.1 (s), 135.9 (s), 136.3 (s), 137.6 (s), 148.0 (s), 152.6 (s), 163.9 (s), 170.9 (s). Anal. calcd. for $C_{28}H_{35}O_8SiCl$: C 59.72, H 6.26; found: C 59.78, H 6.36.

cis-*2-Chloro derivative **15**:* foam; ^{1}H NMR, δ (ppm): 0.21 (s, 3H), 0.24 (s, 3H), 1.02 (s, 9H), 2.97–3.36 (m, 3H), 3.80 (s, 6H), 3.83 (s, 3H), 4.18 (d, J = 9.0 Hz, 1H), 4.51 (s, 1H), 4.75 (dd, J = 5.2, 9.0 Hz, 1H), 4.86 (d, J = 1.4 Hz, 1H), 5.88 (d, J = 1.4 Hz, 1H), 6.35 (s, 1H), 6.47 (s, 2H); M^+ = 562 (calcd. for $C_{28}H_{35}O_8Si$; ^{35}Cl 562. This material was not further characterized.

Desilylation experiments. Method A. Typical procedure

To a solution of 575 mg (0.90 mmol) of **9** in 5 mL of dry THF at 0°C was added 4 equivalents of TBAF (3.6 mL, 1.0 M in THF). The solution was stirred for 1 min and then quenched with saturated NH_3Cl solution. Usual work-up followed by PTLC separation (2:1 ethyl acetate – hexane) afforded 319 mg (86%) of 2-methyl-α-peltatin (*cis*) as fine beige needles, mp 108–111°C; $[\alpha]_D$ (CH_3OH, c = 1.0) = −104; ^{1}H NMR, δ (ppm): 1.20 (s, 3H), 2.81–3.25 (m, 3H), 3.79 (s, 6H), 4.09 (dd, J = 2.2, 9.4 Hz, 1H), 4.14 (s, 1H), 4.58 (dd, J = 6.2, 9.2 Hz, 1H), 5.62 (s, 2H), 5.84 (d, J = 1.0 Hz, 1H), 5.85 (d, J = 1.2 Hz, 1H), 6.29 (s, 1H), 6.54 (s, 2H). Anal. calcd. for $C_{22}H_{22}O_8$: C 63.76, H 5.35; found: C 64.08, H 4.92.

*2-Methyl-α-peltatin (*trans*)*

Treatment of 220 mg (0.34 mmol) of **8** with equivalents of TBAF according to method A above afforded 100 mg (71%) of 2-methyl-α-peltanin (*trans*) after PTLC (2:1 ethyl acetate – hexanes), mp 228–233°C; $[\alpha]_D$ (CH_3OH, c = 0.97) = −123; ^{1}H NMR, δ (ppm): 1.25 (s, 3H), 2.46 (dd, J = 12.4, 16.2 Hz, 1H), 2.70–2.96 (m, 1H), 3.08 (dd, J = 5.4, 16.2 Hz, 1H), 4.25 (s, 1H), 4.30–4.40 (m, 1H), 5.37 (s, 1H), 5.44 (s, 1H), 5.94 (s, 2H), 6.36 (s, 2H). Anal. calcd. for $C_{22}H_{22}O_8$: C 63.76, H 5.35; found: C 63.48, H 5.28.

*2-Methyl-β-peltatin (*trans*)*

Prepared in 75% yield from 169 mg (0.31 mmol) of **10** and 2 equivalents of TBAF following procedure A, mp 242–244°C; ^{1}H NMR, δ (ppm): 1.26 (s, 3H), 2.48 (dd, J = 12.4, 16.4 Hz, 1H), 2.75–2.98 (m, 1H), 3.08 (dd, J = 5.3, 16.1 Hz, 1H), 3.75 (s, 6H), 3.801 (s, 3H), 4.02 (dd, J = 8.7, 11.3 Hz, 1H), 4.26 (s, 1H), 4.31–4.45 (m, 1H), 5.48 (bs, 1H), 5.92 (s, 1H), 6.19 (s, 1H), 6.36 (s, 1H). Anal. calcd. for $C_{23}H_{24}O_8$: C 64.48, H 5.65; found: C 64.59, H 5.82.

*2-Methyl-β-peltatin (*cis*)*

This compound was obtained in 67% yield as yellowish prisms, mp 127–130°C (from CH_2Cl_2–hexanes) from **11** following the desilylation procedure A; $[\alpha]_D$ ($CHCl_3$) = −113; ^{1}H NMR, δ (ppm): 1.24 (s, 3H), 2.67–3.11 (m, 3H), 3.79 (s, 6H), 3.82 (s, 3H), 4.08 (dd, J = 2.8, 9.4 Hz, 1H), 4.20 (s, 1H), 4.52 (dd, J = 6.7, 9.1 Hz, 1H), 5.30 (s, 1H), 5.85 (dd, J = 1.4 Hz, 1H), 5.87 (d, J = 1.4 Hz, 1H), 6.31 (s, 1H), 6.36 (s, 2H). Anal. calcd. for $C_{23}H_{24}O_8$: C 64.48, H 5.65; found: C 64.01, H 5.57.

*2-Chloro-α-peltatin (*trans*)*

A solution of 60 mg (0.091 mmol) of **12** was dissolved in 5 mL of dry THF at 0°C. Acetic acid (0.1 mL) was added followed by 4 equivalents of TBAF (0.35 mL, 1.0 M in THF). After several minutes the reaction mixture was quenched with saturated NH_4Cl solution and then washed with sodium bicarbonate. Further work-up followed by PTLC (3:2 ethyl acetate – hexanes) afforded 38 mg (96%) of 2-chloro-α-peltatin (*trans*) as pale yellow prisms, mp 208–210°C; $[\alpha]_D$ (CH_3OH, c = 0.97) = −136; ^{1}H NMR, δ (ppm): 2.74 (dd, J = 11.0, 15.4 Hz, 1H), 2.84–3.06 (m, 1H), 3.157 (dd, J = 5.4, 15.6 Hz, 1H), 3.784 (s, 6H), 4.19 (dd, J = 8.5, 9.7 Hz, 1H), 4.41 (dd, J = 6.8, 8.4 Hz, 1H), 4.75 (s, 1H), 5.93 (d, J = 1.4 Hz, 1H), 5.96 (d, J = 1.4 Hz, 1H), 6.22 (s, 1H), 6.41 (s, 2H). Anal. calcd. for $C_{21}H_{19}O_8Cl$: C 58.01, H 4.40; found: C 57.70, H 4.41.

*2-Chloro-β-peltatin (*trans*)*

This compound was prepared from the monosilylated chloro derivative **14** using 2 equivalents of TBAF in THF/acetic acid, as above. The yield of product was 91%; mp 182–184°C (pale yellow flakes); $[\alpha]_D$ (CH_3OH, c = 1.01) = −129; ^{1}H NMR, δ (ppm): 2.76 (dd, J = 11.1, 15.7 Hz, 1H), 2.85–3.05 (m, 1H), 3.16 (dd, J = 5.3, 15.7 Hz, 1H), 3.76 (s, 6H), 3.81 (s, 3H), 4.20 (dd, J = 8.7, 9.5 Hz, 1H), 4.43 (dd, J = 6.7, 8.5 Hz, 1H), 4.77 (s, 1H), 5.97 (d, J = 1.4 Hz, 1H), 5.95 (d, J = 1.4 Hz, 1H), 6.22 (s, 1H), 6.40 (s, 2H). Anal. calcd. for $C_{22}H_{21}O_8Cl$: C 58.87, H 4.72; found: C 58.85, H 4.67.

*Reaction of the enolate of **4** with saturated NH_4Cl solution*

The enolate prepared from 202 mg of **4** was prepared as usual at −78°C and then quenched with saturated NH_4Cl solution. Work-up followed by PTLC chromatography (1:6 ethyl acetate – hexanes) afforded 39 mg (19%) of α-peltatin-bis-TBDMS and 158 mg (78%) of the less mobile *cis*-fused isomer **6** as a pale yellow foam, mp 140–142°C; ^{1}H NMR, δ: 0.11 (s, 6H), 0.18 (s, 3H), 0.21 (s, 3H), 0.99 (s, 9H), 1.00 (s, 9H), 2.63–2.71 (m, 2H), 2.92–3.05 (m, 1H), 3.33 (dd, J = 2.8, 9.6 Hz, 1H), 3.69 (s, 6H), 3.89 (dd, J = 3.4, 9.0 Hz, 1H), 4.35 (d, J = 2.6 Hz, 1H), 4.42 (dd, J = 7.6, 9.2 Hz, 1H), 5.86 (d, J = 1.4 Hz, 1H), 5.90 (d, J = 1.4 Hz, 1H), 6.27 (s, 2H), 6.33 (s, 1H); ^{13}C NMR, δ (ppm): −4.6 (q), −4.2 (q), 1.85 (s), 18.7 (s), 24.8 (t), 25.8 (q), 32.6 (d), 45.4 (d), 46.3 (d), 55.9 (q), 73.1 (t), 100.7 (t), 103.9 (d), 105.0 (d), 121.0 (s), 131.6 (s), 133.1 (s), 134.9 (s), 136.0 (s), 136.7 (s), 147.3 (s), 151.6 (s), 178.6 (s). Anal. calcd. for $C_{33}H_{48}O_8Si_2$: C 63.02, H 7.69; found: C 63.06, H 7.49.

*Reaction of the enolate of **5** with saturated NH_4Cl solution. Preparation of **7***

The enolate of **5** was quenched at −78°C with NH_4Cl solution as above. Work-up and PTLC (1:6 ethyl acetate – hexanes) afforded 40 mg (20%) of β-peltatin and 130 mg (64%) of the slightly less polar *cis* isomer **7**; mp 55–60°C; ^{1}H NMR, δ (ppm): 0.21 (s, 3H), 0.27 (s, 3H), 1.01 (s, 9H), 2.71 (dd, J = 1.5, 5.9 Hz, 2H), 2.92–3.10 (m, 1H), 3.32 (dd, J = 2.8, 9.6 Hz, 1H), 3.78 (s, 6H), 3.83 (s, 3H), 3.93 (dd, J = 3.4, 9.0 Hz, 1H), 4.36 (d, J = 3.0, 1H), 4.43 (dd, J = 7.3, 9.3 Hz, 1H), 5.87 (d, J = 1.4 Hz, 1H), 5.92 (d, J = 1.4 Hz, 1H), 6.31 (s, 1H), 6.34 (s, 2H); ^{13}C NMR, δ (ppm): 4.24 (q), 18.5 (s), 24.8 (t), 25.8 (q), 32.6 (d), 45.5 (d), 46.4 (d), 56.2 (d), 60.9 (d), 73.0 (t), 100.7 (t), 103.8 (d), 104.9 (d), 121.0 (s), 131.1 (s), 136.1 (s), 136.7 (s), 136.8 (s), 138.4 (s), 153.3 (s), 178.4 (s). Anal. calcd. for $C_{28}H_{36}O_8Si$: C 63.61, H 6.86; found: 63.89, H 6.69.

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