

CHEMBIOCHEM

Supporting Information

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Supporting Information

for

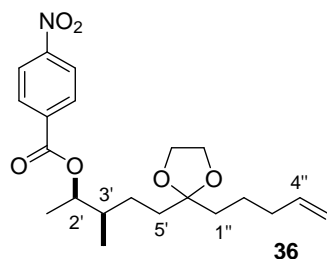
Propionate Analogues of Zearalenone Bind to Hsp90

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Daumantas Matulis, and Martin E. Maier*

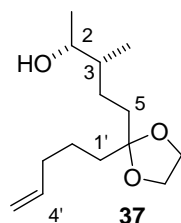
Experimental Section

General. Unless otherwise noted, all reactions were performed in oven-dried glassware. All solvents used in the reactions were purified before use. Dry diethyl ether, tetrahydrofuran, and toluene were distilled from sodium and benzophenone, whereas dry CH_2Cl_2 , dimethylformamide, methanol, ethyl acetate, benzene, and triethylamine were distilled from CaH_2 . Petroleum ether with a boiling range of 40–60 °C was used. Reactions were generally run under argon or nitrogen atmosphere. All commercially available compounds (Acros, Aldrich, Fluka, Merck) were used without purification. ^1H and ^{13}C NMR: Bruker Avance 400, spectra were recorded at 295 K either in CDCl_3 or CD_3OD ; chemical shifts are calibrated to the residual proton and carbon resonance of the solvent: CDCl_3 (δH 7.25, δC 77.0 ppm), CD_3OD (δH 3.30, δC 49.0 ppm). Melting points: Büchi Melting Point B-540, uncorrected. HRMS (FT-ICR): Bruker Daltonic APEX 2 with electron spray ionization (ESI). Analytical LC-MS: HP 1100 Series connected with an ESI MS detector Agilent G1946C, positive mode with fragmentor voltage of 40 eV, column: Nucleosil 100-5, C-18 HD, 5 mm, 70 × 3 mm Machery Nagel, eluent: NaCl solution (5 mM)/acetonitrile, gradient: 0-10-15-17-20 min with 20-80-80-99-99% acetonitrile, flow: 0.5 mL min^{-1} . Flash chromatography: J. T. Baker silica gel 43–60 mm. Thin-layer chromatography Machery-Nagel Polygram Sil G/UV254. Optical rotations: Perkin-Elmer Polarimeter 341, sodium D line (589 nm), $c = \text{g per 100 mL}$.

Procedures for Scheme 7

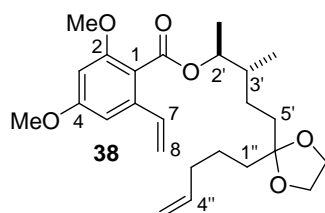


(2R,3R)-3-Methyl-5-(2-(pent-4-enyl)-1,3-dioxolan-2-yl)pentan-2-yl 4-nitrobenzoate (36). To a solution of alcohol **30** (260 mg, 1.073 mmol) in dry THF (8 mL) were added *p*-NBA (538 mg, 3.219 mmol) and PPh₃ (718 mg, 2.738 mmol). The mixture was cooled to 0 °C followed by the addition of DEAD (467 mg, 2.681 mmol). Then the mixture was allowed to reach room temperature and stirred for 1 h. The solvent was removed in vacuo and the residue purified by flash chromatography (petroleum ether/ethyl acetate, 9:1) to give ester **36** (369 mg, 88%) as a colorless oil. $R_f = 0.3$ (petroleum ether/ethyl acetate, 9:1); $[\alpha]_D^{20} = -22.5$ ($c = 1.0$, CH₂Cl₂). ¹H NMR (400 MHz, CDCl₃): $\delta = 1.01$ (d, $J = 6.6$ Hz, 3H, 3'-CH₃), 1.25-1.28 (m, 1H, 3'-H), 1.32 (d, $J = 6.1$ Hz, 3H, 1'-H), 1.39-1.45 (m, 2H, 5'-H), 1.55-1.73 (m, 6H, 4'-H, 1''-H, 2''-H), 2.01 (d, $J = 6.6$ Hz, 2H, 3''-H), 3.90 (s, 4H, OCH₂CH₂O), 4.91 (d, $J = 10.2$ Hz, 1H, 5''-H), 4.97 (d, $J = 17.3$ Hz, 1H, 5''-H), 5.10-5.17 (m, 1H, 2'-H), 5.69-5.80 (m, 1H, 4''-H), 8.18 (d, $J = 7.4$ Hz, 2H, CH-arom.), 8.27 (d, $J = 8.1$ Hz, 2H, CH-arom.); ¹³C NMR (100 MHz, CDCl₃): $\delta = 14.9$ (3'-CH₃), 17.0 (C-1'), 23.1 (C-2''), 26.6 (C-4'), 33.8 (C-5'), 34.6 (C-1''), 36.5 (C-3''), 38.0 (C-3'), 64.9 (OCH₂CH₂O), 75.8 (C-2'), 111.6 (acetal C), 114.7 (C-5''), 123.5 (CH-arom.), 130.6 (CH-arom.), 136.3 (C-arom.), 138.5 (C-4''), 150.4 (C-arom.), 164.2 (CO₂); HRMS (ESI): $[M+Na]^+$ calcd for C₂₁H₂₉NO₆ 414.18871, found 414.18885.

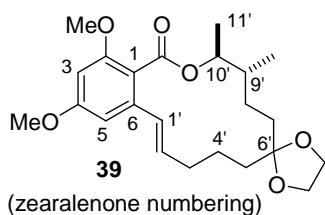


(2R,3R)-3-Methyl-5-(2-(pent-4-enyl)-1,3-dioxolan-2-yl)pentan-2-ol (37). A solution of ester **36** (300 mg, 0.77 mmol) in a mixture of 10% NaOH/MeOH (33 mL, 1:10) was refluxed for 2 h. After cooling, water was added and the mixture extracted with ethyl acetate (3 × 40 mL). The combined organic layers were washed with saturated NaCl solution, dried over MgSO₄, filtered, and concentrated in vacuo. Purification by flash

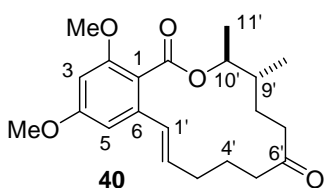
chromatography (petroleum ether/ethyl acetate, 1:1) provided alcohol **37** (194 mg, 85%) as a colorless oil. $R_f = 0.24$ (petroleum ether/ethyl acetate, 3:1); $[\alpha]_D^{20} = +17.0$ ($c = 1.0$, CH_2Cl_2); $^1\text{H NMR}$ (400 MHz, CDCl_3): $\delta = 0.87$ (d, $J = 6.9$ Hz, 3H, 3- CH_3), 1.14 (d, $J = 6.4$ Hz, 3H, 1-H), 1.11-1.22 (m, 1H, 3-H), 1.38-1.72 (m, 8H, 4-H, 5-H, 1'-H, 2'-H), 2.04 (q, $J = 7.0$ Hz, 2H, 3'-H), 3.70 (s, 1H, 2-H), 3.91 (s, 4H, $\text{OCH}_2\text{CH}_2\text{O}$), 4.93 (d, $J = 10.2$ Hz, 1H, 5'-H), 4.99 (d, $J = 17.3$ Hz, 1H, 5'-H), 5.72-5.85 (m, 1H, 4'-H); $^{13}\text{C NMR}$ (100 MHz, CDCl_3): $\delta = 14.1$ (3- CH_3), 20.2 (C-1), 23.1 (C-2'), 26.5 (C-4), 33.8 (C-5), 34.8 (C-1'), 36.5 (C-3'), 39.9 (C-3), 64.9 ($\text{OCH}_2\text{CH}_2\text{O}$), 71.1 (C-2), 111.8 (acetal C), 114.6 (C-5'), 138.6 (C-4'); HRMS (ESI): $[M+\text{Na}]^+$ calcd for $\text{C}_{14}\text{H}_{26}\text{O}_3$ 265.17742, found 265.17742.



Ester 38. For the preparation of this compound the procedure described for ester **31** was used. Thus, to a solution of alcohol **37** (130 mg, 0.54 mmol), acid **15** (151 mg, 0.72 mmol) and PPh_3 (440 mg, 1.68 mmol) in dry THF (7 mL), was added DEAD (290 mg, 1.66 mmol, 40% in toluene) dropwise). After complete addition, the mixture was stirred for 3 h at room temperature before the solvent was removed under reduced pressure. The resulting oil was purified by flash chromatography (petroleum ether/ethyl acetate, 4:1) to give the pure ester **38** (200 mg, 70%) as a colorless oil. $R_f = 0.33$ (petroleum ether/ethyl acetate, 4:1); $[\alpha]_D^{20} = +9.9$ ($c = 1.0$, CH_2Cl_2); $^1\text{H NMR}$ (400 MHz, CDCl_3): $\delta = 0.92$ (d, $J = 6.4$ Hz, 3H, 3'- CH_3), 1.12-1.19 (m, 1H, 3'-H), 1.25 (d, $J = 6.4$ Hz, 3H, 1'-H), 1.40-1.75 (m, 8H, 4'-H, 5'-H, 1''-H, 2''-H), 2.03 (q, $J = 7.0$ Hz, 2H, 3''-H), 3.78 (s, 3H, OCH_3), 3.82 (s, 3H, OCH_3), 3.90 (s, 4H, $\text{OCH}_2\text{CH}_2\text{O}$), 4.93 (d, $J = 9.9$ Hz, 1H, 5''-H), 4.98 (d, $J = 17.0$ Hz, 1H, 5''-H), 5.07 (ddd, $J = 12.7, 6.2, 6.0$ Hz, 1H, 2'-H), 5.30 (d, $J = 10.9$ Hz, 1H, 8-H), 5.69 (d, $J = 17.3$ Hz, 1H, 8-H), 5.73-5.88 (m, 1H, 4''-H), 6.38 (s, 1H, 3-H), 6.63 (s, 1H, 5-H), 6.69-6.76 (m, 1H, 7-H); $^{13}\text{C NMR}$ (100 MHz, CDCl_3): $\delta = 14.2$ (3'- CH_3), 15.5 (C-1'), 22.7 (C-2''), 26.1 (C-4'), 33.5 (C-5'), 34.2 (C-1''), 36.2 (C-3''), 37.0 (C-3'), 55.0 (OCH_3), 55.5 (OCH_3), 64.5 ($\text{OCH}_2\text{CH}_2\text{O}$), 74.8 (C-2'), 97.8 (C-3), 100.9 (C-5), 111.3 (C-1), 114.2 (C-8), 114.8 (C-6'), 116.4 (C-5''), 133.4 (C-7), 137.0 (C-4''), 138.3 (C-6), 157.6 (CO_2), 160.8 (C-2), 167.1 (C-4); HRMS (ESI): $[M+\text{Na}]^+$ calcd for $\text{C}_{25}\text{H}_{36}\text{O}_6$ 455.24041, found 455.24045.

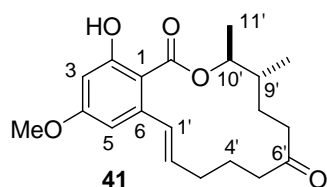


Macrolactone 39. For the preparation of this compound the procedure described for lactone **32** was used. Thus, diene **38** (170 mg, 0.39 mmol) was dissolved in dry toluene (100 mL), then Grubb's^{2nd}-catalyst (16.4 mg, 0.02 mmol) was added and the mixture heated for 5 h at 80 °C. After cooling, the reaction mixture was concentrated under reduced pressure and the resulting residue purified by flash chromatography (petroleum ether/ethyl acetate, 3:1). The pure lactone **38** (159 mg, 88%) was obtained as a slightly brown oil. $R_f = 0.3$ (petroleum ether/ethyl acetate, 3:1), $[\alpha]_D^{20} = +92.0$ ($c = 2.0$, CH_2Cl_2); $^1\text{H NMR}$ (400 MHz, CDCl_3): $\delta = 0.91$ (d, $J = 6.4$ Hz, 3H, 9'- CH_3), 1.31 (d, $J = 7.4$ Hz, 3H, 10'- CH_3), 1.38-1.44 (m, 2H, 7'-H), 1.50-1.84 (m, 7H, 4'-H, 5'-H, 8'-H, 9'-H), 2.12-2.19 (m, 1H, 3'-H), 2.30-2.38 (m, 1H, 3'-H), 3.78 (s, 3H, OCH_3), 3.80 (s, 3H, OCH_3), 3.89 (s, 4H, $\text{OCH}_2\text{CH}_2\text{O}$), 4.86-4.93 (m, 1H, 10'-H), 6.21-6.27 (m, 1H, 2'-H), 6.33 (s, 1H, 3-H), 6.47 (d, $J = 16.0$ Hz, 1H, 1'-H), 6.57 (s, 1H, 5-H); $^{13}\text{C NMR}$ (100 MHz, CDCl_3): $\delta = 15.2$ (9'- CH_3), 18.8 (10'- CH_3), 21.1 (C-4'), 27.6 (C-8'), 30.3 (C-3'), 31.6 (C-9'), 33.0 (C-7'), 37.8 (C-5'), 55.4 (OCH_3), 55.9 (OCH_3), 64.0, 64.3 ($\text{OCH}_2\text{CH}_2\text{O}$), 75.5 (C-10'), 97.5 (C-3), 100.9 (C-5), 112.0 (C-6'), 117.0 (C-1), 126.4 (C-1'), 133.2 (C-2'), 136.6 (C-6), 157.6 (C-2), 161.1 (C-4), 168.1 (CO_2); HRMS (ESI): $[M+\text{Na}]^+$ calcd for $\text{C}_{23}\text{H}_{32}\text{O}_6$ 427.20911, found 427.20908.

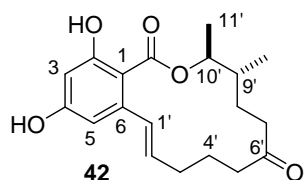


Macrolactone 40. Prepared in analogy to lactone **33**. A solution of lactone **39** (60 mg, 0.15 mmol) in acetone/ H_2O (3 mL, 10:1) containing $p\text{TsoH}$ (7.5 mg, 0.03 mmol) was refluxed for 12 h. After cooling, saturated NaHCO_3 solution was added and the mixture extracted with CH_2Cl_2 (3 \times 30 mL). The combined organic layers were dried over MgSO_4 , filtered, and concentrated under reduced pressure. The residue was purified by flash chromatography (petroleum ether/ethyl acetate, 3:1) to afford the pure ketone **40** (43 mg, 80%) as a colorless oil. $R_f = 0.31$ (petroleum ether/ethyl acetate, 3:1), $[\alpha]_D^{20} = +68.5$ ($c = 1.0$, CH_2Cl_2); $^1\text{H NMR}$ (400 MHz, CDCl_3): $\delta = 0.92$ (d, $J = 6.4$ Hz,

3H, 9'-CH₃), 1.20-1.25 (m, 1H, 8'-H), 1.33 (d, $J = 6.1$ Hz, 3H, 10'-CH₃), 1.48-1.53 (m, 1H, 4'-H), 1.59-1.65 (m, 1H, 9'-H), 1.90-2.02 (m, 2H, 10-H, 8'-H), 2.11-2.22 (m, 3H, 4'-H, 5'-H, 7'-H), 2.31-2.36 (m, 2H, 3'-H, 5'-H), 2.68-2.76 (m, 1H, 7'-H), 3.78 (s, 3H, OCH₃), 3.81 (s, 3H, OCH₃), 4.84-4.91 (m, 1H, 10'-H), 5.95-6.02 (m, 1H, 2'-H), 6.35 (s, 1H, 3-H), 6.37 (d, $J = 14.2$ Hz, 1H, 1'-H), 6.58 (s, 1H, 5-H); ¹³C NMR (100 MHz, CDCl₃): $\delta = 15.5$ (9'-CH₃), 18.7 (10'-CH₃), 21.7 (C-4'), 28.9 (C-8'), 31.4 (C-3'), 38.2 (C-7'), 39.5 (C-9'), 42.4 (C-5'), 55.4 (OCH₃), 55.9 (OCH₃), 75.8 (C-10'), 97.7 (C-3), 101.2 (C-5), 116.6 (C-1), 128.9 (C-1'), 133.6 (C-2'), 136.5 (C-6), 157.5 (C-2), 161.3 (C-4), 167.7 (CO₂), 211.5 (C-6'); HRMS (ESI): $[M+Na]^+$ calcd for C₂₁H₂₈O₅ 383.18290, found 383.18303.

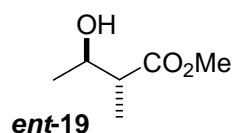


Macrolactone 41. Prepared in analogy to lactone **34**. To a solution of lactone **40** (8.0 mg, 0.023 mmol) in dry CH₂Cl₂ (3 mL) was added BCl₃ (0.11 mL, 1M in CH₂Cl₂, 0.11 mmol) dropwise at -60 °C. The mixture was allowed to warm to -20 °C and stirred for 30 min. Then the mixture was cooled to -50 °C before MeOH (1 mL) was added and the mixture allowed to reach room temperature. After removal of the solvents under reduced pressure the residue was purified by flash chromatography (petroleum ether/ethyl acetate, 3:1) to give lactone **41** (7.4 mg, 93%) as a colorless oil. $R_f = 0.49$ (petroleum ether/ethyl acetate, 3:1); $[\alpha]_D^{20} = -69.7$ ($c = 0.5$, CH₂Cl₂); ¹H NMR (400 MHz, CDCl₃): $\delta = 0.98$ (d, $J = 6.6$ Hz, 3H, 9'-CH₃), 1.24-1.32 (m, 2H, 4'-H), 1.36 (d, $J = 6.1$ Hz, 3H, 10'-CH₃), 1.64-1.72 (m, 1H, 9'-H), 2.00-2.38 (m, 6H, CH₂), 2.51-2.54 (m, 1H, 3'-H), 2.82-2.89 (m, 1H, 7'-H), 3.81 (s, 3H, OCH₃), 4.71-4.78 (m, 1H, 10'-H), 5.63-5.72 (m, 1H, 2'-H), 6.39 (s, 1H, 3-H), 6.45 (s, 1H, 5-H), 7.02 (d, $J = 15.2$ Hz, 1H, 1'-H), 12.18 (s, 1H, OH); ¹³C NMR (100 MHz, CDCl₃): $\delta = 14.9$ (9'-CH₃), 19.0 (10'-CH₃), 21.0 (C-4'), 29.0 (C-8'), 30.9 (C-3'), 36.3 (C-9'), 37.2 (C-7'), 40.1 (C-5'), 55.4 (OCH₃), 77.2 (C-10'), 99.9 (C-5), 103.5 (C-3), 108.1 (C-1), 132.7 (C-1'), 133.3 (C-2'), 143.4 (C-6), 164.1 (C-2), 165.8 (C-4), 171.4 (CO₂), 211.1 (C-6'); HRMS (ESI): $[M+Na]^+$ calcd for C₂₀H₂₆O₅ 369.16725, found 369.16716.



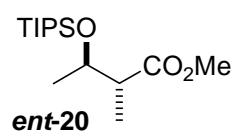
Macrolactone 42. Prepared in analogy to lactone **35**. A flask was charged with aluminium powder (25.8 mg, 0.954 mmol) and iodine (90.0 mg, 0.355 mmol). Then benzene (3 mL) was added and the mixture heated to reflux until the purple color disappeared. After that the mixture was cooled to 0 °C, then TBAI (1.0 mg) and the lactone **40** (8.0 mg, 0.023 mmol), dissolved in benzene (1 mL) was added. After complete addition the mixture was stirred for 3 min, followed by the addition of 2N HCl. The mixture was extracted with ethyl acetate (3 x 25 mL). The combined organic layers were dried over MgSO₄, filtered, and concentrated in vacuo. The residue was purified by flash chromatography (petroleum ether/ethyl acetate, 3:1) providing zearlenone analogue **42** (6.3 mg, 85%) as a colorless oil. $R_f = 0.29$ (petroleum ether/ethyl acetate, 3:1); $[\alpha]_D^{20} = -60.0$ ($c = 0.5$, CH₂Cl₂); ¹H NMR (400 MHz, CDCl₃): $\delta = 0.98$ (d, $J = 6.6$ Hz, 3H, 9'-CH₃), 1.20-1.33 (m, 2H, 4'-H), 1.36 (d, $J = 6.1$ Hz, 3H, 10'-CH₃), 1.63-1.70 (m, 1H, 9'-H), 1.99-2.38 (m, 6H, 3'-H, 5'-H, 7'-H, 8'-H), 2.50-2.56 (m, 1H, 7'-H), 2.82-2.90 (m, 1H, 5'-H), 4.70-4.77 (m, 1H, 10'-H), 5.63-5.71 (m, 1H, 2'-H), 6.33 (d, $J = 2.5$ Hz, 1H, 5-H), 6.39 (d, $J = 2.8$ Hz, 1H, 3-H), 7.02 (dd, $J = 15.3, 1.8$ Hz, 1H, 1'-H), 12.15 (s, 1H, OH); ¹³C NMR (100 MHz, CDCl₃): $\delta = 14.9$ (9'-CH₃), 19.0 (10'-CH₃), 21.0 (C-4'), 29.0 (C-8'), 30.9 (C-3'), 36.3 (C-9'), 37.3 (C-7'), 40.1 (C-5'), 77.2 (C-10'), 102.4 (C-3), 103.9 (C-1), 108.2 (C-5), 132.9 (C-2'), 133.1 (C-1'), 144.1 (C-6), 160.4 (C-4), 165.7 (C-2), 171.3 (CO₂), 211.3 (C-6'); HRMS (ESI): $[M+Na]^+$ calcd for C₁₉H₂₄O₅ 355.15159, found 355.15163.

Procedures for Scheme 8

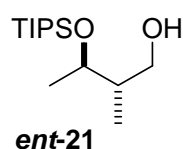


(2R,3R)-Methyl 3-hydroxy-2-methylbutanoate (ent-19). A solution of LDA was prepared by adding *n*BuLi (23.8 mL, 60.0 mmol) at -30 °C to a solution of *i*Pr₂NH (5.76 g, 8.04 mL, 57.0 mmol) in dry THF (35 mL). After 30 min of stirring the solution was cooled to -60 °C and the hydroxyester ent-**18** (3.20 g, 27.1 mmol) was added dropwise to the LDA solution and the mixture stirred for 45 min at -60 °C. Then a solution of MeI (3.85 g, 1.69 mL, 27.1 mmol) in HMPT (8 mL) was added dropwise to the

cooled solution. After complete addition the solution was allowed to reach room temperature. Then saturated NH_4Cl solution was added and the mixture extracted with Et_2O (3 \times 50 mL). The combined organic layers were dried over MgSO_4 , filtered, and concentrated in vacuo. Purification of the residue by flash chromatography (petroleum ether/ethyl acetate, 2:1) gave hydroxy ester **ent-19** (2.63 g, 73%) as a colorless oil. $R_f = 0.46$ (petroleum ether/ethyl acetate, 1:1); $[\alpha]_D^{20} = -27.7$ ($c = 1.0$, CH_2Cl_2); ^1H NMR (400 MHz, CDCl_3): $\delta = 1.15$ (d, $J = 7.1$ Hz, 3H, 2- CH_3), 1.18 (d, $J = 6.4$ Hz, 3H, 4-H), 2.37-2.50 (m, 1H, 2-H), 2.71 (d, $J = 4.3$ Hz, 1H, OH), 3.68 (s, 3H, OCH_3), 3.82-3.90 (m, 1H, 3-H); ^{13}C NMR (100 MHz, CDCl_3): $\delta = 14.0$ (2- CH_3), 20.7 (C-4), 46.9 (C-2), 51.7 (OCH_3), 69.4 (C-3), 176.3 (C-1).

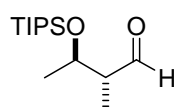


(2R,3R)-Methyl 2-methyl-3-(triisopropylsilyloxy)butanoate (ent-20). To a solution of hydroxyester **ent-19** (2.2 g, 16.8 mmol) in CH_2Cl_2 (40 mL) was added 2,6-lutidine (2.95 mL, 25.2 mmol) at 0 $^\circ\text{C}$ followed by the addition of TIPSOTf (5.42 mL, 20.6 mmol). The mixture was stirred for 8 h at room temperature. Then saturated NH_4Cl solution was added and the mixture extracted with CH_2Cl_2 (3 \times 60 mL). The combined CH_2Cl_2 layers were dried over MgSO_4 , filtered, and concentrated under reduced pressure. Purification of the residue by flash chromatography (petroleum ether/ethyl acetate, 35:1) provided 4.70 g (97%) of the ester **ent-20**. $R_f = 0.24$ (petroleum ether/ethyl acetate, 35:1); $[\alpha]_D^{20} = -28.0$ ($c = 1.0$, CH_2Cl_2); ^1H NMR (400 MHz, CDCl_3): $\delta = 1.04$ (s, 21H, $((\text{CH}_3)_2\text{CH})_3\text{Si}$), 1.10 (d, $J = 7.1$ Hz, 3H, 2- CH_3), 1.12 (d, $J = 6.1$ Hz, 3H, 4-H), 2.57-2.64 (m, 1H, 2-H), 3.65 (s, 3H, OCH_3), 4.22-4.28 (m, 1H, 3-H); ^{13}C NMR (100 MHz, CDCl_3): $\delta = 11.2$ (CHSi), 12.5 (2- CH_3), 18.0, 18.1 $((\text{CH}_3)_2\text{CH})_3\text{Si}$, 19.9 (C-4), 47.7 (C-2), 51.4 (OCH_3), 69.6 (C-3), 175.3 (C-1).



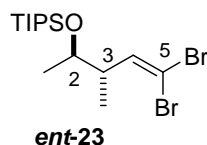
(2S,3R)-2-Methyl-3-(triisopropylsilyloxy)butan-1-ol (ent-21). To a solution of ester **ent-20** (2.16 g, 7.48 mmol) in dry CH_2Cl_2 (60 mL), cooled to -80 $^\circ\text{C}$, was added DIBAL-H (16.5 mL, 16.471 mmol) in a dropwise fashion. After addition the mixture was stirred for 30 min at -80 $^\circ\text{C}$ and then treated with saturated NH_4Cl solution. The

mixture was extracted with CH₂Cl₂ (3 × 40 mL), dried over MgSO₄, filtered, and concentrated in vacuo. The residue was purified by flash chromatography (petroleum ether/ethyl acetate, 9:1) to give 1.74 g (89%) of the pure alcohol **ent-21** as a colorless oil. $R_f = 0.33$ (petroleum ether/ethyl acetate, 9:1); $[\alpha]_D^{20} = -8.5$ ($c = 1.0$, CH₂Cl₂); ¹H NMR (400 MHz, CDCl₃): $\delta = 0.98$ (d, $J = 7.1$ Hz, 1H, 2-CH₃), 1.07 (s, 21H, ((CH₃)₂CH)₃Si) 1.22 (d, $J = 6.4$ Hz, 3H, 4-H), 1.62-1.70 (m, 1H, 2-H), 2.68 (br, 1H, OH), 3.53-3.58 (m, 1H, 1-H), 3.72-3.74 (m, 1H, 1-H), 3.97-4.03 (m, 1H, 3-H); ¹³C NMR (100 MHz, CDCl₃): $\delta = 12.7$ (CHSi), 13.9 (2-CH₃), 18.1, 18.2 ((CH₃)₂CH)₃Si, 21.6 (C-4), 42.2 (C-2), 66.0 (C-1), 73.4 (C-3).



ent-22

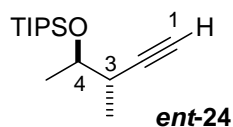
(2R,3R)-2-Methyl-3-(triisopropylsilyloxy)butanal (ent-22). To a solution of alcohol **ent-21** (2.83 g, 10.9 mmol) in dry CH₂Cl₂ (50 mL) were added PhI(OAc)₂ (4.56 g, 14.1 mmol) and TEMPO (340 mg, 2.18 mmol) followed by stirring of the mixture for 2 h at room temperature. Then a 10% solution of Na₂S₂O₃ (10 mL) was added, the mixture stirred for 10 min, before it was extracted with CH₂Cl₂ (3 × 40 mL). The combined CH₂Cl₂ layers were dried over MgSO₄, filtered, and concentrated in vacuo. The residue was purified by flash chromatography (petroleum ether/ethyl acetate, 50:1) to give 2.53 g (90%) of pure aldehyde **ent-22**. $R_f = 0.25$ (petroleum ether/ethyl acetate, 50:1); $[\alpha]_D^{20} = -33.0$ ($c = 1.0$, CH₂Cl₂); ¹H NMR (400 MHz, CDCl₃): $\delta = 0.99$ (s, 21H, ((CH₃)₂CH)₂Si), 1.05 (d, $J = 7.1$ Hz, 3H, 2-CH₃), 1.16 (d, $J = 6.1$ Hz, 3H, 4-H), 2.42-2.50 (m, 1H, 2-H), 4.24-4.30 (m, 1H, 3-H), 9.71 (s, 1H, CHO); ¹³C NMR (100 MHz, CDCl₃): $\delta = 9.7$ (2-CH₃), 12.5 (CHSi), 18.08, 18.13 ((CH₃)₂CH)₃Si, 21.3 (C-4), 54.0 (C-2), 69.4 (C-3), 204.9 (C-1).



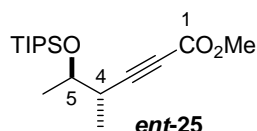
ent-23

Dibromoalkene ent-23. To a solution of aldehyde **ent-22** (1.33 g, 5.15 mmol) in CH₂Cl₂ (12 mL) was added triethylamine (2.24 mL, 15.9 mmol) at 0 °C. Then a solution (prepared at 0 °C) containing PPh₃ (6.58 g, 25.1 mmol) and CBr₄ (4.05 g, 12.2 mmol) in CH₂Cl₂ (25 mL) was added via canula to the aldehyde/amine solution at 0 °C. The mixture was stirred for 45 min at the same temperature. After that, silica

gel was added to the reaction mixture which was then evaporated. The loaded silica gel was applied to a flash column and the product eluted (petroleum ether/ethyl acetate, 50:1) to give 2.05 g (96%) of the dibromide **ent-23**. $R_f = 0.83$ (petroleum ether/ethyl acetate, 25:1). The dibromide was used directly in the subsequent alkyne formation reaction.

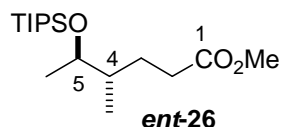


(3S,4R)-4-Triisopropylsilyloxy-3-methylpent-1-yne (ent-24). To a solution of dibromide **ent-23** (2.05 g, 4.94 mmol) in THF (70 mL) was added *n*BuLi (7.9 mL, 19.75 mmol) at $-80\text{ }^\circ\text{C}$. This mixture was stirred for 2 h at $-80\text{ }^\circ\text{C}$, before the reaction was quenched by the addition of water (30 mL). Then the mixture was extracted with Et₂O (3 × 30 mL). The combined organic layers were dried over MgSO₄, filtered, and concentrated. The residue was purified by flash chromatography (petroleum ether/ethyl acetate, 50:1) to give 1.24 g (99%) of pure alkyne **ent-24**. $R_f = 0.4$ (petroleum ether); $[\alpha]_D^{20} = -0.3$ ($c = 0.45$, CH₂Cl₂); ¹H NMR (400 MHz, CDCl₃): $\delta = 1.06$ (s, 21H, ((CH₃)₂CH)₃Si), 1.16 (d, $J = 7.1$ Hz, 3H, 3-CH₃), 1.20 (d, $J = 6.1$ Hz, 3H, 5-H), 2.04 (d, $J = 2.3$ Hz, 1H, 1-H), 2.58-2.65 (m, 1H, 3-H), 4.05-4.11 (m, 1H, 4-H); ¹³C NMR (100 MHz, CDCl₃): $\delta = 12.4$ (CHSi), 13.7 (3-CH₃), 18.08, 18.11 ((CH₃)₂CH)₃Si, 18.7 (C-5), 33.4 (C-3), 69.5 (C-4), 70.1 (C-1), 86.9 (C-2).

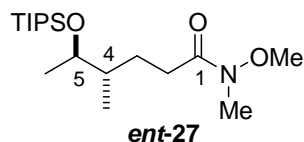


(4S,5R)-Methyl 4-methyl-5-(triisopropylsilyloxy)hex-2-ynoate (ent-25). To a solution of alkyne **ent-24** (820 mg, 3.22 mmol) in dry THF (10 mL), was added *n*BuLi (1.5 mL, 4.03 mmol) at $-80\text{ }^\circ\text{C}$ and the mixture stirred for 2 h at $-80\text{ }^\circ\text{C}$. After that methylchloroformate (1.0 mL, 12.9 mmol) was added, the mixture stirred for 1 h at $-80\text{ }^\circ\text{C}$ and then warmed to $0\text{ }^\circ\text{C}$. Thereafter, saturated NH₄Cl solution was added and the mixture extracted with Et₂O (3 × 60 mL). The combined organic layers were washed with saturated NaCl solution, dried over MgSO₄, filtered and concentrated in vacuo. The residue was purified by flash chromatography (petroleum ether/ethylacetate, 30:1) providing ester **ent-25** as a colorless oil (985 mg, 98%). $R_f = 0.41$ (petroleum ether/ethyl acetate, 25:1); $[\alpha]_D^{20} = -3.4$ ($c = 1.0$, CH₂Cl₂); ¹H NMR (400 MHz, CDCl₃): $\delta = 1.04$ (s, 21H, Si(CH(CH₃)₂)₃), 1.21 (dd, $J = 6.6, 3.3$ Hz, 6H, 6-H, 4-CH₃), 2.70-

2.76 (m, 1H, 4-H), 3.74 (s, 3H, OCH₃), 4.08-4.14 (m, 1H, 5-H); ¹³C NMR (100 MHz, CDCl₃): δ = 12.4 (CHSi), 13.2 (4-CH₃), 18.0, 18.1 ((CH₃)₂CH)₃Si, 19.4 (C-6), 33.8 (C-4), 52.5 (OCH₃), 69.9 (C-5), 74.1 (C-2), 91.4 (C-3), 154.2 (C-1).

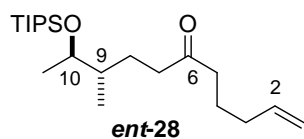


(4S,5R)-Methyl 4-methyl-5-(triisopropylsilyloxy)hexanoate (ent-26). The alkyne **ent-25** (1.02 g, 3.26 mmol) was dissolved in MeOH (10 mL), Pd/C (20 mg) was added and the suspension stirred for 3 h under a H₂-atmosphere. After that the suspension was filtered through a short pad of celite and washed with Et₂O. The filtrate was concentrated under reduced pressure to give **ent-26** as a colorless oil (990 mg, 96%). *R*_f = 0.33 (petroleum ether/ethyl acetate, 25:1); [α]²⁰_D = -4.4 (*c* = 1.0, CH₂Cl₂); ¹H NMR (400 MHz, CDCl₃): δ = 0.88 (d, *J* = 6.8 Hz, 3H, 6-H), 1.05 (s, 24H, 4-CH₃, ((CH₃)₂CH)₃Si), 1.34-1.42 (m, 1H, 4-H), 1.55-1.73 (m, 2H, 3-H), 2.32 (m, 2H, 2-H), 3.65 (s, 3H, OCH₃), 3.84-3.89 (m, 1H, 5-H); ¹³C NMR (100 MHz, CDCl₃): δ = 12.9 (CHSi), 13.6 (4-CH₃), 18.6 ((CH₃)₂CH)₃Si, 18.8 (C-6), 28.6 (C-3), 32.8 (C-2), 40.3 (C-4), 51.9 (OCH₃), 71.8 (C-5), 174.7 (C-1).

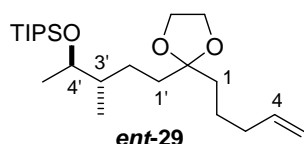


(4S,5R)-N-Methoxy-N,4-dimethyl-5-(triisopropylsilyloxy)hexanamide (ent-27). To a solution of ester **ent-26** (988 mg, 3.12 mmol) in THF (15 mL) was added *N,O*-dimethylhydroxylamine (518 mg, 5.31 mmol) in one portion at -20 °C followed by the dropwise addition of *i*PrMgCl (5.5 mL, 11.0 mmol, 2M in THF). Then the solution was allowed to warm to -10 °C and stirred for 30 min. Now saturated NH₄Cl solution was added and the mixture extracted with Et₂O (3 × 50 mL). The combined organic layers were dried over MgSO₄, filtered, and concentrated under reduced pressure. The residue was purified by flash chromatography (petroleum ether/ethyl acetate, 5:1) yielding pure amide **ent-27** as a colorless oil (1.0 g, 93%). *R*_f = 0.27 (petroleum ether/ethyl acetate, 5:1). [α]²⁰_D = -4.2 (*c* = 1.0, CH₂Cl₂); ¹H NMR (400 MHz, CDCl₃): δ = 0.89 (d, *J* = 6.6 Hz, 3H, 6-H), 1.03-1.05 (m, 24H, 4-CH₃, ((CH₃)₂CH)₃Si), 1.33-1.41 (m, 1H, 4-H), 1.57-1.71 (m, 2H, 3-H), 2.31-2.52 (m, 2H, 2-H), 3.16 (s, 3H, NCH₃), 3.67 (OCH₃), 3.86-3.92 (m, 1H, 5-H); ¹³C NMR (100 MHz, CDCl₃): δ = 11.9 (CH-Si), 12.7 (C-6),

17.6 ((CH₃)₂CH)Si), 17.7 (4-CH₃), 27.4 (C-3), 29.7 (C-2), 31.6 (NCH₃), 39.6 (C-4), 60.6 (OCH₃), 70.9 (C-5), 174.3 (C-1, weak signal).

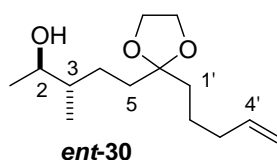


(9S,10R)-9-Methyl-10-(triisopropylsilyloxy)undec-1-en-6-one (ent-28). Mg turnings (98 mg, 4.04 mmol) were placed in a flask with a reflux-condenser and a septum. Then they were covered with dry Et₂O (1 mL) and some drops of 1-bromopentene were added to start the reaction. After the reaction has started the remaining 1-bromopentene (583 mg, 3.91 mmol) in Et₂O (10 mL) was added slowly. After complete addition, the mixture was stirred for 45 min at room temperature. In a separate flask a solution of amide **ent-27** (450 mg, 1.3 mmol) in dry THF (5 mL) was cooled to -80 °C. To this solution the prepared Grignard solution was added dropwise, followed by stirring of the mixture for 15 min at -80 °C before it was slowly warmed to room temperature. Now 1N HCl (2 mL) was added till the formed precipitate disappeared. After dilution with water, the mixture was extracted with Et₂O (3 × 40 mL) and the combined organic layers were washed with saturated NaCl, dried over MgSO₄, filtered, and concentrated in vacuo. Purification of the crude ketone by flash chromatography (petroleum ether/ethyl acetate, 40:1) gave ketone **ent-28** as a colorless oil (452 mg, 98 %). *R*_f = 0.43 (petroleum ether/ethyl acetate, 25:1); [α]_D²⁰ = -5.2 (c = 1.0, CH₂Cl₂); ¹H NMR (400 MHz, CDCl₃): δ = 0.86 (d, *J* = 6.6 Hz, 3H, 11-H), 1.04-1.05 (m, 24H, 9-CH₃, ((CH₃)₂CH)₃Si), 1.24-1.34 (m, 2H, 8-H), 1.49-1.56 (m, 1H, 9-H), 1.63-1.70 (m, 4H, 8-H, 4-H), 2.04 (q, *J* = 7.2 Hz, 2H, 3-H), 2.33-2.45 (m, 4H, 5-H, 7-H), 3.82-3.88 (m, 1H, 10-H), 4.96 (d, *J* = 11.2 Hz, 1H, 1-H), 5.00 (d, *J* = 17.0 Hz, 1H, 1-H), 5.71-5.81 (m, 1H, 2-H); ¹³C NMR (100 MHz, CDCl₃): δ = 12.5 (10-CH₃), 13.4 (9-CH₃), 18.1, 18.2 ((CH₃)₂CH)₃Si, 18.4 (C-5), 22.8 (C-11), 27.0 (C-8), 33.1 (C-3), 40.0 (C-7), 41.1 (C-9), 41.8 (C-5), 71.5 (C-10), 115.2 (C-1), 138.0 (C-2), 211.1 (C-6).

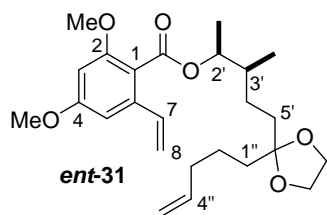


2-((3S,4R)-4-Triisopropylsilyloxy-3-methylpentyl)-2-(pent-4-enyl)-1,3-dioxolane (ent-29). The ketone **ent-28** (960 mg, 2.71 mmol) was dissolved in ethane-1,2-diol (4.53 mL, 81.3 mmol), then triethylorthoformate (2.25 mL, 13.5 mmol) and *p*TsOH

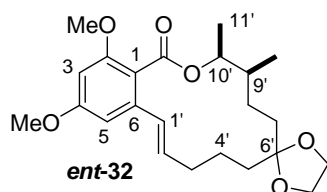
(103 mg, 0.43 mmol) were added and the mixture was stirred for 8 h at room temperature. After this saturated NaHCO₃ solution (20 mL) was added and the mixture extracted with Et₂O (3 × 50 mL). The combined organic layers were dried over MgSO₄, filtered, and concentrated in vacuo. Purification of the residue by flash chromatography (petroleum ether/ethyl acetate, 25:1) afforded the pure ketal **ent-29** (1.02 g, 94%) as a colorless oil. $R_f = 0.29$ (petroleum ether/ethyl acetate = 25/1). $[\alpha]_D^{20} = -3.9$ ($c = 1.0$, CH₂Cl₂); ¹H NMR (400 MHz, CDCl₃): $\delta = 0.86$ (d, $J = 6.9$ Hz, 3H, 5'-H), 1.03 (d, $J = 6.9$ Hz, 3H, 3'-CH₃), 1.04 (s, 21H, ((CH₃)₂CH)₃Si), 1.35-1.71 (m, 8H, 2-H, 1-H, 1'-H, 2'-H), 2.04 (q, $J = 7.0$ Hz, 2H, 3-H), 3.83-3.88 (m, 1H, 4'-H), 3.91 (s, 4H, OCH₂-CH₂O), 4.94 (d, $J = 10.2$ Hz, 5-H), 5.00 (d, $J = 17.3$ Hz, 5-H), 5.73-5.84 (m, 1H, 4-H); ¹³C NMR (100 MHz, CDCl₃): $\delta = 12.5$ (C-5'), 13.4 (3'-CH₃), 18.16, 18.19 ((CH₃)₂CH)₃-Si), 23.1 (C-2), 27.3 (C-2'), 33.9 (C-1'), 35.2 (C-1), 36.5 (C-3), 40.6 (C-3'), 64.9 (OCH₂CH₂O), 71.6 (C-4'), 111.8 (C(OR)₂), 114.6 (C-5), 138.7 (C-4).



(2R,3S)-3-Methyl-5-(2-(pent-4-enyl)-1,3-dioxolan-2-yl)pentan-2-ol (ent-30). To a solution of silyl ether **ent-29** (995 mg, 2.5 mmol) in THF (10 mL) was added TBAF (1.58 g, 5.0 mmol) at room temperature. The mixture was stirred for 6 h at room temperature before it was treated with saturated NH₄Cl solution (20 mL) and extracted with Et₂O (3 × 50 mL). The combined organic layers were dried over MgSO₄, filtered, and concentrated in vacuo. The crude product was purified by flash chromatography (petroleum ether/ethyl acetate, 2:1) to provide alcohol **30** (588 mg, 97%) as a colorless oil. $R_f = 0.2$ (petroleum ether/ethyl acetate, 3:1). $[\alpha]_D^{20} = -19.6$ ($c = 2.0$, CH₂Cl₂). ¹H NMR (400 MHz, CDCl₃): $\delta = 0.85$ (d, $J = 6.9$ Hz, 3H, 1-H), 1.11 (d, $J = 6.4$ Hz, 3H, 3-CH₃), 1.39-1.61 (m, 8H, 4-H, 5-H, 1'-H, 2'-H), 2.03 (q, $J = 7.1$ Hz, 2H, 3'-H), 3.59-3.66 (m, 1H, 3-H), 3.91 (s, 4H, OCH₂CH₂O), 4.93 (d, $J = 10.1$ Hz, 1H, 5'-H), 4.98 (dd, $J = 17.0, 1.5$ Hz, 1H, 5'-H), 5.72-5.83 (m, 1H, 4'-H); ¹³C NMR (100 MHz, CDCl₃): $\delta = 14.7$ (3-CH₃), 19.5 (C-1), 23.1 (C-2'), 26.4 (C-4), 33.8 (C-5), 34.5 (C-1'), 36.5 (C-3'), 40.2 (C-3), 64.9 (OCH₂CH₂O), 71.5 (C-2), 111.8 (C(OR)₂), 114.6 (C-5'), 138.6 (C-4').

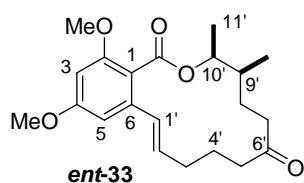


(2S,3S)-3-Methyl-5-(2-(pent-4-enyl)-1,3-dioxolan-2-yl)pentan-2-yl 2,4-dimethoxy-6-vinylbenzoate (*ent*-31). To a solution of alcohol *ent*-30 (21 mg, 0.087 mmol), acid **15** (24 mg, 0.115 mmol) and PPh₃ (73 mg, 0.277 mmol) in dry THF (3 mL), was added DEAD (46 mg, 0.266 mmol, 40% in toluene) dropwise. After complete addition, the mixture was stirred for 3 h at room temperature before the solvent was removed under reduced pressure. The resulting oil was purified by flash chromatography (petroleum ether/ethyl acetate, 4:1) to give the pure ester *ent*-31 (31 mg, 82%) as a colorless oil. $R_f = 0.34$ (petroleum ether/ethyl acetate, 4:1); $[\alpha]_D^{20} = +0.6$ ($c = 1.0$, CH₂Cl₂); ¹H NMR (400 MHz, CDCl₃): $\delta = 0.94$ (d, $J = 6.9$ Hz, 3H, 3'-CH₃), 1.19-1.30 (m, 1H, 4'-H), 1.28 (d, $J = 6.4$ Hz, 3H, 1'-H), 1.40-1.47 (m, 2H, 2''-H), 1.56-1.67 (m, 6H, 3'-H, 4'-H, 5'-H, 1''-H), 2.03 (q, $J = 7.1$ Hz, 2H, 3''-H), 3.78 (s, 3H, OCH₃), 3.82 (s, 3H, OCH₃), 3.90 (s, 4H, OCH₂CH₂O), 4.93 (d, $J = 10.2$ Hz, 1H, 5''-H), 4.98 (dd, $J = 17.2, 1.7$ Hz, 1H, 5''-H), 5.07-5.13 (m, 1H, 2'-H), 5.30 (d, $J = 10.9$ Hz, 1H, 8-H), 5.69 (d, $J = 17.6$ Hz, 1H, 8-H), 5.74-5.82 (m, 1H, 4''-H), 6.38 (d, $J = 2.0$ Hz, 1H, 3-H), 6.63 (d, $J = 2.0$ Hz, 1H, 5-H), 6.72 (dd, $J = 17.3, 10.9$ Hz, 1H, 7-H); ¹³C NMR (100 MHz, CDCl₃): $\delta = 14.8$ (3'-CH₃), 17.0 (C-1'), 23.0 (C-2''), 26.4 (C-4'), 33.9 (C-5'), 34.6 (C-1''), 36.6 (C-3''), 37.9 (C-3'), 55.4 (OCH₃), 55.8 (OCH₃), 64.9 (OCH₂CH₂O), 74.9 (C-2'), 98.2 (C-3), 101.2 (C-5), 111.7 (C-1), 114.6 (C(OR)₂), 116.8 (C-8), 116.9 (C-5''), 133.9 (C-7), 137.4 (C-4''), 138.6 (C-6), 157.9 (CO₂), 161.2 (C-2), 167.6 (C-4).

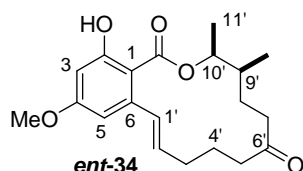


Macrolactone *ent*-32. The diene *ent*-31 (109 mg, 0.252 mmol) was dissolved in dry toluene (50 mL), then Grubb's^{2nd}-catalyst (9.1 mg, 0.012 mmol) was added and the mixture heated for 5 h at 80 °C. After cooling, the reaction mixture was concentrated under reduced pressure and the resulting residue purified by flash chromatography (petroleum ether/ethyl acetate, 3:1). The pure lactone *ent*-32 was obtained as a slightly brown oil in a yield of 90% (82 mg). $R_f = 0.3$ (petroleum ether/ethyl acetate,

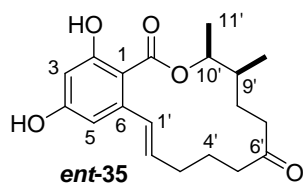
3:1); $[\alpha]_D^{20} = +71.5$ ($c = 2.0$, CH_2Cl_2); $^1\text{H NMR}$ (400 MHz, CDCl_3): $\delta = 0.93$ (d, $J = 6.9$ Hz, 3H, 9'- CH_3), 1.21 (d, $J = 6.6$ Hz, 3H, 10'- CH_3), 1.24-1.32 (m, 1H, 3'-H), 1.40-1.48 (m, 1H, 5'-H), 1.55-1.72 (m, 5H, 3'-H, 4'-H, 8'-H), 1.85-1.92 (m, 1H, 5'-H), 1.95-2.00 (m, 1H, 7'-H), 2.10-2.17 (m, 1H, 7'-H), 2.30-2.39 (m, 1H, 9'-H), 3.77 (s, 3H, OCH_3), 3.80 (s, 3H, OCH_3), 3.90 (dd, $J = 3.5, 1.3$ Hz, 4H, $\text{OCH}_2\text{CH}_2\text{O}$), 5.14-5.20 (m, 1H, 10'-H), 6.22-6.29 (m, 1H, 2'-H), 6.32 (d, $J = 2.0$ Hz, 1H, 3-H), 6.37 (d, $J = 16.1$ Hz, 1H, 1'-H), 6.59 (d, $J = 2.0$ Hz, 1H, 5-H); $^{13}\text{C NMR}$ (100 MHz, CDCl_3): $\delta = 14.8$ (9'- CH_3), 15.2 (10'- CH_3), 21.1 (C-4'), 25.1 (C-8'), 30.2 (C-3'), 32.7 (C-9'), 33.6 (C-7'), 37.1 (C-5'), 55.4 (OCH_3), 56.0 (OCH_3), 64.3 ($\text{OCH}_2\text{CH}_2\text{O}$), 64.4 ($\text{OCH}_2\text{CH}_2\text{O}$), 74.3 (C-10'), 97.5 (C-3), 100.7 (C-5), 112.1 (C-6'), 117.1 (C-1), 126.0 (C-1'), 132.8 (C-2'), 136.6 (C-6), 157.5 (C-2), 161.0 (C-4), 168.0 (CO_2).



Macrolactone *ent*-33. A solution of lactone *ent*-32 (99 mg, 0.244 mmol) in acetone/ H_2O (6 mL, 10:1) containing $p\text{TsOH}$ (5 mg, 0.024 mmol) was refluxed for 12 h. After cooling, saturated NaHCO_3 solution was added and the mixture extracted with CH_2Cl_2 (3 \times 30 mL). The combined organic layers were dried over MgSO_4 , filtered, and concentrated under reduced pressure. The residue was purified by flash chromatography (petroleum ether/ethyl acetate, 3:1) to afford the pure ketone *ent*-33 (73 mg, 83%) as a colorless oil. $R_f = 0.33$ (petroleum ether/ethyl acetate, 3:1); $[\alpha]_D^{20} = +13.0$ ($c = 1.0$, CH_2Cl_2); $^1\text{H NMR}$ (400 MHz, CDCl_3): $\delta = 0.95$ (d, $J = 6.9$ Hz, 3H, 9'- CH_3), 1.21 (d, $J = 6.6$ Hz, 3H, 10'- CH_3), 1.46-1.55 (m, 2H, 4'-H, 8'-H), 1.73-1.87 (m, 2H, 9'-H, 8'-H), 2.02-2.18 (m, 3H, 3'-H, 4'-H, 7'-H), 2.28-2.33 (m, 2H, 3'-H, 5'-H), 2.47-2.53 (m, 1H, 5'-H), 2.67-2.75 (m, 1H, 7'-H), 3.78 (s, 3H, OCH_3), 3.81 (s, 3H, OCH_3), 5.24 (ddd, $J = 13.2, 6.6, 3.0$ Hz, 1H, 10'-H), 5.94-6.02 (m, 1H, 2'-H), 6.28 (dd, $J = 15.8, 1.0$ Hz, 1H, 1'-H), 6.38 (d, $J = 2.0$ Hz, 1H, 3-H), 6.58 (d, $J = 2.0$, 1H, 5-H); $^{13}\text{C NMR}$ (100 MHz, CDCl_3): $\delta = 14.5$ (9'- CH_3), 14.7 (10'- CH_3), 21.1 (C-4'), 26.5 (C-8'), 31.0 (C-3'), 35.4 (C-9'), 37.0 (C-7'), 40.9 (C-5'), 55.4 (OCH_3), 55.9 (OCH_3), 74.3 (C-10'), 97.7 (C-3), 100.9 (C-5), 116.2 (C-1), 128.9 (C-1'), 132.7 (C-2'), 136.8 (C-6), 157.8 (C-3), 161.3 (C-4), 167.2 (CO_2), 211.3 (C-6').

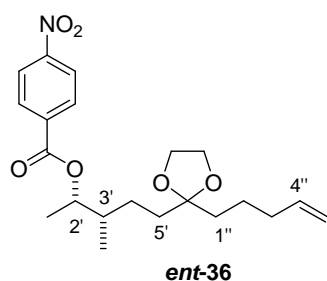


Macrolactone *ent-34*. To a solution of lactone ***ent-33*** (9.0 mg, 0.024 mmol) in dry CH_2Cl_2 (2.5 mL) was added BCl_3 (0.125 mL, 1M in CH_2Cl_2 , 0.125 mmol) dropwise at $-60\text{ }^\circ\text{C}$. The mixture was allowed to warm to $-20\text{ }^\circ\text{C}$ and stirred for 30 min. Then the mixture was cooled to $-50\text{ }^\circ\text{C}$ before MeOH (1 mL) was added and the mixture allowed to reach room temperature. After removal of the solvents under reduced pressure the residue was purified by flash chromatography (petroleum ether/ethyl acetate, 3:1) to give lactone ***ent-34*** (7.8 mg, 90%) as a colorless oil. $R_f = 0.45$ (petroleum ether/ethyl acetate, 3:1); $[\alpha]_D^{20} = -43.3$ ($c = 0.5$, CH_2Cl_2); $^1\text{H NMR}$ (400 MHz, CDCl_3): $\delta = 0.98$ (d, $J = 6.9$ Hz, 3H, 9'- CH_3), 1.27 (d, $J = 6.4$ Hz, 3H, 10'- CH_3), 1.42-1.48 (m, 1H 4'-H), 1.75-1.86 (m, 1H, 8'-H), 1.87-1.94 (m, 2H, 8'-H, 9'-H), 2.02-2.07 (m, 1H, 5'-H), 2.10-2.15 (m, 1H, 5'-H), 2.30-2.36 (m, 2H, 3'-H, 4'-H), 2.43-2.56 (m, 3H, 3'-H, 7'-H), 3.81 (s, 3H, OCH_3), 5.11-5.16 (m, 1H, 10'-H), 5.81-5.88 (m, 1H, 2'-H), 6.38 (s, 1H, 3-H), 6.50 (s, 1H, 5-H), 6.84 (d, $J = 15.5$ Hz, 1H, 1'-H), 11.48 (s, 1H, OH); $^{13}\text{C NMR}$ (100 MHz, CDCl_3): $\delta = 13.4$ (9'- CH_3), 15.8 (10'- CH_3), 22.2 (C-4'), 24.9 (C-8'), 31.3 (C-3'), 37.0 (C-9'), 38.0 (C-7'), 41.8 (C-5'), 55.4 (OCH_3), 76.9 (C-10'), 100.0 (C-5), 104.7 (C-3), 107.5 (C-1), 132.0 (C-1'), 132.5 (C-2'), 142.0 (C-6), 163.8 (C-2), 164.7 (C-4), 170.8 (CO_2), 211.6 (C-6').

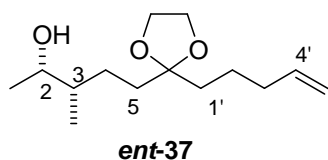


Macrolactone *ent-35*. A flask was charged with aluminium powder (6.5 mg, 0.239 mmol) and iodine (22.5 mg, 0.089 mmol). Then benzene (2 mL) was added and the mixture heated to reflux until the purple color disappeared. After that the mixture was cooled to $0\text{ }^\circ\text{C}$, then TBAI (0.2 mg) and the lactone ***ent-33*** (2.0 mg, 0.005 mmol), dissolved in benzene (1 mL) was added. After complete addition the mixture was stirred for 3 min, followed by the addition of 2N HCl (2 mL). After dilution with water, the mixture was extracted with ethyl acetate (3×20 mL). The combined organic layers were dried over MgSO_4 , filtered, and concentrated in vacuo. The residue was purified by flash chromatography (petroleum ether/ethyl acetate, 3:1) providing zearlenone ana-

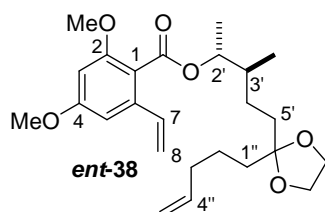
logue **ent-35** (1.34 mg, 82%) as a colorless oil. $R_f = 0.28$ (petroleum ether/ethyl acetate, 3:1); $[\alpha]_D^{20} = -10.5$ ($c = 0.2$, MeOH). $^1\text{H NMR}$ (400 MHz, CDCl_3): $\delta = 0.98$ (d, $J = 6.4$ Hz, 3H, 9'- CH_3), 1.27 (d, $J = 6.4$ Hz, 3H, 10'- CH_3), 1.40-1.80 (m, 1H, 4'-H), 1.74-1.95 (m, 3H, 8'-H, 9'-H), 2.00-2.17 (m, 2H, 4'-H, 5'-H), 2.28-2.36 (m, 2H, 3'-H, 5'-H), 2.42-2.56 (m, 3H, 3'-H, 7'-H), 5.12-5.14 (m, 1H, 10'-H), 5.81-5.88 (m, 1H, 2'-H), 6.32 (s, 1H, 3-H), 6.45 (s, 1H, 5-H), 6.83 (d, $J = 15.5$ Hz, 1H, 1'-H), 11.42 (s, 1H, OH); $^{13}\text{C NMR}$ (100 MHz, CDCl_3): $\delta = 13.4$ (9'- CH_3), 15.8 (10'- CH_3), 22.1 (C-4'), 24.9 (C-8'), 31.3 (C-3'), 36.9 (C-9'), 38.0 (C-7'), 41.8 (C-5'), 77.2 (C-10'), 102.4 (C-3), 105.2 (C-1), 107.7 (C-5), 132.2 (C-2'), 132.3 (C-1'), 142.7 (C-6), 160.1 (C-4), 164.5 (C-2), 170.7 (CO_2), 211.7 (C-6').



(2S,3S)-3-Methyl-5-(2-(pent-4-enyl)-1,3-dioxolan-2-yl)pentan-2-yl 4-nitrobenzoate (ent-36). To a solution of alcohol **ent-30** (240 mg, 1.0 mmol) in dry THF (10 mL) were added *p*-NBA (497 mg, 2.97 mmol) and PPh_3 (663 mg, 2.53 mmol). The mixture was cooled to 0 °C followed by the addition of DEAD (432 mg, 1.1 mL, 2.48 mmol, 40% in toluene). Then the mixture was allowed to reach room temperature and stirred for 1 h. The solvent was removed in vacuo and the residue purified by flash chromatography (petroleum ether/ethyl acetate, 9:1) to give ester **ent-36** (302 mg, 78%) as a colorless oil. $R_f = 0.3$ (petroleum ether/ethyl acetate, 9:1); $[\alpha]_D^{20} = +21.5$ ($c = 1.0$, CH_2Cl_2). $^1\text{H NMR}$ (400 MHz, CDCl_3): $\delta = 1.01$ (d, $J = 6.6$ Hz, 3H, 3'- CH_3), 1.25-1.28 (m, 1H, 3'-H), 1.32 (d, $J = 6.1$ Hz, 3H, 1'-H), 1.39-1.45 (m, 2H, 5'-H), 1.55-1.73 (m, 6H, 4'-H, 1''-H, 2''-H), 2.01 (d, $J = 6.6$ Hz, 2H, 3''-H), 3.90 (s, 4H, $\text{OCH}_2\text{CH}_2\text{O}$), 4.94 (m, 2H, 5''-H), 5.14 (m, 1H, 2'-H), 5.74 (m, 1H, 4''-H), 8.18 (d, $J = 7.4$ Hz, 2H, CH-arom.), 8.27 (d, $J = 8.1$ Hz, 2H, CH-arom.); $^{13}\text{C NMR}$ (100 MHz, CDCl_3): $\delta = 14.9$ (3'- CH_3), 17.0 (C-1'), 23.1 (C-2''), 26.6 (C-4'), 33.8 (C-5'), 34.6 (C-1''), 36.5 (C-3''), 38.0 (C-3'), 64.9 ($\text{OCH}_2\text{CH}_2\text{O}$), 75.8 (C-2'), 111.6 (acetal C), 114.7 (C-5''), 123.5 (CH-arom.), 130.6 (CH-arom.), 136.3 (C-arom.), 138.5 (C-4''), 150.4 (C-arom.), 164.2 (CO_2).

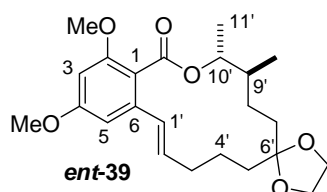


(2S,3S)-3-Methyl-5-(2-(pent-4-enyl)-1,3-dioxolan-2-yl)pentan-2-ol (ent-37). A solution of ester **ent-36** (300 mg, 0.76 mmol) in a mixture of 5% NaOH/MeOH (10 mL, 1:1) was refluxed for 2 h. After cooling, water was added and the mixture extracted with ethyl acetate (3 x 40 mL). The combined organic layers were washed with saturated NaCl solution, dried over MgSO₄, filtered, and concentrated in vacuo. Purification by flash chromatography (petroleum ether/ethyl acetate, 1:1) provided alcohol **ent-37** (158 mg, 85%) as a colorless oil. $R_f = 0.24$ (petroleum ether/ethyl acetate, 3:1); $[\alpha]_D^{20} = -6.0$ ($c = 1.0$, CH₂Cl₂); ¹H NMR (400 MHz, CDCl₃): $\delta = 0.87$ (d, $J = 6.9$ Hz, 3H, 3-CH₃), 1.14 (d, $J = 6.4$ Hz, 3H, 1-H), 1.11-1.22 (m, 1H, 3-H), 1.38-1.72 (m, 8H, 4-H, 5-H, 1'-H, 2'-H), 2.04 (q, $J = 7.0$ Hz, 2H, 3'-H), 3.70 (s, 1H, 2-H), 3.91 (s, 4H, OCH₂CH₂O), 4.96 (m, 2H, 5'-H), 5.78 (m, 1H, 4'-H); ¹³C NMR (100 MHz, CDCl₃): $\delta = 14.1$ (3-CH₃), 20.2 (C-1), 23.1 (C-2'), 26.5 (C-4), 33.8 (C-5), 34.8 (C-1'), 36.5 (C-3'), 39.9 (C-3), 64.9 (OCH₂CH₂O), 71.1 (C-2), 111.8 (acetal C), 114.6 (C-5'), 138.6 (C-4').

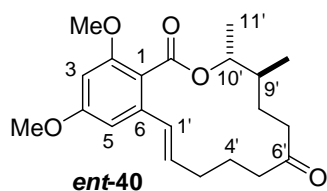


Ester ent-38. For the preparation of this compound the procedure described for ester **31** was used. Thus, to a solution of alcohol **ent-37** (125 mg, 0.52 mmol), acid **15** (236 mg, 1.14 mmol) and PPh₃ (345 mg, 1.32 mmol) in dry THF (6 mL), was added DEAD (225 mg, 0.56 mL, 1.29 mmol, 40% in toluene) dropwise. After complete addition, the mixture was stirred for 3 h at room temperature before the solvent was removed under reduced pressure. The resulting oil was purified by flash chromatography (petroleum ether/ethyl acetate, 4:1) to give the pure ester **ent-38** (173 mg, 74%) as a colorless oil. $R_f = 0.33$ (petroleum ether/ethyl acetate, 4:1); $[\alpha]_D^{20} = -6.4$ ($c = 1.0$, CH₂Cl₂); ¹H NMR (400 MHz, CDCl₃): $\delta = 0.92$ (d, $J = 6.4$ Hz, 3H, 3'-CH₃), 1.12-1.19 (m, 1H, 3'-H), 1.25 (d, $J = 6.4$ Hz, 3H, 1'-H), 1.40-1.75 (m, 8H, 4'-H, 5'-H, 1''-H, 2''-H), 2.03 (q, $J = 7.0$ Hz, 2H, 3''-H), 3.78 (s, 3H, OCH₃), 3.82 (s, 3H, OCH₃), 3.90 (s, 4H, OCH₂-CH₂O), 4.93 (d, $J = 9.9$ Hz, 1H, 5''-H), 4.98 (d, $J = 17.0$ Hz, 1H, 5''-H), 5.07 (ddd, $J = 12.7, 6.2, 6.0$ Hz, 1H, 2'-H), 5.30 (d, $J = 10.9$ Hz, 1H, 8-H), 5.69 (d, $J = 17.3$ Hz, 1H,

8-H), 5.73-5.88 (m, 1H, 4''-H), 6.38 (s, 1H, 3-H), 6.63 (s, 1H, 5-H), 6.69-6.76 (m, 1H, 7-H); ^{13}C NMR (100 MHz, CDCl_3): δ = 14.2 (3'- CH_3), 15.5 (C-1'), 22.7 (C-2''), 26.1 (C-4'), 33.5 (C-5'), 34.2 (C-1''), 36.2 (C-3''), 37.0 (C-3'), 55.0 (OCH_3), 55.5 (OCH_3), 64.5 ($\text{OCH}_2\text{CH}_2\text{O}$), 74.8 (C-2'), 97.8 (C-3), 100.9 (C-5), 111.3 (C-1), 114.2 (C-8), 114.8 (C-6'), 116.4 (C-5''), 133.4 (C-7), 137.0 (C-4''), 138.3 (C-6), 157.6 (CO_2), 160.8 (C-2), 167.1 (C-4).

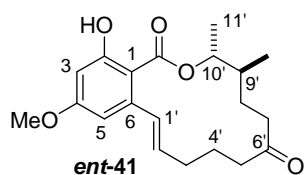


Macrolactone ent-39. For the preparation of this compound the procedure described for lactone **32** was used. Thus, diene **ent-38** (40 mg, 0.09 mmol) was dissolved in dry toluene (25 mL), then Grubb's^{2nd}-catalyst (4.2 mg, 0.0054 mmol) was added and the mixture heated for 5 h at 80 °C. After cooling, the reaction mixture was concentrated under reduced pressure and the resulting residue purified by flash chromatography (petroleum ether/ethyl acetate, 3:1). The pure lactone **ent-39** (33.5 mg, 90%) was obtained as a slightly brown oil. R_f = 0.3 (petroleum ether/ethyl acetate, 3:1), $[\alpha]_D^{20}$ = -91.1 (c = 2.0, CH_2Cl_2); ^1H NMR (400 MHz, CDCl_3): δ = 0.91 (d, J = 6.4 Hz, 3H, 9'- CH_3), 1.31 (d, J = 7.4 Hz, 3H, 10'- CH_3), 1.38-1.44 (m, 2H, 7'-H), 1.50-1.84 (m, 7H, 4'-H, 5'-H, 8'-H, 9'-H), 2.12-2.19 (m, 1H, 3'-H), 2.30-2.38 (m, 1H, 3'-H), 3.78 (s, 3H, OCH_3), 3.80 (s, 3H, OCH_3), 3.89 (s, 4H, $\text{OCH}_2\text{CH}_2\text{O}$), 4.86-4.93 (m, 1H, 10'-H), 6.21-6.27 (m, 1H, 2'-H), 6.33 (s, 1H, 3-H), 6.47 (d, J = 16.0 Hz, 1H, 1'-H), 6.57 (s, 1H, 5-H); ^{13}C NMR (100 MHz, CDCl_3): δ = 15.2 (9'- CH_3), 18.8 (10'- CH_3), 21.1 (C-4'), 27.6 (C-8'), 30.3 (C-3'), 31.6 (C-9'), 33.0 (C-7'), 37.8 (C-5'), 55.4 (OCH_3), 55.9 (OCH_3), 64.0, 64.3 ($\text{OCH}_2\text{CH}_2\text{O}$), 75.5 (C-10'), 97.5 (C-3), 100.9 (C-5), 112.0 (C-6'), 117.0 (C-1), 126.4 (C-1'), 133.2 (C-2'), 136.6 (C-6), 157.6 (C-2), 161.1 (C-4), 168.1 (CO_2).

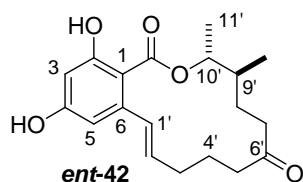


Macrolactone ent-40. Prepared in analogy to lactone **33**. A solution of lactone **ent-39** (120 mg, 0.30 mmol) in acetone/ H_2O (4.5 mL, 10:1) containing $p\text{TsOH}$ (3 mg, 0.015 mmol) was refluxed for 12 h. After cooling, saturated NaHCO_3 solution was

added and the mixture extracted with CH_2Cl_2 (3 × 30 mL). The combined organic layers were dried over MgSO_4 , filtered, and concentrated under reduced pressure. The residue was purified by flash chromatography (petroleum ether/ethyl acetate, 3:1) to afford the pure ketone **ent-40** (89 mg, 83%) as a colorless oil. $R_f = 0.31$ (petroleum ether/ethyl acetate, 3:1), $[\alpha]_D^{20} = -64.2$ ($c = 1.0$, CH_2Cl_2); $^1\text{H NMR}$ (400 MHz, CDCl_3): $\delta = 0.92$ (d, $J = 6.4$ Hz, 3H, 9'- CH_3), 1.20-1.25 (m, 1H, 8'-H), 1.33 (d, $J = 6.1$ Hz, 3H, 10'- CH_3), 1.48-1.53 (m, 1H, 4'-H), 1.59-1.65 (m, 1H, 9'-H), 1.90-2.02 (m, 2H, 3'-H, 8'-H), 2.11-2.22 (m, 3H, 4'-H, 5'-H, 7'-H), 2.31-2.36 (m, 2H, 3'-H, 5'-H), 2.68-2.76 (m, 1H, 7'-H), 3.78 (s, 3H, OCH_3), 3.81 (s, 3H, OCH_3), 4.84-4.91 (m, 1H, 10'-H), 5.95-6.02 (m, 1H, 2'-H), 6.35 (s, 1H, 3-H), 6.37 (d, $J = 14.2$ Hz, 1H, 1'-H), 6.58 (s, 1H, 5-H); $^{13}\text{C NMR}$ (100 MHz, CDCl_3): $\delta = 15.5$ (9'- CH_3), 18.7 (10'- CH_3), 21.7 (C-4'), 28.9 (C-8'), 31.4 (C-3'), 38.2 (C-7'), 39.5 (C-9'), 42.4 (C-5'), 55.4 (OCH_3), 55.9 (OCH_3), 75.8 (C-10'), 97.7 (C-3), 101.2 (C-5), 116.6 (C-1), 128.9 (C-1'), 133.6 (C-2'), 136.5 (C-6), 157.5 (C-2), 161.3 (C-4), 167.7 (CO_2), 211.5 (C-6').



Macrolactone ent-41. Prepared in analogy to lactone **34**. To a solution of lactone **ent-40** (12.0 mg, 0.033 mmol) in dry CH_2Cl_2 (2.5 mL) was added BCl_3 (0.27 ml, 1M in CH_2Cl_2 , 0.27 mmol) dropwise at -60 °C. The mixture was allowed to warm to -20 °C and stirred for 30 min. Then the mixture was cooled to -50 °C before MeOH (1 mL) was added and the mixture allowed to reach room temperature. After removal of the solvents under reduced pressure the residue was purified by flash chromatography (petroleum ether/ethyl acetate, 3:1) to give lactone **ent-41** (11 mg, 96%) as a colorless oil. $R_f = 0.51$ (petroleum ether/ethyl acetate, 3:1); $[\alpha]_D^{20} = +55.2$ ($c = 0.5$, CH_2Cl_2); $^1\text{H NMR}$ (400 MHz, CDCl_3): $\delta = 0.98$ (d, $J = 6.6$ Hz, 3H, 9'- CH_3), 1.24-1.32 (m, 2H, 4'-H), 1.36 (d, $J = 6.1$ Hz, 3H, 10'- CH_3), 1.64-1.72 (m, 1H, 9'-H), 2.00-2.38 (m, 6H, CH_2), 2.51-2.54 (m, 1H, 3'-H), 2.82-2.89 (m, 1H, 7'-H), 3.81 (s, 3H, OCH_3), 4.71-4.78 (m, 1H, 10'-H), 5.63-5.72 (m, 1H, 2'-H), 6.39 (s, 1H, 3-H), 6.45 (s, 1H, 5-H), 7.02 (d, $J = 15.2$ Hz, 1H, 1'-H), 12.18 (s, 1H, OH); $^{13}\text{C NMR}$ (100 MHz, CDCl_3): $\delta = 14.9$ (9'- CH_3), 19.0 (10'- CH_3), 21.0 (C-4'), 29.0 (C-8'), 30.9 (C-3'), 36.3 (C-9'), 37.2 (C-7'), 40.1 (C-5'), 55.4 (OCH_3), 77.2 (C-10'), 99.9 (C-5), 103.5 (C-3), 108.1 (C-1), 132.7 (C-1'), 133.3 (C-2'), 143.4 (C-6), 164.1 (C-2), 165.8 (C-4), 171.4 (CO_2), 211.1 (C-6').



Macrolactone ent-42. Prepared in analogy to lactone **35**. A flask was charged with aluminium powder (49 mg, 1.79 mmol) and iodine (168 mg, 0.67 mmol). Then benzene (3 mL) was added and the mixture heated to reflux until the purple color disappeared. After that the mixture was cooled to 0 °C, then TBAI (4 mg) and the lactone **ent-40** (15 mg, 0.042 mmol), dissolved in benzene (1 mL) was added. After complete addition the mixture was stirred for 3 min, followed by the addition of 2N HCl (2 mL). After addition of water, the mixture was extracted with ethyl acetate (3 × 25 mL). The combined organic layers were dried over MgSO₄, filtered, and concentrated in vacuo. The residue was purified by flash chromatography (petroleum ether/ethyl acetate, 3:1) providing zearlenone analogue **ent-42** (12.3 mg, 88%) as a colorless oil. $R_f = 0.32$ (petroleum ether/ethyl acetate, 3:1); $[\alpha]_D^{20} = +106.4$ ($c = 1.0$, CH₂Cl₂); ¹H NMR (400 MHz, CDCl₃): $\delta = 0.98$ (d, $J = 6.6$ Hz, 3H, 9'-CH₃), 1.20-1.33 (m, 2H, 4'-H), 1.36 (d, $J = 6.1$ Hz, 3H, 10'-CH₃), 1.63-1.70 (m, 1H, 9'-H), 1.99-2.38 (m, 6H, 3'-H, 5'-H, 7'-H, 8'-H), 2.50-2.56 (m, 1H, 7'-H), 2.82-2.90 (m, 1H, 5'-H), 4.70-4.77 (m, 1H, 10'-H), 5.63-5.71 (m, 1H, 2'-H), 6.33 (d, $J = 2.5$ Hz, 1H, 5-H), 6.39 (d, $J = 2.8$ Hz, 1H, 3-H), 7.02 (dd, $J = 15.3, 1.8$ Hz, 1H, 1'-H), 12.15 (s, 1H, OH); ¹³C NMR (100 MHz, CDCl₃): $\delta = 14.9$ (9'-CH₃), 19.0 (10'-CH₃), 21.0 (C-4'), 29.0 (C-8'), 30.9 (C-3'), 36.3 (C-9'), 37.3 (C-7'), 40.1 (C-5'), 77.2 (C-10'), 102.4 (C-3), 103.9 (C-1), 108.2 (C-5), 132.9 (C-2'), 133.1 (C-1'), 144.1 (C-6), 160.4 (C-4), 165.7 (C-2), 171.3 (CO₂), 211.3 (C-6').

Compound binding to human α -Hsp90 N-terminal domain (Hsp90N) by thermal shift assay: The thermal shift assay was performed using the Corbett Rotor-Gene 6000 (Qiagen Rotor-Gene Q) RT-PCR. The temperature repeatability was measured to be with an error of ± 0.2 °C. Protein concentration was measured spectrophotometrically (ϵ_{280} (Hsp90N) = 15 930 M⁻¹ cm⁻¹). Protein concentration was 10 μ M and compound concentrations were from 0 to 200 μ M. DMSO was added to make 1% (v/v) in the ligand solution. Protein unfolding was monitored by measuring the fluorescence of the solvatochromic fluorescent dye ANS added at 50 μ M. The total volume of the reaction was 10 μ L. Samples were overlaid with 2.5 μ L of silicone oil DC 200. Heating of the samples was carried out at a speed of 1 °C min⁻¹. Raw data (fluorescence

intensity vs temperature, Figures S1-S3) were fitted to standard equations describing protein thermal stability.^[23] Graphs were prepared using the software TFAlyst®.

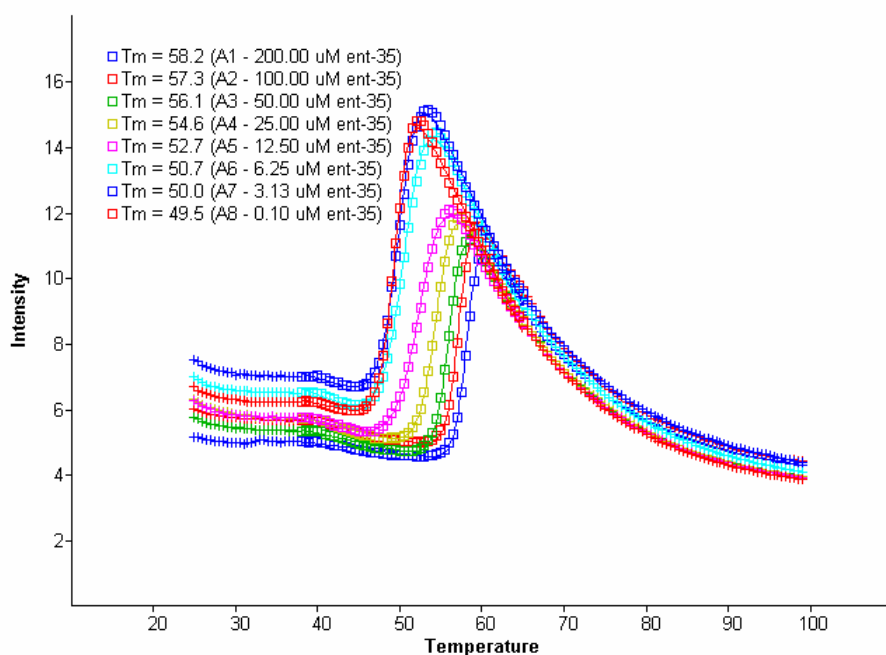


Figure S1. Hsp90N thermal denaturation curves. Addition of 200 μ M compound **ent-35** shifts the melting temperature of Hsp90N from 49.5 to 58.2 $^{\circ}$ C.

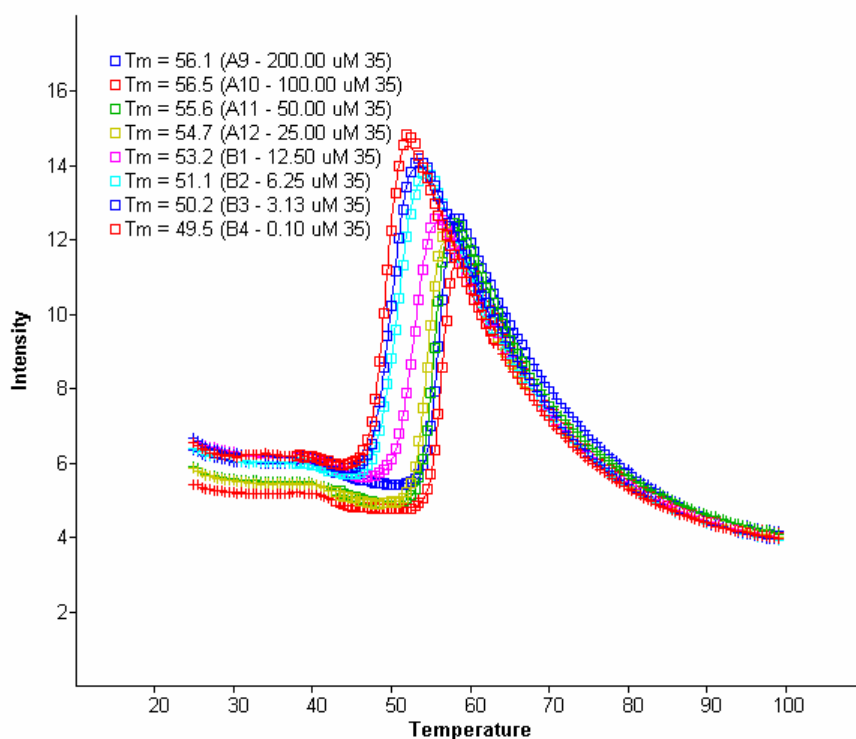


Figure S2. Hsp90N thermal denaturation curves. Addition of 200 μ M compound **35** shifts the melting temperature of Hsp90N from 49.5 to 56.1 $^{\circ}$ C.

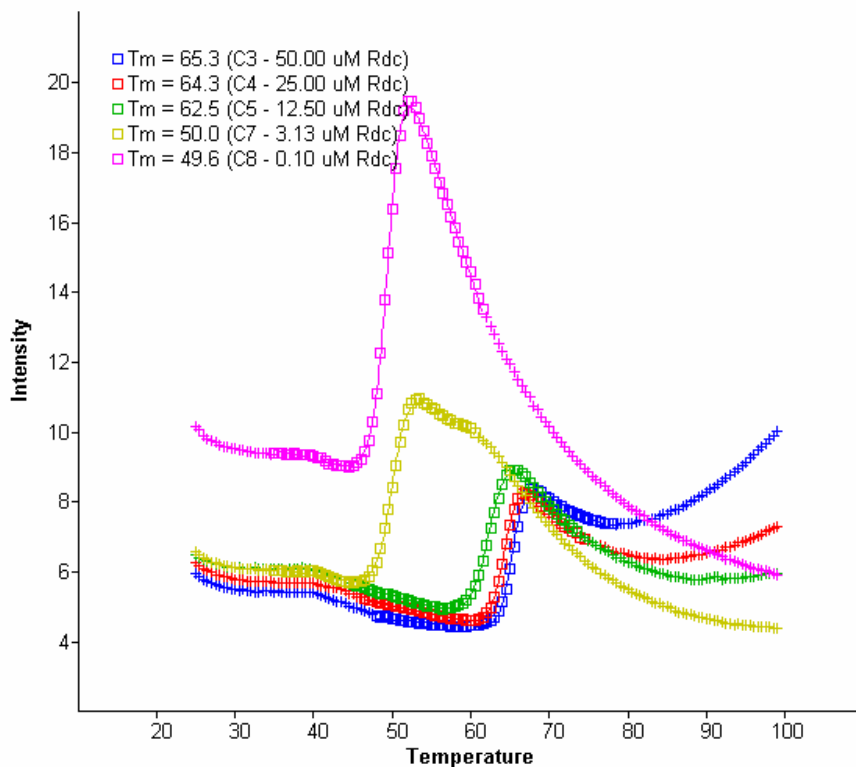


Figure S3. Hsp90N thermal denaturation curves. Addition of 50 μM radicicol shifts the melting temperature of Hsp90N from 49.6 to 65.3 $^{\circ}\text{C}$.

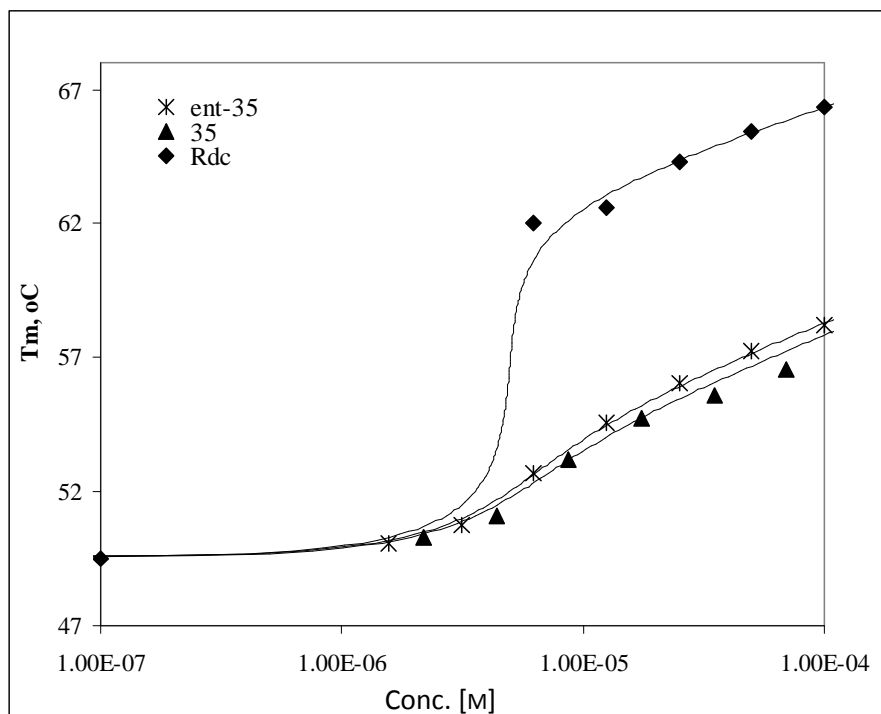


Figure S4. Hsp90N melting temperature T_m data as a function of added ligand concentration L_t . Datapoints are obtained from raw data in Figures S1-S3 and the lines are fitted according to the model.^[23] Rdc = radicicol.

Table S1. Compound-Hsp90N dissociation constants K_d by thermal shift assay

compound	K_d [μM]
<i>ent</i> - 35	0.25
35	0.33
radicol	0.001

