Supplementary Information

Screening of a virtual mirror-image library of natural products

Taro Noguchi, Shinya Oishi,* Kaori Honda, Yasumitsu Kondoh, Tamio Saito, Hiroaki Ohno, Hiroyuki Osada, and Nobutaka Fujii*

Graduate School of Pharmaceutical Sciences, Kyoto University
RIKEN CSRS

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Experimental Procedures

General Methods. $^1$H NMR and $^{13}$C NMR spectra were recorded using a JEOL ECA-500 spectrometer. Chemical shifts are reported in $\delta$ (ppm) relative to Me$_4$Si (in CDCl$_3$) and residual THF (in THF-$d_8$). $^{13}$C NMR spectra were referenced to the residual CHCl$_3$ (in CDCl$_3$) and THF (in THF-$d_8$). $^1$H NMR spectra were tabulated as follows: chemical shift, multiplicity (br: broad, s: singlet, d: doublet, t: triplet, q: quartet, m: multiplet), number of protons, and coupling constants. Exact mass (HRMS) spectra were recorded on a JMS-HX/HX 110A mass spectrometer or Shimadzu LC-ESI-IT-TOF-MS equipment. Melting points were measured by a hot stage melting point apparatus (uncorrected). For flash chromatography, Wakogel C-300E (Wako) was employed. For analytical HPLC for MDM2 proteins, Cosmosil 5C18-AR300 (4.6 × 250 mm, Nacalai Tesque Inc.) was employed with a linear gradient of CH$_3$CN containing 0.1% (v/v) TFA aq. at a flow rate of 1 cm$^3$ min$^{-1}$. Cosmosil 5C18-ARII column (4.6 × 250 mm, Nacalai Tesque Inc.) was employed for the analysis of the other peptides.

Synthesis of MDM2$^{25-109}$ and MDM2$^{TMR}$ Proteins

L-MDM2$^{25-109}$, L-MDM2$^{TMR}$, D-MDM2$^{25-109}$ and D-MDM2$^{TMR}$ proteins were synthesized by the identical protocol in our previous report.$^1$ Briefly, protected MDM2$^{25-109}$ was synthesized by a standard protocol of Fmoc-based SPPS using automatic peptide synthesizer (PSSM-8, Shimadzu Corporation Ltd.) on H-Rink Amide-ChemMatrix resin (0.4-0.6 mmol g$^{-1}$) using HBTU/HOBt/(iPr)$_2$NEt activation. The resulting protected peptide resin was treated with TFA/thio anisole/m-cresol/1,2-ethanedithiol/H$_2$O (80:5:5:5:5) at room temperature for 2 h. After removal of the resin by filtration, the filtrate was poured into ice-cold dry Et$_2$O. The resulting powder was collected by centrifugation and then washed with ice-cold dry Et$_2$O three times. The crude product was purified by HPLC to provide MDM2$^{25-109}$ proteins as white powder.

For the synthesis of MDM2$^{TMR}$ proteins, two glycines and 5-hexynoic acid were coupled with HBTU/HOBt/(iPr)$_2$NEt. The resulting protected peptides were treated with the deprotection cocktail as above to provide alkyne-conjugated proteins. Subsequently, treatment of the proteins with TMR-(PEG)$_3$-azide in the presence of Cu(I) provided TMR-labeled MDM2$^{TMR}$ proteins as pink powder.

ESI-TOF MS for L-MDM2$^{25-109}$: Calcd for C$_{462}$H$_{739}$N$_{117}$O$_{123}$S$_4$: 10027.42; observed: [M+12H]$^{12+}$ m/z = 836.74, [M+11H]$^{11+}$ m/z = 912.76, [M+10H]$^{10+}$ m/z = 1003.86, [M+9H]$^{9+}$ m/z = 1115.39, [M+8H]$^{8+}$ m/z = 1254.76, [M+7H]$^{7+}$ m/z = 1434.00.$^1$

ESI-TOF MS for D-MDM2$^{25-109}$: Calcd for C$_{462}$H$_{739}$N$_{117}$O$_{123}$S$_4$: 10027.42; observed: [M+12H]$^{12+}$ m/z = 836.68, [M+11H]$^{11+}$ m/z = 912.77, [M+10H]$^{10+}$ m/z = 1003.94, [M+9H]$^{9+}$ m/z = 1115.46, [M+8H]$^{8+}$ m/z = 1254.85, [M+7H]$^{7+}$ m/z = 1433.42.$^1$

ESI-TOF MS for L-MDM2$^{TMR}$: Calcd for C$_{505}$H$_{789}$N$_{125}$O$_{133}$S$_4$: 10865.79; observed: [M+13H]$^{13+}$ m/z = 836.94, [M+12H]$^{12+}$ m/z = 906.78, [M+11H]$^{11+}$ m/z = 989.03, [M+10H]$^{10+}$ m/z = 1087.90, [M+9H]$^{9+}$ m/z = 1208.38, [M+8H]$^{8+}$ m/z = 1359.59.$^1$

ESI-TOF MS for D-MDM2$^{TMR}$: Calcd for C$_{505}$H$_{789}$N$_{125}$O$_{133}$S$_4$: 10865.79; observed: [M+13H]$^{13+}$ m/z = 836.65, [M+12H]$^{12+}$ m/z = 906.65, [M+11H]$^{11+}$ m/z = 989.11, [M+10H]$^{10+}$ m/z = 1088.17, [M+9H]$^{9+}$ m/z = 1208.45, [M+8H]$^{8+}$ m/z = 1359.80.
Folding of Synthetic MDM2^{25–109} and MDM2^{TMR} Proteins

Folding of synthetic MDM2 proteins were carried out by the identical protocol in our previous report.\textsuperscript{1} Lyophilized polypeptide was dissolved in 6 M Gu-HCl (1 mg cm\textsuperscript{-3}) followed by 100-fold dilution with PBS (pH 7.4) containing 0.5 mmol dm\textsuperscript{-3} TCEP-HCl and 0.005% Tween-20 at 4 °C. The solution was stored at 4 °C overnight, and the solution was concentrated using a MWCO 3000 centrifugal filtration membrane (Millipore, Amicon-Ultra 3 kDa) (3 times).

Synthesis of p53 Peptides and Their Derivatives

p53 peptides were synthesized by Fmoc-SPPS on Rink-amide resin (0.66 mmol g\textsuperscript{-1}, 45.5 mg, 0.025 mmol) according to the identical protocol in our previous report.\textsuperscript{1} Fmoc-protected amino acids (3 eq) were coupled by using DIC (3 eq) and HOBt (3 eq) in DMF. The Fmoc-protecting group was removed by treatment of the resin with 20% piperidine in DMF. Coupling of biotin (0.125 mmol) was carried out with HBTU (5 eq), HOBt (5 eq) and (Pr\textsubscript{2})NEt (10 eq) in DMF. Coupling of 5-carboxyfluorescein (5 eq) was carried out with DIC (5 eq) and HOBt (10 eq) in DMF. The resulting protected peptide resin was treated with TFA/thioanisole/m-cresol/1,2-ethanedithiol/H\textsubscript{2}O (80:5:5:5:5) at room temperature for 2 h. After removal of the resin by filtration, the filtrate was poured into ice-cold dry Et\textsubscript{2}O. The resulting powder was collected by centrifugation and then washed with ice-cold dry Et\textsubscript{2}O three times. The crude product was purified by HPLC on a Cosmosil 5C18-ARII preparative column (Nacalai Tesque, 20 × 250 mm). All peptides were characterized by ESI-MS or MALDI-TOF-MS.

Biotin-labeled L-p53 peptide (biotinyl-aminocaproyl-GSGSSQETFSDLWKLLPEN-NH\textsubscript{2}): MS (MALDI-TOF) calcd for \textit{C}_{108}\textit{H}_{166}\textit{N}_{27}\textit{O}_{35}\textit{S} [M+H]\textsuperscript{+} 2434.19; found: 2434.13.\textsuperscript{1}


L-P4 (H-LTFEHYWAQLTS-NH\textsubscript{2}): MS (MALDI-TOF) calcd for \textit{C}_{71}\textit{H}_{100}\textit{N}_{17}\textit{O}_{19} [M+H]\textsuperscript{+} 1494.74; found: 1494.75.\textsuperscript{1}

D-P4: MS (MALDI-TOF) calcd for \textit{C}_{71}\textit{H}_{100}\textit{N}_{17}\textit{O}_{19} [M+H]\textsuperscript{+} 1494.74; found: 1494.73.\textsuperscript{11}

5-Carboxyfluorescein-labeled L-P4 (5-FAM-LTFEHYWAQLTS-NH\textsubscript{2}): MS (MALDI-TOF) calcd for \textit{C}_{92}\textit{H}_{110}\textit{N}_{17}\textit{O}_{25} [M+H]\textsuperscript{+} 1852.79; found: 1852.80.\textsuperscript{1}

5-Carboxyfluorescein-labeled D-P4: MS (MALDI-TOF) calcd for \textit{C}_{92}\textit{H}_{110}\textit{N}_{17}\textit{O}_{25} [M+H]\textsuperscript{+} 1852.79; found: 1852.76.
Synthesis of NP843 Derivatives 12a-d

Methyl (R)-2-Methyl-3-(trityloxy)propanoate (S2). To a stirred solution TrtCl (23.5 g, 169 mmol) in CH₂Cl₂ (150 cm³) were added Et₃N (17.9 cm³, 186 mmol), DMAP (1.03 g, 16.9 mmol), (R)-Roche ester (S1, 4.69 cm³, 42.3 mmol) at room temperature. After the mixture was stirred overnight, saturated NH₄Cl was added. The whole was extracted with CHCl₃ and the extract was washed with H₂O and brine, and dried over MgSO₄. The filtrate was concentrated under reduced pressure. Recrystallization from EtOH gave the title compound S2 as colorless crystals (9.55 g, 63%): mp 96–98 °C (from EtOH); [α]²⁵D –16.8 (c 1.10, CHCl₃); IR (neat) ν max/cm⁻¹: 1737 (C=O); δH (500 MHz, CDCl₃) 1.15 (3H, d, J 6.9, CH₃), 2.71-2.77 (1H, m, CH), 3.17 (1H, dd, J₁ 8.9, J₂ 6.0, CH₂), 3.30 (1H, dd, J₁ 8.6, J₂ 6.9, CH₂), 3.70 (3H, s, CH₃) 7.21-7.42 (15H, m, Ar); δC(125 MHz, CDCl₃) 14.0, 40.4, 51.6, 65.3, 86.3, 127.0 (3C), 127.7 (6C), 128.7 (6C), 143.9 (3C), 175.4. Anal. calcd. for C₂₄H₂₄O₃: C, 79.97; H, 6.71. Found: C, 79.76; H, 6.86.

Methyl (S)-2-Methyl-3-(trityloxy)propanoate (ent-S2). According to the procedure described for the preparation of S2, (S)-Roche ester (ent-S1, 4.69 cm³, 42.3 mmol) was converted into ent-S2 as colorless crystals (9.56 g, 63%): mp 96–98 °C (from EtOH); [α]²⁵D +16.6 (c 1.05, CHCl₃); IR (neat) ν max/cm⁻¹: 1739 (C=O); δH (500 MHz, CDCl₃) 1.15 (3H, d, J 6.9, CH₃), 2.71-2.77 (1H, m, CH), 3.17 (1H, dd, J₁ 8.6, J₂ 5.7, CH₂), 3.30 (1H, dd, J₁ 8.6, J₂ 6.9, CH₂), 3.70 (3H, s, CH₃), 7.21-7.42 (15H, m, Ar); δC(125 MHz, CDCl₃) 14.0, 40.4, 51.6, 65.3, 86.3, 127.0 (3C), 127.7 (6C), 128.7 (6C), 143.9 (3C), 175.4. Anal. calcd. for C₂₄H₂₄O₃: C, 79.97; H, 6.71. Found: C, 80.05; H, 6.77.

(R)-2-Methyl-3-(trityloxy)propyl 4-Methylbenzenesulfonate (2). To a stirred solution of compound S2 (9.00 g, 25.0 mmol) in THF (200 cm³) under argon was added LiAlH₄ (1.42 g, 37.4 mmol) at 0 °C. After the mixture was stirred for 30 min, saturated aqueous solution of sodium potassium tartrate was slowly added at 0 °C, and the mixture was stirred overnight at room temperature. The whole was extracted with Et₂O and the extract was washed with H₂O and brine, and dried over MgSO₄. Filtration through a short pad of silica gel and concentration under reduced pressure gave a crude alcohol, which was used without further purification. TsCl (5.96 g, 31.3 mmol) was added to a solution of the alcohol in pyridine (18.2 cm³) at 0 °C. After the mixture was stirred overnight, the reaction was quenched with H₂O at 0 °C. The whole was extracted with Et₂O and the extract was washed with H₂O and brine, and dried over MgSO₄. Filtration through a short pad of silica gel and concentration gave a crude sulfonate. Recrystallization from CHCl₃/hexane gave the title compound 2 as colorless crystals (11.6 g, 96%): mp 146–148 °C (from CHCl₃/hexane); [α]²⁵D −11.4 (c 1.15, CHCl₃); IR (neat) ν max/cm⁻¹: 1360 (OSO₂), 1176 (OSO₂); δH(500 MHz, CDCl₃) 0.89 (3H, d, J 8.6, CH₃), 2.01-2.08 (1H, m, CH), 2.43 (3H, s, CH₃), 2.93 (1H, dd, J₁ 9.2, J₂ 6.9, CH₂), 3.02 (1H, dd, J₁ 9.2, J₂ 5.2, CH₂), 3.99 (1H, dd, J₁ 9.2, J₂ 6.3, CH₂), 4.12 (1H, dd, J₁ 9.5, J₂ 5.4, CH₂), 7.21-7.34 (17H, m, Ar), 7.74-7.76 (2H, m, Ar); δC(125 MHz, CDCl₃) 13.9, 21.6, 34.0, 64.2, 72.5, 86.4, 127.0 (3C), 133.9 (3C), 153.4. Anal. calcd. for C₂₄H₂₄O₃S: C, 79.97; H, 6.71. Found: C, 80.05; H, 6.77.
127.7 (6C), 127.9 (2C), 128.6 (6C), 129.8 (2C), 133.0, 143.9 (3C), 144.6. Anal. calcd. for C₃₀H₃₀O₄S: C, 74.05; H, 6.21. Found: C, 73.91; H, 6.26.

(S)-2-Methyl-3-(trityloxy)propyl 4-Methylbenzenesulfonate (ent-2). According to the procedure described for the preparation of 2, compound ent-S₂ (8.00 g, 22.2 mmol) was converted into ent-2 as colorless crystals (9.05 g, 84%): mp 145–148 °C (from CHCl₃/hexane); [α]²⁵D +11.4 (c 1.08, CHCl₃); IR (neat) νmax/cm⁻¹: 1361 (OSO₂), 1175 (OSO₂); δH(500 MHz, CDCl₃) 0.89 (3H, d, J 8.6, CH₃), 2.01-2.08 (1H, m, CH), 2.43 (3H, s, CH₃), 2.93 (1H, dd, J₁ 9.2, J₂ 6.9, CH₂), 3.02 (1H, dd, J₁ 9.2, J₂ 5.2, CH₂), 3.99 (1H, dd, J₁ 9.2, J₂ 6.3, CH₂), 4.12 (1H, dd, J₁ 9.5, J₂ 5.4, CH₂), 7.21-7.34 (17H, m, Ar), 7.74-7.76 (2H, m, Ar); δC(125 MHz, CDCl₃) 13.9, 21.6, 34.0, 64.2, 72.5, 86.4, 127.0 (3C), 127.7 (6C), 127.9 (2C), 128.6 (6C), 129.8 (2C), 133.0, 143.9 (3C), 144.6. Anal. calcd. for C₃₀H₃₀O₄S: C, 74.05; H, 6.21. Found: C, 73.86; H, 6.33.

(2S,6R)-2,6,10-Trimethylundec-9-en-1-ol (4). To a stirred mixture of Mg (1.00 g, 41.2 mmol) in THF (6.6 cm³) was added dropwise (S)-citronellyl bromide (S₃, 6.00 g, 27.4 mmol) in THF (16.8 cm³) over 2 h using syringe pump at 60 °C to give Grignard reagent 3. To a stirred solution of 2 (6.9 g, 14.2 mmol) in THF (31.0 cm³) were added the reagent 3 and Li₂CuCl₄ in THF (0.1 mol dm⁻³, 27.0 cm³, 2.70 mmol) at –40 °C. The resulting mixture was stirred at –40 °C overnight and the reaction was quenched with saturated NH₄Cl at 0 °C. The whole was extracted with Et₂O and the extract was washed with H₂O and brine, and dried over MgSO₄. The filtrate was concentrated under reduced pressure. The oily residue was dissolved in hexane and the solution was filtrated through a short pad of silica gel to give crude S₄. To a stirred solution of the compound S₄ in dry Et₂O (120 cm³) was added HCO₂H (120 cm³) dropwise at 0 °C under argon, and the stirring was continued for 1 h. After toluene (150 cm³) was added, the solution was concentrated under reduced pressure. 1N NaOH in MeOH/H₂O (1:1, 100 cm³) was added to the residue and the mixture was stirred for 10 min. The mixture was concentrated under reduced pressure. The residue was extracted with Et₂O and the extract was dried over MgSO₄. The filtrate was concentrated under reduced pressure, and the residue was purified by column chromatography to give the title compound 4 as a colorless oil (1.67 g, 55%). [α]²⁵D -7.5 (c 1.10, CHCl₃); IR (neat) νmax/cm⁻¹: 3319 (OH), 1035 (C-O); δH(500 MHz, CDCl₃) 0.86 (3H, d, J 6.3, CH₃), 0.93 (3H, d, J 6.3, CH₃), 1.04-1.42 (9H, m), 1.60 (3H, s, CH₃), 1.58-1.65 (1H , m), 1.68 (3H, s, CH₃), 1.89-2.03 (2H, m, CH₂), 3.42 (1H, dd, J₁ 10.3, J₂ 6.3, CH₂), 3.51 (1H, dd, J₁ 10.6, J₂ 5.4, CH₂), 3.99 (1H, dd, J₁ 10.6, J₂ 6.0, CH₂), 5.09-5.11 (1H, m, CH); δC(125 MHz, CDCl₃) 16.6, 17.6, 19.6, 24.3, 25.5, 25.7, 32.4, 33.4, 35.8, 37.1, 37.2, 68.4, 125.0, 131.0. HRMS (FAB) calcd for C₁₄H₂₉O (MH⁺): 213.2213; found: 213.2217.

(2R,6S)-2,6,10-Trimethylundec-9-en-1-ol (ent-4). According to the procedure described for the preparation of 4, compound ent-2 (5.00 g, 10.3 mmol) was converted into ent-4 with (R)-citronellyl bromide (6.40 g, 29.2 mmol) as a colorless oil (996 mg, 46%). [α]²⁵D +7.4 (c 1.17, CHCl₃); IR
(3S,7R)-3,7,11-Trimethylidodec-10-enenitrile (5). To a stirred solution of compound 4 (1.26 g, 5.93 mmol) in pyridine (4.3 cm³) was added TsCl (1.40 g, 7.72 mmol) at 0 °C. After the mixture was stirred for 2 h, saturated aqueous solution of citric acid was added. The whole was extracted with Et₂O and the extract was washed with H₂O and brine, and dried over MgSO₄. The extract was concentrated under reduced pressure after filtration through a short pad of silica gel to give a crude sulfonate. To a stirred solution of the sulfonate in DMSO (13.0 cm³) was added NaCN (0.581 g, 11.8 mmol) at room temperature. After the mixture was stirred overnight, saturated aqueous solution of NH₄Cl was added. The whole was extracted with CHCl₃ and the extract was washed with H₂O and brine, and dried over MgSO₄. The filtrate was concentrated under reduced pressure, and the residue was purified by column chromatography to give the title cyanide 5 as a colorless oil (1.17 g, 89%). [α]₂⁵±5.2 (c 1.02, CHCl₃); IR (neat)  ν max/cm⁻¹: 2247 (C≡N); δ H(500 MHz, CDCl₃) 0.87 (3H, d, J 6.3, CH₃), 1.07-1.17 (2H, m), 1.23-1.43 (7H, m), 1.61 (3H, s, CH₃), 1.81-1.89 (1H, m), 2.24 (1H, dd, J 16.6, J 6.9, CH₂), 2.32 (1H, dd, J 16.6, J 5.7, CH₂), 5.09-5.11 (1H, m, CH); δC(125 MHz, CDCl₃) 17.6, 19.5 (2C), 24.2, 24.4, 25.5, 25.7, 30.5, 32.3, 36.2, 36.8, 37.0, 119.0, 124.9, 131.1. HRMS (FAB) calcd for C₁₅H₂₈N (MH⁺): 222.2216; found: 222.2224.

(3S,7R)-3,7,11-Trimethylidodec-10-en-1-ol (6).³ To a stirred solution of 5 (1.00 g, 4.52 mmol) in CH₂Cl₂ (5.0 cm³) was added dropwise DIBAL-H in THF (1.0 mol dm⁻³, 4.97 cm³, 4.97 mmol) at −78 °C under argon. After the mixture was stirred for 2 h, saturated aqueous solution of sodium potassium tartrate was added and the mixture was warmed to room temperature. The whole was extracted with Et₂O and the extract was dried over MgSO₄. The extract was concentrated under reduced pressure after filtration through a short pad of silica gel to give crude aldehyde. To a stirred solution of the aldehyde in CH₂Cl₂ (5.0 cm³) was added dropwise DIBAL-H in THF (1.0 mol dm⁻³, 4.97 cm³, 4.97 mmol) at −78 °C under argon. After the mixture was stirred for 2 h, saturated aqueous solution of sodium potassium tartrate was added. The whole was extracted with Et₂O and the extract was dried
over MgSO₄. The extract was concentrated under reduced pressure, and the residue was purified by column chromatography to give the title alcohol 6 as a colorless oil (0.848 g, 83%). \([\alpha]^{25}_D -2.3 \ (c \ 0.90, \ \text{CHCl}_3)\); IR (neat) \(\nu_{\text{max}}/\text{cm}^{-1}: 3299 \ (\text{OH}), 1058 \ (\text{C}-\text{O})\); \(\delta\text{H}(500 \ \text{MHz, CDCl}_3) 0.86 \ (3\ H, \ d, \ J \ 6.3, \ \text{CH}_3), 0.90 \ (3\ H, \ d, \ J \ 6.3, \ \text{CH}_3), 1.07-1.41 \ (11\ H, \ m), 1.54-1.64 \ (1\ H, \ m), 1.61 \ (3\ H, \ s, \ \text{CH}_3), 1.69 \ (3\ H, \ s, \ \text{CH}_3), 1.89-2.03 \ (2\ H, \ m, \ \text{CH}_2), 3.64-3.73 \ (2\ H, \ m, \ \text{CH}_2), 5.09-5.11 \ (1\ H, \ m, \ \text{CH}); \delta\text{C}(125 \ \text{MHz, CDCl}_3) 17.6, 19.6, 19.7, 24.3, 25.6, 25.7, 29.5, 32.4, 37.0, 37.2, 37.5, 40.0, 61.3, 125.0, 124.9, 131.0. \text{HRMS} \ (\text{FAB}) \text{ calcd for C}_{15}\text{H}_{31}\text{O} (\text{MH}^+): 227.2369; \text{found: 227.2363.}

\((3R,7S)-3,7,11\text{-Trimethyl}dodec-10\text{-en}-1\text{-ol (ent-6).} \) According to the procedure described for the preparation of 6, compound \textit{ent-}5 (900 mg, 4.07 mmol) was converted into \textit{ent-}6 as a colorless oil (606 mg, 66%). \([\alpha]^{25}_D +2.0 \ (c \ 1.05, \ \text{CHCl}_3)\); IR (neat) \(\nu_{\text{max}}/\text{cm}^{-1}: 3299 \ (\text{OH}), 1055 \ (\text{C}-\text{O})\); \(\delta\text{H}(500 \ \text{MHz, CDCl}_3) 0.86 \ (3\ H, \ d, \ J \ 6.3, \ \text{CH}_3), 0.90 \ (3\ H, \ d, \ J \ 6.3, \ \text{CH}_3), 1.04-1.41 \ (11\ H, \ m), 1.52-1.63 \ (1\ H, \ m), 1.60 \ (3\ H, \ s, \ \text{CH}_3), 1.68 \ (3\ H, \ s, \ \text{CH}_3), 1.89-2.03 \ (2\ H, \ m, \ \text{CH}_2), 3.64-3.73 \ (2\ H, \ m, \ \text{CH}_2), 5.08-5.12 \ (1\ H, \ m, \ \text{CH}); \delta\text{C}(125 \ \text{MHz, CDCl}_3) 17.6, 19.6, 19.7, 24.3, 25.6, 25.7, 29.5, 32.4, 37.0, 37.2, 37.5, 39.9, 61.3, 125.0, 124.9, 131.0. \text{HRMS} \ (\text{FAB}) \text{ calcd for C}_{15}\text{H}_{31}\text{O} (\text{MH}^+): 227.2369; \text{found: 227.2363.}

\((6R,10S)-12\text{-Bromo}-2,6,10\text{-trimethyl}dodec-2\text{-ene (7).} \) To a stirred solution of compound 6 (800 mg, 3.53 mmol) in pyridine (2.70 cm³) was added TsCl (876 mg, 4.60 mmol) at 0 °C. After the mixture was stirred for 2 h, saturated aqueous solution of citric acid was added. The whole was extracted with \text{Et}_2\text{O} and the extract was washed with \text{H}_2\text{O} and brine, and dried over MgSO₄. The extract was concentrated under reduced pressure after filtration through a short pad of silica gel to give a crude sulfonate. To a stirred solution of the sulfonate in acetone (5 cm³) was added LiBr (1.80 g, 17.7 mmol). After the mixture was stirred for 1 h under reflux, the whole was concentrated under reduced pressure. To the residue was added \text{H}_2\text{O} and the whole was extracted with \text{Et}_2\text{O} and dried over MgSO₄. The filtrate was concentrated under reduced pressure, and the residue was purified by column chromatography to give the title bromide 7 as a colorless oil (923 mg, 90%). \([\alpha]^{25}_D +5.8 \ (c \ 1.00, \ \text{CHCl}_3)\); \(\delta\text{H}(500 \ \text{MHz, CDCl}_3) 0.86 \ (3\ H, \ d, \ J \ 6.9, \ \text{CH}_3), 0.89 \ (3\ H, \ d, \ J \ 6.9, \ \text{CH}_3), 1.04-1.41 \ (9\ H, \ m), 1.61 \ (3\ H, \ s, \ \text{CH}_3), 1.68 \ (3\ H, \ s, \ \text{CH}_3), 1.61-1.68 \ (2\ H, \ m), 1.85-2.03 \ (3\ H, \ m), 3.38-3.49 \ (2\ H, \ m, \ \text{CH}_2), 5.09-5.11 \ (1\ H, \ m, \ \text{CH}); \delta\text{C}(125 \ \text{MHz, CDCl}_3) 17.6, 19.0, 19.6, 24.1, 25.6, 25.7, 31.6, 32.3, 32.4, 36.8, 37.1 (2C), 40.0, 125.0, 131.0. \text{Anal. calcd. for C}_{15}\text{H}_{29}\text{Br: C, 62.28; H, 10.10. Found: C, 62.31; H, 10.32.}

\((6S,10R)-12\text{-Bromo}-2,6,10\text{-trimethyl}dodec-2\text{-ene (ent-7).} \) According to the procedure described for the preparation of 7, compound \textit{ent-}6 (580 mg, 2.56 mmol) was converted into \textit{ent-}7 as a colorless oil (669 mg, 90%). \([\alpha]^{25}_D -5.8 \ (c \ 1.01, \ \text{CHCl}_3)\); \(\delta\text{H}(500 \ \text{MHz, CDCl}_3) 0.86 \ (3\ H, \ d, \ J \ 6.9, \ \text{CH}_3), 0.89 \ (3\ H, \ d, \ J \ 6.9, \ \text{CH}_3), 1.07-1.41 \ (9\ H, \ m), 1.61 \ (3\ H, \ s, \ \text{CH}_3), 1.68 \ (3\ H, \ s, \ \text{CH}_3), 1.61-1.68 (2\ H, \ m), 1.84-2.03 \ (3\ H, \ m), 3.38-3.49 \ (2\ H, \ m, \ \text{CH}_2), 5.09-5.11 \ (1\ H, \ m, \ \text{CH}); \delta\text{C}(125 \ \text{MHz, CDCl}_3) 17.6, 19.0, 19.6, 24.1, 25.6, 25.7, 31.6, 32.2, 32.4, 36.8, 37.1 (2C), 40.0, 125.0, 131.0. \text{Anal. calcd. for C}_{15}\text{H}_{29}\text{Br: C, 62.28; H, 10.10. Found: C, 62.22; H, 10.08.} \)
Triphenyl[(3S,7R)-3,7,11-trimethyldodec-10-en-1-yl]phosphonium Bromide (8). A mixture of the bromide 7 (757 mg, 2.61 mmol) and PPh₃ (750 mg, 2.87 mmol) was heated to 100 °C and the mixture was stirred for 15 h. After cooling, Et₂O was added to the mixture and the resulting white precipitate was washed with Et₂O to remove the excess PPh₃. The residue was dried under vacuum to give the title phosphonium salt 8, which was used without further purification.

Triphenyl[(3R,7S)-3,7,11-trimethyldodec-10-en-1-yl]phosphonium Bromide (ent-8). According to the procedure described for the preparation of 8, compound ent-7 (600 mg, 2.07 mmol) was converted into ent-8.

Benzyl (R)-6-(Benzyl oxy)-2,5,7,8-tetramethylchromane-2-carboxylate (S6). To a stirred solution of (R)-Trolox (S5, 1.90 g, 7.59 mmol) in DMF (13.0 cm³) were added K₂CO₃ (8.38 g, 60.8 mmol) and BnBr (3.61 cm³, 30.4 mmol) at room temperature. After the mixture was stirred overnight, the reaction was quenched with H₂O. The whole was extracted with Et₂O and the extract was washed with H₂O and brine, and dried over MgSO₄. The filtrate was concentrated under reduced pressure, and the residue was purified by column chromatography to give the title compound S6 as a colorless oil (3.24 g, 99%). [α]²⁵D +38.6 (c 1.05, CHCl₃); IR (neat) νmax/cm⁻¹: 1731 (C=O); δH(500 MHz, CDCl₃) 1.64 (3H, s, CH₃), 1.83-1.91 (1H, m, CH₂), 2.09 (3H, s, CH₃), 2.15 (3H, s, CH₃), 2.23 (3H, s, CH₃), 2.38-2.48 (2H, m, CH₂), 2.54-2.62 (1H, m, CH₂), 4.68 (2H, dd, J₁ 12.6, J₂ 11.5, CH₂), 7.09-7.51 (10H, m, Ar); δC(125 MHz, CDCl₃) 11.8, 11.9, 12.9, 20.9, 25.5, 30.6, 66.6, 74.7, 77.2, 117.3, 123.0, 126.0, 127.6 (2C), 127.7 (2C), 127.8, 128.0, 128.2, 128.4 (2C), 128.5 (2C), 135.7, 137.9, 148.0, 148.9, 173.7. HRMS (ESI) calcd for C₂₈H₃₀NaO₄ (MNa⁺): 453.2036; found: 453.2026.

Benzyl (S)-6-(Benzyl oxy)-2,5,7,8-tetramethylchromane-2-carboxylate (ent-S6). According to the procedure described for the preparation of S6, (S)-Trolox (ent-S5, 2.5 g, 10.0 mmol) was converted into ent-S6 as a colorless oil (4.28 g, 99%). [α]²⁵D –38.4 (c 1.27, CHCl₃); IR (neat) νmax/cm⁻¹: 1731 (C=O); δH(500 MHz, CDCl₃) 1.64 (3H, s, CH₃), 1.83-1.91 (1H, m, CH₂), 2.09 (3H, s, CH₃), 2.15 (3H, s, CH₃), 2.23 (3H, s, CH₃), 2.38-2.48 (2H, m, CH₂), 2.54-2.62 (1H, m, CH₂), 4.68 (2H, dd, J₁ 13.2, J₂ 11.2 Hz, CH₂), 5.04 (1H, d, J 12.6, CH₂), 5.15 (1H, d, J 12.6, CH₂), 7.09-7.51 (10H, m, Ar); δC(125 MHz, CDCl₃) 11.8, 11.9, 12.9, 20.9, 25.5, 30.6, 66.6, 74.7, 77.2, 117.3, 123.0, 126.0, 127.6 (2C), 127.7 (2C), 127.8, 128.0, 128.2, 128.4 (2C), 128.5 (2C), 135.7, 137.9, 148.0, 148.9, 173.7. HRMS (ESI) calcd for C₂₈H₃₀NaO₄ (MNa⁺): 453.2036; found: 453.2039.

(R)-6-(Benzyloxy)-2,5,7,8-tetramethylchromane-2-carbaldehyde (9). To a stirred solution of compound S6 (3.24 g, 7.53 mmol) in CH₂Cl₂ (8.2 cm³) was added dropwise DIBAL-H in THF (1.0 M,
15.2 cm³, 15.2 mmol) at –78 °C under argon. After the mixture was stirred for 2 h, saturated aqueous solution of sodium potassium tartrate was added. The whole was extracted with Et₂O and the extract was dried over MgSO₄. Filtration through a short pad of silica gel and concentration under reduced pressure gave a crude alcohol. To a stirred solution of (COCl)₂ (0.881 cm³, 15.2 mmol) in CH₂Cl₂ (60.0 cm³) was slowly added DMSO (1.97 cm³, 30.4 mmol) in CH₂Cl₂ (9.0 cm³) at –78 °C under argon. After the mixture was stirred for 15 min, the above alcohol in CH₂Cl₂ (41.5 cm³) was added dropwise, and stirred for 30 min. Et₃N (6.36 cm³, 45.6 mmol) was slowly added to the mixture and the mixture was warmed to 0 °C. After the mixture was stirred for 30 min, saturated NH₄Cl aq. was added. The whole was extracted with Et₂O, the extract was washed with H₂O and brine, and was dried over Na₂SO₄. The filtrate was concentrated under reduced pressure, and the residue was purified by column chromatography. Recrystallization from CHCl₃/hexane at –20 °C gave the title compound 9 as white solid (1.85 g, 75%): mp 60–61 °C (from CHCl₃/hexane); [α]²⁵D –13.6 (c 0.90, CHCl₃); IR (neat) νmax/cm⁻¹: 1737 (C=O); δH(500 MHz, CDCl₃) 1.41 (3H, s, CH₃), 1.80-1.86 (1H, m, CH₂), 2.13 (3H, s, CH₃), 2.20 (3H, s, CH₃), 2.24 (3H, s, CH₃), 2.25-2.29 (1H, m, CH₂), 2.50-2.63 (2H, m, CH₂), 4.68 (2H, s, CH₂), 7.33-7.50 (5H, m, Ar), 9.64 (1H, d, J 1.1, CHO); δC(125 MHz, CDCl₃) 11.9, 12.0, 12.9, 20.2, 21.6, 27.7, 74.7, 80.4, 117.7, 123.1, 126.4, 127.7 (2C), 127.9, 128.5 (2C), 128.6, 137.7, 147.4, 149.1, 204.4. Anal. calcd. for C₂₁H₂₄O₃: C, 77.75; H, 7.46. Found: C, 77.50; H, 7.49.

(S)-6-(Benzyloxy)-2,5,7,8-tetramethylchroman-2-carbaldehyde (ent-9). According to the procedure described for the preparation of 9, ent-S6 (3.90 g, 9.06 mmol) was converted into ent-9 as white solid (2.10 g, 71%): mp 59–61 °C (from CHCl₃/hexane); [α]²⁵D +12.5 (c 1.00, CHCl₃); IR (neat) νmax/cm⁻¹: 1737 (C=O); δH(500 MHz, CDCl₃) 1.41 (3H, s, CH₃), 1.80-1.86 (1H, m, CH₂), 2.13 (3H, s, CH₃), 2.20 (3H, s, CH₃), 2.24 (3H, s, CH₃), 2.25-2.29 (1H, m, CH₂), 2.50-2.63 (2H, m, CH₂), 4.68 (2H, s, CH₂), 7.34-7.50 (5H, m, Ar), 9.64 (1H, d, J 1.1, CHO); δC(125 MHz, CDCl₃) 11.9, 12.0, 12.9, 20.3, 21.6, 27.7, 74.7, 80.4, 117.7, 123.1, 126.4, 127.7 (2C), 127.9, 128.5 (2C), 128.6, 137.7, 147.4, 149.1, 204.4. Anal. calcd. for C₂₁H₂₄O₃: C, 77.75; H, 7.46. Found: C, 77.52; H, 7.55.

(5R,8S)-2,5,7,8-Tetramethyl-2-((4S,8S)-4,8,12-trimethyltridecyl)chroman-6-ol (10b). To a stirred solution of phosphonium salt 8 (ca. 2.61 mmol) in THF (15.0 cm³) was added LiHMDS (1.0 mol dm⁻³, 2.30 cm³, 2.30 mmol) in THF dropwise at –40 °C under argon. After the mixture was stirred for 30 min, the aldehyde 9 (675 mg, 2.08 mmol) in THF (5.0 cm³) was added to the mixture. The stirring was continued for 30 min at the same temperature and for 1 h at 0 °C. After the reaction was quenched with saturated aqueous solution of NH₄Cl, the whole was extracted with Et₂O and the extract was washed with H₂O and brine, and dried over Na₂SO₄. The filtrate was concentrated under reduced pressure. The oily residue was dissolved in hexane and the solution was filtered through a short pad of silica gel and concentrated down to give crude S7. To a stirred solution of S7 in TBME (9.5 cm³) was added PtO₂ (37.8 mg, 0.167 mmol). The mixture was stirred under an atmosphere of H₂ at room temperature. After
30 min, the reaction mixture was filtrated through Celite and the filtrate was concentrated. To a stirred solution of the residue in MeOH (35.0 cm$^3$) was added 10% Pd/C (222 mg, 0.208 mmol). The mixture was stirred under an atmosphere of H$_2$ at room temperature. After 30 min, the reaction mixture was filtrated through Celite. The filtrate was concentrated under reduced pressure, and the residue was purified by column chromatography to give the title compound 10b as a pale yellow oil (759 mg, 84%).

$[\alpha]^{25}_D$ $-0.7$ (c 1.01, CHCl$_3$); $\delta_H$(500 MHz, CDCl$_3$) 0.83-0.87 (12H, m, 4CH$_3$), 1.02-1.53 (21H, m), 1.23 (3H, s, CH$_3$), 1.73-1.84 (2H, m, CH$_2$), 2.11 (6H, s, 2CH$_3$), 2.16 (3H, s, CH$_3$), 2.60 (2H, t, $J$ 6.9, CH$_2$), 4.17 (1H, s, OH); $\delta_C$(125 MHz, CDCl$_3$) 11.3, 11.8, 12.2, 19.6, 19.7, 20.7, 21.0, 22.6, 22.7, 23.8, 24.4, 24.8, 28.0, 31.5, 32.7, 32.9, 37.3, 37.4 (3C), 39.4, 39.8, 74.5, 117.3, 118.4, 121.0, 122.6, 144.5, 145.5. HRMS (ESI) calcd for C$_{29}$H$_{50}$O$_2$ (M$^+$): 430.3811; found: 430.3806.

(R)-2,5,7,8-Tetramethyl-2-((4S,8S)-4,8,12-trimethyltridecyl)chroman-6-ol (10c). According to the procedure described for the preparation of 10b, compound ent-9 (675 mg, 2.07 mmol) was converted into compound 10c by the reaction with 8 (ca. 2.61 mmol) as a pale yellow oil (573 mg, 57%). $[\alpha]^{25}_D$ $+1.0$ (c 1.10, CHCl$_3$); $\delta_H$(500 MHz, CDCl$_3$) 0.83-0.87 (12H, m, 4CH$_3$), 1.03-1.58 (21H, m), 1.23 (3H, s, CH$_3$), 1.73-1.84 (2H, m, CH$_2$), 2.11 (6H, s, 2CH$_3$), 2.16 (3H, s, CH$_3$), 2.60 (2H, t, $J$ 6.9, CH$_2$), 4.17 (1H, s, OH); $\delta_C$(125 MHz, CDCl$_3$) 11.3, 11.8, 12.2, 19.7 (2C), 20.8, 21.0, 22.6, 22.7, 23.8, 24.4, 24.8, 28.0, 31.4, 32.7, 32.8, 37.3, 37.4 (2C), 37.5, 39.4, 39.8, 74.5, 117.3, 118.4, 121.0, 122.6, 144.5, 145.5. HRMS (ESI) calcd for C$_{29}$H$_{50}$O$_2$ (M$^+$): 430.3811; found: 430.3803.

(S)-2,5,7,8-Tetramethyl-2-((4R,8R)-4,8,12-trimethyltridecyl)chroman-6-ol (10d). According to the procedure described for the preparation of 10b, compound 9 (540 mg, 1.66 mmol) was converted into compound 10d by the reaction with ent-8 (ca. 2.08 mmol) as a pale yellow oil (573 mg, 80%). $[\alpha]^{25}_D$ $-1.0$ (c 1.00, CHCl$_3$); $\delta_H$(500 MHz, CDCl$_3$) 0.83-0.87 (12H, m, 4CH$_3$), 1.03-1.58 (21H, m), 1.23 (3H, s, CH$_3$), 1.73-1.84 (2H, m, CH$_2$), 2.11 (6H, s, 2CH$_3$), 2.16 (3H, s, CH$_3$), 2.60 (2H, t, $J$ 6.9, CH$_2$), 4.17 (1H, s, OH); $\delta_C$(125 MHz, CDCl$_3$) 11.3, 11.8, 12.2, 19.7 (2C), 20.8, 21.0, 22.6, 22.7, 23.8, 24.4, 24.8, 28.0, 31.5, 32.7, 32.9, 37.3, 37.4 (2C), 37.5, 39.4, 39.8, 74.5, 117.3, 118.4, 121.0, 122.6, 144.5, 145.5. HRMS(ESI) calcd for C$_{29}$H$_{50}$O$_2$ (M$^+$): 430.3811; found: 430.3801.

1-[2,4-Bis(benzyloxy)phenyl]ethan-1-one (S9). To a stirred solution of 2,4-dihydroxyacetophenone (S8, 5.00 g, 32.9 mmol) in MeCN (100 cm$^3$) were added K$_2$CO$_3$ (13.6 g, 98.7 mmol) and
BnBr (8.59 cm³, 72.3 mmol). The mixture was stirred under reflux. After the mixture was stirred overnight, the reaction mixture was filtrated through Celite. After concentration, the residue was dissolved in Et₂O. The whole was washed with H₂O and brine, and dried over MgSO₄. The filtrate was concentrated under reduced pressure. Recrystallization from CHCl₃/hexane gave the title compound S9 as colorless crystals (9.5 g, 90%): mp 75–77 °C (from CHCl₃/hexane); IR (neat) ν max/cm⁻¹: 1661 (C=O); δH(500 MHz, CDCl₃) 2.55 (3H, s, CH₃), 5.09 (2H, s, CH₂), 5.11 (2H, s, CH₂), 6.60-6.63 (2H, m, Ar), 7.34-7.44 (10H, m, Ar), 7.85 (1H, dd, J₁ 7.7, J₂ 1.4, Ar); δc(125 MHz, CDCl₃) 32.2, 70.2, 70.7, 100.3, 106.2, 121.7, 127.5 (2C), 127.6 (2C), 128.3 (2C), 128.7 (4C), 132.7, 136.0, 136.1, 160.1, 163.5, 197.8. Anal. calcd. for C₂₂H₂₀O₃: C, 79.50; H, 6.07. Found: C, 79.56; H, 6.13.

1-[2,4-Bis(benzyloxy)phenyl]-2-bromoethan-1-one (11). A solution of phenyltrimethylammonium tribromide (PTT; 4.51 g, 12.0 mmol) in THF (14.6 cm³) was slowly added to compound S9 (4.00 g, 12.0 mmol) at 0 °C. After the mixture was stirred for 2.5 h at room temperature, the reaction was quenched with H₂O. The whole was extracted with Et₂O and the extract was washed with H₂O and brine, and dried over MgSO₄. The filtrate was concentrated under reduced pressure. Recrystallization from CHCl₃/hexane gave the title compound 11 as pink crystals (3.98 g, 80%): mp 78–80 °C (from CHCl₃/hexane); IR (neat) ν max/cm⁻¹: 1671 (C=O); δH(500 MHz, CDCl₃) 4.50 (2H, s, CH₂), 5.09 (2H, s, CH₂), 5.13 (2H, s, CH₂), 6.61 (1H, d, J 2.3, Ar), 6.65 (1H, dd, J₁ 8.6, J₂ 2.3, Ar), 7.34-7.45 (10H, m, Ar), 7.91 (1H, d, J 8.6, Ar); δc(125 MHz, CDCl₃) 37.8, 70.4, 71.1, 100.1, 107.0, 118.3, 127.5 (2C), 127.8 (2C), 128.4, 128.6, 128.7 (2C), 128.9 (2C), 133.9, 135.4, 135.9, 160.0, 164.3. Anal. calcd. for C₂₂H₁₉BrO₃: C, 64.25; H, 4.66. Found: C, 64.32; H, 4.75.

1-(2,4-Dihydroxyphenyl)-2-[(S)-2,5,7,8-tetramethyl-2-[(4(S),8S)-4,8,12-trimethyltridecyl] chroman-6-yl]oxy]ethan-1-one (12b). To a stirred solution of compound 10b (600 mg, 1.39 mmol) in DMF (1.4 cm³) were added K₂CO₃ (385 mg, 2.78 mmol) and compound 11 (745 mg, 1.81 mmol) at room temperature. After the mixture was stirred overnight, the reaction was quenched with H₂O. The whole was extracted with Et₂O and the extract was washed with H₂O and brine, and dried over MgSO₄. The filtrate was concentrated under reduced pressure. To a stirred solution of the residue in EtOAc (15.0 cm³) was added 10% Pd/C (149 mg, 0.140 mmol). The mixture was stirred under an atmosphere of H₂ at room temperature. After the reaction was completed, the reaction mixture was filtrated through Celite. Purification by column chromatography gave the title compound 12b as pale yellow solid (583 mg, 72%): mp 87–88 °C (from EtOAc/hexane); [α]D²⁵ –4.0 (c 1.01, CHCl₃); IR (neat) ν max/cm⁻¹: 3315 (OH), 1627 (C=O); δH(500 MHz, CDCl₃) 0.84-0.87 (12H, m, CH₃), 1.04-1.57 (21H, m), 1.24 (3H, s, CH₃), 1.73-1.85 (2H, m, CH₂), 2.09 (3H, s, CH₃), 2.14 (3H, s, CH₃), 2.18 (3H, s, CH₃), 2.58 (2H, t, J 6.6, CH₂), 4.92 (2H, s, CH₂), 5.80 (1H, br s, OH), 6.36 (1H, dd, J₁ 8.6, J₂ 2.3 Hz, Ar), 6.42 (1H, d, J 2.3, Ar), 7.52 (1H, d, J 7.7, Ar), 12.40 (1H, s, OH); δc(125 MHz, CDCl₃) 11.8, 11.9, 12.8, 19.7 (2C), 20.6, 21.0, 22.6, 22.7, 23.8, 24.4, 24.8, 28.0, 31.2, 32.7, 32.8, 37.3, 37.4 (3C), 39.4, 40.0, 74.0, 75.0,
103.8, 108.0, 111.9, 117.8, 123.2, 125.7, 127.6, 130.9, 147.8, 148.3, 162.8, 165.3, 198.0. *Anal. calcd.* for C$_{37}$H$_{56}$O$_5$: C, 76.51; H, 9.72. Found: C, 76.41; H, 9.89.

1-(2,4-Dihydroxyphenyl)-2-{{[(R)-2,5,7,8-tetramethyl-2-((4S,8S)-4,8,12-trimethyltridecyl)chroman-6-yl]oxy}ethan-1-one (12c). According to the procedure described for the preparation of 12b, compound 10c (100 mg, 0.232 mmol) was converted into compound 12c as pale yellow solid (88.9 mg, 66%): mp 86–87 °C (from EtOAc/hexane); [α]$_D^{25}$ +3.7 (c 0.55, CHCl$_3$); IR (neat) $\nu_{\text{max}}$/cm$^{-1}$: 3351 (OH), 1626 (C=O); δ$_H$(500 MHz, CDCl$_3$) 0.84-0.87 (12H, m, 4CH$_3$), 1.04-1.61 (21H, m), 1.24 (3H, s, CH$_3$), 1.73-1.85 (2H, m, CH$_2$), 2.08 (3H, s, CH$_3$), 2.14 (3H, s, CH$_3$), 2.18 (3H, s, CH$_3$), 2.58 (2H, t, $J$ 6.6, CH$_2$), 4.92 (2H, s, CH$_2$), 6.36 (1H, dd, $J$ 8.9, $J$ 2.6, Ar), 6.42 (1H, d, $J$ 2.3, Ar), 7.51 (1H, d, $J$ 7.7, Ar), 12.40 (1H, s, OH); δ$_C$(125 MHz, CDCl$_3$) 11.8, 11.9, 12.8, 19.7 (2C), 20.6, 21.0, 22.6, 22.7, 23.8, 24.4, 24.8, 28.0, 31.1, 32.7, 32.8, 37.3, 37.4 (2C), 37.5, 39.4, 40.1, 74.0, 75.0, 103.8, 108.1, 111.9, 117.8, 123.2, 125.7, 127.6, 130.9, 147.8, 148.3, 162.9, 165.3, 198.0. *Anal. calcd.* for C$_{37}$H$_{56}$O$_5$: C, 76.51; H, 9.72. Found: C, 76.35; H, 9.79.

1-(2,4-Dihydroxyphenyl)-2-{{[(S)-2,5,7,8-tetramethyl-2-((4R,8R)-4,8,12-trimethyltridecyl)chroman-6-yl]oxy}ethan-1-one (12d). According to the procedure described for the preparation of 12b, compound 10d (500 mg, 1.16 mmol) was converted into compound 12d as pale yellow solid (483 mg, 72%): mp 86–87 °C (from EtOAc/hexane); [α]$_D^{25}$ −3.9 (c 1.11, CHCl$_3$); IR (neat) $\nu_{\text{max}}$/cm$^{-1}$: 3350 (OH), 1626 (C=O); δ$_H$(500 MHz, CDCl$_3$) 0.84-0.87 (12H, m, 4CH$_3$), 1.04-1.61 (21H, m), 1.24 (3H, s, CH$_3$), 1.73-1.85 (2H, m, CH$_2$), 2.08 (3H, s, CH$_3$), 2.14 (3H, s, CH$_3$), 2.18 (3H, s, CH$_3$), 2.58 (2H, t, $J$ 6.9, CH$_2$), 4.92 (2H, s, CH$_2$), 6.36 (1H, dd, $J$ 8.9, $J$ 2.6, Ar), 6.42 (1H, d, $J$ 2.9, Ar), 7.51 (1H, d, $J$ 9.2, Ar), 12.40 (1H, s, OH); δ$_C$(125 MHz, CDCl$_3$) 11.8, 11.9, 12.8, 19.7 (2C), 20.6, 21.0, 22.6, 22.7, 23.8, 24.4, 24.8, 28.0, 31.1, 32.7, 32.8, 37.3, 37.4 (2C), 37.5, 39.4, 40.1, 74.0, 75.0, 103.8, 108.1, 111.8, 117.8, 123.2, 125.7, 127.6, 130.9, 147.8, 148.3, 162.9, 165.3, 198.0. HRMS (ESI) calcd for C$_{37}$H$_{57}$O$_5$ (MH$^+$): 581.4201; found: 581.4207.

7-Hydroxy-3-{{[(S)-2,5,7,8-tetramethyl-2-((4S,8S)-4,8,12-trimethyltridecyl)chroman-6-yl]oxy-}4H-chromen-4-one (1b: *ent*-NP843). To a stirred solution of compound 12b (100 mg, 0.172 mmol) in THF (0.76 cm$^3$) was added DMF-DMA (0.0277 cm$^3$, 0.260 mmol) under argon, and the mixture was stirred under reflux for 4 h. Then, aqueous 1N HCl (1 cm$^3$) and MeOH (1 cm$^3$) were added and the mixture was stirred overnight. The whole was extracted with Et$_2$O and dried over Na$_2$SO$_4$. The filtrate was concentrated under reduced pressure, and the residue was purified by column chromatography to give the title compound 1b as white solid (23.3 mg, 23%): mp 228–229 °C (from
EtOAc); [α]$^\text{D}_2$ –5.3 (c 1.00, CHCl$_3$); IR (neat) $\nu_{\text{max}}$/cm$^{-1}$: 3133 (OH); δH(500 MHz, CDCl$_3$) 0.84-0.87 (12H, m, 4CH$_3$), 1.03-1.59 (21H, m), 1.25 (3H, s, CH$_3$), 1.75-1.85 (2H, m, CH$_2$), 2.02 (3H, s, CH$_3$), 2.06 (3H, s, CH$_3$), 2.09 (3H, s, CH$_3$), 2.58 (2H, t, $J$ 6.9, CH$_2$), 6.86 (1H, d, $J$ 2.3, Ar), 7.07-7.10 (2H, m, Ar), 8.22 (1H, d, $J$ 9.2, Ar), 8.55 (1H, br s, OH); δc(125 MHz, CDCl$_3$) 11.8 (2C), 12.6, 19.6, 19.7, 20.6, 21.0, 22.6, 22.7, 23.4, 24.8, 28.0, 31.2, 32.7, 32.8, 37.3, 37.4 (3C), 39.4, 40.0, 75.2, 102.7, 115.5, 117.0, 118.2, 123.7, 125.3, 127.1, 127.6, 140.0, 142.8, 143.4, 149.0, 157.8, 162.1, 172.6. Anal. calcd. for C$_{38}$H$_{54}$O$_5$: C, 77.25; H, 9.21. Found: C, 77.22; H, 9.43.

7-Hydroxy-3-[(R)-2,5,7,8-tetramethyl-2-((4S,8R)-4,8,12-trimethyltridecyl)chroman-6-yl]oxy]-4H-chromen-4-one (1a, NP843). mp 228–229 °C (from EtOAc); [α]$^\text{D}_2$ +5.0 (c 1.00, CHCl$_3$); IR (neat) $\nu_{\text{max}}$/cm$^{-1}$: 3163 (OH); δH(500 MHz, CDCl$_3$) 0.84-0.87 (12H, m, 4CH$_3$), 1.03-1.64 (21H, m), 1.25 (3H, s, CH$_3$), 1.75-1.85 (2H, m, CH$_2$), 2.58 (2H, t, $J$ 6.9, CH$_2$), 6.86 (1H, d, $J$ 2.3, Ar), 7.07-7.10 (2H, m, Ar), 8.22 (1H, d, $J$ 9.2, Ar), 8.67 (1H, br s, OH); δc(125 MHz, CDCl$_3$) 11.8 (2C), 12.6, 19.6, 19.7, 20.6, 21.0, 22.6, 22.7, 23.7, 24.4, 24.8, 28.0, 31.2, 32.7, 32.8, 37.3, 37.4 (3C), 39.4, 40.0, 75.3, 102.7, 115.5, 117.0, 118.2, 123.7, 125.3, 127.1, 127.6, 140.0, 142.8, 143.4, 149.0, 157.8, 162.2, 172.6. Anal. calcd. for C$_{38}$H$_{54}$O$_5$: C, 77.25; H, 9.21. Found: C, 77.13; H, 9.40.

7-Hydroxy-3-[(S)-2,5,7,8-tetramethyl-2-((4R,8S)-4,8,12-trimethyltridecyl)chroman-6-yl]oxy]-4H-chromen-4-one (1c). According to the procedure described for the preparation of 1b, compound 1c (50.0 mg, 0.0861 mmol) was converted into compound 1c as white solid (12.2 mg, 24%): mp 229–231 °C (from EtOAc); [α]$^\text{D}_2$ +5.4 (c 1.01, CHCl$_3$); IR (neat) $\nu_{\text{max}}$/cm$^{-1}$: 3125 (OH); δH(500 MHz, CDCl$_3$) 0.84-0.87 (12H, m, 4CH$_3$), 1.03-1.64 (21H, m), 1.25 (3H, s, CH$_3$), 1.75-1.85 (2H, m, CH$_2$), 2.58 (2H, t, $J$ 6.9, CH$_2$), 6.86 (1H, d, $J$ 2.3, Ar), 7.07-7.10 (2H, m, Ar), 8.57 (1H, br s, OH); δc(125 MHz, CDCl$_3$) 11.8 (2C), 12.6, 19.6, 19.7, 20.6, 21.0, 22.6, 22.7, 23.7, 24.4, 24.8, 28.0, 31.1, 32.7, 32.8, 37.3, 37.4 (3C), 39.4, 40.0, 75.3, 102.7, 115.5, 117.0, 118.2, 123.7, 125.3, 127.1, 127.6, 140.0, 142.8, 143.4, 149.0, 157.8, 162.1, 172.6. Anal. calcd. for C$_{38}$H$_{54}$O$_5$·0.15EtOAc: C, 76.75; H, 9.21. Found: C, 77.13; H, 9.40.

7-Hydroxy-3-[(S,R)-2,5,7,8-tetramethyl-2-((4S,8R)-4,8,12-trimethyltridecyl)chroman-6-yl]oxy]-4H-chromen-4-one (1d). According to the procedure described for the preparation of 1b, compound 1d (100 mg, 0.172 mmol) was converted into compound 1d as white solid (23.5 mg, 23%): mp 229–231 °C (from EtOAc); [α]$^\text{D}_2$ +5.4 (c 0.95, CHCl$_3$); IR (neat) $\nu_{\text{max}}$/cm$^{-1}$: 3126 (OH); δH(500 MHz, CDCl$_3$) 0.84-0.87 (12H, m, 4CH$_3$), 1.03-1.64 (21H, m), 1.25 (3H, s, CH$_3$), 1.75-1.85 (2H, m, CH$_2$), 2.02 (3H, s, CH$_3$), 2.06 (3H, s, CH$_3$), 2.58 (2H, t, $J$ 6.9, CH$_2$), 6.86 (1H, d, $J$ 2.3, Ar), 7.07-7.10 (2H, m, Ar), 8.57 (1H, br s, OH); δc(125 MHz, CDCl$_3$) 11.8 (2C), 12.6, 19.7 (2C), 20.6, 21.0, 22.6, 22.7, 23.7, 24.4, 24.8, 28.0, 31.1, 32.7, 32.8, 37.3, 37.4 (3C), 39.4, 40.0, 75.3, 102.7, 115.5, 117.0, 118.3, 123.7, 125.3, 127.1, 127.6, 140.0, 142.8, 143.4, 149.1, 157.8, 162.1, 172.6. Anal. calcd. for C$_{38}$H$_{54}$O$_5$: C, 77.25; H, 9.21. Found: C, 77.40; H, 9.21.
Synthesis of Compounds 13a,b

(R)-(3,7-Dimethyl-6-en-1-yl)triphenylphosphonium Bromide (S10). According to the procedure described for the preparation of 8, (R)-citronellyl bromide (3.99 g, 18.2 mmol) was converted into compound S10, which was used without further purification.

(R)-2-((S)-4,8-Dimethylnonyl)-2,5,7,8-tetramethylchroman-6-ol (S11a). According to the procedure described for the preparation of 10b, compound ent-9 (800 mg, 2.47 mmol) was converted into compound S11a using the phosphonium salt S10 (ca. 4.93 mmol) as a pale yellow oil (735 mg, 83%). \([\alpha]_D^{25} +0.4 \quad (c \ 1.00, \ \text{CHCl}_3)\); \(\delta_H(500 \ \text{MHz}, \ \text{CDCl}_3) \ 0.83-0.88 \quad (9H, \ \text{m}, \ \text{CH}_3), \ 1.05-1.53 \quad (14H, \ \text{m}), \ 1.23 \quad (3H, \ \text{s}, \ \text{CH}_3), \ 1.73-1.84 \quad (2H, \ \text{m}, \ \text{CH}_2), \ 2.11 \quad (6H, \ \text{s}, \ 2\text{CH}_3), \ 2.16 \quad (3\text{H}, \ \text{s}, \ \text{CH}_3), \ 2.60 \quad (2H, \ t, \ J \ 6.9, \ \text{CH}_2), \ 4.18 \quad (1H, \ \text{s}, \ \text{OH}); \ \delta_C(125 \ \text{MHz}, \ \text{CDCl}_3) \ 11.3, \ 11.8, \ 12.2, \ 19.7 \quad (2C), \ 21.0, \ 22.6, \ 22.7, \ 23.8, \ 24.7, \ 28.0, \ 31.5, \ 32.7, \ 37.2, \ 37.5, \ 39.3, \ 39.8, \ 74.5, \ 117.3, \ 118.4, \ 121.0, \ 122.6, \ 144.5, \ 145.5.\ \text{HRMS (ESI) c}alcd \quad \text{for} \ \text{C}_{24}\text{H}_{40}\text{O}_2 \quad (\text{M}^+) \ : \ 360.3028; \ \text{found:} \ 360.3026.

(S)-2-((S)-4,8-Dimethylnonyl)-2,5,7,8-tetramethylchroman-6-ol (S11b). According to the procedure described for the preparation of 10b, compound 9 (800 mg, 2.47 mmol) was converted into compound S11b using phosphonium salt S10 (ca. 4.93 mmol) as a pale yellow oil (735 mg, 82%). \([\alpha]_D^{25} +1.5 \quad (c \ 1.00, \ \text{CHCl}_3)\); \(\delta_H(500 \ \text{MHz}, \ \text{CDCl}_3) \ 0.83-0.88 \quad (9H, \ \text{m}, \ 3\text{CH}_3), \ 1.04-1.53 \quad (14H), \ 1.23 \quad (3\text{H}, \ \text{s}, \ \text{CH}_3), \ 1.73-1.84 \quad (2\text{H}), \ 2.11 \quad (6\text{H}, \ \text{s}, \ \text{CH}_3), \ 2.16 \quad (3\text{H}, \ \text{s}, \ \text{CH}_3), \ 2.60 \quad (2\text{H}, \ t, \ J \ 6.9, \ \text{CH}_2), \ 4.18 \quad (1\text{H}, \ \text{s}, \ \text{OH}); \ \delta_C(125 \ \text{MHz}, \ \text{CDCl}_3) \ 11.3, \ 11.8, \ 12.2, \ 19.6, \ 20.7, \ 21.0, \ 22.6, \ 22.7, \ 23.8, \ 24.8, \ 28.0, \ 31.5, \ 32.7, \ 37.3, \ 37.5, \ 39.3, \ 39.7, \ 74.5, \ 117.3, \ 118.4, \ 121.0, \ 122.6, \ 144.5, \ 145.5.\ \text{HRMS (ESI) c}alcd \quad \text{for} \ \text{C}_{24}\text{H}_{40}\text{O}_2 \quad (\text{M}^+) \ : \ 360.3028; \ \text{found:} \ 360.3035.

1-(2,4-Dihydoxyphenyl)-2-([(R)-2-((S)-4,8-dimethylnonyl)-2,5,7,8-tetramethylchroman-6-yl]-oxy)ethan-1-one (S12a). According to the procedure described for the preparation of 12b, compound S11a (700 mg, 1.94 mmol) was converted into compound S12a as a pale yellow solid (916 mg, 84%): mp 89–91 °C (from EtOAc/hexane); \([\alpha]_D^{25} +3.8 \quad (c \ 0.70, \ \text{CHCl}_3)\); IR (neat) \(v_{\text{max}}/\text{cm}^{-1}: \ 3341 \quad (\text{OH}), \ 1626 \quad (\text{C}=\text{O}); \ \delta_H(500 \ \text{MHz}, \ \text{CDCl}_3) \ 0.84-0.87 \quad (9\text{H}, \ \text{m}, \ \text{CH}_3), \ 1.06-1.61 \quad (14\text{H}, \ \text{m}), \ 1.24 \quad (3\text{H}, \ \text{s}, \ \text{CH}_3), \ 1.73-1.85 \quad (2\text{H}, \ \text{m}, \ \text{CH}_2), \ 2.09 \quad (3\text{H}, \ \text{s}, \ \text{CH}_3), \ 2.14 \quad (3\text{H}, \ \text{s}, \ \text{CH}_3), \ 2.18 \quad (3\text{H}, \ \text{s}, \ \text{CH}_3), \ 2.57 \quad (2\text{H}, \ t, \ J \ 6.6, \ \text{CH}_2), \ 4.93 \quad (2\text{H}, \ \text{s}, \ \text{CH}_2), \ 6.37 \quad (1\text{H}, \ \text{dd}, \ J_1 \ 8.9, \ J_2 \ 2.6, \ \text{Ar}), \ 6.42 \quad (1\text{H}, \ d, \ J \ 2.9, \ \text{Ar}), \ 7.49 \quad (1\text{H}, \ d, \ J \ 8.6, \ \text{Ar}), \ 12.37 \quad (1\text{H}, \ \text{s}, \ \text{OH}); \ \delta_C(125 \ \text{MHz}, \ \text{CDCl}_3) \ 11.8, \ 11.9, \ 12.8, \ 19.6, \ 20.6, \ 21.0, \ 22.6, \ 22.7, \ 23.8, \ 24.7, \ 28.0,
31.1, 32.7, 37.2, 37.5, 39.3, 40.0, 73.9, 75.0, 103.8, 108.2, 111.7, 117.8, 123.2, 125.7, 127.6, 130.8, 147.7, 148.3, 163.2, 165.3, 198.0. HRMS (ESI) calcd for C₃₂H₄₇O₅ (MH⁺): 511.3418; found: 511.3414.

1-(2,4-Dihydroxyphenyl)-2-{(S)-2-((S)-4,8-dimethylnonyl)-2,5,7,8-tetramethylchroman-6-yl|oxy}ethan-1-one (S₁₂b). According to the procedure described for the preparation of S₁₂b, compound S₁₁b (700 mg, 1.94 mmol) was converted into compound S₁₂b as a pale yellow solid (766 mg, 79%): mp 89–91 °C (from EtOAc/hexane); [α]₂₅D –3.7 (c 0.62, CHCl₃); IR (neat) ν_max/cm⁻¹: 3363 (OH), 1626 (C=O); δ_H(500 MHz, CDCl₃) 0.84-0.87 (9H, m, 3CH₃), 1.03-1.60 (14H, m), 1.24 (3H, s, CH₃), 1.73-1.85 (2H, m, CH₂), 2.09 (3H, s, CH₃), 2.14 (3H, s, CH₃), 2.18 (3H, s, CH₃), 2.57 (2H, t, J 6.6, CH₂), 4.93 (2H, s, CH₂), 6.26 (s, 1H), 6.37 (1H, dd, J₁ 9.2, J₂ 2.3, Ar), 6.42 (1H, d, J 2.9, Ar), 7.50 (1H, s, CH₃), 12.38 (1H, s, OH); δ_C(125 MHz, CDCl₃) 11.8, 11.9, 12.8, 19.6, 20.6, 21.0, 22.6, 22.7, 23.8, 24.7, 28.0, 31.2, 32.7, 37.3, 37.5, 39.3, 40.0, 73.9, 75.0, 103.8, 108.2, 111.7, 117.8, 123.2, 125.7, 127.6, 130.9, 147.7, 148.3, 163.1, 165.3, 198.0. HRMS (ESI) calcd for C₃₂H₄₇O₅ (MH⁺): 511.3418; found: 511.3419.

3-{(R)-2-((S)-4,8-Dimethylnonyl)-2,5,7,8-tetramethylchroman-6-yl|oxy}-7-hydroxy-4H-chromen-4-one (13a). According to the procedure described for the preparation of 1b, compound S₁₂a (100 mg, 0.196 mmol) was converted into compound 13a as a white solid (20.5 mg, 20%): mp 239–241 °C (from EtOAc); [α]₂₅D +5.4 (c 0.33, CHCl₃); IR (neat) ν_max/cm⁻¹: 3116 (OH); δ_H(500 MHz, CDCl₃) 0.84-0.87 (9H, m, 3CH₃), 1.06-1.61 (14H, m), 1.75-1.85 (2H, m, CH₂), 2.02 (3H, s, CH₃), 2.05 (3H, s, CH₃), 2.09 (3H, s, CH₃), 2.58 (2H, t, J 6.7, CH₂), 6.86 (1H, d, J 2.3, Ar), 7.07-7.10 (2H, m, Ar), 8.22 (1H, d, J 8.7, Ar), 8.59 (1H, br s, OH); δ_C(125 MHz, CDCl₃) 11.8 (2C), 12.7, 19.6, 20.6, 21.0, 22.6, 22.7, 23.8, 24.7, 28.0, 31.1, 32.7, 37.2, 37.5, 39.3, 39.9, 75.3, 102.7, 115.5, 117.0, 118.3, 123, 125.3, 127.1, 127.6, 140.0, 142.8, 143.4, 149.0, 157.8, 162.1, 172.6. Anal. calcd. for C₃₃H₄₄O₅·0.25EtOAc: C, 75.24; H, 8.54. Found: C, 75.08; H, 8.60.

3-{(S)-2-((S)-4,8-Dimethylnonyl)-2,5,7,8-tetramethylchroman-6-yl|oxy}-7-hydroxy-4H-chromen-4-one (13b). According to the procedure described for the preparation of 1b, compound S₁₂b (100 mg, 0.196 mmol) was converted into compound 13b as a white solid (23.4 mg, 23%): mp 239–241 °C (from EtOAc); [α]₂₅D –5.6 (c 0.39, CHCl₃); IR (neat) ν_max/cm⁻¹: 3131 (OH); δ_H(500 MHz, CDCl₃) 0.86 (9H, d, J 6.3, 3CH₃), 1.05-1.59 (14H, m), 1.25 (3H, s, CH₃), 1.76-1.84 (2H, m, CH₂), 2.02 (3H, s, CH₃), 2.06 (3H, s, CH₃), 2.09 (3H, s, CH₃), 2.58 (2H, t, J 6.9, CH₂), 6.85 (1H, d, J 1.7, Ar), 7.07 (1H, dd, J₁ 9.2, J₂ 2.3, Ar), 7.09 (1H, s, Ar), 8.22 (1H, d, J 8.6, Ar), 8.38 (1H, br s, OH); δ_C(125 MHz, CDCl₃) 11.8 (2C), 12.6, 19.6, 20.6, 21.0, 22.6, 22.7, 23.7, 24.7, 28.0, 31.2, 32.7, 37.3, 37.5, 39.3, 39.9, 75.3, 102.7, 115.7, 118.3, 123.6, 125.3, 127.1, 127.4, 140.2, 142.8, 143.3, 149.0, 157.9, 162.5, 172.7. Anal. calcd. for C₃₃H₄₄O₅·0.1EtOAc: C, 75.76; H, 8.53. Found: C, 75.65; H, 8.58.
Synthesis of Compounds 14a,b

(Isopentyl)triphenylphosphonium Bromide (S14). According to the procedure described for the preparation of 8, 1-bromo-3-methylbutane (S13, 1.99 g, 13.2 mmol) was converted into compound S14, which was used without further purification.

(R)-2,5,7,8-Tetramethyl-2-(4-methylpentyl)chroman-6-ol (S15a). According to the procedure described for the preparation of 10b, compound ent-9 (800 mg, 2.47 mmol) was converted into compound S15a using the phosphonium salt S14 (ca. 4.93 mmol) as a pale yellow oil (675 mg, 94%). [α]25D –0.9 (c 1.00, CHCl3); δH(500 MHz, CDCl3) 0.87 (6H, d, J6.9, 2CH3), 1.14-1.19 (2H, m), 1.22 (3H, s, CH3), 1.37-1.58 (5H, m), 1.73-1.84 (2H, m, CH2), 2.11 (6H, s, 2CH3), 2.16 (3H, s, CH3), 2.60 (2H, t, J6.9, CH2), 4.17 (1H, s, OH); δc(125 MHz, CDCl3) 11.3, 11.8, 12.2, 20.7, 21.4, 22.6 (2C), 23.8, 27.9, 31.5, 39.4, 39.7, 74.5, 117.3, 118.4, 121.0, 122.6, 144.5, 145.5. HRMS (ESI) calcd for C19H30O2 (M+): 290.2246; found: 290.2245.

(S)-2,5,7,8-Tetramethyl-2-(4-methylpentyl)chroman-6-ol (S15b). According to the procedure described for the preparation of 10b, compound 9 (800 mg, 2.47 mmol) was converted into compound S15b using compound S14 (ca. 4.93 mmol) as a pale yellow oil (621 mg, 86%). [α]25D +0.9 (c 1.00, CHCl3); δH(500 MHz, CDCl3) 0.87 (6H, d, J6.9, 2CH3), 1.14-1.19 (2H, m), 1.22 (3H, s, CH3), 1.37-1.58 (5H, m), 1.73-1.84 (2H, m, CH2), 2.11 (6H, s, 2CH3), 2.16 (3H, s, CH3), 2.60 (2H, t, J6.9, CH2), 4.17 (1H, s, OH); δc(125 MHz, CDCl3) 11.3, 11.8, 12.2, 20.7, 21.4, 22.6 (2C), 23.8, 27.9, 31.5, 39.4, 39.7, 74.5, 117.3, 118.4, 121.0, 122.6, 144.5, 145.5. HRMS (ESI) calcd for C19H30O2 (M+): 290.2246; found: 290.2239.

(R)-1-(2,4-Dihydroxyphenyl)-2-[[2,5,7,8-tetramethyl-2-(4-methylpentyl)chroman-6-yl]oxy]ethan-1-one (S16a). According to the procedure described for the preparation of 12b, compound S15a (600 mg, 2.07 mmol) was converted into compound S16a as a pale yellow solid (765 mg, 84%): mp 86–87 °C (from EtOAc/hexane); [α]25D +3.5 (c 0.78, CHCl3); IR (neat) νmax/cm⁻¹: 3351 (OH), 1626 (C=O); δH(500 MHz, CDCl3) 0.87 (6H, d, J6.9, 2CH3), 1.18 (2H, dd, J1 14.9, J2 6.9, CH2), 1.24 (3H, s, CH3), 1.37-1.61 (5H, m), 1.73-1.85 (2H, m, CH2), 2.08 (3H, s, CH3), 2.14 (3H, s, CH3), 2.18 (3H, s, CH3), 2.57 (2H, t, J 6.6, CH2), 4.93 (2H, s, CH2), 6.36 (1H, dd, J1 8.9, J2 2.3, Ar), 6.42 (1H, d, J 2.3, Ar), 6.52 (1H, br s, OH), 7.49 (1H, d, J 9.2 Hz, Ar), 12.37 (1H, s, OH); δc(125 MHz, CDCl3) 11.8, 11.9, 12.8, 20.6, 21.3, 22.6 (2C), 23.8, 27.8, 31.1, 39.3, 39.9, 73.9, 75.0, 103.8, 108.1, 111.7, 117.8, 123.2, 125.7, 127.6, 130.8, 147.7, 148.3, 163.3, 165.3, 198.0. HRMS (ESI) calcd for C27H37O5 (MH+): 441.2636; found: 441.2633.
(S)-1-(2,4-Dihydroxyphenyl)-2-[[2,5,7,8-tetramethyl-2-(4-methylpentyl)chroman-6-yl]oxy]ethan-1-one (S16b). According to the procedure described for the preparation of 12b, compound S15b (600 mg, 2.07 mmol) was converted into compound S16b as pale yellow solid (505 mg, 55%): mp 86–87 °C (from EtOAc/hexane); [α]D 25 –3.9 (c 1.11, CHCl3); IR (neat) νmax/cm⁻¹: 3351 (OH), 1626 (C=O); δH(500 MHz, CDCl3) 0.87 (6H, d, J 6.9, 2CH3), 1.17 (2H, dd, J1 14.9, J2 6.9, CH2), 1.24 (3H, s, CH3), 1.37-1.61 (5H, m), 1.73-1.85 (2H, m, CH2), 2.09 (3H, s, CH3), 2.14 (3H, s, CH3), 2.18 (3H, s, CH3), 2.57 (2H, t, J 6.6, CH2), 4.93 (2H, s, CH2), 6.28 (1H, br s, OH), 6.37 (1H, dd, J 8.9, J 2.3, Ar), 6.42 (1H, d, J 2.3, Ar), 7.50 (1H, d, J 9.2 Hz, Ar), 12.38 (1H, s, OH); δC(125 MHz, CDCl3) 11.8, 11.9, 12.8, 20.6, 21.3, 22.6 (2C), 23.8, 27.8, 31.1, 39.3, 39.9, 73.9, 75.0, 103.8, 108.1, 111.7, 117.8, 123.2, 125.7, 127.6, 130.8, 147.7, 148.3, 163.1, 165.3, 198.0. Anal. calcd. for C27H36O5: C, 73.61; H, 8.24. Found: C, 73.39; H, 8.49.

(R)-7-Hydroxy-3-[[2,5,7,8-tetramethyl-2-(4-methylpentyl)chroman-6-yl]oxy]-4H-chromen-4-one (14a). According to the procedure described for the preparation of 1b, compound S16a (100 mg, 0.227 mmol) was converted into compound 14a as white solid (23.0 mg, 23%): mp 255–258 °C (from EtOAc); [α]D 25 +5.3 (c 0.40, THF); IR (neat) νmax/cm⁻¹: 3265 (OH); δH(500 MHz, THF-d8) 2.69 (3H, d, J 2.9, CH3), 2.70 (3H, d, J 2.9, CH3), 3.01 (2H, dd, J1 14.9, J2 6.9, CH2), 3.06 (3H, s, CH3), 3.26-3.43 (5H, m), 3.56-3.68 (2H, m, CH2), 3.82 (3H, s, CH3), 3.86 (3H, s, CH3), 3.89 (3H, s, CH3), 4.44 (2H, t, J 6.9, CH2), 8.47 (1H, d, J 2.3, Ar), 8.64 (1H, dd, J 8.6, J 2.3, Ar), 8.84 (1H, s, Ar), 9.87 (1H, d, J 8.6, Ar), 11.19 (1H, s, OH); δC(125 MHz, THF-d8) 10.9, 11.1, 11.8, 20.4, 21.4, 22.0 (2C), 23.2, 27.9, 31.1, 39.5, 39.9, 74.9, 101.9, 114.1, 117.5, 118.0, 123.2, 125.2, 126.9, 127.3, 139.2, 143.4, 143.8, 148.9, 157.6, 162.4, 170.0. Anal. calcd. for C28H34O5·0.1EtOAc: C, 74.25; H, 7.64. Found: C, 74.12; H, 7.61.

(S)-7-Hydroxy-3-[[2,5,7,8-tetramethyl-2-(4-methylpentyl)chroman-6-yl]oxy]-4H-chromen-4-one (14b). According to the procedure described for the preparation of 1b, compound S16b (100 mg, 0.227 mmol) was converted into compound 14b as white solid (22.7 mg, 22%): mp 255–258 °C (from EtOAc); [α]D 25 –5.1 (c 0.52, THF); IR (neat) νmax/cm⁻¹: 3147 (OH); δH(500 MHz, THF-d8) 2.69 (3H, d, J 2.9, CH3), 2.70 (3H, d, J 2.9, CH3), 3.01 (2H, dd, J1 14.9, J2 6.9, CH2), 3.06 (3H, s, CH3), 3.26-3.43 (5H, m), 3.56-3.68 (2H, m, CH2), 3.82 (3H, s, CH3), 3.86 (3H, s, CH3), 3.89 (3H, s, CH3), 4.44 (2H, t, J 6.9, CH2), 8.47 (1H, d, J 2.3, Ar), 8.64 (1H, dd, J 8.6, J 2.3, Ar), 8.84 (1H, s, Ar), 9.87 (1H, d, J 8.6, Ar), 11.20 (1H, s, OH); δC(125 MHz, THF-d8) 10.9, 11.1, 11.8, 20.4, 21.4, 22.0 (2C), 23.2, 27.9, 31.1, 39.5, 39.9, 74.9, 101.9, 114.1, 117.5, 118.0, 123.2, 125.2, 126.9, 127.3, 139.2, 143.4, 143.8, 148.9, 157.6, 162.4, 170.0. Anal. calcd. for C28H34O5: C, 74.64; H, 7.64. Found: C, 74.33; H, 7.74.

Screening by the Chemical Array
Photoaffinity linker-coated (PALC) slides were prepared according to previous reports using amine-coated slides and the photoaffinity proline linker. A solution of compounds (2.5 mg cm⁻³ in DMSO) from the in-house chemical library (NPDepo, RIKEN) was immobilized onto the PALC glass slides with a chemical arrayer equipped with 24 stamping pins. The slides were exposed to UV irradiation of 4 J cm⁻² at 365 nm using a CL-1000L UV crosslinker (UVP, CA). The slides were washed successively with DMSO, DMF, acetonitrile, THF, dichloromethane, EtOH, and ultra-pure water (5 min, 3 times each), and dried. D- or L-MDM2TMR (3 µmol dm⁻³ in 1% skim-milk-TBS-T) was incubated with the glass slide for 1 h, and then washed with TBS-T (10 mmol dm⁻³ Tris-HCl (pH 8.0), 150 mmol dm⁻³ NaCl, 0.05% Tween-20) (5 min, 3 times). The slides were dried and scanned at 532 nm on a GenePix scanner. The fluorescence signals were quantified with GenePixPro (Table S1).

**Fluorescent Polarization Assay**

Fluorescence polarization (FP) assays were carried out in PBS containing 2% DMSO and 0.005% Tween-20 using a fluorescein-labeled p53 (P4) peptide (0.5-1.0 nmol dm⁻³) and MDM2²⁵⁻¹⁰⁹ (10 nmol dm⁻³) in black 96-well non-binding surface assay plates (Corning). The potential inhibitors and fluorescein-labeled P4 peptide in DMSO were diluted five-fold with PBS in advance. The protein (0.090 cm³) was preincubated with the compound solution (0.005 cm³) for 30 min. Then, the fluorescein-labeled P4 peptide (0.005 cm³) was added and incubated for 30 min. The P4 peptide was used as the positive control. FP signals were analyzed using an EnVision Xcite plate reader (Perkin Elmer) with a 480-nm excitation filter and a 535-nm emission filter. The mP values of the assay were calculated according to the report by Czarna et al (Table 1, Table S2, Figure S6).

**SPR Analysis**

SPR Analyses of p53 binding to synthetic MDM2²⁵⁻¹⁰⁹ and MDM2TMR were carried out using Biacore T200 SPR instrument. PBS (Nacalai Tesque, pH 7.4) containing 0.05 % Tween-20 was used as the running buffer at 25 °C. Biotinylated wild-type p53 peptides (biotinyl-aminocaproyl-GSGS-SQETFSDLWKLLPEN-NH₂) were immobilized on a SA sensor chip (L-p53: 45.1 RU, D-p53: 46.4 RU). All analytes were evaluated for 2 min as contact time, followed by 2 min dissociation at a flow rate of 30 µL min⁻¹ (Figure S4).

For competitive inhibition assays, L-MDM2²⁵⁻¹⁰⁹ (30 nmol dm⁻³) in the presence of varying concentration of inhibitors in PBS containing 0.05% Tween-20 and 1% DMSO were injected on SA sensor chip, where biotinylated wild-type L-p53 peptide was immobilized (127.3 RU) (Table S3, Figure S6).

**Competitive Binding Inhibition Assay by a Standard ELISA**

ELISA assays were carried out in HEPES buffer [20 mmol dm⁻³ HEPES (pH7.4), 100 mmol dm⁻³ NaCl, 0.05% Tween-20, 0.1% BSA]. Precoated streptavidin 96-well plates (Nunc) were incubated with HEPES buffer containing 3% BSA (0.300 cm³ well⁻¹) for 1h. After three washes, biotinylated wild-type p53 peptide (100 nmol dm⁻³) in HEPES buffer (0.100 cm³ well⁻¹) was added and incubated for 2 h. After three washes, 100 nmol dm⁻³ MDM2 (recombinant human MDM2, untagged, Sigma) in
the presence of varying concentration of inhibitors in HEPES buffer containing 1% DMSO (0.100 cm$^3$ well$^{-1}$) was added and incubated for 1 h. After three washes, 1:100 dilution of anti-MDM2 rabbit IgG antibody (N-20, Santa Cruz) in HEPES buffer (0.100 cm$^3$ well$^{-1}$) was added and incubated for 1 h. After three washes, 1:5000 dilution of HRP-conjugated anti-rabbit IgG antibody (Promega) in HEPES buffer (0.100 cm$^3$ well$^{-1}$) was added and incubated for 1 h. After three washes, TMB (3,3',5,5'-Tetramethylbenzidine) solution (WAKO, 0.100 cm$^3$ well$^{-1}$) was added and incubated for 1 h. Then, aqueous 2 N H$_2$SO$_4$ (0.010 cm$^3$ well$^{-1}$) was added. Absorbance at 450 nm was measured for each well using an EnVision Xcite plate reader. The IC$_{50}$ values were calculated by using GraphPad Prizm (GraphPad software, San Diego, CA) (Table S3, Figure S6).

**Cell Growth Inhibition Assay**

SJSA-1 and H1299 cells were cultured in RPMI-1640 medium (high glucose) (WAKO) supplemented with 10% (v/v) FBS at 37 °C in a 5% CO$_2$-incubator. Cell-based assays using SJSA-1 and H1299 cells were performed in 96-well plates (BD Falcon). Both cells were seeded at 500 cells well$^{-1}$ in 0.050 cm$^3$ of DMEM, and placed for 6 h. Chemical compounds in DMSO were diluted 250-fold with the culture medium in advance. Following the addition of the fresh culture medium (0.040 cm$^3$), the chemical diluents (0.030 cm$^3$) were also added to the cell cultures. The final volume of DMSO in the medium was equal to 0.1% (v/v). The cells under chemical treatment were incubated for a further 72 h. The wells in the plates were washed with the cultured medium without phenol-red twice. After 1 h of incubation with 0.10 cm$^3$ of the medium, the cell culture in each well was supplemented with the MTS reagent (0.020 cm$^3$, Promega), followed by incubation for an additional 40 min. Absorbance at 490 nm was measured for each well using an EnVision Xcite plate reader. The GI$_{50}$ values were calculated by using GraphPad Prizm (GraphPad software, San Diego, CA) (Figure S8).
References
**Fig. S1.** LC–MS Chromatograms of Purified Synthetic MDM2<sup>25–109</sup> and MDM2<sup>TMR</sup> Proteins.

**HPLC Conditions:** HPLC analysis was performed at 25 °C on a Cosmosil SC18-AR300 preparative column (Nacalai Tesque, 4.5 × 250 mm) with a linear gradient of 30–50% CH<sub>3</sub>CN containing 0.1% TFA at a flow rate of 1 cm<sup>3</sup> min<sup>−1</sup> over 20 min.
Fig. S2. Mass Spectrometry Data of Synthetic MDM2^{25-109} and MDM2^{TMR} Proteins.
Fig. S3. CD Spectra of Synthetic L-MDM2 and D-MDM2 Proteins. Spectra of L-MDM2$^{25-109}$ and D-MDM2$^{25-109}$ were measured at 25 °C in PBS containing 0.5 mmol dm$^{-3}$ TCEP and 0.005% Tween-20 (pH 7.4).

Fig. S4. SPR Analysis of p53 Binding to Synthetic MDM2$^{25-109}$.

<table>
<thead>
<tr>
<th>Analyte/Ligand</th>
<th>$k_a$ (1 Ms$^{-1}$)</th>
<th>$k_d$ (1 s$^{-1}$)</th>
<th>$K_d$ (mol dm$^{-3}$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>L-MDM2$^{25-109}$/L-p53</td>
<td>1.1 ± 0.75 E$^{-6}$</td>
<td>8.5 ± 0.16 E$^{-1}$</td>
<td>7.5 ± 0.40 E$^{-7}$</td>
</tr>
<tr>
<td>D-MDM2$^{25-109}$/D-p53</td>
<td>5.6 ± 0.69 E$^{-5}$</td>
<td>2.8 ± 0.24 E$^{-1}$</td>
<td>5.1 ± 0.21 E$^{-7}$</td>
</tr>
</tbody>
</table>
Table S1. Data of the Initial Chemical Array Screening.

<table>
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<tr>
<th>Code name</th>
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<th>D-MDM2</th>
<th>Code name</th>
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<th>D-MDM2</th>
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</thead>
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<td>NPD9453</td>
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<tr>
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<td>NPD9481</td>
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<td>NPD9515</td>
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<td>–</td>
<td>NPD10099</td>
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<td>++</td>
</tr>
<tr>
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Hit compounds by chemical array screening had selective binding activity to L-MDM2TMR or D-MDM2TMR protein. Fluorescent intensity $I$ of each spot was measured at 532 nm. The relative intensities ($\Delta I$) of each compound were calculated from the difference between the fluorescence signals of L-MDM2TMR and D-MDM2TMR ($\Delta I = |I_L - I_D|$). The average ($\Delta I_{ave}$) and standard deviation (SD) for all spots were also calculated. The compounds with a larger $\Delta I$ value than ‘$\Delta I_{ave} + 1SD$’ were determined to be selective compounds for L-MDM2 or D-MDM2. [+; >+1SD; ++; >+2SD; +++; >+3SD]. Because of the heterogeneous system of chemical array screening, the result of chemical array screening was not always consistent with that of homogeneous fluorescence polarization assay. Therefore, we should consider the possibilities that false positive signals (non-specific binding between the protein and array surface) and false negative signals (weak binding of the protein to randomly immobilized compounds) occurred in chemical array screening.
Fig. S5. Structures of Hit Compounds.

Table S2. Inhibitory Activities of Reference Compounds by a Fluorescence Polarization Assay.

<table>
<thead>
<tr>
<th>Compound</th>
<th>IC₅₀ (µM)</th>
<th>L-MDM2</th>
<th>D-MDM2</th>
</tr>
</thead>
<tbody>
<tr>
<td>L-P4</td>
<td>0.030 ± 0.016</td>
<td>&gt;1.0</td>
<td>&gt;1.0</td>
</tr>
<tr>
<td>D-P4</td>
<td>&gt;1.0</td>
<td>0.019 ± 0.004</td>
<td></td>
</tr>
<tr>
<td>Nutlin-3a</td>
<td>0.32 ± 0.03</td>
<td>&gt;30</td>
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</table>

Table S3. Inhibitory Activities of NP843 (1a) and ent-NP843 (1b) by SPR Analysis and ELISA.

<table>
<thead>
<tr>
<th>Compound</th>
<th>IC₅₀ (µM)</th>
<th>SPR</th>
<th>ELISA</th>
</tr>
</thead>
<tbody>
<tr>
<td>L-P4</td>
<td>0.011 ± 0.001</td>
<td>0.066 ± 0.007</td>
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</tr>
<tr>
<td>D-P4</td>
<td>&gt;1.0</td>
<td>&gt;1.0</td>
<td></td>
</tr>
<tr>
<td>NP843 (1a)</td>
<td>&gt;30</td>
<td>&gt;30</td>
<td></td>
</tr>
<tr>
<td>ent-NP843 (1b)</td>
<td>3.1 ± 0.5</td>
<td>16.7 ± 1.1</td>
<td></td>
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</tbody>
</table>
Fig. S6. The Representative Dose-response Curves in the Binding Inhibition Assays: (A) FP Assay; (B) SPR Analysis; (C) ELISA.
**Fig. S7.** Results of Cell Growth Inhibition Assay.

<table>
<thead>
<tr>
<th>Compounds</th>
<th>GI$_{50}$ (µM)</th>
<th>SJSA-1 cells$^{12}$</th>
<th>H1299 cells$^{13}$</th>
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<tbody>
<tr>
<td>Nutlin-3a</td>
<td>5.2 ± 0.19</td>
<td>&gt;30</td>
<td>&gt;30</td>
</tr>
<tr>
<td>NP843 (1a)</td>
<td>&gt;30</td>
<td>&gt;30</td>
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<tr>
<td>ent-NP843 (1b)</td>
<td>&gt;30</td>
<td>&gt;30</td>
<td>&gt;30</td>
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</table>
### Spectrum 1

**Technical Details**
- **Date**: 2015-03-03 12:45:18
- **Sample**: G176_diOH_500H
- **Mode**: Single Pulse
- **FID File**: G176_diOH_500H-1.als
- **Reference**: 0.00 ppm
- **Temperature**: 20.8°C
- **Spin System**: CDCL3
- **Gain**: 44

**Spectroscopic Parameters**
- **Frequency**: 7507.39 Hz
- **Scans**: 8
- **Acquisition Time**: 1.7459 sec
- **Power Delay**: 5.000 sec
- **Pulse Width**: 6.82 µsec
- **Chemical Shifts**:
  - 7.5 ppm
  - 6.4 ppm
  - 5.8 ppm
  - 4.9 ppm
  - 2.6 ppm

### Spectrum 2

**Technical Details**
- **Date**: 2015-03-03 13:33:12
- **Sample**: G176_diOH_500C
- **Mode**: Single Pulse Decoupled Gated NOE
- **FID File**: G176_diOH_500C-1.als
- **Reference**: 77.00 ppm
- **Temperature**: 21.3°C
- **Spin System**: CDCL3
- **Gain**: 44

**Spectroscopic Parameters**
- **Frequency**: 31446.06 Hz
- **Scans**: 1000
- **Acquisition Time**: 0.8336 sec
- **Power Delay**: 2.000 sec
- **Pulse Width**: 3.50 µsec
- **Chemical Shifts**:
  - 198.0 ppm
  - 165.3 ppm
  - 162.8 ppm
  - 148.3 ppm
  - 130.9 ppm

**Additional Information**
- **Molecular Structure**
- **Structural Analysis**
- **Chemical Connectivity**
**TMS** single_pulse
**DATIM** 2015-03-06 11:09:42
**OBNUC** 1H
**EXMOD** single_pulse.ex2
**OBFRQ** 500.16 MHz
**OBSET** 2.41 KHz
**OBFIN** 6.01 Hz
**POINT** 13107
**FREQU** 7507.39 Hz
**SCANS** 8
**ACQTM** 1.7459 sec
**PD** 5.0000 sec
**PW1** 6.82 usec
**CTEMP** 21.5 c
**SLVNT** CDCL3
**EXREF** 0.00 ppm
**RGAIN** 40

**TMS** single_pulse decoupled gated NOS
**DATIM** 2015-03-06 11:57:37
**OBNUC** 13C
**EXMOD** single_pulse_dec
**OBFRQ** 125.77 MHz
**OBSET** 7.87 KHz
**OBFIN** 4.21 Hz
**POINT** 26214
**FREQU** 31446.06 Hz
**SCANS** 1000
**ACQTM** 0.8336 sec
**PD** 2.0000 sec
**PW1** 3.50 usec
**CTEMP** 21.8 c
**SLVNT** CDCL3
**EXREF** 77.00 ppm
**RGAIN** 40

**TMS** single_pulse decoupled gated NOS
**DATIM** 2015-03-06 11:57:37
**OBNUC** 13C
**EXMOD** single_pulse_dec
**OBFRQ** 125.77 MHz
**OBSET** 7.87 KHz
**OBFIN** 4.21 Hz
**POINT** 26214
**FREQU** 31446.06 Hz
**SCANS** 1000
**ACQTM** 0.8336 sec
**PD** 2.0000 sec
**PW1** 3.50 usec
**CTEMP** 21.8 c
**SLVNT** CDCL3
**EXREF** 77.00 ppm
**RGAIN** 40
**Single Pulse Experiment**

Experiment details:
- **DFILE**: G162re-1.als
- **COMMT**: single_pulse
- **DATIM**: 2015-05-07 18:52:37
- **OBNUC**: 1H
- **EXMOD**: single_pulse.ex2
- **OBFRQ**: 500.16 MHz
- **OBSET**: 2.41 KHz
- **OBFIN**: 6.01 Hz
- **POINT**: 13107
- **FREQU**: 7507.39 Hz
- **SCANS**: 8
- **ACQTM**: 1.7459 sec
- **PD**: 5.0000 sec
- **PW1**: 6.82 usec
- **IRNUC**: 1H
- **CTEMP**: 21.4 c
- **SLVNT**: CDCL3
- **EXREF**: 0.00 ppm
- **BF**: 0.12 Hz
- **RGAIN**: 44

![NMR Spectrum](C:\Documents and Settings\-%\My Documents\NMRdata\G162re-1.als)

**Single Pulse Decoupled Experiment**

Experiment details:
- **DFILE**: g162 PRODUCT_500C-1.als
- **COMMT**: single_pulse_dec gated NOE
- **DATIM**: 2015-02-12 12:31:47
- **OBNUC**: 13C
- **EXMOD**: single_pulse_dec gated NOE
- **OBFRQ**: 125.77 MHz
- **OBSET**: 7.87 KHz
- **OBFIN**: 4.21 Hz
- **POINT**: 26214
- **FREQU**: 31446.06 Hz
- **SCANS**: 1000
- **ACQTM**: 0.8336 sec
- **PD**: 2.0000 sec
- **PW1**: 3.50 usec
- **IRNUC**: 1H
- **CTEMP**: 21.1 c
- **SLVNT**: CDCL3
- **EXREF**: 77.00 ppm
- **BF**: 1.20 Hz
- **RGAIN**: 54

![NMR Spectrum](C:\Documents and Settings\-%\My Documents\NMRdata\g162 PRODUCT_500C-1.als)
**S64**

**PPM**

1. **DFILE G155DiOH_500H-1.als**
   - **COMNT** single pulse
   - **DATIM** 2015-04-14 19:09:53
   - **OBNUC** 1H
   - **EXMOD** single_pulse.ex2
   - **OBFRQ** 500.16 MHz
   - **OBSET** 2.41 KHz
   - **OBFIN** 6.01 Hz
   - **POINT** 13107
   - **FREQU** 7507.39 Hz
   - **SCANS** 8
   - **ACQTM** 1.7459 sec
   - **PD** 5.0000 sec
   - **PW1** 6.82 usec
   - **CTEMP** 22.2 c
   - **SLVNT CDCL3**
   - **EXREF** 0.00 ppm
   - **BF** 0.12 Hz
   - **RGAIN** 40

2. **DFILE G155DiOH_500C-1.als**
   - **COMNT** single pulse decoupled gated NO
   - **DATIM** 2015-04-14 19:58:11
   - **OBNUC** 13C
   - **EXMOD** single_pulse_dec
   - **OBFRQ** 125.77 MHz
   - **OBSET** 7.87 KHz
   - **OBFIN** 4.21 Hz
   - **POINT** 26214
   - **FREQU** 31446.06 Hz
   - **SCANS** 1000
   - **ACQTM** 0.8336 sec
   - **PD** 5.0000 sec
   - **PW1** 3.50 usec
   - **CTEMP** 22.2 c
   - **SLVNT CDECl3**
   - **EXREF** 0.00 ppm
   - **BF** 0.12 Hz
   - **RGAIN** 40

**Chemicals**

- HO
- O
- S16a

**Graphical Representation**

- Chemical structures with PPM values.