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# Supporting Information

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for

## A Central Strategy for Converting Natural Products into Fluorescent Probes

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### General Synthetic Procedure

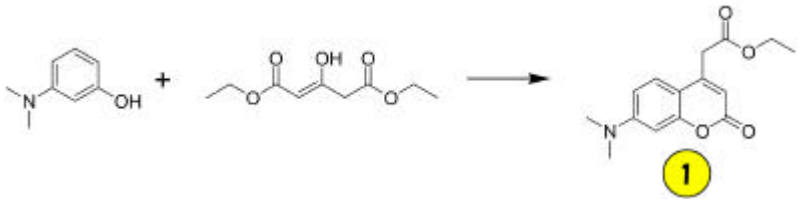
Unless otherwise noted, all reagents and chemical compounds were purchased from commercial sources and used without further purification. Anhydrous solvents: tetrahydrofuran, dichloromethane, and *N*,*N*-dimethylformamide were purified by passing through a solvent column composed of activated A-1 alumina, or distilled. Triethylamine and ethyl-*N*,*N*-diisopropylamine (*i*-$Pr_2NEt$) were dried over sodium and freshly distilled. All reactions were performed in oven-dried glassware under a positive pressure of argon and magnetically stirred with a Teflon-coated stir bar. Flash chromatography was performed on Silica Gel 60 (230-400 mesh) according to Still's method (W. C. Still, M. Kahn, A. Mitra, *J. Org. Chem.* **1978**, *43*, 2923). Analytical TLC was performed on Silica Gel 60 F254 glass plates that were precoated with a 250 μm layer of silica. Visualization

was achieved with UV and/or an appropriate stain ($I_2$ on $SiO_2$, $KMnO_4$, bromocresol green, dinitrophenylhydrazine, ninhydrin, and ceric ammonium molybdate).

Proton NMR ($^1$H NMR) spectra were recorded on a Bruker MSL400 at 400 MHz or Varian XL-400 NMR at 400 MHz, or Varian VXR-400 at 400 MHz and carbon-13 NMR ($^{13}$C NMR) spectra were recorded at 133 MHz. Chemical shifts for $^1$H and $^{13}$C NMR analyses were reported using the standard of Gottlieb, Kotlyar, and Nudelman (H. E. Gottlieb, V. Kotlyar, A. Nudelman, *J. Org. Chem.* **1997**, *62*, 7512). Melting points were recorded on a Thomas Scientific Unimelt apparatus and are uncorrected. Electrospray (ESI) and fast atom bombardment (FAB) mass spectra were obtained at the UCSD Mass Spectrometry Facility using a Finnigan LCQDECA mass spectrometer and a ThermoFinnigan MAT-900XL mass spectrometer, respectively.

**Label Synthesis**

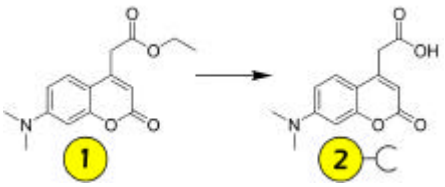
Ethyl 7-dimethylaminocoumarin-4-acetate (**1**)




Compound **1** was synthesized from *m*-dimethylaminophenol and diethyl 1,3-acetonedicarboxylate as described (P. Portonovo, X. Ding, M. S. Leonard, M. M. Joullie, *Tetrahedron* **2000**, *56*, 3687). The starting materials were heated at reflux in EtOH with zinc chloride for 12-15 h, diluted with EtOAc, washed with water, dried ($Na_2SO_4$), evaporated, and recrystallized from EtOH. Additionally, it was found that *m*-dimethylaminophenol is unstable and commercial material often contains significant contamination. The yield of **1** was greatly increased when the phenol starting material was first purified by flash chromatography such that compound **1** was reproducibly produced in 50–60% yield on 1-100 g scale: $^1$H NMR (400 MHz, $CDCl_3$) $d$=7.38 (d, $J$=9.2 Hz, 1 H), 6.59 (dd, $J$=2.4, 9.2 Hz, 1 H), 6.48 (d, $J$=2.4 Hz, 1 H), 6.03 (s, 1 H), 4.16 (q, $J$=7.6 Hz, 2 H), 3.65 (s, 1 H), 3.03 (s, 6 H), 1.23 (t, $J$=7.2 Hz, 3 H); $^{13}$C NMR (100 MHz, $CDCl_3$) $d$=169.2, 162.0, 155.9, 153.0, 148.7, 125.4, 110.6, 109.1, 108.5, 98.3, 61.7, 40.1, 38.3, 14.2; MS

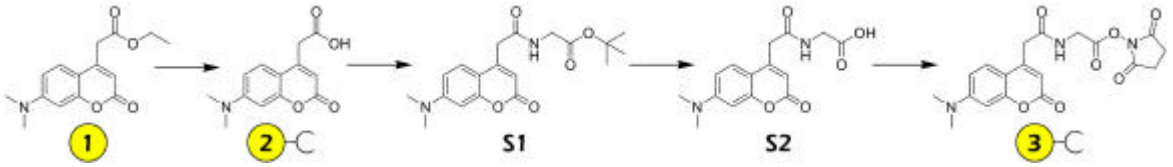
(FAB) *m*/*z* 276.2 $[M+H]^+$ (100%); HRMS (FAB) calcd for $C_{15}H_{17}NO_4$ $[M+H]^+$ 276.1230, found 276.1234 *m*/*z*.

7-Dimethylaminocoumarin-4-acetic acid (**2**)




Ethyl ester **1** (2.86 g, 10.30 mmol) was dissolved in THF/$H_2O$ (3:1, 50 mL) and cooled to 0 °C before 1 M LiOH (20.6 mL, 20.6 mmol) was added and the resulting solution stirred at RT for 3 h. At this point in the reaction, compound **1** was barely detectable by TLC analysis (50:1 $CHCl_3$-EtOAc; **1**, $R_f$=0.14; **2**, $R_f$=0.00). The reaction was washed with $Et_2O$ and the solution was adjusted to pH 2 with 2 M HCl, which resulted in the precipitation of **2** as a light yellow solid (2.57 g, 100%) that was collected by filtration: $^1$H NMR (400 MHz, $CD_3OD$) *d*=7.53 (d, *J*=8.8 Hz, 1 H), 6.76 (dd, *J*=2.4, 8.8 Hz, 1 H), 6.56 (d, *J*=2.4 Hz, 1 H), 6.05 (s, 1 H), 3.78 (s, 2 H), 3.06 (s, 6 H); $^{13}$C NMR (100 MHz, [$D_6$]DMSO) *d* 171.1, 168.4, 160.7, 155.4, 152.8, 151.2, 126.2, 109.4, 109.0, 108.2, 97.5, 42.3, 40.9, 38.4. MS (ESI) *m*/*z* 248.2 $[M+H]^+$ (100%); HRMS (FAB) calcd for $C_{13}H_{14}NO_4$ $[M+H]^+$ 248.0917, found 248.0920 *m*/*z*.

Label **3**



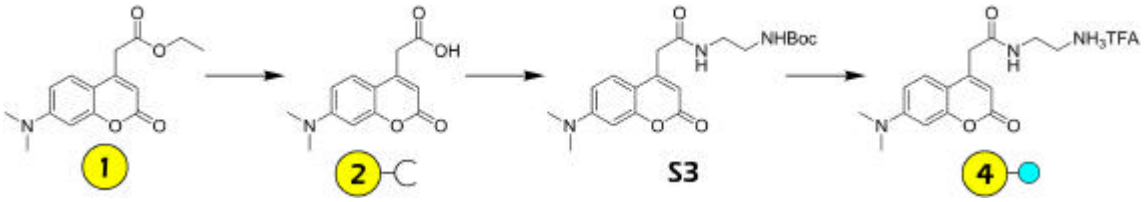

EDAC (1.84 g, 9.60 mmol) and *i*-$Pr_2$NEt (1.34 mL, 7.68 mmol) were added sequentially to a mixture of coumarin **2** (1.58 g, 6.40 mmol), *tert*-butyl glycine hydrochloride (1.29 g, 7.68 mmol), and HOBt (1.18 g, 7.68 mmol) in DMF (20 mL) cooled to 0 °C. The cold bath was removed 1 h after addition, and the mixture was kept for 18 h at RT. At this point, a voluminous precipitate had formed and TLC (3:1 hexanes-EtOAc) indicated that **2** ($R_f$=0.01) had been consumed and *N*-7-dimethylamino-coumarin-4-acetyl glycine, *tert*-butyl ester **S1** ($R_f$=0.28) had formed. The reaction was diluted with EtOAc (20 mL) and filtered to provide **S1** (1.59 g, 69%) as a light yellow solid. $^1$H NMR (400 MHz,

[$D_6$]DMSO) *d*=8.57 (t, *J*=6.0 Hz, 1 H), 7.56 (d, *J*=8.8 Hz, 1 H), 6.71 (dd, *J*=2.4, 9.0 Hz, 1 H), 6.55 (d, *J*=2.8 Hz, 1 H), 6.05 (s, 1 H), 3.74 (d, *J*=5.6 Hz, 2 H), 3.66 (s, 2 H), 3.02 (s, 6 H), 1.38 (s, 9 H); $^{13}$C NMR (100 MHz, 1:1 $CDCl_3$-$CD_3OD$) *d* 169.5, 169.1, 163.2, 156.0, 153.4, 150.9, 126.0, 109.8, 109.6, 108.7, 98.1, 82.5, 42.4, 40.1, 39.5, 28.0; MS (FAB) *m*/*z* 360.2 $[M]^+$ (100%); HRMS (FAB) calcd for $C_{19}H_{24}N_2O_5$ $[M]^+$ 360.1680, found 360.1684 *m*/*z*.

TFA (10 mL) was added dropwise to a stirred solution of compound **S1** (1.59 g, 4.41 mmol) in $CH_2Cl_2$ (10 mL) at 0 °C. After 8 hr at RT, the reaction was complete according to TLC analysis (93:5:2 $CH_2Cl_2$-MeOH-AcOH; **S1** $R_f$=0.42; *N*-7-dimethylaminocoumarin-4-acetyl glycine **S2**, $R_f$=0.13). The flask was placed on the rotary evaporator and the $CH_2Cl_2$ and TFA were removed at reduced pressure. The resulting solid was resuspended in EtOAc and the evaporation repeated (3x10 mL) to afford pure compound **S2** as a yellow solid (1.34 g, 100%): $^1$H NMR (400 MHz, [$D_6$]DMSO) *d* 8.57 (t, *J*=6.0 Hz, 1 H), 7.56 (d, *J*=8.8 Hz, 1 H), 6.69 (dd, *J*=2.4, 9.0 Hz, 1 H), 6.54 (d, *J*=2.4 Hz, 1 H), 6.06 (s, 1 H), 3.78 (d, *J*=6.0 Hz, 2 H), 3.67 (s, 2 H), 3.01 (s, 6 H); $^{13}$C NMR (100 MHz, [$D_6$]DMSO) *d* 171.1, 168.4, 160.7, 155.4, 152.8, 151.2, 126.2, 109.4, 109.0, 108.2, 97.5, 42.3, 40.9, 38.4; MS (FAB) *m*/*z* 305.2 $[M+H]^+$ (100%); HRMS (FAB) calcd for $C_{15}H_{17}N_2O_5$ $[M+H]^+$ 305.1132, found 305.1138 *m*/*z*.

EDC (11.2 mg, 0.058 mmol) was added to a solution of **S2** (12 mg, 0.039 mmol) *N*-hydroxysuccinimide (6.7 mg, 0.058 mmol) in DMF (1 mL). TLC monitor ($CHCl_3$/MeOH/AcOH 90:5:5; **S2**, $R_f$=0.01; **3**, $R_f$=0.49) indicated that the reaction was complete after stirring at RT for 18 h. The reaction mixture was diluted with 2:1 EtOAc-hexanes and the resulting solid collected by filtration to yield 5.7 mg (36%) of pure **3** as a white powder: $^1$H NMR (400 MHz, [$D_6$]DMSO) *d* 8.87 (t, *J*=5.8 Hz, 1 H), 7.53 (d, *J*=8.8 Hz, 1 H), 6.71 (dd, *J*=2.6, 9.0 Hz, 1 H), 6.55 (d, *J*=2.8 Hz, 1 H), 6.04 (s, 1 H), 4.30 (d, *J*=6.0 Hz, 2 H), 3.71 (s, 2 H), 3.02 (s, 6 H); $^{13}$C NMR (100 MHz, 1:1 $CDCl_3$/$CD_3OD$) *d* 169.9, 168.8, 166.2, 160.6, 155.3, 152.8, 150.6, 126.0, 109.4, 109.0, 108.1, 97.5, 38.4; MS (FAB) *m*/*z* 401.2 $[M]^+$ (100%); HRMS (FAB) calcd for $C_{19}H_{19}N_3O_7$ $[M]^+$ 401.1218, found 401.1224 *m*/*z*.

Label **4**

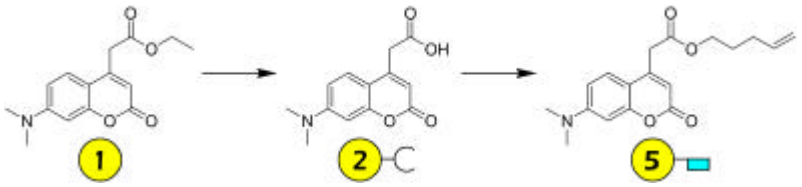



Coumarin acid **2** (1.68 g, 6.79 mmol), HOBt (1.35 g, 8.83 mmol), and EDAC (1.69 g, 8.83 mmol) were dissolved in DMF (20 mL) and cooled to 0 °C. A solution of *N*-Boc-ethylenediame (1.20 g, 7.47 mmol) in DMF (1 mL) was added and the reaction stirred overnight, at which point a voluminous white precipitate had formed. The solid (1.13 g) was isolated by filtration and the mother liquor was diluted with $CHCl_3$, which resulted in the precipitation of an additional 522 mg. The solvent was removed from the mother liquor by rotary evaporation and the desired product was purified from the residue by flash chromatography (2:1 to 1:3 hexanes-EtOAc) to afford another 334 mg of **S3** ($R_f$= 0.26, EtOAc) for a combined yield of 1.97 g (76%): $^1$H NMR (400 MHz, [$D_6$]DMSO) $d$= 8.20 (t, $J$=5.2 Hz, 1 H), 7.52 (d, $J$=9.2 Hz, 1 H), 6.81 (t, $J$=5.2 Hz, 1 H), 6.72 (dd, $J$=2.8, 9.2 Hz, 1 H), 6.54 (d, $J$=2.8 Hz, 1 H), 5.99 (s, 1 H), 3.58 (s, 2 H), 3.09 (m, 2 H), 3.01 (s, 6 H), 2.97 (m, 2 H), 1.37 (s, 9 H); $^{13}$C NMR (100 MHz, [$D_6$]DMSO) $d$ 168.0, 160.7, 155.6, 155.4, 152.8, 151.2, 126.0, 109.5, 109.0, 108.2, 97.5, 77.7, 40.1, 39.7, 39.0, 38.8, 28.2; MS (FAB) $m/z$ 389.2 $[M]^+$ (100%); HRMS (FAB) calcd for $C_{20}H_{27}N_3O_5$ $[M]^+$ 389.1945, found 389.1943 $m/z$.

*N*-Boc-ethylenediamine-containing coumarin **S3** (500 mg, 1.28 mmol) was suspended in $CH_2Cl_2$ (10 mL), cooled to 0 °C, and TFA (1 mL) was added. The solid material immediately dissolved and after 30 min, the starting material was no longer detectable by TLC (EtOAc; **S3**, $R_f$=0.26; **4**, $R_f$=0.00). The flask was placed on the rotary evaporator at reduced pressure to remove the solvent and excess TFA. The residue was diluted with EtOAc and the evaporation procedure repeated (3x) to ensure complete removal of TFA. The pure TFA salt, **4** (520 mg, 100%), was isolated as a yellow-orange solid: $^1$H NMR (400 MHz, [$D_6$]DMSO) $d$=8.41 (br s, 1 H), 7.88 (br s, 3 H), 7.53 (d, $J$=8.8 Hz, 1 H), 6.71 (dd, $J$=2.4, 9.2 Hz, 1 H), 6.55 (d, $J$=2.4 Hz, 1 H), 6.01 (s, 1 H), 3.63 (s, 2 H), 3.30 (m, 2 H), 3.02 (s, 6 H), 2.87 (m, 2 H); $^{13}$C NMR (100 MHz, [$D_6$]DMSO) $d$ 168.7, 160.7, 155.4, 152.8, 150.9, 126.1, 109.5, 109.0, 108.2, 97.5, 39.7, 38.6, 38.4, 36.7; MS (FAB)

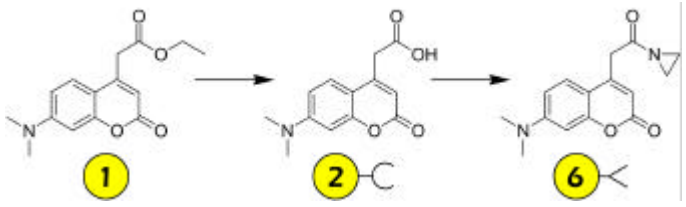
*m/z* 290.2 $[M]^+$ (100%); HRMS (FAB) calcd for $C_{15}H_{20}N_3O_3$ $[M]^+$ 290.1499, found 290.1506.

Label **5**




Coumarin acid **2** (20 mg, 0.081 mmol), pent-4-en-1-ol (9 µL, 0.081 mmol), and EDAC (19 mg, 0.097 mmol) were dissolved in DMF (1 mL) and the solution was stirred overnight, at which point the reaction was complete according to TLC analysis (EtOAc/hexanes 3:1; **2**, $R_f$=0.00; **5**, $R_f$=0.58). The DMF was removed at low pressure and the residue was redissolved in EtOAc, washed with water and brine, and dried ($Na_2SO_4$). Removal of the solvent by rotary evaporation yielded a yellow oil that crystallized on standing (23 mg, 92 %): $^1$H NMR (400 MHz, $CDCl_3$) *d* 7.39 (d, *J*=8.8 Hz, 1 H), 6.60 (dd, *J*=2.4, 8.8 Hz, 1 H), 6.50 (d, *J*=2.4 Hz, 1 H), 6.04 (s, 1 H), 5.73 (ddt, *J*=16.8, 10.8, 6.4 Hz, 1 H), 4.97 (dq, *J*=7.8, 1.6 Hz, 1 H), 4.94 (t, *J*=1.6 Hz, 1 H), 4.12 (t, *J*=6.6 Hz, 2 H), 3.67 (s, 2 H), 3.04 (s, 6 H), 2.03 (m, 2 H), 1.69 (m, 2 H); $^{13}$C NMR (100 MHz, $CDCl_3$) *d*= 169.2, 161.8, 156.1, 153.1, 148.5, 137.3, 125.4, 115.6, 110.8, 109.1, 108.6, 98.5, 65.1, 40.2, 38.4, 30.0, 27.7; MS (FAB) *m/z* 316.2 $[M+H]^+$ (100%); HRMS (FAB) calcd for $C_{18}H_{22}NO_4$ $[M+H]^+$ 316.1543, found 316.1548.

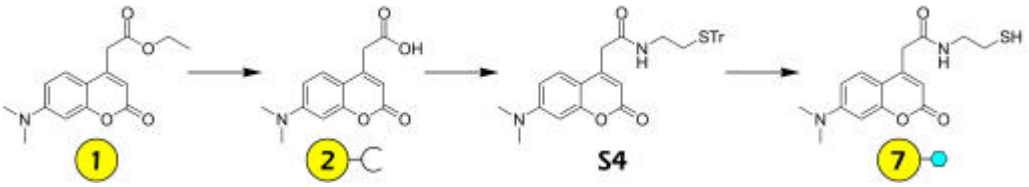
Label **6**




Coumarin **2** (45 mg, 0.18 mmol) and EDAC (52 mg, 0.27 mmol) were dissolved in DMF (1 mL) at 0 °C and 4 and *N*-hydroxysuccinimide (36 mg, 0.24 mmol) was added. The reaction was stirred overnight, at which point the starting material was no longer detectable by TLC (EtOAc; **8**, $R_f$=0.24; **2**, $R_f$=0.00). Aziridine (36 mg, 0.24 mmol) was added dropwise over 10 min. The solvent was removed by high vacuum pump to yield a yellow

semi-solid. Purification of this material by flash chromatography (EtOAc/hexanes 1:1 to EtOAc) afforded **8** (36 mg, 55%). $^1$H NMR (400 MHz, 1% [D$_5$]pyridine in CDCl$_3$) *d*=7.64 (d, *J*=8.8 Hz, 1 H), 6.31 (dd, *J*=2.4, 9.2 Hz, 1 H), 6.52 (d, *J*=2.4 Hz, 1 H), 6.06 (s, 1 H), 3.59 (s, 2 H), 3.02 (s, 6 H), 2.61 (s, 4 H); $^{13}$C NMR (100 MHz, 10% [D$_5$]pyridine in [D$_6$]DMSO) *d*=168.4, 162.3, 156.5, 153.2, 151.1, 127.2, 108.3, 108.9, 108.1, 96.6, 39.8, 38.2, 29.7; MS (FAB) *m*/*z* 290.2 [*M*+NH$_4$]$^+$ (100%); HRMS (FAB) calcd for C$_{15}$H$_{20}$N$_3$O$_3$ [*M*+NH$_4$]$^+$ 290.1505, found 290.1506 *m*/*z*.

Label **7**

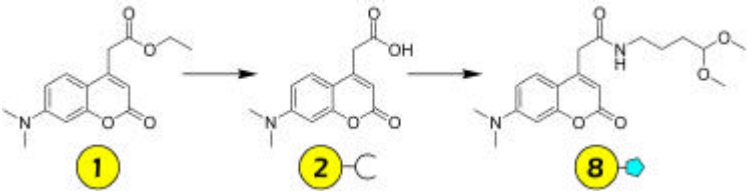



A solution of coumarin **2** (40 mg, 0.16 mmol) in DMF (2 mL) was cooled to 0 °C and *S*-trityl-cysteamine (58 mg, 0.16 mmol) was added followed by HOBt (32 mg, 0.24 mmol), EDAC (46 mg, 0.24 mmol), and *i*-Pr$_2$NEt (28 µL, 0.16 mmol). After 12 h, the reaction was complete according to TLC analysis (3:1 EtOAc-hexanes; **2**, $R_f$=0.00; **S4**, $R_f$=0.37). The reaction was diluted with Et$_2$O which resulted in the precipitation of the product. Flash chromatography (1:1 to 3:1 EtOAc-hexanes) followed by crystallization (CH$_2$Cl$_2$/hexanes) yielded **S4** as a white solid (37 mg, 42%): $^1$H NMR (400 MHz, CDCl$_3$) *d* 7.42 (d, *J*=9.2 Hz, 1 H), 7.33 – 7.36 (m, 6 H), 7.17 – 7.27 (m, 9 H), 6.57 (dd, *J*=2.4, 9.2 Hz, 1 H), 6.49 (d, *J*=2.8 Hz, 1 H), 5.99 (s, 1 H), 5.71 (bt, *J*=5.6 Hz, 1 H), 3.52 (s, 2 H), 3.04 (t, *J*=6.2 Hz, 2 H), 3.02 (s, 6 H), 2.36 (t, *J*=6.4 Hz, 2 H); $^{13}$C NMR (100 MHz, CDCl$_3$) *d*= 167.8, 161.7, 156.1, 153.1, 149.6, 144.7, 129.6, 128.1, 126.9, 125.9, 110.7, 109.5, 109.5, 108.6, 98.5, 66.8, 40.8, 40.3, 38.7, 31.7; MS (FAB) *m*/*z* 548.6 [*M*]$^+$ (100%); HRMS (FAB) calcd for C$_{34}$H$_{32}$N$_2$O$_3$S [*M*]$^+$ 548.2128, found 548.2133 *m*/*z*.

The trityl protected thiol **S4** (15 mg, 0.027 mmol) was dissolved in CH$_2$Cl$_2$ (0.5 mL) and Et$_3$SiH (28 µL, 0.14 mmol) was added followed by TFA (0.5 mL). The reaction was stirred for 45 min, at which point TLC analysis indicated the reaction was complete (EtOAc; **S4**, $R_f$=0.68; **7**, $R_f$=0.30). The volatile reaction components were removed at reduced pressure and compound **7** was purified by flash chromatography (EtOAc/hexanes 1:1 to EtOAc) to yield a yellow solid (8 mg, 97%): $^1$H NMR (400 MHz, CDCl$_3$) *d*=

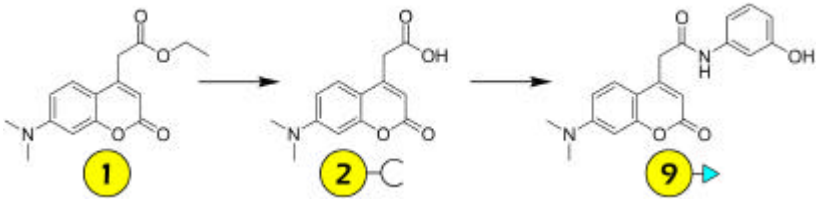
7.48 (d, *J*=8.8 Hz, 1 H), 6.64 (dd, *J*=2.8, 8.8 Hz, 1 H), 6.50 (d, *J*=2.8 Hz, 1 H), 6.27 (br s, 1 H), 6.06 (s, 1 H), 3.64 (s, 2 H), 3.40 (q, *J*=6.4 Hz, 2 H), 3.05 (s, 6 H), 2.61 (dt, *J*=8.4, 6.4 Hz, 2 H), 1.22 (t, *J*=8.4 Hz, 1 H); $^{13}$C NMR (100 MHz, $CDCl_3$) *d*=168.2, 161.8, 156.2, 153.1, 149.7, 125.8, 110.8, 109.6, 108.6, 98.6, 42.8, 41.0, 40.4, 24.5; MS (FAB) *m/z* 307.2 $[M+H]^+$ (100%); HRMS (FAB) calcd for $C_{15}H_{19}N_2O_3S$ $[M+H]^+$ 307.1111, found 307.1114 *m/z*.

Label **8**




Coumarin **2** (45 mg, 0.18 mmol), EDAC (52 mg, 0.27 mmol), and HOBt (36 mg, 0.24 mmol) were dissolved in DMF (1 mL) at 0 °C and 4,4-dimethoxybutylamine (48 mg, 0.36 mmol) was added. The reaction was stirred overnight, at which point the starting material was no longer detectable by TLC (EtOAc; **8**, $R_f$=0.24; **2**, $R_f$=0.00). The solvent was removed by high vacuum pump to yield a yellow semi-solid. Purification of this material by flash chromatography (EtOAc/hexanes 1:1 to EtOAc) afforded **8** (36 mg, 55%) as a light yellow solid: $^1$H NMR (400 MHz, $CDCl_3$) *d*=7.44 (d, *J*=8.8 Hz, 1 H), 6.55 (dd, *J*=2.8, 9.0 Hz, 1 H), 6.47 (br s, 1 H), 6.38 (d, *J*=2.4 Hz, 1 H), 5.98 (s, 1 H), 4.24 (bm, 1 H), 3.56 (s, 2 H), 3.20 (s, 6 H), 3.18 (m, 2 H), 2.99 (s, 6 H), 1.60 – 1.48 (m, 4 H); $^{13}$C NMR (100 MHz, $[D_6]$DMSO) *d*=168.0, 161.9, 155.9, 153.1, 150.2, 125.7, 110.2, 109.2, 108.4, 104.2, 98.1, 52.9, 40.7, 40.1, 39.6, 29.8, 24.3; MS (FAB) *m/z* 362.2 $[M]^+$ (100%); HRMS (FAB) calcd for $C_{19}H_{26}N_2O_5$ $[M]^+$ 362.1836, found 362.1843 *m/z*.

Label **9**

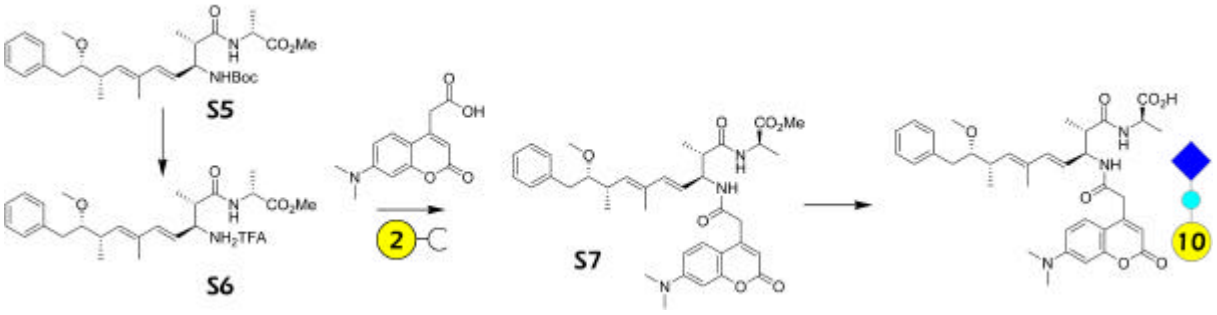



Coumarin acid **2** (20 mg, 0.081 mmol), 3-aminophenol (9 mg, 0.081 mmol), and EDAC (19 mg, 0.097 mmol) were dissolved in DMF (1 mL) and the reaction was stirred over-

night, at which point the reaction was complete according to TLC analysis (EtOAc/hexanes 3:1; **2**, $R_f$=0.00; **9**, $R_f$=0.19). The DMF was removed at low pressure and the gummy residue was crystallized from MeOH–$H_2O$ to yield the phenolic product as a yellow powder (24 mg, 88 %): $^1$H NMR (400 MHz, [$D_6$]DMSO) *d* 10.17 (br s, 1 H), 9.39 (br s, 1 H), 7.58 (d, *J*=8.8 Hz, 1 H), 7.15 (s, 1 H), 7.07 (t, *J*=8.2 Hz, 1 H), 6.95 (d, *J*=8.0 Hz, 1 H), 6.75 (dd, *J*=2.2, 9.0 Hz, 1 H), 6.56 (d, *J*=2.8 Hz, 1 H), 6.46 (dd, *J*=1.4, 8.2 Hz, 1 H), 6.06 (s, 1 H), 3.82 (s, 2 H), 3.01 (s, 6 H); $^{13}$C NMR (100 MHz, [$D_6$]DMSO) *d*=166.6, 160.7, 157.6, 155.4, 152.8, 151.1, 139.9, 129.4, 126.0, 110.6, 109.9, 109.5, 109.1, 108.2, 106.4, 97.5, 39.7, 39.7.

**Analogue Synthesis**

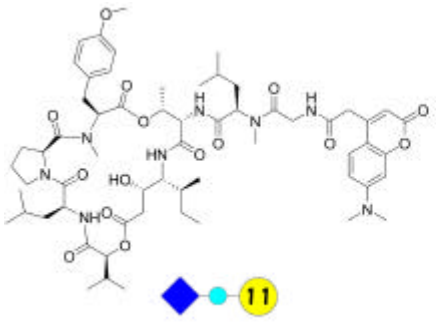
Microcystin analogue **10**

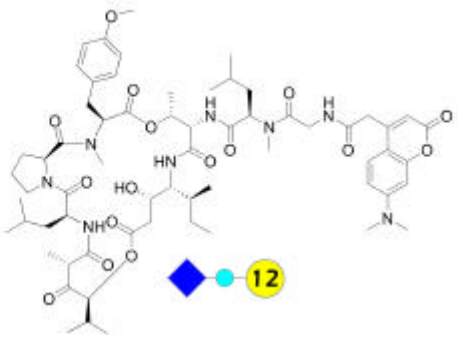



The synthesis of analogue **10** began with *N*-Boc-Adda-D-Ala-OMe **S5** (B. M. Gulledge, J. B. Aggen, H. Eng, K. Sweimeh, A. R. Chamberlin, *Bioorg. Med. Chem. Lett.* **2003**, *13*, 2907). *N*-Boc-Adda-D-Ala-OMe **S5** (0.0177 g, 0.0343 mmol) was dissolved in $Et_3SiH$ (0.05 mL, 0.31 mmol) and 5.0 mL of $CH_2Cl_2$. To this solution was added 0.26 mL (3.4 mmol) of TFA, and the reaction mixture was stirred for 1.5 h. The solution was concentrated in vacuo from hexanes (3x20 mL) to afford **S6**. The crude oil was dissolved in DMF (3.0 mL) and treated with coumarin **2** (0.0232 g, 0.0938 mmol) and collidine (0.04 mL, 0.30 mmol) before cooling to 0 ?C. HATU (0.0534 g, 0.140 mmol) was added and the resultant solution was stirred for 3 h before warming to RT. After 14h, saturated aqueous $NaHCO_3$ (15 mL) was added and the aqueous mixture was extracted with EtOAc (4x10 mL). The combined organic extracts were washed with 1 N HCl (10 mL) and brine (15 mL), dried over $MgSO_4$, and concentrated *in vacuo*. The coumarin-labeled dipeptide **S7** was purified via column chromatography (0% isopropanol to 20% isopropanol gradient in EtOAc) to give a yellow oil that was dissolved in 3.0 mL of THF and cooled to

0 °C. The solution was treated with 0.24 mL (0.024 mmol) of LiOH (0.1 N in $H_2O$) and stirred for 15 h, after which an additional 0.24 mL (0.24 mmol) of LiOH (1 N in $H_2O$) was added and stirring continued for an additional hour before 15 mL of 1 N HCl was added. The resultant aqueous mixture was extracted with EtOAc (4x10 mL). The combined organic extracts were washed with brine (10 mL), dried over $MgSO_4$, and concentrated in vacuo. Product **10** was purified by reverse-phase HPLC (5% to 95% $CH_3CN$ gradient in 0.1% aqueous $HCO_2H$) to give a light yellow oil (4.7 mg, 22% from **S4**): $^1$H NMR (500 MHz, [$D_6$]DMSO) *d* 7.70 (br s, 1H), 7.63 (br s, 1H), 7.58 (d, *J*=8.9, 1H), 7.26 (app t, *J*= 7.3, 2H), 7.22–7.12 (m, 3H), 6.53 (d, *J*=5.9, 1H), 6.53 (s, 1H), 6.08–6.00 (m, 2H), 5.53 (dd, *J*=14.8, 8.9, 1H), 5.35–5.30 (m, 1H), 4.52–4.45 (m, 1H), 4.30–4.23 (m, 1H), 3.66 (s, 2H), 3.30–3.21 (m, 1H), 3.20 (s, 3H), 3.01 (s, 6H), 2.79–2.70 (m, 1H), 2.69–2.62 (m, 2H), 2.60–2.55 (m, 1H), 1.57 (s, 3H), 1.24 (d, *J*=7.2, 3H), 1.04 (d, *J*=6.7, 3H), 0.98 (d, *J*=6.6, 3H); HRMS (CI/$NH_3$) *m/z* calcd for $C_{36}H_{45}N_3O_7Na$ [*M*+Na]$^+$ 654.3155, found 654.3141 *m/z*.

Tamandarin **11** and Didemnin **12** analogues

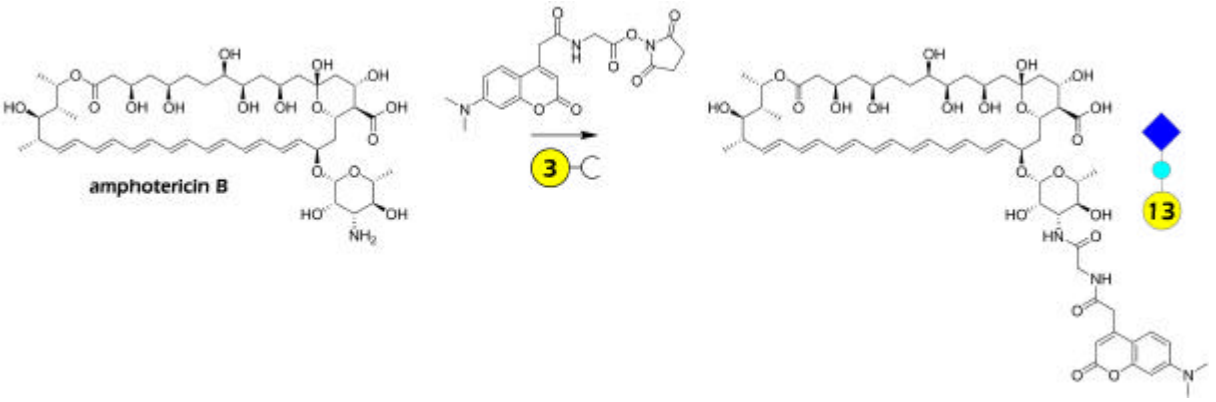



The synthesis of analogues **11**-**12** and their characterization has been published (M. M. Joullie, M. S. Leonard, P. Portonovo, B. Liang, X. Ding, J. J. La Clair, *Bioconjug. Chem*. **2003**, *14*, 30-37).

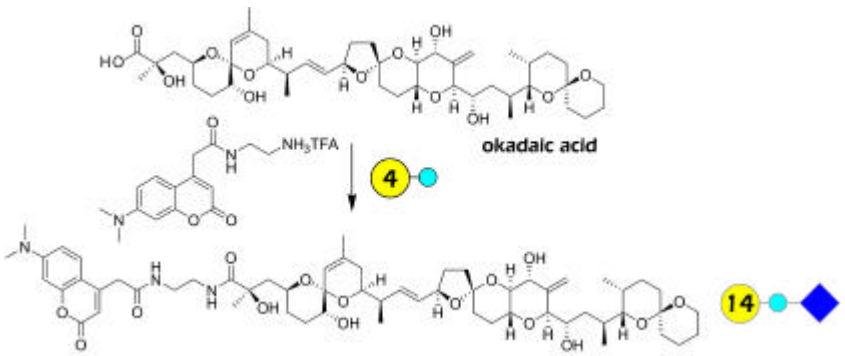
Amphotericin analogue **13**

*N*-Hydroxysuccinimide ester **3** (5.7 mg, 0.014 mmol) was dissolved in DMSO (0.5 mL) under argon and cooled to 0 °C. A solution of amphotericin B (10.5 mg, 0.011 mmol) in DMSO (0.5 mL) was added over 5 min, followed by *i*-$Pr_2NEt$ (2 µL, 0.011 mmol). After 1h, the reaction was complete as judged by TLC analysis ($CHCl_3$/EtOAc 2:1, 5% AcOH;



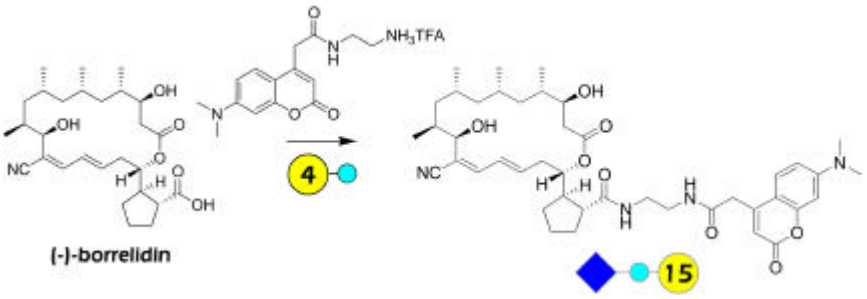

**3**, $R_f$=0.86; 13, $R_f$=0.53; amphotericin B, $R_f$=0.08). The product was recrystallized from $H_2O$/MeOH/DMF/$CH_2Cl_2$ (2:4:2:1) to afford **13** as a yellow solid (3.6 mg, 27 %): $^1$H NMR (400 MHz, [$D_6$]DMSO with 1% $D_2O$) *d*=8.41 (br s, 1 H), 7.70 (d, *J*=8.4 Hz, 1 H), 7.57 (d, *J*= 8.4 Hz, 1 H), 7.23 (dt, *J*=7.2, 12.2 Hz, 1 H), 6.72 (dd, *J*=8.4, 2.3 Hz, 1 H), 6.55 (d, *J*= 2.3 Hz, 1 H), 6.37 (m, 2 H), 6.32 (m, 3 H), 6.16 (t, *J*=3.2, 3.2 Hz, 2 H), 6.12 (m, 1 H), 6.07 (s, 1 H), 5.93 (dd, *J*=8.4, 15.2 Hz, 1 H), 5.88 (s, 1 H), 5.44 (dd, *J*=9.8, 10.4, 1 H), 5.34 (s, 1 H), 5.20 (m, 1 H), 4.79 (d, *J*=4.0 Hz, 1 H), 4.78 (d, *J*=7.2 Hz, 1 H), 4.74 (d, *J*= 3.6, 1 H), 4.70 (d, *J*=5.2 Hz, 1 H), 4.62 (d, *J* =5.6 Hz, 1 H), 4.43 (d, *J*=5.6 Hz, 1 H), 4.40 (s, 2 H), 4.21 (dd, *J*=9.6, 9.6 Hz, 1 H), 4.04 (m, 1 H), 4.00 (m, 1 H), 3.80 (br s, 2 H), 3.70 (m, 1 H), 3.69 (s, 3 H), 3.53 (m, 1 H), 3.52-3.06 cm, 9 H), 3.02 (s, 6 H), 2.30 (s, 1 H), 2.29 (m, 1 H), 2.16 (d, *J*=6.2 Hz, 1 H), 1.98 (t, *J*=10.4, 1 H), 1.88 (m, 1 H), 1.73 (m, 1 H), 1.57 (m, 4 H), 1.49-1.18 (cm, 7 H), 1.16 (m, 1 H), 1.15 (d, *J*=5.6 Hz 3 H), 1.11 (d, *J*=5.4 Hz, 3 H), 1.03 (d, *J*=6.4 Hz, 3 H), 0.92 (d, *J*=6.8 Hz, 3 H); HRMS (FAB) *m/z* calcd for $C_{62}H_{87}N_3O_{21}Na$ [*M*+Na]$^+$ 1232.5730, found 1232.5679 *m/z*.

Okadaic acid analogue **14**



A 1 mM stock of *N*-DMACA--ethylenediamine **4** was prepared in DMF. A 260 µL aliquot of this stock, containing *N*-DMACA--ethylenediamine **4** (75.5 µg, 0.260 µmol), was added to okadaic acid (200 µg, 0.248 µmol). The mixture was dried by evaporation of DMF (2x1 mL) and pyridine (1 x1 mL), redissolved in 0.5 mL of DMF and cooled to 0 C. A 1 mM stock of EDAC containing 0.1 mM DMAP was prepared in DMF. A 0.5 mL aliquot of this reagent was added providing EDAC (0.0958 mg, 0.5 µmol) and DMAP (6.1 µg, 0.05 ?mol) over the period of 2 min. The reaction mixture was warmed to RT over 2 h. After stirring at RT for 12 h, 20 µL of MeOH was added and the reaction mixture was concentrated in vacuo. Product **14** was purified by reverse-phase HPLC (5% to 95% $CH_3CN$ gradient in 0.1% aqueous $NH_4^+HCO_2^-$) to provided a residue with a light fluorescent pink hue (0.163 mg, 61% from **4**): $^1$H NMR (400 MHz, CDCl3, 383 K) *d*=7.45 (d, *J*=8.9,1H), 6.63 (dd, *J*=2.8, 8.9, 1H), 6.46 (d, *J*=2.8, 1H), 6.02 (s, 1H), 5.48 (dd, *J*=15.0, 8.0 Hz, 1H), 5.38 (s, 1H), 5.36 (dd, *J*=15.0, 8.0 Hz, 1H), 5.32 (s, 1H), 5.04 (s, 1H), 4.50 (m, 1H), 4.06 (m, 2H), 3.95 (m, 2H), 3.62 (m, 2H), 3.54 (s, 2H), 3.31 (m, 4H), 3.26 (dd, *J*=10.5, 2.0 Hz, 1 H), 3.25 (t, *J*=6.0 Hz, 2H), 3.15 (t, *J*=6.0, 2H), 3.01 (s, 6H), 2.3-1.2 (m, 47H), 1.04 (d, *J*=6.5 Hz, 3H), 0.94 (d, *J*=6.5 Hz, 3H), 0.90 (d, *J*=7.0 Hz, 3H); HRMS (FAB) *m/z* calcd for $C_{59}H_{85}N_3O_{15}Na$ $[M+Na]^+$ 1076.5981, found 1076.5996 *m/z*.

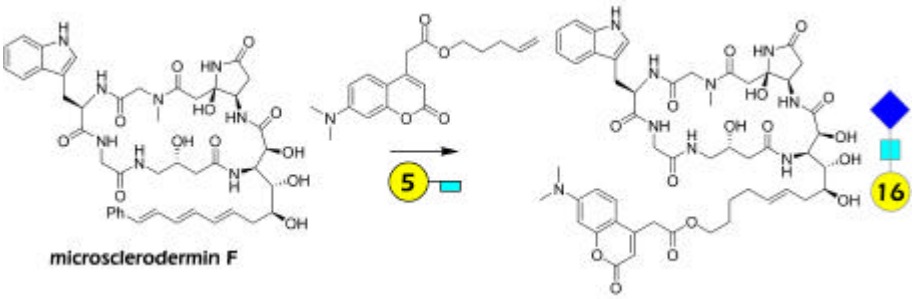
(-)-Borrelidin analogue **15**




(-)-Borrelidin (3 mg, 0.006 mmol), coumarin **4** (3 mg, 0.007 mmol), EDAC (2 mg, 0.01 mmol) and triethylamine (2 mg, 0.02 mmol) were stirred in $CH_2Cl_2$ (0.5 mL) for 12 h at RT. The solvent was evaporated and the residue was purified on silica gel chromatography (3% MeOH in $CH_2Cl_2$) to yield 3.9 mg of the fluorescent natural product amide (86%). $R_f$=0.3 (7% MeOH in $CH_2Cl_2$). $^1$H NMR (400 MHz, $CD_3CN$) *d*=7.47 (1H, d, *J*=9.2 Hz), 6.83 (1H, d, *J*=11.2 Hz), 6.80 (1H, m), 6.68 (1H, dd, *J*=8.8 Hz, 2.4 Hz), 6.54 (1H, d, *J*=2.4 Hz), 6.51 (2H, m), 6.30 (1H, m), 5.96 (1H, s), 4.84 (1H, d, *J*=11.2 Hz), 4.10 (1H,

dd, *J*=10 Hz, 3.6 Hz), 3.78 (1H, m), 3.55 (2H, s), 3.47 (1H, d, *J*=4.4 Hz), 3.29-3.23 (3H, m), 3.19-3.15 (2H ,m), 3.09 (2H,m), 3.02 (6H,s), 2.63-2.42 (3H,m), 2.49-2.22 (3H, m), 1.8-0.6 (26H, m); $^{13}$C NMR (100 MHz, $CD_3CN$) *d*=177.9, 172.5, 169.4, 162.1, 156.9, 154.1, 151.2, 144.4, 140.0, 128.1, 126.7, 111.1, 109.8, 109.2, 98.6, 77.5, 72.6, 70.0, 50.9, 48.6, 45.1, 43.9, 40.6, 40.5, 40.4, 40.3, 39.7, 38.1, 36.5, 36.4, 35.3, 32.5, 30.1, 27.0, 25.8, 20.4, 18.4, 17.2, 15.1.

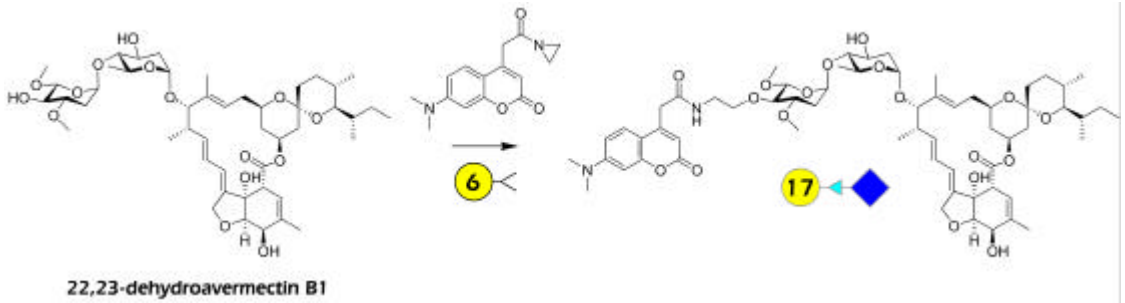
Microsclerodermin analogue **16**




Microsclerodermin F (1.0 mg, 1.1 µmol) and coumarin **5** (1.5 mg, 4.8 µmol) were dissolved in DMF (3 mL). The reaction flask was degassed and filled with argon, and bis-(tricyclohexylphosphine) benzylidine ruthenium(IV) chloride (0.092 mg, 0.11 µmol) was added from a 100 mM stock solution in $CH_2Cl_2$ (1.1 mL). The reaction was warmed to 40 °C for 6 hours at which point microsclerodermin F (50% aq. MeOH, $R_f$=0.62) had been consumed, and a predominant new spot was observed (50% aq. MeOH, $R_f$=0.45). The crude reaction mixture was concentrated and subjected to purification by reversed-phase HPLC (5% to 95% $CH_3CN$ gradient) to yield 1.3 mg of analogue **16**: $^1$H NMR (500 MHz, [$D_6$]DMSO) *d*=10.92 (s, 1 H), 8.66 (d , *J*=4 Hz, 1 H), 8.59 (t, *J*=6.6 Hz, 1 H), 8.01 (s, 1 H), 7.56 (d, *J*=8.5 Hz, 1 H), 7.54 (d, *J*=8.0, 1 H), 7.54 (m, 1 H), 7.41 (d, *J*=8.9 Hz, 1 H), 7.31 (m, 1 H), 7.21 (m, 1 H), 7.02 (dd, *J*=7.5,7.5 Hz, 1 H), 6.92 (dd, *J*=7.5,7.5 Hz, 1 H), 6.61 (dd, *J*=2.5, 8.9 Hz, 1 H), 6.49 (d, *J*=2.5, 1 H), 6.09 (m, 1 H), 6.03 (s, 1 H), 5.98 (bm, 4H), 5.88 (br s, 1 H), 4.89 (d, *J*=4.5, 1 H), 4.62 (d, *J*=8.5, 1 H),4.47 (dd, *J*=18,9 Hz, 1 H),4.39 (d, *J*=6 Hz, 1 H), 4.33 (d, *J*=4.51 Hz, 1 H),4.22 (m, 1 H), 4.12 (m, 1 H), 4.11 (m, 1 H), 4.08 (t, *J*=6.5 Hz, 2 H), 3.84 (m, 1 H), 3.79 (d, *J*=7 Hz, 1 H), 3.77 (m, 1 H), 3.69 (s, 2 H), 3.49 (dd, *J*=9.5,9.5 Hz, 1 H), 3.43 (m, 1 H), 3.39 (m, 1 H), 3.16 (m, 1 H), 3.13 (br s, 1 H), 3.08 (s, 6H), 3.01 (m, 1 H), 2.96 (s, 3 H), 2.87 (d, *J*=17 Hz, 1 H), 2.72 (d, *J*= 16.9 Hz, 1 H), 2.67 (m, 1 H), 2.45 (m, 1 H), 2.45 (m, 1 H), 2.3 (m, 2 H), 2.14 (m, 1 H),

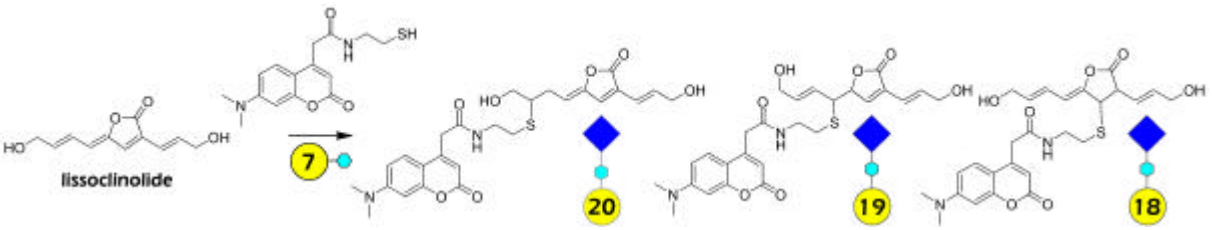
2.11 (m, 2 H), 1.65 (m, 2 H), 0.95 (d *J*=6.5 Hz, 3H); HRMS (FAB) *m/z* calcd for $C_{50}H_{63}N_9O_{16}Na$ $[M+Na]^+$ 1068.4290, found 1068.4312 *m/z*.

22,23-Dehydroavermectin B1 analogue **17**




22,23-dehydroavermectin B1 (4.3 mg, 5.1 mmol) was dried by evaporation of pyridine (2x2 mL) and dissolved in pyridine (0.5 mL). A solution of **6** (3.2 mg, 11.7 mmol) was added in $CH_2Cl_2$ (0.5 mL) and stirred at RT for 16 h. The solvent was evaporated in vacuo and the reaction contents were purified by PTLC using sequential elution with 50% $CH_2Cl_2$ in hexanes, $CH_2Cl_2$, and 10% $CH_3OH$ in $CH_2Cl_2$ to provide 1.1 mg of **17** (33%). The yield of this process could be increased to 89% by protection of the C-5 hydroxyl group as its TBDMS ether (K. Nagai, T. Sunazuka, K. Shiomi, A. Harder, A. Turberg, S. Omura, *Bioorg. Med. Chem. Lett.* **2003**, *13*, 3943-3946). Analogue **17**: $^1$H NMR (500 MHz, 1% $CD_3OD$ in $CDCl_3$) *d*=9.56 (br s, NH), 7.49 (d, *J*=9.1 Hz, 1 H), 6.69 (dd, *J*= 2.4,9.1 Hz, 1 H), 6.50 (dd, *J*=2.4 Hz, 1 H), 6.02 (s, 1 H), 5.92 (m, 1 H), 5.78 (m, 1 H), 5.70 (m, 1 H), 5.44 (m, 1 H), 5.40 (m, 1 H), 5.36 (m, 1 H), 4.99 (m, 1 H), 4.71 (dd, *J*= 4.2,1.5 Hz, 1 H), 4.69 (dd, *J*=14.0,2.4 Hz, 1 H), 4.60 (dd, *J*=14.1, 2.4 Hz, 1 H), 4.12 (d, *J*=5.4 Hz, 1 H), 3.99 (m, 1 H), 3.89 (m, 1 H), 3.84 (dq, *J*=9.4, 9.4 Hz, 1 H), 3.78 (dq, *J*= 9.4, 9.4 Hz 1 H), 3.71 (m, 1 H), 3.62 (m, 1 H), 3.61 (m, 2 H), 3.58 (s, 2 H), 3.48 (m, 1 H), 3.45 (s, 3 H), 3.45 (s, 3 H), 3.32 (m, 2 H), 3.27 (m, 1 H), 3.22 (dd, *J*=9.4, 9.3 Hz, 1 H), 3.20 (m, 1 H), 3.17 (dd, *J*=9.4, 9.3 Hz, 1 H), 3.05 (s, 6 H), 2.51 (m, 1 H), 2.30 (m, 1 H), 2.28 (m, 1 H), 2.25 (m, 1 H), 1.98 (ddd, *J*=11.8, 5.2, 1.5 Hz, 1 H), 1.88 (m, 1 H), 1.77 (m, 1 H), 1.66 (m, 1 H), 1.52-1.49 (cm, 8 H),1.36 (dd, *J*=11.8, 11.8 Hz,1 H), 1.28 (d, 6 1 H), 1.24 (d, 6.1 1 H), 1.17 (d, *J*=7.0 Hz, 1 H), 0.98 (dd, *J*=7.5, 7.5 Hz, 1 H), 0.85 (m, 1 H), 0.82 (d, *J*=6.6 Hz, 1 H), 0.78 (d, 6.4 1 H); HRMS (FAB) *m/z* calcd for $C_{62}H_{88}N_2O_{18}Na$ $[M+Na]^+$ 1171.5930, found 1171.5899 *m/z*.

Lissoclinolide analogues **18**, **19**, **20**



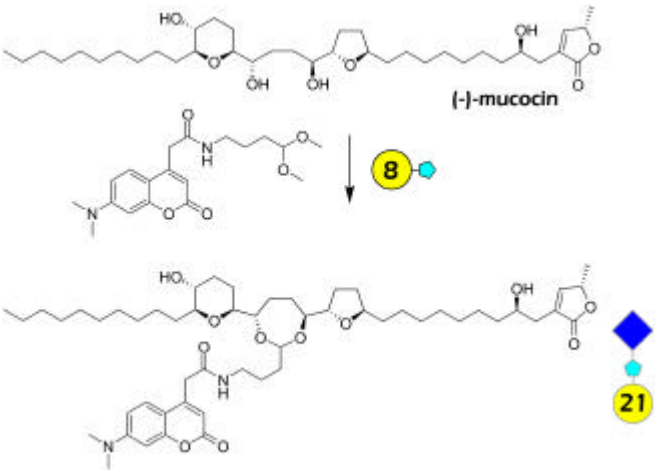

A stock solution of **7** (0.6 mg, 2.9 μmol) and *i*Pr$_2$NEt (1.5 μg, 8.6 μmol) in THF (2 mL) was added to lissoclinolide (0.6 mg, 2.8 μmol) dissolved in THF (2 mL). The reaction was maintained under a strict argon atmosphere. After incubation at RT for 1.5 h, TLC evidence indicated the conversion of lissoclinolide ($R_f$ =0.8, 80% $CH_2Cl_2$ in hexanes) to **18** ($R_f$ =0.32, 80% $CH_2Cl_2$ in hexanes), **19** ($R_f$ =0.23, 80% $CH_2Cl_2$ in hexanes) and **20** ($R_f$ =0.41, 80% $CH_2Cl_2$ in hexanes). Analogs were isolated by PTLC purification using sequential elution with 50% $CH_2Cl_2$ in hexanes followed by 10% EtOAc in hexanes to provide 0.18 mg of **18** (12%), 0.37 mg of **19** (25%), and 0.58 of **20** (39%). The yields of these analogues were determined spectrophotometrically in a 5 mL volumentric flask using the absorption or fluorescence from the coumarin tag ($?_{ex}$=370 nm, $e$=22 000 $M^{-1}cm^{-1}$, $?_{em}$=459 nm).

Adduct **18**: $^1$H NMR (400 MHz, 20% $CDCl_3$ in $CD_3OD$) $d$=7.39 (d, $J$=9.1 Hz, 1 H), 6.75 (dt, $J$=15.6, 5.2 Hz, 1 H), 6.60 (dd, $J$=2.4, 9.1 Hz, 1 H), 6.51 (d, $J$=2.4 Hz, 1 H), 6.41 (ddt, $J$=15.8, 11.2, 1.7 Hz, 1 H), 6.25 (br s, NH), 6.21 (dt, $J$=15.6, 3.4, 1.8 Hz, 1 H), 6.04 (s, 1 H), 5.92 (dt, $J$=15.8, 5.1 Hz, 1 H), 5.89 (dd, $J$=11.2,1.6 Hz, 1 H), 4.21 (m, 4 H), 3.68 (m, 1H), 3.59 (s, 2 H), 3.46 (t, $J$=6.4 Hz, 2 H), 3.21 (m, 1 H), 3.05 (s, 6 H), 2.74 (t, $J$=6.4 Hz, 2 H); HRMS (FAB) *m/z* calcd for $C_{26}H_{30}N_2O_7SNa$ $[M+Na]^+$ 537.1671, found 537.1701 *m/z*.

Adduct **19**: $^1$H NMR (400 MHz, 20% $CDCl_3$ in $CD_3OD$) $d$=7.45 (d, $J$=8.9 Hz, 1 H), 7.41 (s, 1 H), 6.86 (dt, $J$=16.0, 4.9 Hz, 1 H), 6.63 (dd, $J$=2.4, 9.0 Hz, 1 H), 6.51 (d, $J$=2.4 Hz, 1 H), 6.45 (dt, $J$=16.0, 1.8 Hz, 1 H), 6.18 (br s, NH), 6.02 (s, 1 H), 5.92 (ddt, $J$=15.2, 4.3, 1.8 Hz, 1 H), 5.67 (dt, $J$=15.2, 5.1 Hz, 1 H), 5.23 (m, 1 H), 4.21 (m, 4 H), 3.84 (m, 1 H), 3.69 (s, 2 H), 3.45 (t, $J$=6.2 Hz, 2 H), 3.05 (s, 6 H), 2.71 (t, $J$=6.2 Hz, 2 H); HRMS (FAB) *m/z* calcd for $C_{26}H_{30}N_2O_7SNa$ $[M+Na]^+$ 537.1671, found 537.1641 *m/z*.

Adduct **20**: $^1$H NMR (400 MHz, 20% $CDCl_3$ in $CD_3OD$) *d*=7.48 (d, *J*=8.8 Hz, 1 H), 7.31 (s, 1 H), 6.79 (dt, *J*=15.5, 5.1 Hz, 1 H), 6.63 (dd, *J*=2.4, 8.8 Hz, 1 H), 6.50 (d, *J*=2.4 Hz, 1 H), 6.41 (dd, *J*=15.5, 1.8 Hz, 1 H), 6.27 (br s, NH), 6.06 (s, 1 H), 5.49 (m, 1 H), 4.21 (dd, *J*=5.0, 1.8 Hz, 2 H), 3.84 (d, *J*=4.5 Hz, 2 H), 3.66 (s, 2 H), 3.51 (t, *J*=6.4 Hz, 2 H), 3.06 (s, 6 H), 2.69 (t, *J*=6.4 Hz, 2 H), 2.61 (m, 1 H), 2.29 (m, 2 H); HRMS (FAB) *m/z* calcd for $C_{26}H_{30}N_2O_7SNa$ $[M+Na]^+$ 537.1671, found 537.1632 *m/z*.

Mucosin analogue **21**



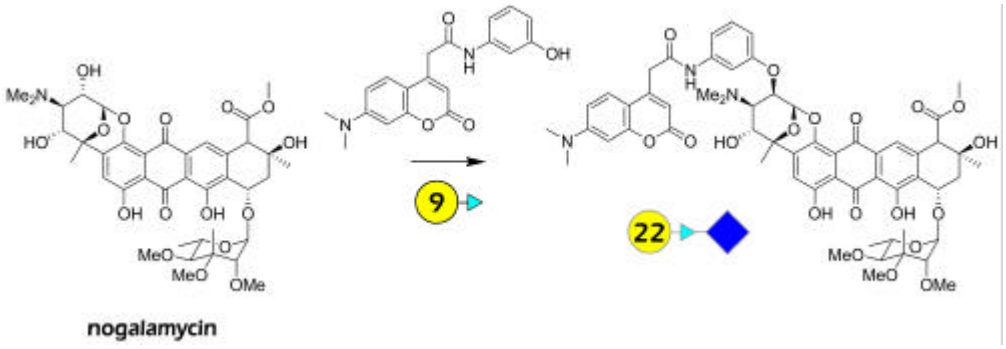

A mixture of **8** (0.5 mg, 1.4 µmol) and (-)-mucosin (0.4 mg, 0.62 µmol) was dried by evaporation of toluene (3x5 mL). The residue was taken up in dry $CH_2Cl_2$ (2 mL) and treated with dried (1*S*)-(+)-10-camphorsulfonic acid (14 µg, 63 nmol, 10 mol%) dissolved in $CH_2Cl_2$ (100 µL) at 0 °C. The reaction mixture was warmed over 1 hr to RT. After incubation at RT for 3 h, solid sodium bicarbonate (10 mg) was added and the mixture was subjected to flash chromatography (hexanes to $CH_2Cl_2$) to yield 0.44 mg (75% yield) of a 5:1 mixture of acetal isomers **21**. The yield of **21** was determined spectrophotometrically in a 5 mL volumentric flask using the absorption or fluorescence from the coumarin tag ($\lambda_{ex}$=370 nm, $\varepsilon$=22 000 $M^{-1}cm^{-1}$, $\lambda_{em}$=459 nm). Each isomer was separable via reversed-phase HPLC using a gradient of 80% aq. $CH_3CN$ to $CH_3CN$.

*Major isomer*: $^1$H NMR (400 MHz, 25% $CD_3OD$ in $CDCl_3$) *d*=7.36 (d, *J*=10.4 Hz, 1 H), 7.26 (s, 1 H), 6.46 (dd, *J*=2.4, 10.4 Hz, 1 H), 6.34 (d, *J*=2.4, 1 H), 5.92 (m, 1 H), 5.86 (s, 1 H), 5.27 (s, 1 H), 4.80 (m, 1 H), 4.76 (s, 1 H), 3.45 (s, 1 H), 3.30 (m, 1 H), 3.17 (s, 6 H), 3.16 (m, 1 H), 3.02 (m, 1 H), 2.84 (s, 2 H), 2.82 (ddd, *J*=8.1,8.1,8.1 Hz 1 H), 2.68 (m, 2

H), 2.38 (d, *J*=9.6 Hz 1 H), 2.15 (dd, *J*=4.8, 13.6 Hz, 1 H), 1.85 (m, 2 H), 1.62-1.05 (cm, 50 H), 1.46 (d, 5.6 3 H), 0.67 (m, 3 H); HRMS (FAB) *m/z* calcd for $C_{54}H_{84}N_2O_{11}Na$ $[M+Na]^+$ 959.5973, found 959.5911 *m/z*.

*Minor isomer*: $^1$H NMR (400 MHz, 25% $CD_3OD$ in $CDCl_3$) *d*=7.37 (d, *J*=9.2 Hz, 1 H), 7.26 (s, 1 H), 6.50 (dd, *J*=2.4, 9.2 Hz, 1 H), 5.92 (m, 1 H),.27 (s, 1 H), 4.80 (m, 1 H), 4.76 (s, 1 H), 3.45 (s, 1 H), 3.30 (m, 1 H), 3.17 (s, 6 H), 3.16 (m, 1 H), 3.02 (m, 1 H), 2.84 (s, 2 H), 2.82 (ddd, *J*=8.1,8.1,8.1 Hz 1 H), 2.68 (m, 2 H), 2.38 (d, *J*=9.6 Hz 1 H), 2.15 (dd, *J*=4.8, 13.6 Hz, 1 H), 1.85 (m, 2 H), 1.62-1.05 (cm, 50 H), 146 (d, 5.6 3 H), 0.67 (m, 3 H); HRMS (FAB) *m/z* calcd for $C_{54}H_{84}N_2O_{11}Na$ $(M+Na)^+$ 959.5973, found 959.5992 *m/z*.

Nogalamycin analogue **22**




A mixture nogalamycin (1.2 mg, 1.5 μmol), label **9** (0.8 mg, 2.3 μmol), and $Ph_3P$ (1.2 mg, 4.6 μmol) were mixed and dried by evaporation of toluene (3x3 mL). The residue was dissolved in dry $CH_2Cl_2$ (1 mL) and DEAD (0.8 mg, 4.6 μmol) was added dropwise in toluene (150 μL). After 12 h, the solution was applied to flash chromatography (hexanes to 10% MeOH in $CH_2Cl_2$) to afford 0.59 mg of **22** (35%). The yield of analogue **22** was determined spectrophotometrically in a 5 mL volumentric flask using the absorption or fluorescence from the coumarin tag ($?_{ex}$=370 nm, *e*=22 000 $M^{-1}cm^{-1}$, $?_{em}$=459 nm). $^1$H NMR (400 MHz, 30% $CD_3OD$ in $CDCl_3$) *d*=9.41 (br s, NH), 7.63 (d, *J*=8.8 Hz, 1 H), 7.54 (s, 1 H), 7.19 (s, 1 H), 7.06 (s, 1 H), 7.04 (t, *J*=8.2 Hz, 1 H), 6.99 (d, *J*=8.0, 1 H), 6.71 (dd, *J*=2.2, 9.0 Hz, 1 H), 6.52 (d, *J*=2.8 Hz, 1 H), 6.39 (dd, *J*=1.4, 8.2 Hz 1 H), 6.11 (s, 1 H), 5.87 (d, *J*=3.8 Hz, 1 H), 5.39 (br s, 1 H), 5.09 (m, 1 H), 4.42 (s, 1 H), 3.99 (s, 1 H), 3.93 (d, *J*=4.5 Hz, 1 H), 3.79 (s, 3 H), 3.72 (m, , 1 H), 3.61 (s, 2 H), 3.53 (s, 3 H), 3.49 (s, 3 H), 3.27 (d, *J*=3.9 Hz, 1 H), 3.21 (s, 3 H), 3.09 (d, *J*=6.8 Hz, 1 H), 3.03 (s, 6 H), 2.71 (d, *J*=3.8 Hz, 1 H), 2.32 (s, 6 H), 1.82 (m, 2 H), 1.68 (s, 3 H), 1.52 (s, 3 H), 1.23 (d, *J*=5.8

Hz, 3 H), 1.09 (s, 3 H); HRMS (FAB) *m/z* calcd for $C_{56}H_{65}N_3O_{19}Na$ $[M+Na]^+$ 1130.4110, found 1130.4085 *m/z*.