Electronic Supplementary Information

Programmable Chemical Switch based on Triggerable Michael Acceptors

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Materials and Instrumentation

All reagents were used as received from commercial sources unless otherwise mentioned. $^1$H NMR, $^{13}$C NMR spectra were recorded on a Brucker 400 MHz NMR spectrometer. UV-Vis spectra were recorded on PerkinElmer Lambda 35 UV/Vis spectrometer. Fluorescent spectra were obtained from PerkinElmer LS 55 fluorescence spectrometer. Protein labeling and quantification were carried out on Biodrop microvolume UV-Vis spectrophotometer. Mass spectra were recorded on a Bruker AmaZon quadrupole ion trap mass spectrometer coupled with electrospray ionization source or a Thermo Orbitrap Fusion tribrid (quadrupole, orbitrap, and ion trap) mass spectrometer coupled with Easy nLC 1000 nanoLC system. Rheological measurements were performed by a Malvern Kinexus Pro rheometer.

Synthesis of compound 1 to 38

**Synthesis of compound 1b:** Compound 1b was synthesized followed the reported procedure. Briefly, compound 1a (2.0 g, 17.23 mmol) was dissolved in 20 mL of dry DCM and cooled to 0 °C in ice bath. PBr$_3$ (2.332 g, 8.615 mmol) was added slowly to the solution. Then the reaction mixture was stirred at room temperature. The completion of reaction was monitored by TLC. The reaction was then quenched by the addition of ice. The reaction solution was neutralized by sodium bicarbonate solution and then extracted with DCM three times. Organic layers were collected and dried over anhydrous sodium sulfate, then concentrated. Compound 1b was obtained by flash chromatography. Yield: 2.52 g, 82%. $^1$H-NMR (400 MHz, CDCl$_3$): δ 6.34 (s, 1H), 5.96 (s, 1H), 4.18 (s, 2H), 3.82 (s, 3H).

**Synthesis of compound 1:** Compound 1b (50 mg, 0.28 mmol) was dissolved in 200 uL of dry THF. To the solution, triethylamine (57 mg, 0.56 mmol) was added. The reaction was allowed to stirred at room temperature for 2 hours. Then mixture was concentrated and precipitated in dry diethyl ether for 3 times. The precipitate was collected and dried to afford compound 1. Yield: 65 mg, 83%. $^1$H-NMR (400 MHz, MeOH-d$_4$): δ (ppm) 6.90 (s, H), 6.40 (s, H), 4.23 (s, 2H), 3.85 (s, 3H), 3.30 (q, 6H), 1.37 (t, 9H). $^{13}$C-NMR (100 MHz, MeOH-d$_4$): δ (ppm) 167.42, 140.52, 130.66, 56.97, 54.30, 53.44, 8.08. MS (m/z): [M]$^+$ calcd. for C$_{11}$H$_{22}$BrNO$_2$, 279.08; found, 200.4 for [M-Br]$^+$.

**Synthesis of compound 2.** Compound 1b (25 mg, 0.14 mmol) was weighed to glass vial. To the vial, tripropylamine (40 mg, 0.28 mmol) was added. The reaction was kept for 2 hours at room temperature. Then the white precipitate was washed with diethyl ether for three times. The precipitate was collected and dried to achieve compound 2. Yield: 32.1 mg, 71%. $^1$H-NMR (400 MHz, MeOH-d$_4$): δ (ppm) 6.90 (s, 1H), 6.36 (s, 1H), 4.27 (s, 2H), 3.85 (s, 3H), 3.15 (p, 6H), 1.81 (m, 6H), 0.99 (t, 9H). $^{13}$C-NMR (100 MHz, MeOH-d$_4$): δ (ppm) 166.04, 139.16, 129.44, 60.00, 57.31, 52.09, 15.27, 9.31. MS (m/z): [M]$^+$ calcd. for C$_{14}$H$_{28}$BrNO$_2$, 321.13; found, 242.3 for [M-Br]$^+$.
Synthesis of compound 3. Compound 1b (25 mg, 0.14 mmol) was weighed to glass vial. To the vial, N, N-diisopropylethylamine (36.2 mg, 0.28 mmol) was added. The reaction was kept for 2 hours at room temperature. Then the white precipitate was washed with diethyl ether for three times. The precipitate was collected and dried to achieve compound 3. Yield: 35.7, 83%. $^1$H-NMR (400 MHz, MeOH-d4): $\delta$(ppm) 6.83 (s, 1H), 6.46 (s, 1H), 4.23 (s, 2H), 4.16 (m, 2H), 3.87 (s, 3H), 3.46 (q, 2H), 1.51 (q, 12 H), 1.40 (t, 3H). $^{13}$C-NMR (100 MHz, MeOH-d4): $\delta$(ppm) 166.48, 138.62, 131.19, 62.66, 57.00, 52. 16, 17.65, 17.21, 8.76. MS (m/z): [M]$^+$ calcd. for C$_{13}$H$_{26}$BrNO$_2$, 307.11; found, 228.4 for [M-Br]$^+$.

Synthesis of compound 4. Compound 1b (25 mg, 0.14 mmol) was weighed to glass vial. To the vial, pyridine (22.1 mg, 0.28 mmol) was added. The reaction was kept for 2 hours at room temperature. Then the white precipitate was washed with diethyl ether for three times. The precipitate was collected and dried to achieve compound 4. Yield: 30.1 mg, 83%. $^1$H-NMR (400 MHz, MeOH-d4): $\delta$(ppm) 9.06 (d, $J=5.6$ Hz, 2H), 8.63 (t, $J=7.8$ Hz, 1H), 8.14 (t, 2H), 6.67(s, 1H), 6.36 (s, 1H), 5.53(s, 2H) 3.75 (s, 1H). $^{13}$C-NMR (100 MHz, MeOH-d4): $\delta$(ppm) 166.29, 147.56, 146.52, 134.89, 134.83, 62.94, 53.03. MS (m/z): [M]$^+$ calcd. for C$_{10}$H$_{12}$BrNO, 257.01; found, 178.4 for [M-Br]$^+$.

Synthesis of compound 5. 4-(dimethylamino) pyridine (24.4 mg, 0.2 mmol) was dissolved in 100 uL of dry THF. To the solution, compound 1b (50 mg, 0.28 mmol) was added. The reaction was kept for 2 hours at room temperature. Then the white precipitate was washed with diethyl ether for three times. The precipitate was collected and dried to achieve compound 5. Yield: 53.2 mg, 89%. $^1$H-NMR (400 MHz, MeOH-d4): $\delta$ 8.16 (d, 2H), 6.99 (d, 2H), 6.52(s, 1H), 6.08(s, 1H), 5.02(s, 2H), 3.76(s, 3H), 3.26 (s, 6H). $^{13}$C-NMR (100 MHz, MeOH-d4): $\delta$(ppm) 166.53, 158.10, 143.43, 136.17, 132.28, 108.76, 58.93, 52.87, 40.36. MS (m/z): [M]$^+$ calcd. for C$_{12}$H$_{17}$BrNO, 300.05; found, 221.3 for [M-Br]$^+$.

Synthesis of compound 6. Compound 1b (232 mg, 2 mmol) was dissolved in 5 mL of DCM with TEA (220 mg, 2 mmol) and cooled to 0 °C. To the solution, acetyl chloride (157 mg, 2 mmol) was added dropwise. The reaction mixture was stirred at room temperature after addition. After reaction, the reaction solution was collected by filtration and extracted with water for three times. The organic layers was collected and dried over anhydrous sodium sulfate. The solution was further filtered and concentrated to afford crude product which was subjected to flash chromatography to obtain pure compound 6. Yield: 224.4 mg, 71%. $^1$H-NMR (400 MHz, CDCl$_3$): $\delta$ 6.36 (s, 1H), 5.85 (s, 2H), 4.81 (s, 2H), 3.79 (s, 3H), 2.10 (s, 3H). $^{13}$C-NMR (100 MHz, CDCl$_3$): $\delta$(ppm) 170.41, 165.68, 135.21, 127.60, 62.47, 52.06, 20.88. The compound was previously reported.²
Synthesis of compound 7. Compound 1b (116 mg, 1 mmol) was dissolved in 5 mL of THF with 4-nitrophenyl isocyanate (164.1 mg, 1 mmol) and 2,6-Di-tert-butyl-4-methylphenol (2.2 mg, 0.01 mmol). The reaction mixture was heated at 60 °C under argon atmosphere. The completion of reaction was followed by TLC. Compound 7 was obtained by flash chromatography. Yield: 134 mg, 48%.

$^1$H-NMR (400 MHz, MeOH-d$_4$): $\delta$ (ppm) 8.19 (d, 2H), 7.67 (d, 2H), 6.37 (d, 1H), 5.98 (d, 1H), 4.91 (s, 2H), 3.79 (s, 3H).

$^{13}$C-NMR (100 MHz, MeOH-d$_4$): $\delta$ (ppm) 165.71, 153.21, 145.29, 142.54, 135.75, 126.75, 124.45, 117.50, 62.88, 51.13.

MS (m/z): [M]$^+$ calcd. for C$_{12}$H$_{12}$N$_2$O$_6$, 280.07; found, 303.1 for [M+Na]$^+$.

Synthesis of compound 8, 9, 10.

Synthesis of compound 8a. Compound 1a (3.48 g, 30 mmol) was dissolved in 50 mL of dry THF with imidazole (2.250 g, 33 mmol). TBDPSCI (9.070 g, 33 mmol) was added to the solution dropwise at 0 °C. The reaction was stirred overnight at room temperature. Then, precipitate was removed by filtration to achieve clear solution which was further extracted with saturated sodium bicarbonate solution, water and brine. The organic layers were combined and dried over anhydrous sodium sulfate. The solution was collected, concentrated and subjected to flash chromatography to afford compound 8a. Yield, 6.99 g, 66%.

$^1$H-NMR (400 MHz, CDCl$_3$): $\delta$ (ppm) 7.64-7.69 (m, 4H), 7.34-7.47 (m, 6H), 6.33 (q, 1H), 6.11 (q, 1H), 4.42 (t, 2H), 3.70 (s, 3H), 1.08 (s, 9H).

$^{13}$C-NMR (100 MHz, CDCl$_3$): $\delta$ (ppm) 166.25, 139.33, 135.46, 133.24, 129.78, 127.77, 124.09, 62.21, 51.63, 26.82, 19.30. MS (m/z): [M]$^+$ calcd. for C$_{21}$H$_{26}$O$_3$Si, 354.17; found, 377.1 for [M+Na]$^+$.

Synthesis of compound 8b. Compound 8b (5 g, 14.15 mmol) was dissolved in 50 mL of THF/H$_2$O mixture (1:1). Lithium hydroxide (1.015 g, 42.45 mmol) in 2 mL of water was added to the solution. The reaction was stirred overnight. Then, the solution was acidified with 2M HCl solution to pH 3 and extracted with DCM. Organic layers were collected and dried over anhydrous sodium sulfate. The organic layer was further concentrated to obtain crude product, compound 8b which was subjected for reaction without further purification. Yield: 3.925 g, 82%. $^1$H-NMR (400 MHz, CDCl$_3$): $\delta$ (ppm) 7.62-7.68 (m, 4H), 7.34-7.46 (m, 6H), 6.43 (s, 1H) 6.18 (s, 1H), 4.40 (q, 0.92Hz, 2H), 1.08 (s, 9H). $^{13}$C-NMR (100 MHz, CDCl$_3$): $\delta$ (ppm) 169.70, 138.42, 135.44, 133.00, 129.86, 127.80, 126.47, 62.06, 26.80. 19.30. MS (m/z): [M]$^+$ calcd. for C$_{20}$H$_{24}$O$_3$Si, 340.15; found, 363.1 for [M+Na]$^+$.

Synthesis of compound 8c. To 50 mL of compound 8b (2.620 g, 8.13 mmol) solution in THF, thionyl chloride (19.4 g, 160.26 mmol) was added. The mixture was refluxed for 3 hours. Then volatiles were removed under vacuum. The residue was dissolved in 10 mL of dry DCM in a round bottom flask. To the
flask, a solution of methylamine (4.13 mL 2M THF solution, 8.13 mmol) and TEA (0.828 g, 8.13 mmol) in 10 mL DCM was added dropwise at 0 °C. The reaction was stirred overnight at room temperature. The white precipitate was removed by filtration. Solvent was removed and the residue was extracted with sodium bicarbonate, and water using ethyl acetate. The organic layers were combined and dried over anhydrous sodium sulfate. The solution was collected and dried to achieve compound 8c. Then the white precipitate was washed with diethyl ether for three times. The precipitate was collected and subjected to flash chromatography. Pure compound 8c was obtained after purification. Yield: 2.60 g, 95%. \(^1\)H-NMR (400 MHz, CDCl\(_3\)): 7.62-7.69 (m, 4H), 7.36-7.48 (m, 6H), 6.84 (bs, NH), 5.98 (s, 1H), 5.36 (d, 1H), 4.41 (s, 2H), 2.89 (d, 3H), 1.07 (s, 9H).

**Synthesis of compound 8d.** To 10 mL of compound 8e (2.50 g, 7.08 mmol) solution, 7.79 mL of TBAF solution (1M in THF) was added dropwise at 0 °C. The reaction completed in 20 minutes. Compound 8d was obtained by flash chromatography. Yield: 513 mg, 63%. \(^1\)H-NMR (400 MHz, MeOH-d4): 5.80 (d, 1H), 5.58 (d, 1H), 4.27 (s, 2H), 2.79 (s, 3H). \(^13\)C-NMR (100 MHz, MeOH-d4): 169.06, 143.76, 118.13, 61.22, 24.94.

**Synthesis of compound 8e.** To the solution of compound 8d (230 mg, 2 mmol) in 5 mL of dry DCM, PBr\(_3\) (270 mg, 1 mmol) was added slowly at 0 °C. The reaction was stirred at room temperature after addition. The completion of reaction was followed by TLC. Then reaction was quenched by addition of 2 M sodium bicarbonate solution. The solution was further extracted with DCM and water for two more times, and organic layers were combined and dried over anhydrous sodium sulfate. The solution was collected and dried to afford compound 8e for further reaction.

**Synthesis of compound 8.** Compound 8e (35.6 mg, 0.2 mmol) was weighed into a small glass vial. To the vial, triethylamine (44.48 mg, 0.44 mmol) was added. The reaction was kept for 2 hours at room temperature. Then the white precipitate was washed with diethyl ether for three times. The precipitate was collected and dried to afford compound 8. Yield: 42 mg, 75%. \(^1\)H-NMR (400 MHz, MeOH-d4): 6.24 (s, 1H), 6.07 (s, 1H), 4.19 (s, 2H), 3.26 (q, 6H), 2.83 (s, 3H), 1.34 (t, J=7.20 Hz, 9H). \(^13\)C-NMR (100 MHz, MeOH-d4): 168.41, 134.17, 131.35, 56.41, 52.78, 25.18, 6.65. MS (m/z): [M]\(^+\) calcd. for C\(_{11}\)H\(_7\)BrN\(_2\)O, 278.10; found, 199.4 for [M-Br]\(^+\).

**Synthesis of compound 9.** Compound 8d (115 mg, 1 mmol) was dissolved in 1 mL of dry DCM with triethylamine (101 mg, 1 mmol). The reaction mixture was cooled to 0 °C followed by slow addition of acetyl chloride (78 mg, 1 mmol). Then reaction was stirred at room temperature for 6 hours. The mixture was dried and subjected to flash chromatography to afford pure compound 8d. Yield: 120 mg, 76%. \(^1\)H-NMR (400 MHz, CDCl\(_3\)): \(\delta\) (ppm) 6.12 (bs, NH), 5.94 (s, 1H), 5.61 (s, 1H), 4.82 (d, 2H), 2.88 (d, 3H), 2.10 (s, 3H). \(^13\)C-NMR (100 MHz, MeOH-d4): \(\delta\) (ppm) 170.80, 166.88, 139.39, 122.97, 63.59, 26.61, 21.07. MS (m/z): [M]\(^+\) calcd. for C\(_{11}\)H\(_{11}\)NO\(_3\), 157.07; found, 180.4 for [M+Na]\(^+\).

**Synthesis of compound 10.** Compound 8d (115 mg, 1 mmol) was dissolved in 5 mL of THF with 4-nitrophenyl isocyanate (164.1 mg, 1 mmol) and 2,6-Di-tert-butyl-4-methylphenol (2.2 mg, 0.01 mmol). The reaction mixture was heated at 60 °C under argon atmosphere. The completion of reaction was followed by TLC. Compound 7 was obtained by flash chromatography. Yield: 166 mg, 59%. \(^1\)H-NMR (400 MHz, MeOH-d4): \(\delta\) (ppm) 8.18 (m, 2H), 7.67 (m, 2H), 5.91 (s, 1H), 5.72 (t, 1H), 4.91 (dd, 2H), 2.80 (s, 3H). \(^13\)C-NMR (100 MHz, MeOH-d4): \(\delta\) (ppm) 168.09, 153.23, 145.32, 142.52, 139.66, 124.46, 120.84, 117.49, 63.78, 25.09. MS (m/z): [M]\(^+\) calcd. for C\(_{12}\)H\(_{13}\)N\(_3\)O\(_3\), 279.09; found, 302.1 for [M+Na]\(^+\).
Synthesis of compound 11. 2-(Hydroxy-phenyl-methyl)-acrylic acid methyl ester (403.4 mg, 2.1 mmol) was dissolved in 5 mL dry DCM and cooled to 0 °C. To the solution, PBr₃ (285 mg, 1.05 mmol) was added slowly. The solution was quenched by addition of 2 M sodium bicarbonate solution. The mixture was extracted using DCM, then washed with water for 2 more times. The organic layer was collected and dried over anhydrous sodium sulfate. The product was obtained after filtration and concentration followed by drying over vacuum. The product was used for next step without purification. Then compound 11a (25.5 mg, 0.1 mmol) was weighed into a small vial followed by addition of triethylamine (20.2 mg, 0.2 mmol). The reaction was kept for 2 hours. The white precipitate was washed with diethyl ether for 3 times and dried to afford compound 11. Yield: 29 mg, 79%. ¹H-NMR (400 MHz, MeOH-d₄): δ(ppm) 8.44 (s, 1H), 7.41-7.58 (m, 4H), 4.47 (s, 2H), 3.92 (s, 3H), 3.11 (q, 6H), 1.06 (t, 3H). ¹³C-NMR (100 MHz, MeOH-d₄): δ(ppm) 168.65, 153.25, 135.49, 130.99, 130.62, 129.66, 123.61, 54.32, 53.61, 51.88, 7.91. MS (m/z): [M⁺] calcd. for C₁₁H₂₆BrO₂, 355.11; found, 276.2 for [M-Br]⁺.

Synthesis of compound 12. Methyl 2-[hydroxy (4-methoxyphenyl) methyl]acrylate (222.2 mg, 1.0 mmol) was dissolved in 5 mL dry DCM and cooled to 0 °C. To the solution, PBr₃ (142 mg, 0.5 mmol) was added slowly. The solution was quenched by addition of 2 M sodium bicarbonate solution. The mixture was extracted using DCM, then washed with water for 2 more times. The organic layer was collected and dried over anhydrous sodium sulfate. The product was obtained after filtration and concentration followed by drying over vacuum. The product was used for next step without purification. Then compound 12a (28.5 mg, 0.1 mmol) was weighed into a small vial followed by addition of triethylamine (20.2 mg, 0.2 mmol). The reaction was kept for 2 hours. The white precipitate was washed with diethyl ether for 3 times and dried to afford compound 12. Yield: 29 mg, 75%. ¹H-NMR (400 MHz, MeOH-d₄): δ(ppm) 8.35 (s, 1H), 7.45(m, 2H), 7.08(m, 2H), 4.52 (s, 2H), 3.90 (s, 3H), 3.86 (s, 3H), 3.14 (q, 6H), 1.12 (t, Hz, 3H). ¹³C-NMR (100 MHz, MeOH-d₄): δ(ppm) 168.96, 162.72, 153.01, 131.88, 127.39, 121.51, 116.01, 55.99, 54.31, 53.47, 52.20, 8.05. MS (m/z): [M⁺] calcd. for C₁₈H₂₆BrO₃, 385.13; found, 306.1 for [M-Br]⁺.

Synthesis of compound 13. Methyl 2-[hydroxy (4-bromophenyl) methyl]acrylate (271.1 mg, 1.0 mmol) was dissolved in 5 mL dry DCM and cooled to 0 °C. To the solution, PBr₃ (142 mg, 0.5 mmol) was added slowly. The solution was quenched by addition of 2 M sodium bicarbonate solution. The mixture was extracted using DCM, then washed with water for 2 more times. The organic layer was collected and dried over anhydrous sodium sulfate. The product was obtained after filtration and concentration followed by
drying over vacuum. The product was used for next step without purification. Then compound 13a (33.4 mg, 0.1 mmol) was weighed into a small vial followed by addition of triethylamine (20.2 mg, 0.2 mmol). The reaction was kept for 2 hours. The white precipitate was washed with diethyl ether for 3 times and dried to afford compound 11. Yield: 38 mg, 88%. 

1H-NMR (400 MHz, MeOH-d4): δ 8.34 (s, 1H), 7.71 (m, 2H), 7.39 (m, 2H), 4.45 (s, 2H), 3.92 (s, 3H), 3.13 (q, 6H), 1.09 (t, 9H). 13C-NMR (100 MHz, MeOH-d4): 168.48, 151.88, 134.42, 133.81, 131.61, 125.19, 124.13, 54.39, 53.68, 51.93, 7.97. MS (m/z): [M]+ calcd. for C17H25Br2NO2, 331.90; found, 354.8 for [M+Na]+.

Synthesis of compound 14. 2-(Hydroxy-phenyl-methyl)-acrylic acid methyl ester (192 mg, 1.0 mmol) was dissolved in 1 mL of DCM with triethylamine (101.2 mg, 1.0 mmol) and cooled to 0°C. Acetyl chloride (78.5 mg, 1.0 mmol) was added to the solution slowly. Completion of reaction was monitored by TLC. The reaction mixture was extracted using DCM and water. The organic layer was collected and dried over anhydrous sodium sulfate followed by filtration. The solution was then concentrated and subjected to flash chromatography to afford compound 14. Yield: 162 mg, 69%. 1H-NMR (400 MHz, CDCl3): δ (ppm) 7.27-7.41 (m, 5H), 6.68 (s, 1H), 6.40 (s, 1H), 5.86 (s, 1H), 3.71 (s, 3H), 2.11 (s, 3H).

13C-NMR (100 MHz, CDCl3): δ (ppm) 169.57, 165.57, 139.80, 137.93, 128.61, 128.54, 127.82, 125.93, 73.26, 52.15, 21.25.

MS (m/z): [M]+ calcd. for C14H16O5, 264.1; found, 287.1 for [M+Na]+.

Synthesis of compound 15. Methyl 2-[hydroxy (4-methoxyphenyl) methyl]acrylate (444.4 mg, 2.0 mmol) was dissolved in 4 mL of DCM with triethylamine (202.4 mg, 2.0 mmol) and cooled to 0°C. Acetyl chloride (157 mg, 2.0 mmol) was added to the solution slowly. Completion of reaction was monitored by TLC. The reaction mixture was extracted using DCM and water. The organic layer was collected and dried over anhydrous sodium sulfate followed by filtration. The solution was then concentrated and subjected to flash chromatography to afford compound 15. Yield: 196 mg, 74%. 1H-NMR (400 MHz, CDCl3): δ (ppm) 7.30 (d, J=4.85Hz, 2H), 6.86 (d, J=4.85Hz, 2H), 6.63 (s, 1H), 6.34 (s, 1H), 5.87 (t, 1H), 3.79 (s, 1H), 3.70 (s, 1H). 13C-NMR (100 MHz, CDCl3): δ (ppm) 169.62, 165.62, 159.78, 139.89, 129.98, 129.32, 125.27, 114.00, 73.00, 55.41, 52.13, 21.30. MS (m/z): [M]+ calcd. for C14H16O5, 264.1; found, 287.1 for [M+Na]+.

Synthesis of compound 16. Methyl 2-[hydroxy (4-bromophenyl) methyl]acrylate (542 mg, 2.0 mmol) was dissolved in 4 mL of DCM with triethylamine (202.4 mg, 2.0 mmol) and cooled to 0°C. Acetyl chloride (157 mg, 2.0 mmol) was added to the solution slowly. Completion of reaction was monitored by TLC. The reaction mixture was extracted using DCM and water. The organic layer was collected and dried over anhydrous sodium sulfate followed by filtration. The solution was then concentrated and subjected to flash
chromatography to afford compound 16. Yield: 197 mg, 63%. 1H-NMR (400 MHz, CDCl3): 7.30 (m, 2H), 6.86 (m, 2H), 6.63 (s, 1H), 6.40 (t, 1H), 5.87 (dd, 1H), 3.79 (s, 1H), 3.70 (s, 1H). 13C-NMR (100 MHz, CDCl3): δ(ppm) 169.45, 165.35, 139.64, 137.57, 129.57, 126.06, 122.66, 72.65, 52.22, 21.20. MS (m/z): [M]+ calcd. for C13H7BrO4, 312.00; found, 335.0 for [M+Na]+.

1H-NMR (400 MHz, CDCl3): 7.30 (m, 2H), 6.86 (m, 2H), 6.63 (s, 1H), 6.40 (t, 1H), 5.87 (dd, 1H), 3.79 (s, 1H), 3.70 (s, 1H). 13C-NMR (100 MHz, CDCl3): δ(ppm) 169.45, 165.35, 139.64, 137.57, 129.57, 126.06, 122.66, 72.65, 52.22, 21.20. MS (m/z): [M]+ calcd. for C13H7BrO4, 312.00; found, 335.0 for [M+Na]+.

Synthesis of compound 17. Compound 7 (56 mg, 0.2 mmol) was dissolved in 1 mL of MeOH. To the solution, 2-(2-methoxyethoxy) ethanethiol (26 mg, 0.19 mmol) in 0.5 mL water was slowly added. Solvent was removed after reaction. The residue was extracted with DCM for three times. Organic layers were combined and dried over anhydrous sodium sulfate. The solution was dried after filtration and then subjected to flash chromatography afford compound 17. Yield: 40 mg, 90%. 1H-NMR (400 MHz, MeOH-d4): δ(ppm) 7.98 (m, 2H), 6.62 (m, 2H), 6.15 (d, 1H), 5.72 (d, 1H), 3.76 (s, 3H), 3.63 (t, 2H), 3.57-3.61 (m, 2H), 3.51-3.56 (m, 2H), 3.44 (d, 2H), 3.36 (s, 3H), 2.63 (t, 3H). 13C-NMR (100 MHz, CDCl3): δ(ppm) 166.46, 136.78, 126.32, 71.94, 70.95, 70.28, 59.09, 52.09, 33.06, 30.66. MS (m/z): [M]+ calcd. for C10H18O4S, 234.09; found, 257.1 for [M+Na]+.

Synthesis of compound 18. Compound 8d (140 mg, 0.5 mmol) was dissolved in 2 mL of water. To the solution, ethanethiol (28.2 mg, 0.45 mmol) was slowly added. Completion of reaction was monitored by TLC. Then the reaction mixture was extracted with DCM for three times. Organic layers were combined and dried over anhydrous sodium sulfate. The solution was dried after filtration and then subjected to flash chromatography afford compound 18. Yield: 72 mg, 50%. 1H-NMR (400 MHz, CDCl3): δ(ppm) 6.20 (d, 1H), 5.65 (q, 1H), 3.79 (s, 3H), 3.40 (s, 2H), 2.48 (q, 2H), 1.24 (t, 3H). 13C-NMR (100 MHz, CDCl3): δ(ppm) 166.71, 136.90, 125.80, 52.08, 32.34, 25.41, 14.28. The compound was previously reported.

Synthesis of compound 19. Compound 8e (53.4 mg, 0.3 mmol) and 2-(2-methoxyethoxy) ethanethiol (39.0 mg, 0.29 mmol) was dissolved in 1 mL of DCM. To the solution, triethylamine (30.5 mg, 0.3 mmol) was added. Completion of reaction was monitored by TLC. Then the reaction mixture was extracted using DCM and water for three times. Organic layers were combined. Organic layers were combined and dried over anhydrous sodium sulfate. The solution was dried after filtration and then subjected to flash chromatography afford compound 19. Yield: 58 mg, 85%. 1H-NMR (400 MHz, D2O): δ(ppm) 5.74 (s, 1H), 5.54 (s, 1H), 3.58-3.72 (m, 6H), 3.48 (s, 2H), 3.39 (s, 3H), 2.82 (s, 3H), 2.71 (t, 2H). 1H-NMR (100 MHz, MeOH-d4): δ(ppm) 169.32, 141.14, 119.15, 71.57, 70.60, 69.69, 57.69, 32.86, 29.75, 25.15. MS (m/z): [M]+ calcd. for C10H19NO3S, 233.11; found, 256.1 for [M+Na]+.
**Synthesis of compound 20.** Compound 13a (70 mg, 0.21 mmol) and triethylamine (21.2 mg, 0.21 mmol) was mixed in a glass vial and kept for 30 minutes. Then ethanethiol (6.8 mg, 0.11 mmol) in 1 mL of MeOH was added to the vial. Completion of reaction was monitored by TLC. Then the reaction mixture was extracted using DCM and water for three times. Organic layers were combined and dried over anhydrous sodium sulfate. The solution was dried after filtration and then subjected to flash chromatography to afford compound 20. Yield: 30 mg, 87%. 

\[ \text{ Compound 13a (70 mg, 0.21 mmol)} \]

\[ \begin{align*} \text{Synthesis of compound 21.} \end{align*} \]

MeOH solution (1 mL) of compound 16 (100 mg, 0.32 mmol) was added with 1 mL of saturate NaHCO\(_3\) aqueous solution. To the mixture ethanethiol (19.8 mg, 0.32 mmol) was added. The was stirred at room temperature until reaction completed. MeOH was then removed. The residue was extracted using DCM and water for 3 times. Organic layers were combined and dried over anhydrous sodium sulfate. The solution was dried after filtration and then subjected to flash chromatography to afford compound 21. Yield: 85 mg, 84%. The product contains 13% cis and 87% trans isomers. 

\[ \text{ Compound 16 (100 mg, 0.32 mmol)} \]

\[ \begin{align*} \text{Synthesis of compound 22.} \end{align*} \]

MeOH solution (1 mL) of compound 15 (132 mg, 0.50 mmol) was added with 1 mL of saturate NaHCO\(_3\) aqueous solution. To the mixture ethanethiol (31 mg, 0.50 mmol) was added. The was stirred at room temperature until reaction completed. MeOH was then removed. The residue was extracted using DCM and water for 3 times. Organic layers were combined and dried over anhydrous sodium sulfate. The solution was dried after filtration and then subjected to flash chromatography to afford compound 22. Yield: 85 mg, 84%. The product contains 7% cis and 93% trans isomers. 

\[ \text{ Compound 15 (132 mg, 0.50 mmol)} \]
168.39, 160.39, 140.61, 127.66, 127.37, 114.26, 55.47, 52.32, 28.68, 140.61, 131.75, 127.66, 127.37, 114.26, 55.47, 52.32, 28.68, 27.15, 14.85. MS (m/z): [M]+ calcd. for C_{13}H_{15}BrO_{2}S, 266.1; found, 289.1 for [M+Na]+

**Synthesis of compound 23.** Triethylamine (55.7 mg, 0.55 mmol) was added to compound 1b (89 mg, 0.50 mmol) at room temperature. Then, benzylamine (48.2 mg, 0.45 mmol) in 0.5 mL of THF was added to the reaction mixture followed by the addition of 0.5 mL of water. The reaction mixture was extracted with ethyl acetate for 3 times after reaction completed. Organic layers was collected and dried over anhydrous sodium sulfate. Product, compound 23 was obtained after flash chromatography. Yield: 100.6 mg, 84%. 

1H-NMR (400 MHz, MeOH-d₄/D₂O (1:1)): δ (ppm): 7.21-7.37 (m, 5H), 6.20 (s, 1H), 5.86 (d, 1H), 3.71 (s, 6H), 3.55 (s, 2H), 3.25 (s, 4H).

13C-NMR (100 MHz, CDCl₃): δ (ppm) 167.31, 139.08, 137.99, 128.54, 128.37, 126.99, 126.11, 58.34, 54.26, 51.73. MS (m/z): [M]+ calcd. for C_{14}H_{18}BrO_{3}S, 266.1; found, 289.1 for [M+Na]+

**Synthesis of compound 24.** Compound 15 (52.8 mg, 0.20 mmol) was dissolved in 400 uL of MeOH and 200 uL mixture. Benzyl amine (10.7 mg, 0.10 mmol) was added to the mixture. The reaction was kept overnight. MeOH was then removed. The residue was extracted using DCM and water for 3 times. Organic layers were combined and dried over anhydrous sodium sulfate. The solution was dried after filtration and then subjected to flash chromatography to afford compound 24. Yield: 27 mg, 52%. The product contains 87% trans isomer. 

1H-NMR (400 MHz, CDCl₃): δ (ppm) 7.78 (s, 2H), 7.56 (m, 4H), 7.19-7.35 (m, 5H), 6.63 (m, 4H), 3.73 (s, 3H), 3.72 (s, 3H), 3.57 (s, 4H), 3.52 (s, 2H). 

13C-NMR (100 MHz, CDCl₃): δ (ppm) 169.69, 160.38, 143.09, 138.99, 132.90, 130.24, 128.01, 127.63, 127.04, 126.79, 113.76, 59.45, 55.18, 52.00, 50.63. MS (m/z): [M]+ calcd. for C_{31}H_{33}NO_{6}, 515.2; found, 538.2 for [M+Na]+

**Synthesis of compound 25.** MeOH solution (2 mL) of compound 12 (77.2 mg, 0.20 mmol) was added 1mL of sodium bicarbonate saturated solution followed by the addition of benzyl amine (10.7 mg, 0.10 mmol). The reaction was kept overnight. MeOH was then removed. The residue was extracted using DCM and water for 3 times. Organic layers were combined and dried over anhydrous sodium sulfate. The solution was dried after filtration and then subjected to flash chromatography to afford compound 25. Yield: 23 mg, 73%. 

1H-NMR (400 MHz, CDCl₃): 7.28-7.34 (m, 6H), 7.20-7.26 (m, 1H), 6.85 (m, 2H), 6.34 (d, 1H), 6.00 (t, 1H), 4.67 (s, 1H), 3.79 (s, 3H), 3.70 (q, 2H), 3.68 (s, 3H). 

13C-NMR (100 MHz, CDCl₃): 167.09, 158.96, 142.34, 140.47, 133.72, 128.93, 128.52, 128.28, 127.09, 125.22, 113.90 61.66, 55.38, 51.90, 51.87. MS (m/z): [M]+ calcd. for C_{19}H_{21}NO₃, 311.15; found, 334.1 for [M+Na]+
Synthesis of compound 30. Solution of compound 1a (116.1 mg, 1 mmol) in 2 mL of dry DCM was cooled to 0 °C. p-Toluenesulfonyl isocyanate (205 mg, 1.05 mmol) was added to the solution. Completion of reaction was monitored by TLC. The residue was subjected to flash chromatography after removing the solvent to afford compound 30. Yield, 285 mg, 91%. ¹H-NMR (400 MHz, MeOH-d₄): δ(ppm) 7.85 (m, 2H), 7.39 (m, 2H), 6.28 (s, 1H), 5.81 (s, 1H), 4.75 (s, 2H), 3.71 (s, 3H), 2.44 (s, 3H). ¹³C-NMR (100 MHz, CDCl₃): δ(ppm) 165.38, 144.71, 136.50, 135.00, 129.19, 127.67, 127.34, 125.76, 63.62, 51.10, 20.13. MS (m/z): [M]+ calcd. for C₁₃H₁₅NO₆S, 313.06; found, 336.0 for [M+Na]+.

Synthesis of compound 31. Compound 1b (44 mg, 0.25 mmol) was weighed into a glass vial and was added with triethylamine (50 mg, 0.50 mmol). Benzyl alcohol (265 mg, 2.5 mmol) with 100 uL of water was added to the mixture. The reaction was kept for 6 hours. DCM and water were used to extract the reaction mixture. Organic layers were combined and dried over anhydrous sodium sulfate. The solution was dried after filtration and then subjected to flash chromatography to afford compound 31. Yield: 41mg, 80% ¹H-NMR (400 MHz, CDCl₃): δ(ppm) 7.33-7.38 (m, 4H), 7.30 (m, 1H), 6.33 (q, 1H), 5.94 (s, 1H), 4.59 (s, 2H), 4.24 (t, 2H), 3.77(s, 3H). ¹³C-NMR (100 MHz, CDCl₃): δ(ppm) 166.47, 138.16, 137.25, 128.56, 127.84, 127.79, 126.16, 72.87, 68.45, 51.98. MS (m/z): [M]+ calcd. for C₁₂H₁₄O₃, 206.09; found, 229.2 for [M+Na]+.

Synthesis of compound 32. Compound 1b (100 mg, 0.56 mmol) was weighed into a glass vial and was added with triethylamine (62 mg, 0.62 mmol). N-methylbenzylamine (68mg, 0.56 mmol) in 100 uL of THF/H₂O (1:1) mixed solution was added to the vial. The mixture was extracted using DCM and water for 3 times. Organic layers were combined and dried over anhydrous sodium sulfate. The solution was dried after filtration and then subjected to flash chromatography to afford compound 32. Yield: 91 mg, 74%. ¹H-NMR (400 MHz, CDCl₃): δ(ppm) 7.28-7.35(m, 4H), 7.20-7.27(m, 1H), 6.27(s, 1H), 5.85(q, 1H), 3.75 (s, 3H), 3.55 (s, 2H), 3.23 (s, 2H), 2.20 (s, 3H). ¹³C-NMR (100 MHz, CDCl₃): δ(ppm) 167.63, 139.22, 138.06, 128.95, 128.33, 127.10, 126.63, 62.21, 57.71, 51.93, 42.31. MS (m/z): [M]+ calcd. for C₁₃H₁₇NO₂, 219.13; found, 242.2 for [M+Na]+.
Synthesis of compound 33.

\[
\text{Synthesis of compound 33a. To solution of compound 8b (630 mg, 1.96 mmol) in 10 ml dry DCM, DCC(404 mg, 1.96 mmol) was added. PEG amine (500 mg, 1.63 mmol) was added to the solution after 10 minutes. The reaction was stirred overnight at room temperature. Then the white precipitate was filtered, and the clear solution was subjected to flash chromatography to afford compound 33a. Yield, 635 mg, 62\%.
}\]

\[
^{1}H-NMR (400 MHz, CDCl}_3): \delta (ppm) 7.67 (d, 4H), 7.35-7.47 (m, 6H), 7.08 (t, NH), 5.93 (s, 1H), 5.46 (s, 1H), 4.42 (s, 2H), 3.57-3.72 (m, 20H), 3.54 (q, 2H), 3.38 (t, 2H), 1.07 (s, 9H).
\]

\[
^{13}C-NMR (100 MHz, CDCl}_3): \delta (ppm) 167.05, 142.10, 135.68, 132.99, 130.06, 127.94, 120.80, 70.85, 70.82, 70.77, 70.74, 70.72, 70.71, 70.64, 70.42, 70.18, 69.99, 64.30, 50.82, 39.42, 26.94, 19.34. MS (m/z): [M]^{+} \text{calcd. for C}_{32}\text{H}_{48}\text{N}_{4}\text{O}_{7}\text{Si, 628.33; found, 651.3 for [M+Na]^{+}.}
\]

\[
\text{Synthesis of compound 33b To solution of compound 33a (630 mg, 1.0 mmol) in 2 ml dry THF, 1.15 mL of TBAF solution (1M in THF) was added dropwise at 0 \degree C. The reaction mixture was stirred at room temperature until the reaction completed. Reaction mixture was then dried and subjected to flash chromatography to afford compound 33b. Yield: 296 mg, 76\%.}
\]

\[
^{1}H-NMR (400 MHz, CDCl}_3): \delta (ppm) 7.36 (bs, NH), 5.95 (s, 1H), 5.53 (dd, 1H), 4.34 (d, 2H), 3.56-3.73 (m, 20H), 3.53 (s, 2H), 3.38 (t, 2H), 2.62 (bs, OH).
\]

\[
^{13}C-NMR (100 MHz, CDCl}_3): \delta (ppm) 167.51, 142.27, 121.76, 70.80, 70.76, 70.73, 70.70, 70.63, 70.60, 70.53, 70.17, 69.68, 63.90, 50.81, 39.31. MS (m/z): [M]^{+} \text{calcd. for C}_{16}\text{H}_{30}\text{N}_{4}\text{O}_{7}, 390.21; found, 413.10 for [M+Na]^{+}.}
\]

\[
\text{Synthesis of compound 33bexarotene (34.8 mg, 0.1 mmol) was dissolved in 0.5 mL of DCM. To the solution, DCC (20.6 mg, 0.1 mmol) was added followed by the addition of 1 mL of DCM solution of Compound 33b (39 mg, 0.1 mmol) and DMAP (12.2 mg, 0.1 mmol) after 10 minutes. The reaction as stirred overnight. The mixture was dried and subjected to flash chromatography to afford compound 33. Yield: 31 mg, 43\%.}
\]

\[
^{1}H-NMR (400 MHz, CDCl}_3): \delta (ppm) 7.96 (d, J=8.44Hz, 2H), 7.34 (d, J=8.44Hz, 2H), 7.12 (s, 1H), 7.07 (s, 1H), 6.74 (t, NH), 5.98 (s, 1H), 5.80 (d, 1H), 5.69 (s, 1H), 5.33 (d, 1H), 5.09 (s, 2H), 3.57-3.68 (m, 20H), 3.55 (q, 2H), 3.37 (t, 2H), 1.93 (s, 3H), 1.70 (s, 4H), 1.30 (s, 6H), 1.27 (s, 6H).
\]

\[
^{13}C-NMR (100 MHz, CDCl}_3): \delta (ppm) 166.36, 166.02, 149.24, 146.04, 144.55, 142.50, 139.58, 138.07, 132.81, 129.93, 128.71, 128.19, 126.77, 121.83, 117.12, 70.83, 70.80, 70.75, 70.70, 70.68, 70.64, 70.41, 70.17, 69.85, 63.83, 50.81, 39.60, 35.32, 34.14, 34.04, 32.07, 32.02, 20.07. MS (m/z): [M]^{+} \text{calcd. for C}_{40}\text{H}_{56}\text{N}_{8}\text{O}_{8}, 720.41; found, 743.4 for [M+Na]^{+}.}
\]

\[
\text{Synthesis of compound 34 Bexarotene (34.8 mg, 0.1 mmol) was dissolved in 0.5 mL of DCM. To the solution, DCC (20.6 mg, 0.1 mmol) was added followed by the addition of 1 mL of DCM solution of Compound 33b (39 mg, 0.1 mmol) and DMAP (12.2 mg, 0.1 mmol) after 10 minutes. The reaction as stirred overnight. The mixture was dried and subjected to flash chromatography to afford compound 33. Yield: 31 mg, 43\%.}
\]

\[
^{1}H-NMR (400 MHz, MeOH-d4): \delta (ppm) 7.31-7.40 (m, 2H), 7.07-7.30 (m, 8H), 6.78
(d, 2H), 6.60 (d, 2H), 6.07 (s, 1H), 5.49 (s, 1H), 4.00 (t, 2H), 3.33--3.67 (m, 24H), 2.76 (t, 2H), 2.45 (q, 2H), 2.28 (s, 3H), 0.91 (t, 3H). 13C-NMR (100 MHz, MeOH-d4): δ (ppm) 168.05, 156.84, 143.69, 142.37, 141.33, 139.05, 138.46, 135.66, 131.58, 129.49, 129.03, 127.83, 127.58, 126.32, 125.81, 124.28, 113.19, 70.21, 70.20, 70.18, 70.16, 69.94, 69.74, 69.02, 65.34, 60.40, 55.31, 50.37, 40.66, 38.72, 28.45, 12.43.

MS (m/z): [M]+ calcd. for C41H54N4O8, 730.39; found, 753.2 for [M+Na]+.

Synthesis of compound 36a: Solution of compound 8b (400 mg, 1.24 mmol) in 2 mL DCM was added with DCC (256 mg, 1.24 mmol). After 10 minutes, the reaction mixture was added with PEG alcohol (352 mg, 1.0 mmol) and DMAP (152 mg, 1.24 mmol). The reaction was stirred overnight at room temperature. The solution was collected after removal of white precipitate using filtration. Then, the solution was dried and subjected to flash chromatography to afford compound 36a. Yield: 298 mg, 45%.

1H-NMR (400 MHz, CDCl3): δ (ppm) 7.63--7.71 (d, 4H), 7.33--7.47 (m, 6H), 6.35 (q, 1H), 6.12 (q, 1H), 4.42 (t, 2H), 4.26 (t, 2H), 3.69 (t, 2H), 3.58--3.67 (m, 22H), 3.54--3.67 (m, 2H), 3.38 (s, 3H), 1.08 (s, 9H).

13C-NMR (100 MHz, CDCl3): δ (ppm) 165.81, 139.38, 135.58, 129.92, 127.90, 124.42, 72.08, 70.82, 70.78, 70.72, 70.67, 69.16, 63.86, 62.32, 59.18, 26.95, 19.43. MS (m/z): [M]+ calcd. for C35H54O10Si, 662.35; found, 685.3 for [M+Na]+.

Synthesis of compound 36b: Solution of compound 36a (200 mg, 0.31 mmol) in 1 mL of dry THF was added with 340 uL of TBAF solution (1 M in THF) at 0 °C. After reaction completion, the solution was dried and subjected to flash chromatography to afford compound 36b. Yield: 125 mg, 94%. 1H-NMR (400 MHz, CDCl3): δ (ppm) 7.63--7.71 (d, 4H), 7.33--7.47 (m, 6H), 6.35 (q, 1H), 6.12 (q, 1H), 4.42 (t, 2H), 4.26 (t, 2H), 3.69 (t, 2H), 3.58--3.67 (m, 22H), 3.54 (m, 2H), 3.38 (s, 3H), 1.08 (s, 9H). 13C-NMR (100 MHz, CDCl3): δ (ppm) 165.81, 139.38, 135.58, 129.92, 127.90, 124.42, 72.08, 70.78, 70.72, 70.67, 69.16, 63.86, 62.32, 59.18, 26.95, 19.43. MS (m/z): [M]+ calcd. for C35H54O10Si, 662.35; found, 685.3 for [M+Na]+.

Synthesis of compound 36c: Compound 36b (120 mg, 0.28 mmol) was dissolved in 1 mL of dry DCM and cooled to 0 °C. PBr3 (38 mg, 0.14 mmol) was slowly added to the solution. The reaction completed in 30 minutes. The reaction was quenched by adding saturated sodium bicarbonate solution. The reaction mixture was further extracted using DCM and water for 3 times. Organic layers were combined and dried over anhydrous sodium sulfate. The solution was dried after filtration to afford compound 36c which was directly used for next reaction.

Synthesis of compound 36: Compound 36c (24 mg, 0.2 mmol) was weighed to a glass vial. To the vial triethylamine (81 mg, 0.8 mmol) was added. After 4 hours, 3 mL of diethyl ether was added and sonicated for 1 minutes. Then, supernatant was removed. The operation was repeated for 4 times and the viscous solid
was collected and dried to afford compound 36. Yield: 94 mg, 68%. 

\( ^1 \)H-NMR (400 MHz, MeOH-d4): \( \delta \text{ (ppm)} \) 6.93(s, 1H), 6.40 (s, 1H), 4.40 (m, 2H), 4.24 (s, 2H), 3.79(m, 2H), 3.60-3.69 (m, 22H), 3.54 (m, 2H), 3.36 (s, 3H), 3.28-3.35 (q, 6H), 1.37(t, 9H). 

\( ^{13} \)C-NMR (100 MHz, MeOH-d4): \( \delta \text{ (ppm)} \) 166.86, 140.63, 130.82, 72.98, 71.57, 71.55,71.36, 69.85, 66.18, 59.09, 57.01, 54.35, 8.12. 

MS (m/z): [M] \(^+\) calcd. for C\(_{25}\)H\(_{50}\)BrNO\(_9\), 587.27; found, 508.3 for [M-Br] \(^+\).

**Synthesis of compound 37**

**Synthesis of compound 37a** DABCO (4.9 g, 44 mmol) were dispersed in 10 mL water followed by the addition of paraformaldehyde (1.98 g, 52 mmol) at 0 °C. Acetonitrile (17.5 mL) and 1,6-hexanediol diacrylate (5 g, 22 mmol) was sequentially added to the reaction after 15 minutes. Then the reaction mixture was heated at 45 °C for 3 hours. The reaction mixture was then cooled and volatiles were removed. The crude was extracted using ethyl acetate and water for 3 times. Organic layers were combined and dried over anhydrous sodium sulfate. The solution was dried after filtration and then subjected to flash chromatography to afford compound 37a. Yield: 3.2 g, 51%.

\( ^1 \)H-NMR (400 MHz, CDCl\(_3\)): \( \delta \text{ (ppm)} \) 6.23(s, 2H), 5.82(s, 2H), 4.31(s, 4H), 4.17(t, 4H), 2.51(bs, 2H), 1.69 (m, 4H), 1.42(m, 4H).

\( ^{13} \)C-NMR (100 MHz, CDCl\(_3\)): 166.38, 139.49, 125.72, 64.75, 62.62, 28.44, 25.61.

MS (m/z): [M] \(^+\) calcd. for C\(_{14}\)H\(_{22}\)O\(_6\), 286.14; found, 309.1 for [M+Na] \(^+\).

**Synthesis of compound 37b** To solution of compound 37a (500 mg, 1.75 mmol) in 10 mL drug DCM, PBr\(_3\) (473 mg, 165 µL) was added at 0 °C. The reaction was stirred at room temperature and completed in 30 minutes. The reaction was quenched by addition of 2 mL of saturated sodium bicarbonate solution. The crude was extracted using ethyl acetate and water for 3 times. Organic layers were combined and dried over anhydrous sodium sulfate. The solution was dried after filtration and further subjected chromatography to afford compound 37b. Yield: 360 mg, 50%. 

\( ^1 \)H-NMR (400 MHz, CDCl\(_3\)): \( \delta \text{ (ppm)} \) 6.33(s, 2H), 5.94(s, 2H), 4.22(t, 4H),  4.18(s, 4H), 1.73 (m, 4H), 1.46 (m, 4H).

\( ^{13} \)C-NMR (100 MHz, CDCl\(_3\)): 164.90,137.55, 129.05, 65.20, 28.46, 28.4639, 25.6. MS (m/z): [M] \(^+\) calcd. for C\(_{14}\)H\(_{20}\)Br\(_2\)O\(_4\), 409.97; found, 433.0 for [M+Na] \(^+\).

**Synthesis of compound 37** Compound 37b (100 mg, 0.244 mmol) was dissolved in 3 mL of dry DCM. Then triethylamine (247 mg, 345 µL) was added to the solution. The reaction was stirred for 24 hours. Then volatiles were removed, and the residue was added with 5 mL of diethyl ether to precipitate the product. Then the precipitate was re-dissolved in 100 µL of MeOH and precipitated in 5 mL of diethyl ether. The procedure was repeated for two more times. The precipitate was collected and dried to afford compound 37. Yield: 98 mg, 64%. 

\( ^1 \)H-NMR (400 MHz, MeOH-d4): \( \delta \text{ (ppm)} \) 6.89 (s, 2H), 6.40 (s, 2H), 4.27 (t, 4H), 4.23 (s, 4H), 3.25-3.36 (q, 12H), 1.76 (m, 4H), 1.48 (m, 4H), 1.31-1.42 (t, 18H). 

\( ^{13} \)C-NMR (100 MHz, MeOH-d4): \( \delta \text{ (ppm)} \) 165.94, 138.90, 129.46, 65.73, 53.01, 28.13, 28.10, 25.25, 7.98.

**Synthesis of compound 38** Compound 38 was synthesized following the reported procedures.\(^4\)

**Experimental Section**

**Kinetics study for trigger-to-release process**

10 µmol (2 eq.) of A1 molecules was dissolved in single or mixed deuterated solvents (the volume of solvent depends on the solubility of A1). Then, 5 µmol (1 eq.) of 2-(2-Methoxyethoxy) ethanethiol or
benzylamine was added to the solution. The reaction was immediately followed by NMR. The conversion of thiol or amine was calculated from the integration of corresponding peaks by NMR and plotted against reaction time.

**Thiol-addition condition:**

- **Compound 1, 2, 3, 4, 5, 8, 11, 12, 13:** 10 µmol (2 eq.) of molecule 1, 2, 3, 4, 5, 8, 11, 12, 13 was dissolved in 1000 µL of MeOH-d4. Then, 5 µmol (1 eq.) of thiol was added to the solution. The reaction was immediately followed by NMR.

- **Compound 6, 7, 9:** 10 µmol (2 eq.) of molecule 6, 7, 9 was dissolved in 500 µL of MeOH-d4 and 500 µL of 50 mM pH 7.4 phosphate buffer. Then, 5 µmol (1 eq.) of thiol or amine was added to the solution. The reaction was immediately followed by NMR.

- **Compound 10:** 10 µmol (2 eq.) of molecule 10 was dissolved in 1000 µL of DMSO-d6 and 500 µL of 50 mM pH 7.4 phosphate buffer. Then, 5 µmol (1 eq.) of thiol or amine was added to the solution. The reaction was immediately followed by NMR.

- **Compound 14, 15, 16:** 10 µmol (2 eq.) of molecule 14, 15, 16 was dissolved in 500 µL of MeOH-d4 and 500 µL of 50 mM pH 7.4 phosphate buffer. Then, 5 µmol (1 eq.) of thiol or amine was added to the solution. The reaction was immediately followed by NMR.

**Amine-addition condition:**

- **Compound 1, 2, 3, 4, 5, 6, 7, 8, 9, 11, 12, 13:** 10 µmol (2 eq.) of molecule 1, 2, 3, 4, 5, 6, 7, 8, 9, 11, 12, 13 was dissolved in 500 µL of MeOH-d4 and 500 µL of 50 mM pH 7.4 phosphate buffer. Then, 5 µmol (1 eq.) of benzylamine was added to the solution. The reaction was immediately followed by NMR.

- **Compound 10:** 10 µmol (2 eq.) of molecule 10 was dissolved in 1000 µL of DMSO-d6 and 500 µL of 50 mM pH 7.4 phosphate buffer. Then, 5 µmol (1 eq.) of benzylamine was added to the solution. The reaction was immediately followed by NMR.

- **Compound 14, 15, 16:** 10 µmol (2 eq.) of molecule 14, 15, 16 was dissolved in 800 µL of MeOH-d4 and 200 µL of 50 mM pH 7.4 phosphate buffer. Then, 5 µmol (1 eq.) of benzylamine was added to the solution. The reaction was immediately followed by NMR.

**Trigger-to-reverse reaction**

Typically, product of thiol-A1 or amine-A1 addition product was dissolved in single or mixed deuterated solvents (the volume of solvent depends on the solubility of product). Then, thiol was added to the solution. The reaction was immediately followed by NMR. Detailed information is shown below:

- **Compound 17:** 10 µmol (1 eq.) of 17 was dissolved in 500 µL of MeOH-d4 and 500 µL of 50 mM pH 7.4 phosphate buffer. Then, 80 µmol (8 eq.) of thiol was added to the solution. Final concentration of 17 is 10 mM.

- **Compound 18:** 10 µmol (1 eq.) of 18 was dissolved in 500 µL of MeOH-d4 and 500 µL of 50 mM pH 7.4 phosphate buffer. Then, 5 µmol (0.5 eq.) of thiol was added to the solution. Final concentration of 18 is 10 mM.

- **Compound 19:** 10 µmol (1 eq.) of 19 was dissolved in 500 µL of MeOH-d4 and 500 µL of 50 mM pH 7.4 phosphate buffer. Then, 5 µmol (0.5 eq.) of thiol was added to the solution. Final concentration of 19 is 10 mM.
Compound 20: 10 µmol (1 eq.) of 20 was dissolved in 1000 µL of MeOH-d4 and 500 µL of 50 mM pH7.4 phosphate buffer. Then, 5 µmol (0.5 eq.) of thiol was added to the solution. Final concentration of 20 is 6.67 mM.

Compound 21: 10 µmol (1 eq.) of 21 was dissolved in 500 µL of MeOH-d4 and 500 µL of 50 mM pH7.4 phosphate buffer. Then, 5 µmol (0.5 eq.) of thiol was added to the solution. Final concentration of 21 is 10 mM.

Compound 22: 10 µmol (1 eq.) of 22 was dissolved in 500 µL of MeOH-d4 and 500 µL of 50 mM pH7.4 phosphate buffer. Then, 5 µmol (0.5 eq.) of thiol was added to the solution. Final concentration of 22 is 10 mM.

Compound 23: 10 µmol (1 eq.) of 23 was dissolved in 1000 µL of MeOH-d4 and 500 µL of 50 mM pH7.4 phosphate buffer. Then, 20 µmol (2 eq.) of thiol was added to the solution. Final concentration of 23 is 6.67 mM.

Compound 24: 10 µmol (1 eq.) of 24 was dissolved in 2000 µL of MeOH-d4. Then, 5 µmol (0.5 eq.) of thiol or amine was added to the solution. Final concentration of 24 is 5 mM.

Compound 25: 10 µmol (1 eq.) of 25 was dissolved in 1000 µL of MeOH-d4 and 500 µL of 50 mM pH7.4 phosphate buffer. Then, 5 µmol (0.5 eq.) of thiol was added to the solution. Final concentration of 25 is 6.67 mM.

**Thiol triggered functionality recovery**

*Functionality recovery from small molecules: 1, 4, 6, 7, 17, 30, 31, 32*

Triethylamine recovery: 10 µmol (1 eq.) of 1 was dissolved in 1000 µL of MeOH-d4. Then, 10 µmol (10 eq.) of thiol was added to the solution. The release of triethylamine was followed by NMR.

Pyridine recovery: 10 µmol (1 eq.) of 4 was dissolved in 500 µL of MeOH-d4 and 500 µL of 50 mM pH7.4 phosphate buffer. Then, 10 µmol (1 eq.) of thiol was added to the solution. The release of pyridine was followed by NMR.

Acetic acid recovery: 10 µmol (1 eq.) of 6 was dissolved in 500 µL of MeOH-d4 and 500 µL of 50 mM pH7.4 phosphate buffer. Then, 10 µmol (1 eq.) of thiol was added to the solution. The release of acetic acid was followed by NMR.

4-nitroaniline recovery: 10 µmol (1 eq.) of 7 was dissolved in 500 µL of MeOH-d4 and 500 µL of 50 mM pH7.4 phosphate buffer. Then, 10 µmol (1 eq.) of thiol was added to the solution. The release of 4-nitroaniline was followed by NMR.

2-(2-Methoxyethoxy) ethanethiol recovery: 10 µmol (1 eq.) of 17 was dissolved in 500 µL of MeOH-d4 and 500 µL of 50 mM pH6.8 phosphate buffer. Then, 10 µmol (8 eq.) of thiol was added to the solution. The release of 2-(2-Methoxyethoxy) ethanethiol was followed by NMR.

Sulfonamide recovery: 10 µmol (1 eq.) of 30 was dissolved in 500 µL of MeOH-d4 and 500 µL of 50 mM pH7.4 phosphate buffer. Then, 10 µmol (1 eq.) of thiol was added to the solution. The release of sulfonamide was followed by NMR.

Benzyl alcohol recovery: 10 µmol (1 eq.) of 31 was dissolved in 500 µL of MeOH-d4 and 500 µL of 50 mM pH7.4 phosphate buffer. Then, 10 µmol (1 eq.) of thiol was added to the solution. The release of benzyl alcohol was followed by NMR.
N-methylbenzylamine recovery: 10 µmol (1 eq.) of 32 was dissolved in 500 µL of MeOH-d4 and 500 µL of 50 mM pH7.4 phosphate buffer. Then, 10 µmol (1 eq.) of thiol was added to the solution. The release of N-methylbenzylamine was followed by NMR.

*Functionality recovery from small molecules 33 & 34:*

**Compound 33:**

Procedure A: 5 µmol (1 eq.) of 33 was dissolved in 500 µL of MeOH-d4 followed by addition of 10 mg of Na₂CO₃. 10 µmol (2 eq.) of 2-(2-Methoxyethoxy) ethanethiol was added to solution. The release of Bexarotene was followed by NMR.

Procedure B: To 250 µL of stock solution of compound 33 (1 mM solution in MeOH), 250 µL of 10 mM Glutathione (GSH) solution in 50 mM pH7.4 sodium phosphate buffer was added. The reaction solution was subjected to mass spectral analyses after 6 hours.

**Compound 34:** 5 µmol (1 eq.) of 34 was dissolved in 500 µL of MeOH-d4 and 500 µL of 50 mM pH7.4 phosphate buffer. 10 µmol (2 eq.) of 2-(2-Methoxyethoxy) ethanethiol was added to solution. The release of Desmethyltamoxifen was followed by NMR.

*Aptamer drug conjugation and thiol triggered release of Desmethyltamoxifen*

Stock solution of 34 was first prepared in DMSO. 100 µM DNA aptamer (AS1411: 5’-GGT GGT GGT GGT TGT GGT GGT GGT/3’DBCO/3’, ordered from Integrated DNA Technologies) solution was incubated with 20 equivalents of 34 in a solvent mixture of H₂O and DMSO (V/V=80:20) for 1 h at room temperature. After reaction, the aptamer was purified using GE PD SpinTrap G-25 column (GE Healthcare Bio-Sciences, Pittsburgh, PA) to remove the excess of 34. The collected aptamer was characterized by ESI-MS before further experiments. Drug release was performed by incubating 50 µM conjugated aptamer with 10 mM of 2-(2-Methoxyethoxy) ethanethiol at room temperature for 12 h. After reaction, the mixture was injected in a LC-MS system to separate and detect the released drug molecules.

LC-MS detection of the released small-molecule drug from the aptamer drug conjugates was acquired on a Bruker AmaZon (Billerica, MA) quadrupole ion trap mass spectrometer equipped with an electrospray ionization source. The electrospray needle voltage was kept at 4 kV, and the capillary temperature was set to 250 °C.

**PEG drug conjugation and thiol triggered release of Bexarotene**
Conjugation: mPEG(5000)-DBCO (50mg, 0.01mmol) was dissolved in 1 mL DCM. To the solution, compound 33 (15mg, 0.02 mmol) in 500 uL of DCM was added. The reaction was stirred at room temperature for 2 hours. The reaction mixture was then concentrated and precipitated in diethyl ether for 4 times. The precipitate was collected and dried to afford compound 35. Yield: 51mg, 89%. The successful conjugation was monitored by NMR.

Bexarotene release: PEG-33 conjugate (5.74 mg, 1 µmol) was dissolved in 500 µL of MeOH-d4 and 500 µL of 50 mM pH7.4 phosphate buffer. Then 4 µmol (2 eq.) of 2-(2-Methoxyethoxy) ethanethiol was added to solution. The release of Bexarotene was followed by NMR.

**Protein modification and analysis**

**Protein modification procedures:**

For βLGb labeling reaction, βLGb was first denatured by 8 M of urea to expose the free cysteine. The denatured protein was then diluted with 50 mM pH 8.0 phosphate buffer, which results in a protein solution concentration of 100 µM. This protein solution was incubated with 20 equivalents of 1 for 5 min or with 20 equivalents of 8 for 40 min at room temperature to obtain the fully modified proteins respectively. After labeling, the reaction mixtures were injected immediately into an HPLC to remove excess labeling reagents and phosphate salts and then later analyzed by ESI-MS.

For BCA labeling reaction, BCA was dissolved in 50 mM pH 8.0 phosphate buffer to acquire a 100 µM protein solution. This protein solution was incubated with 20 equivalents of 36 for 1h at room temperature to obtain the modified proteins (>95% of the protein was modified). After labeling, the reaction mixtures were injected immediately into LC-MS for intact protein analyses.

For Myoglobin (Myo) reversible modification reactions, the modified protein solution was prepared by reacting 100 µM of the Myo with 20 equivalents of 1 for 5 min in 50 mM pH 8.0 phosphate buffer at room temperature. The proteins were collected from HPLC purification from previous modification reactions, and the excess labeling reagents were removed from the reaction mixtures. The collected protein solution was characterized by ESI-MS or other biophysical characterizations such as Circular Dichroism (CD) and Ultraviolet-Visible Spectroscopy (UV-Vis). The same protein solution was also buffer-exchanged using 10K NMWL Amicon Ultra centrifugal filters (Millipore, Burlington, MA) with 50 mM pH 7.4 phosphate buffer to yield a 100 µM solution for further reverse reaction. The proteins from previous step were incubated with 10 equivalents of 2-(2-Methoxyethoxy) ethanethiol for 2.5 h at room temperature. After reverse modification reaction, the reaction mixture was injected to a LC-MS system for intact protein analyses.

For kinetic experiments in Figure 4G & 4H, 20 equivalents of 7 were used to react with 100 µM Myo for different time periods. After labeling, the reaction mixtures were injected immediately into an LC-MS for intact protein analyses. The corresponding absorbance spectra of the reaction mixtures were also recorded on a Thermo Scientific NanoDrop 2000c Spectrophotometers (Thermo Scientific, Tewksbury, MA).
**Proteolytic digestion:**

The labeled βLGb protein samples were first buffer-exchanged using 10K NMWL Amicon Ultra centrifugal filters (Millipore, Burlington, MA) with 100 mM triethylamine acetate (pH 8.0), and reconstituted with 1 M urea before enzymatic digestion. To reduce the disulfide bonds in βLGb, TCEP in water was added at a protein:TCEP molar ratio of 1:20, and the sample was incubated at room temperature for 10 min. To alkylate the reduced cysteines, iodoacetamide in water was added at a protein:iodoacetamide molar ratio of 1:80, and the sample was incubated in the dark at room temperature for 30 min. The denatured, reduced, and alkylated protein samples were then digested with trypsin at an enzyme: substrate ratio of 1:10. After 4 h of digestion at 37 °C, the enzyme was separated from the mixture by centrifugation using a 10K NMWL Microcon filter (Millipore, Burlington, MA). The filtrate was then analyzed by LC-MS and LC-MS/MS.

**HPLC separation:**

To quench the labeling reaction and to remove excess labeling reagents and buffer salts, a Thermo Scientific Ultimate 3000 HPLC system (Thermo Scientific, Tewksbury, MA) with an OPTI-TRAP C4 reverse phase column (1 × 8 mm) was used. The protein was eluted using an acetonitrile gradient that increases from 1 to 99% over 12 min at a flow rate of 0.2 mL/min. The labeled protein was collected for proteolytic digestion or intact protein MS characterization.

To analyze the Myo digests from the labeling experiments, a Thermo Scientific EASY-nLC 1000 liquid chromatography system (Thermo Scientific, Tewksbury, MA) with an Acclaim PepMap RSLC C18 reverse phase column (75 µm × 15 cm, 2 µm particle size) from Thermo Scientific (Tewksbury, MA) was used. To achieve efficient separation of the proteolytic peptides, a shallow gradient was used where %B (0.1% formic acid in acetonitrile) was increased from 0% to 40% over 45 min. The column was then flushed by increasing to 95% B over 15 min. The column was then cleaned at 95% B for another 20 min. A flow rate of 300 nL/min was used throughout the run.

**Mass spectrometry:**

Mass spectral analyses of the HPLC separated intact protein samples (Myo and βLGb) from the covalent labeling experiments were acquired on a Bruker AmaZon (Billerica, MA) quadrupole ion trap mass spectrometer equipped with an electrospray ionization source. The electrospray needle voltage was kept at 4 kV, and the capillary temperature was set to 250 °C. Mass spectra of intact BCA protein samples were acquired on a Thermo Orbitrap Fusion Tribrid (Tewksbury, MA) mass spectrometer. The electrospray ionization source was typically operated at a needle voltage of 3800 V, and the ion transfer tube temperature was set to 325 °C.

LC-MS and LC-MS/MS analyses of protein proteolytic fragments were conducted on a Thermo Orbitrap Fusion Tribrid (Tewksbury, MA) mass spectrometer. The electrospray ionization source was typically operated at a needle voltage of 2100 V, and the ion transfer tube temperature was set to 300 °C. Tandem mass spectra were collected using CID with a normalized collision energy of 35%. Due to the large number of detectable peaks, an exclusion limit of 60 s was applied after five spectra had been collected for any given peak. The resolution of the Orbitrap was set to 60000.

**Peptide and modification identification:**

Raw mass spectral data files were analyzed by Thermo Proteome Discoverer 2.2 software. Spectra were searched against the corresponding protein sequence. Variable modification by certain labeling reagents of the residues and the protein N-terminus was added as a dynamic modification. Other dynamic modifications such as oxidation of methionine and carboxyamidomethylation of cysteine were also used in the searches. Trypsin enzyme cleavage was selected, and a precursor mass tolerance of 10 ppm was used.
Identifications of peptides and modifications at high confidence levels were used and were manually checked in all cases.

Circular dichroism:

Far-UV CD analyses were performed on a Jasco J-1500 spectropolarimeter. CD spectra were recorded at room temperature over a scan range of 260 to 200 nm. Protein samples were diluted to 0.1 mg/mL in 50 mM pH 7.4 phosphate buffer prior to analysis. The CD spectrometric parameters were set as follows: a scan resolution (data pitch) of 0.5 nm, a scan rate of 20 nm/min, a band width of 2 nm, and a digital integration time of 1 sec. Triplicate measurements were performed for each sample at room temperature.

Temperature dependent CD measurements were also performed for the protein samples to evaluate the thermal stability (melting temperature) of Myo before and after labeling. Ellipticity at 222 nm was recorded every 1 °C from 25 °C to 90 °C. Prior to individual scans, samples were equilibrated at the new temperature for 1 min. Samples were measured in triplicates and the results were shown in average values.

Orthogonal hydrogel manipulation

Hydrogel formation: Compound 37 (24.48 mg, 40 µmol) was dissolved in 2 mL 50 mM sodium phosphate buffer to make stock solution with desired pH. 4 arm PEG10000-thiol (100 mg, 20 µmol) was dissolved in 2 mL 50 mM sodium phosphate buffer with desired pH to make stock solution. Then, 0.75 mL of compound 37 solution was mixed with 0.75 mL of PEG solution.

Hydrogel post-functionalization using small molecule model: Compound 38 (0.4 mg, 2.5 µmol) and compound 18 (0.665 mg, 2.5 µmol) was respectively dissolved in 2 mL of MeOH to prepare the stock solutions. Then, 1 mL of compound 38 and compound 18 solutions was mixed to form reaction solution. 50 µL of the mixed solution was then diluted with 950 µL of MeOH to form final reaction solution, the absorption and emission spectra was measured before and after UV irradiation at λ300nm for 30 seconds.

Hydrogel photo-patterning: Solution of compound 37 (24.48 mg, 40 µmol) in 2 mL 50 mM pH6.8 sodium phosphate buffer and solution of 4 arm PEG10000-thiol (100 mg, 20 µmol) in 2 mL 50 mM pH6.8 sodium phosphate buffer was prepared. 0.75 mL of each solution was mixed in a petri dish and allowed to cure for 30 minutes. The petri dish was placed on the top of a hand-held UV lamp with a patterned cover in between. Then 1 mL solution of compound 38 with concentration of 1.25 mM in MeOH was added to the middle of the dish followed by the irradiation at λ300nm on the UV lamp. After irradiation, the hydrogel was washed with 2 mL of MeOH for 3 times to remove unreacted compound 38. A video of the photo-patterning was shown separately.

BSA loading hydrogel formation and thiol triggered BSA release. To a glass vial, 50 µL of 1.0 mg/mL FITC labelled BSA was diluted with 150 µL of 50 mM pH 7.4 sodium phosphate buffer followed by addition of
100 µL of 4-arm PEG10000 thiol (100 mg/mL). Then, 100 µL of compound 37 solution (12.24 mg/mL) in 50 mM pH 7.4 sodium phosphate buffer was added to the vial to form BSA loaded hydrogel in seconds. Two identical hydrogels were prepared using the same procedure at the same time. Then, one of hydrogel was added with 3 mL of 50 mM pH 7.4 sodium phosphate buffer, while the other one was added with 3 mL of 50 mM 2-(2-Methoxyethoxy) ethanethiol solution in 50 mM pH 7.4 sodium phosphate buffer. To monitor the release of BSA, 50 µL of liquid from each hydrogel was sampled and diluted to 1mL and subjected for fluorescence measurement at different interval. Here, we assumed that BSA would diffuse to liquid phase once it is released due to dissolution of hydrogel, while the BSA in intact hydrogel should be entrapped in the solid gel phase.

**Hydrogel sample preparation of rheological measurement:** Solution of compound 37 (24.48 mg, 40 µmol) in 2 mL 50 mM pH6.8 sodium phosphate buffer and solution of 4 arm PEG10000-thiol (100 mg, 20 µmol) in 2 mL 50 mM pH6.8 sodium phosphate buffer was prepared respectively. 0.75 mL of each solution was mixed in cylinder mode with diameter of 2.5 centimeters. Then hydrogel sample was placed in parallel plate and cut to fit the dimension of the plate.

**Hydrogel dissolution for rheological measurement:** 10 µL of m-PEG6-thiol (Mw 312.4) was placed and spread in parallel plate. Then hydrogel sample was placed on the top of plate and the measurement was started immediately.

**Rheometry:** Measurements were performed by using the Malvern Kinexus Pro stress-controlled instrument in a small angle oscillation shear-controlled manner. A stainless parallel plate (20 mm diameter) fixture with a solvent trap was used for all experiments. Sample height was fixed at 1 mm. The shear strain amplitude and angular frequency were 1% and 10 rad/s, respectively. The storage (G’) and loss moduli (G’’) were measured as a function of time. The transient complex viscosity profile was calculated via the IRIS software package. All experiments were conducted at 25 °C.

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Figure S1 Reaction of DEGSH with 1 followed by NMR. Bottom: compound 1; top: compound 1 incubated with 0.5 equivalent of thiol for 3 minutes. The reaction was carried out at 50 mM pH 7.4 phosphate buffer.

Figure S2 NMR spectrum of isolated product from reaction of DEGSH and compound 1 in MeOH-d4.
Figure S3 Reaction of benzyl amine with 1 followed by NMR at different timepoints. Compound 1 was incubated 0.5 equivalent of benzyl amine and monitored. The reaction was carried at MeOH-d4 and 50 mM pH 7.4 phosphate buffer mixture (1:1). The spectra were aligned with MeOH-d4 peak. Peak at 4.79 ppm is attributed to H2O.

Figure S4 NMR spectrum of isolated product from reaction of benzyl amine and compound 1 in MeOH-d4. "*" indicates solvent peaks.
Figure S5 Structure-property relationship investigation on trigger-to-release kinetics for thiol and amine. (A) Structure variations of A1. (B) Influence of R¹ substitution on thiol-based trigger-to-release kinetics. Reaction carried out in MeOH-d4 (data points for compound 4 at longer time scales are shown in Fig S5H). (C) Influence of X and R² substitution on thiol-based trigger-to-release kinetics. Reaction for 1 & 8 was carried out in MeOH-d4, while reaction for 6, 9, 7, & 10 performed in MeOH-d4 and pH7.4 phosphate buffer mixture (1:1). (D) Influence of R³ and R⁴ substitution on thiol-based trigger-to-release kinetics. Reaction for 11, 12, 13 was carried out in MeOH-d4, while reaction for 14, 15, & 16 performed in MeOH-d4 and pH 6.2 phosphate buffer mixture (1:1). (E) Influence of R¹ substitution amine-based trigger-to-release kinetics. Reaction carried out in MeOH-d4 and pH7.4 phosphate buffer mixture (1:1). (F) Influence of X and R² substitution on amine-based trigger-to-release kinetics. Reaction carried out in MeOH-d4 and pH7.4 phosphate buffer mixture (1:1). (G) Influence of R³ and R⁴ substitution on amine-based trigger-to-release kinetics. Reaction carried out in MeOH-d4 and pH 7.4 phosphate buffer. Solvent ratio for 11, 12, &13 was 1:1 while that for 14, 15, &16 was 1:4. (H) Influence of R¹ substitution on thiol-based trigger-to-release kinetics for longer time scale. (I) Influence of X and R² substitution on amine-based trigger-to-release kinetics for longer time scale. Note: The original NMR spectra for the kinetics measurement are shown at the end of supplementary information from Figure S52-S103.
Figure S6. Reaction of thiol with 4 enhanced by the addition of DMAP. The reaction was followed by NMR. NMR spectra can be found in Figure S59 & S60.
Figure S7 Kinetics of trigger-to-release reaction of 1, 4, 8 with thiol in MeOH-d4 and phosphate buffer.

Figure S8 Isolated product from amine-addition of 12 with benzyl amine.
Figure S9 Reversion of thiol-addition product, 17 to T2 triggered by T1 followed by NMR. [T1]/[17]=8/1. The reaction was carried out in mixture of MeOH-d4 and 50 mM pH 6.8 phosphate buffer (1:1).
Figure S10 Zoom-in spectra of Figure S9. The integration of the peaks assigned was used to calculate the population of each species.
Thiol-triggered reversion of 17. [T1] / [17] = 3/1. The reaction was carried out in mixture of MeOH-d4 and 50 mM pH 6.8 phosphate buffer (1:1). [17] : 10 mM.

Figure S11 Thiol-triggered reversion of 17. [T1] / [17] = 3/1. The reaction was carried out in mixture of MeOH-d4 and 50 mM pH 6.8 phosphate buffer (1:1). [17] : 10 mM.
Figure S12 Thiol triggered reversion of amine-addition product, 23. The progress of the reaction was followed by NMR. After 120 minutes, benzyl amine (BA) was added to the reaction mixture to identify that the released compound is actually benzyl amine. The reaction was carried out in mixture of MeOH-d4 and 50 mM pH 7.4 phosphate buffer (2:1). The ratio between 23 and DEGSH was 1:2. Concentration of 23 was 6.67mM.
Figure S13 Reversion of thiol-addition product, 18 is triggerable by thiol suggested by NMR. The reaction was carried out at MeOH-d4 and 50 mM pH 7.4 phosphate buffer (1:1). [18]/[DEGSH]=2. [18]: 10 mM. (Reaction at 3 min & 60 min).

Figure S14 Reversion of thiol-addition product, 18 is triggerable by thiol suggested by NMR. Zoom-in spectra of Figure S13. (Reaction at 3 min & 60 min).
Figure S15 Reversion of thiol-addition product, 19, is triggerable by thiol suggested by NMR. The reaction was carried out at MeOH-d4 and 50 mM pH 7.4 phosphate buffer (1:1). [19]/[ethanethiol]=2. [19]: 10 mM. (Reaction at 15 min & 24 h).

Figure S16 Reversion of thiol-addition product, 20, is triggerable by thiol suggested by NMR. The reaction was carried out at MeOH-d4 and 50 mM pH 7.4 phosphate buffer (2:1). [20]/[DEGSH]=2. [20]:6.67 mM. (Reaction at 3 min & 60 min).
Figure S17 Thiol-addition product, 21 is irreversible in the presence of thiol suggested by NMR. The reaction was carried out at MeOH-d4 and 50 mM pH 7.4 phosphate buffer (2:1). [21]/[DEGSH]=2. [21]:10 mM. (*) indicates the isomer of 21. 

Figure S18 Thiol-addition product, 22 is irreversible in the presence of thiol suggested by NMR. The reaction was carried out at MeOH-d4 and 50 mM pH 7.4 phosphate buffer (2:1). [22]/[DEGSH]=2. [22]:6.67 mM. (*) indicates the isomer of 22.)
Figure S19 Amine-addition product, 24 is irreversible in the presence of thiol suggested by NMR. The reaction was carried out at MeOH-d4. [22]/[DEGSH]=2. [22]: 5 mM.

Figure S20 Reversion of amine-addition product, 25 is triggerable by thiol suggested by NMR. The reaction was carried out at MeOH-d4 and 50 mM pH 7.4 phosphate buffer (2:1). [25]/[DEGSH]=2. [22]: 6.67 mM.
Figure S21 Solvent dependent reactivity of compound 1 toward thiol and amine. Aqueous reaction media drastically accelerates the reaction and improves the selectivity among thiol and amine. $[1]/[\text{Nucleophile}]=2:1$.

Figure S22 Mass spectra of compound 1 & 8 modified βLGb. (A) Full mass spectra. Top: βLGb modified with 8. Middle: βLGb modified with 1 Bottom: unmodified βLGb. (B) Corresponding mass spectra of highlighted charge state (+17) in (A). 1 La stands for 1 modification.
Sequence of βLGb:

1-10  11-20  21-30  31-40  41-50
LIVTQTMKGL  DIQKVAGTWY  SLAMAASDIS  LLDAQSAPLR  VYVEELKPTP
51-60  61-70  71-80  81-90  91-100
EGDLEILLQK  WENGECAQKK  IIAEKTKIPA  VFKIDALNEN  KVLVLDTDYK
101-110  111-120  121-130  131-140  141-150
KYLFCMENS  AEPEQSLACQ  CLVRTPEADD  EALEKFDKAL  KALPMHIRLS
151-160  161-162
FNPTQLEEQC  HI

**Figure S23** MS/MS of digested βLGb modified with compound 8 shows selective modification on free cysteine, C121 (green coded) while no modifications on cysteines involved in disulfide bond (yellow coded).
Figure S24 MS/MS of digested βLGb modified with compound 1 shows modification on free cysteine, C121 (green coded) while no modifications on cysteines involved in disulfide bond (yellow coded).

Figure S25 MS/MS of digested βLGb modified with compound 1 shows unselective modification of lysine K91.
Figure S26 MS/MS of digested βLGb modified with compound 1 shows unselective modification of lysine K70

Figure S27 MS/MS of digested βLGb modified with compound 1 shows unselective modification of lysine K138
**Figure S28** MS/MS of digested βLGb modified with compound 1 shows unselective modification of lysine K100

**Figure S29** MS/MS of digested βLGb modified with compound 1 shows unselective modification of lysine T76
Figure S30 MS/MS of digested βLGb modified with compound 1 shows unselective modification of lysine Y42

Figure S31 Mass spectra of compound 1 treated myoglobin followed by incubation with and without thiol. (A) Full mass spectra with all charge states. (B) Corresponding mass spectra of highlighted charge state (+21) in (A). 1 La stands for 1 modification.
Figure S32 Mass spectra of myoglobin modified with compound 1. Left: whole mass spectrum. Right: Mass spectrum on +21 charge state.

Figure S33 Biophysical characterization of myoglobin before and after modification using 1. (A) CD, (B) Melting point; (C) UV-visible absorption spectrum of Heme
Figure S34 Mass spectrum of bovine carbonic anhydrase (BCA) treated with compound 36. (A) Full mass spectrum of BCA modified with 36. (B) Mass spectrum of highlighted m/z region as an example to analyze the modification. Charge states are color coded with the dots. The average modification for each protein is ~3.1.

Figure S35 Time-dependent acetic acid recovered from 6 in the presence of thiol monitored by NMR. After 40 minutes, acetic acid was added to the mixture to confirm the identity of released functionality. The reaction was carried out at MeOH-d4 and 50 mM pH 7.4 phosphate buffer (1:1). [6]/[DEGSH]=1. [6]: 10 mM. Slight shift on peaks were observed due to decreasing pH caused by addition of acetic acid. However, chemical shift of acetic acid is identical to that of released molecule confirming recovery of acetic acid.
Figure S36 Zoom-in NMR spectra of Figure S35.

Figure S37 Time-dependent sulfonamide recovered from 30 in the presence of thiol monitored by NMR. After 240 minutes, p-toluene sulfonamide (TSA) was added to the mixture to confirm the identity of released functionality. The reaction was carried out at MeOH-d4 and 50 mM pH 7.4 phosphate buffer (1:1). [30]/[DEGSH]=1. [30]: 10 mM.
Figure S38 Zoom-in NMR spectra of Figure S37.

Figure S39 Time-dependent benzyl alcohol recovered from 31 in the presence of thiol monitored by NMR. After 120 minutes, benzyl alcohol was added to the mixture to confirm the identity of released functionality. The reaction was carried out at MeOH-d4 and 50 mM pH 7.4 phosphate buffer (1:1). [31]/[DEGSH]=1. [31]: 10 mM.
Figure S40 Zoom-in NMR spectra of Figure S39.

Figure S41 Time-dependent nitroaniline (NA) recovered from 7 in the presence of thiol monitored by NMR. After reaction complete, nitroaniline was added to the mixture to confirm the identity of released functionality. The reaction was carried out at MeOH-d4 and 50 mM pH 7.4 phosphate buffer (1:1). [7]/[DEGSH]=1. [7]: 10 mM.
Figure S42 Time-dependent N-methylbenzylamine (MBA) recovered from 32 in the presence of thiol monitored by NMR. After reaction complete, MBA was added to the mixture to confirm the identity of released functionality. The reaction was carried out at MeOH-d4 and 50 mM pH 7.4 phosphate buffer (1:1). [32]/[DEGSH]=1. [32]: 10 mM.

Figure S43 Time-dependent TEA recovered from 1 in the presence of thiol monitored by NMR. The reaction was carried out at MeOH-d4 and 50 mM pH 7.4 phosphate buffer (1:1). [1]/[DEGSH]=1. [1]: 10 mM.
Figure S44 Time-dependent pyridine recovered from 4 in the presence of thiol monitored by NMR. The reaction was carried out at MeOH-d4 and 50 mM pH 7.4 phosphate buffer (1:1). [4]/[DEGSH]=1. [4]: 10 mM.
Figure S45 Time-dependent bexarotene released from 33 triggered by thiol and monitored by NMR. The reaction was carried out at MeOH-d4 in the presence of 10 mg of solid Na₂CO₃. Note: NMR of 33 was recorded in CDCl₃. [33]/[DEGSH]=1/2. [33]: 10 mM.
Figure S46 Release of active drug from compound 33 triggered by Glutathione (GSH) followed by mass spectrometry. TOP: Positive mode mass spectrometry analysis; Bottom: negative mode mass spectrometry analysis. Observation of active drug signature suggests the release of drug from the prodrug. The reaction was carried out at MeOH and 50 mM pH 7.4 phosphate buffer mixture (1:1). [33] =0.5 mM, [GSH] =5 mM. The sample was analyzed by mass spectrometry after 6-hours incubation.
Figure S47 Time dependent desmethyltamoxifen released from 34 in the presence of thiol monitored by NMR. After 3 days, commercial desmethyltamoxifen (DMT) was added to the reaction mixture to validate the identity of released molecule. Observation of identical chemical shifts of both desmethyltamoxifen and released molecules indicates the release of desmethyltamoxifen from 34. The reaction was carried out at MeOH-d4 and 50 mM pH 7.4 phosphate buffer (1:1). [34]/[DEGSH]=1/2. [34]: 5 mM.

Figure S48 Zoom-in NMR spectra of Figure S47.
Figure S49 Conjugation of compound 33 to PEG-DBCO followed by NMR in CDCl$_3$. (* and # are attributed to DCM and H$_2$O). Top: NMR spectrum of PEG-DBCO. Bottom: NMR spectrum of compound 33. Middle: NMR spectrum of PEG-drug conjugate. Observation of chemical shift shifting of a, b and c protons whose chemical shift should be significantly changed after click reaction and signatures from both precursors suggests successful conjugation.
**Figure S50** Release of bexarotene from PEG-33 conjugate in the presence of thiol monitored by NMR. Bottom: PEG-33 conjugate itself. Middle: PEG-33 conjugate treated with thiol for 16 hours. Top: PEG-33 conjugate treated with thiol for 16 hours followed by addition of commercial bexarotene to validate the identity of released molecule. The fact that newly appeared peaks after thiol incubation is identical to bexarotene proves the release of bexarotene from polymer-drug conjugate. The reaction was carried out at MeOH-d4 and 50 mM pH 7.4 phosphate buffer (1:1). [PEG-33]/[DEGSH]=1:2. [PEG-33]: 1 mM.

**Figure S51** BSA release followed by fluorescence in the presence(A) and absence(B) of thiol.
NMR data for trigger-to-release kinetics measurement (Figure S52-S103)

Figure S52 Thiol-addition with 1.

Figure S53 Amine-addition with 1.
Figure S54 Thiol-addition with 2.

Figure S55 Amine-addition with 2
Figure S56 Thiol-addition with 3.

Figure S57 Amine-addition with 3.
Figure S58 Thiol-addition with 4.

Figure S59 Thiol-addition with 4 accelerated by introduction of DMAP in reaction mixture
**Figure S60.** Zoom-in spectra of Figure S59.

**Figure S61.** Amine-addition with 4
Figure S62 Thiol-addition with 5

Figure S63 Zoom-in spectra of Figure S62
Figure S64 Amine-addition with 5.
Figure S65 Thiol-addition with 6.

Figure S66 Zoom-in spectra of Figure S65.
Figure S67 Amine-addition with 6.

Figure S68 Thiol-addition with 7.
Figure S69 Amine-addition with 7.
Figure S70 Thiol-addition with 8.

Figure S71 Zoon-in spectra of Figure S70.
Figure S72 Amine-addition with 8.
Figure S73 Thiol-addition with 9.

Figure S74 Zoom in spectra of Figure S73.
Figure S75 Amine-addition with 9.
Figure S76 Thiol-addition with 10.

Figure S77 Zoon-in spectra of Figure 76.
Figure S78 Amine-addition with 10.

Figure S79 Zoom-in spectra of Figure S78.
Figure S80 Thiol-addition with 11.

Figure S81 Zoom-in spectra of Figure S80.
Figure S82 Amine-addition reaction with 11.

Figure S83 Zoom-in spectra of Figure S82
Figure S84 Thiol-addition with 12.

Figure S85 Zoom-in spectra of Figure S84.
**Figure S86** Amine-addition reaction with 12.

**Figure S87** Zoom-in spectra of Figure S86.
Figure S88 Thiol-addition with 13.

Figure S89 Zoom-in spectra of Figure S88.
Figure S90 Amine-addition with 13.

Figure S91 Zoom-in spectra of Figure S90.
Figure S92 Thiol-addition with 14.

Figure S93 Zoom-in spectra of Figure S92.
Figure S94 Amine-addition with 14.

Figure S95 Zoom-in spectra of Figure S94.
Figure S96 Thiol-addition with 15.

Figure S97 Zoom-in spectra of Figure S96.
**Figure S98** Amine-addition with 15.

**Figure S99** Zoom-in spectra of Figure S98.
Figure S100 Thiol-addition with 16.

Figure S101 Zoom-in spectra of Figure S100.
Figure S102 Amine-addition with 16.

Figure S103 Zoom-in spectra of Figure S102.
Figure S104 Hydrolytic stability of compound 1 at 50 mM pH7.4 phosphate buffer followed by NMR at 0 hour and 23 hours. The integration ratio between 4' and 4 suggests that 9% of compound 1 hydrolyzes in 23 hours.
Thiol oxidation

Figure S105. Time dependent NMR of PEG2-SH measured in MeOD.

Figure S106. Time dependent NMR of DEGSH measured in MeOH-d4 and pH 7.4 phosphate buffer mixture
Influence of solvent polarity on trigger-to-reverse process

Figure S107. Trigger-to-reverse of compound 18 in pH 7.4 phosphate buffer and MeOH-d4 mixture. [DEGSH]/[18] = 1:2.

Figure S108. Trigger-to-reverse of compound 18 in MeOH-d4 mixture. [DEGSH]/[18] = 1:2.
$^1$H NMR and $^{13}$C NMR spectra of all synthesized compounds

* : MeOD
# : H$_2$O
* : MeOD
#: H₂O
* : MeOD
## : H₂O
* : MeOD
# : H₂O
* : MeOD
#: H₂O
& : THF

& : THF
*: MeOD
#: H₂O
* : CHCl₃
# : H₂O
*: CHCl₃
* : CHCl₃
#: H₂O
* : CHCl₃
# : H₂O
* : MeOD
#: H₂O
* : CHCl₃
#: H₂O
* : MeOD
#: H₂O
* : CHCl₃
# : H₂O
* : MeOD
#: H₂O
*: CHCl₃
#: H₂O
&: TMS

&: TMS
*: CHCl₃
#: H₂O
&: TMS

&: TMS
* : CHCl₃
# : H₂O
& : TMS

& : TMS
* : MeOD
#: H$_2$O
*: MeOD
#: H₂O
* : CHCl₃
#: H₂O
&: TMS
* : CHCl₃
#: H₂O
& : TMS

& : TMS
*: CHCl₃
#: H₂O
&: Toluene sulfonamide
*: CHCl₃
#: H₂O
&: TMS
*: CHCl₃
#: H₂O
&: TMS
*: CHCl₃
#: H₂O
&: TMS
*: CHCl₃
#: H₂O
&: TMS
*: MeOD
#: H$_2$O
*: CHCl₃
#: H₂O
*: CHCl₃
#: H₂O
&: triethylamine-HBr salt
* : CHCl₃
# : H₂O
* : CHCl₃
# : H₂O
&: triethylamine-HBr salt