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Developing Antineoplastic Agents Targeting Peroxisomal Enzymes: Cytisine-linked Isoflavonoids as Inhibitors of Hydroxysteroid 17-beta-dehydrogenase-4 (HSD17B4)

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Supplemental Methods

Chemicals were purchased from Sigma Aldrich (Milwaukee, WI) or Fisher Scientific (Pittsburgh, PA) or were synthesized according to literature procedures. Hydrazide-PEG_4-biotin was purchased from Thermo Fisher Scientific (Florence, KY). Solvents were used from commercial vendors without further purification unless otherwise noted. Nuclear magnetic resonance spectra were determined on a Varian instrument (^1H, 400MHz; ^{13}C, 100Mz). High resolution mass spectra (HRMS) were recorded on a Thermo Scientific Q Exactive Orbitrap mass spectrometer. Resolution was set at 140,000. Samples were introduced through direct infusion using a syringe pump with a flow rate of 5 µL/min. Purity of compounds, which were not acid-sensitive and hence amenable to HPLC analysis using gradients of formic acid-acetonitrile for elution, was greater than 98% as established using high performance liquid chromatography (HPLC) trace and low-resolution electrospray mass spectra (LRMS). These spectra were recorded on a liquid chromatography-mass spectrometry tandem system using an Agilent 1200 series Quaternary LC system equipped with a diode array detector, Eclipse XDB-C18 column (250 mm x 4.6 mm, 5 mm), and an Agilent 6120 Quadrupole MSD mass spectrometer. Method: 0-16 min 5-95% acetonitrile with 0.1% formic acid, 16-20 min 100% acetonitrile with 0.1% formic acid, 20-30 min, 100-5% acetonitrile with 0.1% formic acid. Solvent: H_2O with 0.1% formic acid and acetonitrile with 0.1% formic acid. Purity of all new compounds was greater than 95% as established using combustion analyses determined by Atlantic Microlabs, Inc. (Norcross, GA). Compounds were chromatographed on preparative layer Merck silica gel F254 unless otherwise indicated.

General procedure for the synthesis of isoflavones 3. To a solution of deoxybenzoin (10 mmol) in DMF (7 mL) at 30-40°C under an argon atmosphere was added dropwise 3.7 mL of a 98% solution of boron trifluoride etherate. The mixture was stirred for 30 min, and phosphorous oxytrichloride (2 mL, 21.5 mmol) was added. The mixture was heated at 60°C for 3-5 h, cooled, poured into water and extracted with ethyl acetate. The organic solution was dried over anhydrous $MgSO_4$. The product was isolated by crystallization (from either methanol or ethanol) to afford isoflavones **3**.

7-Hydroxy-3-phenyl-4*H*-chromen-4-one (3a). Yield: 69%; mp 209-210°C (lit[1] mp 210-213°C). ^1H NMR (400 MHz, DMSO-d_6) δ 10.86 (s, 1H), 8.41 (s, 1H), 8 (d, 1H, J = 8.8 Hz), 7.62-7.55 (m, 2H), 7.5-7.36 (m, 3H), 6.97 (dd, 1H, J = 8.8, 2 Hz), 6.9 (d, 1H, J = 2 Hz); ^{13}C NMR (100 MHz, DMSO-d_6) δ 174.4, 162.7, 157.5, 153.9, 132.1, 129, 128.1, 127.7, 127.3, 123.6, 116.6, 115.3, 102.2. HRMS (ESI) Calcd for $C_{15}H_{11}O_3$: 239.0703 $(M+H)^+$. Found: 239.0704. NMR data was consistent with reported data.[1-4]

7-Hydroxy-3-(4-methoxyphenyl)-4*H*-chromen-4-one (3b). Yield: 53%; mp 259-260°C (lit[5] mp 259-261°C). ^1H NMR

(400 MHz, DMSO-d_6) δ 10.8 (s, 1H), 8.34 (1H), 7.97 (d, 1H, J = 8.7), 7.51 (d, 2H, J = 8.8 Hz), 6.99 (d, 2H, J = 8.8 Hz), 6.94 (dd, 1H, J = 8.7, 2.3 Hz), 6.87 (d, 1H, J = 2.3 Hz), 3.78 (s, 3H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 174.6, 162.6, 158.9, 157.4, 153.2, 130.1, 127.3, 124.2, 123.2, 116.6, 115.2, 113.6, 102.1, 55.2. NMR data was consistent with reported data.[5] HRMS (ESI) Calcd for $C_{16}H_{13}O_4$: 269.0808 $(M+H)^+$. Found: 269.0810.

3-(4-Chlorophenyl)-7-hydroxy-4*H*-chromen-4-one (3c). Yield: 59%; mp 260-261°C (lit[6] mp 260°C); ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.86 (s, 1H), 8.45 (s, 1H), 7.98 (d, 1H, J = 8.8 Hz), 7.61 (d, 2H, J = 8.4 Hz), 7.5 (d, 2H, J = 8.4 Hz), 6.96 (dd, 1H, J = 8.8, 2.4 Hz), 6.89 (d, 1H, J = 2.4 Hz); ^{13}C NMR (100 MHz, DMSO-d_6) δ 174.2, 162.8, 157.5, 154.1, 132.5, 131, 130.7, 128.1, 127.3, 122.3, 116.5, 115.4, 102.2. NMR data was consistent with reported data.[7-8] HRMS (ESI) Calcd for $C_{15}H_{10}O_3{}^{35}Cl$: 273.0313 $(M+H)^+$. Found: 273.0316; Calcd for $C_{15}H_{10}O_3{}^{37}Cl$: 275.0283 $(M+H)^+$. Found: 275.0285.

3-(4-Chlorophenyl)-7-hydroxy-2-methyl-4*H*-chromen-4-one (3d). Acetic anhydride (3 mL, 31.7 mmol) was added to a suspension of potassium carbonate (94.2 g, 30.4 mmol) and α-4-chlorophenyl-2,4-dihydroxyacetophenone (2 g, 7.6 mmol) in DMF (20 mL) and the resulting suspension was heated at 120°C for 8 h under an argon atmosphere. The mixture was cooled and poured into water (100 mL). The precipitate was filtered, washed with water (two 100 mL portions) and diethyl ether (100 mL) to afford 1.91 g (88%) of the product as a white solid: mp 277-278°C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.88 (s, 1H), 7.87 (d, 1H, J = 8.6 Hz), 7.48 (d, 2H, J = 8.4 Hz), 7.3 (d, 2H, J = 8.4 Hz), 6.9 (dd, 1H, J = 8.6, 2 Hz), 6.83 (d, 1H, J = 2 Hz), 2.24 (s, 3H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 174.6, 162.8, 162.6, 157.1, 132.5, 132.3, 132.2, 128.1, 127.1, 121, 115.4, 114.9, 101.9, 19.2. HRMS (ESI) Calcd for $C_{16}H_{12}O_3{}^{35}Cl$: 287.0480 $(M+H)^+$. Found: 287.0471; Calcd for $C_{16}H_{12}O_3{}^{37}Cl$: 289.0451 $(M+H)^+$. Found: 289.0441. Anal. Calcd for $C_{16}H_{11}O_3Cl$: C, 67.03; H, 3.87. Found: C, 66.87; H, 4.04.

General procedure of for the synthesis deoxybenzoins 5. To a mixture of resorcinol (60 mmol) and phenylacetic acid (60 mmol) under an argon atmosphere was added 74 mL of 98% solution of boron trifluoride etherate. The mixture was heated to 85°C for 3-5 h. The mixture was poured into cold water and extracted with ethyl acetate. The combined organic layers were washed with brine and dried over anhydrous $MgSO_4$. The product was purified by column chromatography (using 1:20 to 1:3 ethyl acetate-hexanes or using 1:99 to 2:98 methanol-dichloromethane) to afford deoxybenzoins **5**.

1-(2,4-dihydroxyphenyl)-2-phenylethanone (5a). Yield: 60%; mp 111-112°C (lit[1] mp 110-113°C); ^{1}H NMR (400 MHz, $CDCl_3$) δ 12.69 (s, 1H), 7.75 (d, 1H, J = 8.6 Hz), 7.37-7.32 (m, 2H), 7.3-7.24 (m, 3H), 6.4-6.34 (m, 2H), 5.76 (s, 1H), 4.21 (s, 2H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 202.4, 165.7, 162.9, 134.4, 133, 129.5, 128.9, 127.3, 113.7, 108.2, 103.8, 45. NMR data was consistent with reported data in DMSO-d_6.[1-2,9] HRMS (ESI) Calcd for $C_{14}H_{13}O_3$: 229.0859 $(M+H)^+$. Found: 229.0860.

1-(2,4-Dihydroxyphenyl)-2-(4-methoxyphenyl)ethanone (5b). Yield: 63%; mp 154-155°C (lit[10] mp 156-157°C); ^{1}H NMR (400 MHz, DMSO-d_6) δ 12.56 (s, 1H), 10.66 (s, 1H), 7.94 (d, 1H, J = 9 Hz), 7.2 (d, 2H, J = 8.8 Hz), 6.87 (d, 2H, J = 8.8 Hz), 6.39 (dd, 1H, J = 9, 2.3 Hz), 6.25 (d, 1H, J = 2.3 Hz), 4.2 (s, 2H), 3.72 (s, 3H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 202.5, 164.9, 164.7, 158, 133.6, 130.5, 127, 113.8, 112.1, 108.2, 102.5, 55, 43.2. NMR data was consistent with reported data in DMSO-d_6.[9] HRMS (ESI) Calcd for $C_{15}H_{15}O_4$: 259.0965 $(M+H)^+$. Found: 259.0967.

α-4-Chlorophenyl-2,4-dihydroxyacetophenone (5c). Yield: 49%; mp 157-158°C (lit[10] mp 150-150.5°C); ^{1}H NMR (400 MHz, DMSO-d_6) δ 12.41 (s, 1H), 10.71 (s, 1H), 7.93 (d, 1H, J = 9 Hz), 7.37 (d, 2H, J = 8.2 Hz), 7.29 (d, 2H, J = 8.2 Hz), 6.41 (dd, 1H, J = 9, 1.6 Hz), 6.26 (d, 1H, J = 1.6 Hz), 4.33 (s, 2H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 201.6, 165, 164.5, 134.2, 133.5, 131.6, 131.4, 128.3, 112.3, 108.3, 102.5, 43.4. NMR data was consistent with reported data in acetone-d_6[11] and methanol-d_4.[12] HRMS (ESI) Calcd for $C_{14}H_{12}O_3{}^{35}Cl$: 263.0469 $(M+H)^+$. Found: 263.0473; Calcd for $C_{14}H_{12}O_3{}^{37}Cl$: 265.0440 $(M+H)^+$. Found: 265.0444.

General procedure for 7-(2-bromoethoxy)isoflavones 6. To a solution of 2 mmol of 7-hydroxyisoflavone **3** in DMF (10 mL) was added K_2CO_3 (690 mg, 5 mmol) and 1,2-dibromoethane (0.9 mL, 10.4 mmol). The mixture was stirred for 3 h at 80°C under a nitrogen atmosphere. The product was cooled and poured into cold water. The precipitate was filtered, washed successively with water and cold diethyl ether to afford 7-(2-bromoethoxy)isoflavones **6** that were purified by crystallization and/or chromatography on silica gel.

7-(2-Bromoethoxy)-3-phenyl-4*H*-chromen-4-one (6a). Yield: 77%; mp 200-201°C (lit[13] mp 202-204°C); ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.24 (d, 1H, J = 8.8 Hz), 7.95 (s, 1H), 7.56 (d, 2H, J = 7.2 Hz), 7.48-7.34 (m, 3H), 7.02 (dd, 1H, J = 8.8, 2 Hz), 6.88 (d, 1H, J = 2 Hz), 4.4 (t, 2H, J = 6.1 Hz), 3.7 (t, 1H, J = 6.1 Hz); ^{13}C NMR (100 MHz, $CDCl_3$) δ 175.7, 162.5, 157.9, 152.8, 132, 129.1, 128.6, 128.3 (128.31), 128.3 (128.27), 125.5, 119.1, 114.8, 101.3, 68.3, 28.4. NMR data was consistent with reported data in DMSO-d_6.[13] HRMS (ESI) Calcd for $C_{17}H_{14}O_3{}^{79}Br$: 345.0121 $(M+H)^+$.

Found: 345.0125; Calcd for $C_{17}H_{14}O_3{}^{81}Br$: 347.0100 $(M+H)^+$. Found: 347.0101.

7-(2-Bromoethoxy)-3-(4-methoxyphenyl)-4*H*-chromen-4-one (6b). Yield: 80%. mp 174-175°C (lit[13] mp 178-180°C); ^1H NMR (400 MHz, DMSO-d_6) δ 8.44 (s, 1H), 8.05 (d, 1H, J = 8.8 Hz), 7.53 (d, 2H, J = 8.6 Hz), 7.22 (d, 1H, J = 2.4 Hz), 7.12 (dd, 1H, J = 8.8, 2.4 Hz), 7 (d, 2H, J = 8.6 Hz), 4.5 (t, 2H, J = 5.2 Hz), 3.87 (t, 2H, J = 5.2 Hz), 3.79 (s, 3H); ^1H NMR (400 MHz, $CDCl_3$) δ 8.23 (d, 1H, J = 9 Hz), 7.93 (s, 1H), 7.5 (d, 2H, J = 8.8 Hz), 7.01 (dd, 1H, J = 9, 2.4 Hz), 6.97 (d, 2H, J = 8.8 Hz), 6.86 (d, 1H, J = 2.4 Hz), 4.4 (t, 2H, J = 6.4 Hz), 3.84 (s, 3H), 3.7 (t, 2H, J = 6.4 Hz); ^{13}C NMR (100 MHz, $CDCl_3$) δ 175.9, 162.4, 159.7, 157.9, 152.2, 130.2, 128.2, 125.1, 124.2, 119, 114.7, 114.1, 101.2, 68.3, 55.5, 28.5. NMR data was consistent with reported data in DMSO-d_6[13] and $CDCl_3$.[14] HRMS (ESI) Calcd for $C_{18}H_{16}O_4{}^{79}Br$: 375.0226 $(M+H)^+$. Found: 375.0230; Calcd for $C_{17}H_{14}O_3{}^{81}Br$: 377.0206 $(M+H)^+$. Found: 377.0206.

7-(2-Bromoethyloxy)-3-(4-chlorophenyl)-4*H*-chromen-4-one (6c). Yield: 85%; mp 188-189°C; ^1H NMR (400 MHz, DMSO-d_6) δ 8.54 (s, 1H), 8.06 (d, 1H, J = 9 Hz), 7.64 (d, 2H, J = 8.4 Hz), 7.51 (d, 2H, J = 8.4 Hz), 7.25 (d, 1H, J = 2.4 Hz), 7.14 (dd, 1H, J = 9; 2.4 Hz), 4.51 (t, 2H, J = 5.3 Hz), 3.87 (t, 2H, J = 5.3 Hz); ^1H NMR (400 MHz, $CDCl_3$) δ 8.23 (d, 1H, J = 9.2 Hz), 7.95 (s, 1H), 7.51 (d, 2H, J = 8.4 Hz), 7.41 (d, 2H, J = 8.4 Hz), 7.03 (dd, 1H, J = 9.2, 2.4 Hz), 6.88 (d, 1H, J = 2.4 Hz), 4.4 (t, 2H, J = 6.3 Hz), 3.7 (t, 2H, J = 6.3 Hz); ^{13}C NMR (100 MHz, $CDCl_3$) δ 175.4, 162.6, 157.9, 152.8, 134.3, 130.4, 130.3, 128.8, 128.2, 124.5, 118.9, 114.9, 101.3, 68.4, 28.4. HRMS (ESI) Calcd for $C_{17}H_{13}O_3{}^{79}Br^{35}Cl$: 378.9742 $(M+H)^+$. Found: 378.9734; Calcd for $C_{17}H_{13}O_3{}^{79}Br^{37}Cl$: 380.9713 $(M+H)^+$ Found: 380.9711; Calcd for $C_{17}H_{13}O_3{}^{81}Br^{37}Cl$: 382.9692 $(M+H)^+$. Found: 382.9682. Anal. Calcd for $C_{17}H_{12}O_3BrCl$: C, 53.79; H, 3.19. Found: C, 54.09; H, 3.27.

7-(2-Bromoethoxy)-3-(4-chlorophenyl)-2-methyl-4*H*-chromen-4-one (6d). To a solution of 7-hydroxyisoflavone **3d** (573 mg, 2 mmol) in anhydrous DMF (10 mL) was added K_2CO_3 (690 mg, 5 mmol) and 1,2-dibromoethane (0.9 mL, 10.4 mmol). The mixture was stirred at 80°C for 3 h under a nitrogen atmosphere. The mixture was filtered, and DMF was evaporated. The product was isolated by column chromatography using ethyl acetate-hexanes (from 1:9 to 3:7) to give 485 mg (62%) of **6d**: mp 160-161°C (lit[14] mp 165-167°C); ^1H NMR (400 MHz, DMSO-d_6) δ 7.95 (d, 1H, J = 8.6 Hz), 7.5 (d, 2H, J = 8.4 Hz), 7.33 (d, 2H, J = 8.4 Hz), 7.21 (d, 1H, J = 2.4 Hz), 7.09 (dd, 1H, J = 8.6; 2.4 Hz), 4.5 (t, 2H, J = 5.5 Hz), 3.87 (t, 2H, J = 5.5 Hz), 2.27 (s, 3H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 174.6, 163.2, 162.3, 157, 132.5, 132.3, 132, 128.1, 126.9, 121.3, 116.7, 114.8, 101.2, 68.5, 31, 19.2. NMR data was consistent with reported data in $CDCl_3$.[14] HRMS (ESI) Calcd for $C_{18}H_{15}O_3{}^{79}Br^{35}Cl$: 392.9888 $(M+H)^+$. Found: 392.9883; Calcd for $C_{18}H_{15}O_3{}^{79}Br^{37}Cl$: 394.9858 $(M+H)^+$. Found: 394.9858; Calcd for $C_{18}H_{15}O_3{}^{81}Br^{37}Cl$: 396.9838 $(M+H)^+$. Found: 396.9830. Anal. Calcd for $C_{18}H_{14}O_3BrCl$: C, 54.92; H, 3.58. Found: C, 54.85; H, 3.48.

General procedure for the synthesis of piperazinyl-substituted isoflavones 7 and 8. A mixture of 1 mmole of isoflavone **6**, either piperazine or 1-(2-hydroxyethyl)piperazine (1.2 mmol), NaI (1 mmol) and diisopropylethylamine (0.6 mL, 3.5 mmol) in DMF (9 mL) was stirred for 3 h at 60 °C under a nitrogen atmosphere. The mixture was cooled; the solvent was evaporated; and the product was purified by column chromatography using methanol-dichloromethane (1:9 to 1:3) to afford piperazinyl-substituted isoflavones **7** or **8** as white solids.

3-(4-Chlorophenyl)-7-(2-(piperazin-1-yl)ethoxy)-4*H*-chromen-4-one (7c). To a solution of 378 mg (1 mmole) of **6c** in DMF (10 mL) was added piperazine (172 mg, 2 mmol), NaI (150 mg, 1 mmol). and K_2CO_3 (276 mg, 2 mmol). The mixture was stirred for 2 h at 60°C under a nitrogen atmosphere. The mixture was cooled and poured into cold water (100 mL). The precipitated was collected and washed with cold water. The product was recrystallized from methanol to afford 306 mg (79%) of **7c** as a white solid: mp 147-148°C; ^1H NMR (400 MHz, $CDCl_3$) 8.19 (d, 1H, J = 9 Hz), 7.94 (s, 1H), 7.5 (d, 2H, J = 8.6 Hz), 7.4 (d, 2H, J = 8.6 Hz), 7 (dd, 1H, J = 9, 2.4 Hz), 6.86 (d, 1H, J = 2.4 Hz), 4.2 (t, 2H, J = 5.7 Hz), 2.96-2.9 (m, 4H), 2.87 (t, 2H, J = 5.7 Hz), 2.64-2.53 (m, 4H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 175.5, 163.4, 158, 152.7, 134.3, 130.5, 130.4, 128.8, 127.9, 124.4, 118.5, 115.2, 101, 66.7, 57.6, 54.9, 46.1. HRMS (ESI) Calcd for $C_{21}H_{22}O_3N_2{}^{35}Cl$: 385.1324 $(M+H)^+$. Found 385.1327; Calcd for $C_{21}H_{22}O_3N_2{}^{37}Cl$: 387.1295 $(M+H)^+$. Found 387.1297. Rapid air oxidation precluded obtaining a satisfactory combustion analysis.

7-(2-(4-(2-Hydroxyethyl)piperazin-1-yl)ethoxy)-3-phenyl-4*H*-chromen-4-one (8a). Yield: 60%; mp 159-160°C; ^1H NMR (400 MHz, $CDCl_3$) δ 8.21 (d, 1H, J = 8.8 Hz), 7.95 (s, 1H), 7.6-7.52 (m, 2H), 7.48-7.34 (m, 3H), 7 (dd, 1H, J = 8.8, 2 Hz), 6.87 (d, 1H, J = 2 Hz), 4.21 (t, 2H, J = 5.6 Hz), 3.65 (t, 2H, J = 5.6 Hz), 2.89 (t, 2H, J = 5.6 Hz), 2.78-2.52 (m, 8H), 2.6 (t, 2H, J = 5.6 Hz); ^{13}C NMR (100 MHz, DMSO-d_6) δ 174.4, 163, 157.5, 154.2, 132, 128.9, 128.1, 127.8, 126.9, 123.8, 117.6, 115.2, 101.2, 66.5, 60.2, 58.4, 56.4, 53.2, 53; ^{13}C NMR (100 MHz, $CDCl_3$) δ 175.7, 163.2, 158, 152.8, 132.1, 129.1, 128.6, 128.3, 128, 125.5, 118.7, 115, 101, 66.8, 59.4, 57.8, 56.9, 53.7, 52.9. HRMS (ESI) Calcd for $C_{23}H_{27}O_4N_2$: 395.1965 $(M+H)^+$. Found: 395.1957. Anal. Calcd. for $C_{23}H_{26}O_4N_2$: C, 70.03; H, 6.64; N, 7.10. Found: C, 69.79; H, 6.65; N, 7.07.

7-[2-[4-(2-Hydroxyethyl)piperazin-1-yl]ethoxy]-3-(4-methoxyphenyl)-4*H*-chromen-4-one (8b). Yield: 73%; mp 145-146°C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.2 (d, 1H, J = 8.8 Hz), 7.92 (s, 1H), 7.5 (d, 2H, J = 9.2 Hz), 7 (dd, 1H, J = 8.8, 2.4 Hz), 6.97 (d, 2H, J = 9.2 Hz), 6.86 (d, 1H, J = 2.4 Hz), 4.2 (t, 2H, J = 5.8 Hz), 3.84 (s, 3H), 3.63 (t, 2H, J = 5.2 Hz), 2.88 (t, 2H, J = 5.8 Hz), 2.74-2.54 (m, 8H), 2.57 (t, 2H, J = 5.2 Hz); ^{13}C NMR (100 MHz, $CDCl_3$) δ 176, 163.1, 159.7, 158, 152.2, 130.2, 127.9, 125, 124.3, 118.6, 114.9, 114.1, 100.9, 66.7, 59.5, 57.7, 56.9, 55.5, 53.5, 52.9. HRMS (ESI) Calcd for $C_{24}H_{29}O_5N_2$: 425.2071 $(M+H)^+$. Found: 425.2071. Anal. Calcd. for $C_{24}H_{28}O_5N_2$: C, 67.91; H, 6.65; N, 6.60. Found: C, 68.15; H, 6.71; N, 6.56.

3-(4-Chlorophenyl)-7-(2-(4-(2-hydroxyethyl)piperazin-1-yl)ethoxy)-4*H*-chromen-one (8c). Yield: 74%; mp 152-153°C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.2 (d, 1H, J = 8.8 Hz), 7.94 (s, 1H), 7.51 (d, 2H, J = 8.2 Hz), 7.41 (d, 2H, J = 8.2 Hz), 7.02 (dd, 1H, J = 8.8, 2.4 Hz), 6.87 (d, 1H, J = 2.4 Hz), 4.21 (t, 2H, J = 5.6 Hz), 3.62 (t, 2H, J = 5.2 Hz), 2.88 (t, 2H, J = 5.6 Hz), 2.72-2.5 (m, 8H), 2.57 (t, 2H, J = 5.2 Hz); ^{13}C NMR (100 MHz, $CDCl_3$) δ 175.5, 163.4, 158, 152.7, 134.3, 130.5, 130.4, 128.8, 127.9, 124.4, 118.5, 115.2, 101, 66.8, 59.3, 57.8, 56.9, 53.8, 52.9. HRMS (ESI) Calcd for $C_{23}H_{26}O_4N_2{}^{35}Cl$: 429.1576 $(M+H)^+$. Found: 429.1576; Calcd for $C_{23}H_{26}O_4N_2{}^{37}Cl$: 431.1546 $(M+H)^+$. Found: 431.1551. Anal. Calcd. for $C_{23}H_{25}O_4N_2Cl$: C, 64.41; H, 5.88; N, 6.53. Found: C, 64.52; H, 6.01; N, 6.50.

General procedure for the synthesis of cytisine-linked isoflavones 10. A mixture of 0.5 mmol of 7-(2-bromoethoxy)isoflavone **6**, (-)-cytisine (143 mg, 0.75 mmol), NaI (75 mg, 0.5 mmol), and diisopropylethylamine (0.3 mL, 3.5 mmol) in DMF (5 mL) was stirred for 2-4 h at 80°C under a nitrogen atmosphere. The mixture was cooled and poured into cold water. A precipitate was collected and purified by column chromatography using methanol-dichloromethane (2:98 to 5:95) to afford cytisine-linked isoflavones **10** as white solids.

(1*R*,5*S*)-3-(2-((4-Oxo-3-phenyl-4H-chromen-7-yl)oxy)ethyl)-3,4,5,6-tetrahydro-1*H*-1,5-methanopyrido[1,2-*a*][1,5]diazocin-8(2*H*)-one (10a). Yield: 72%; mp 197-198°C (lit[13] mp 195-196°C); ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.46 (s, 1H), 7.99 (d, 1H, J = 9 Hz), 7.62-7.55 (m, 2H), 7.48-7.34 (m, 3H), 7.29 (dd, 1H, J = 8.9, 6.7 Hz), 7.11 (d, 1H, J = 2.4 Hz), 6.98 (dd, 1H, J = 9, 2.4 Hz), 6.18 (dd, 1H, J = 8.9, 1.2 Hz), 6.07 (dd, 1H, J = 6.7, 1.2 Hz), 4.2-4.04 (m, 2H), 3.82-3.64 (m, 2H), 3.06-2.98 (m, 2H), 2.94-2.86 (m, 1H), 2.76-2.62 (m, 2H), 2.52-2.34 (m, 3H), 1.79 (d, 1H, J = 12.5 Hz), 1.7 (d, 1H, J = 12.5 Hz); ^{13}C NMR (100 MHz, DMSO-d_6) δ 174.4, 162.9, 162.2, 157.4, 154.2, 152.1, 138.7, 132, 128.9, 128.2, 127.8, 126.9, 123.7, 117.6, 115.3, 115.1, 103.7, 101.3, 66.3, 60.3, 59.4, 55.7. 49.5, 34.6, 27.3, 25. NMR data was consistent with reported data in DMSO-d_6.[13] Purity by HPLC-LRMS >98% (Fig. 2S, panel A). HRMS (ESI) Calcd for $C_{28}H_{27}O_4N_2$: 455.1965 $(M+H)^+$. Found: 455.1964.

(1*R*,5*S*)-3-(2-((3-(4-Methoxyphenyl)-4-oxo-4*H*-chromen-7-yl)oxy)ethyl)-3,4,5,6-tetrahydro-1*H*-1,5-methanopyrido[1,2-*a*][1,5]diazocin-8(2*H*)-one (10b). Yield: 76%; mp 117-118°C (lit[13] mp 85-86°C); ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.41 (s, 1H), 7.98 (d, 1H, J = 8.8 Hz), 7.52 (d, 2H, J = 8.8 Hz), 7.29 (dd, 1H, J = 8.9, 6.9 Hz), 7.09 (d, 1H, J = 2 Hz), 7.04-6.92 (m, 3H), 6.18 (dd, 1H, J = 8.9, 0.8 Hz), 6.06 (d, 1H, J = 6.9 Hz), 4.18-4.04 (m, 2H), 3.82-3.66 (m, 2H), 3.79 (s, 3H), 3.06-2.98 (m, 2H), 2.94-2.86 (m, 1H), 2.76-2.62 (m, 2H), 2.52-2.32 (m, 3H), 1.84-1.66 (m, 2H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 175.9, 163.7, 162.9, 159.6, 157.9, 152.2, 151.4, 138.7, 130.2, 127.8, 124.8, 124.3, 118.5, 116.7, 114.8, 114, 104.6, 100.8, 66.7, 60.9, 60.4, 56.2, 55.4, 50, 35.6, 28.1, 25.7. NMR data was consistent with reported data in DMSO-d_6.[13] Purity by HPLC-LRMS >98% (Fig. 2S, panel B). HRMS (ESI) Calcd for $C_{29}H_{29}O_5N_2$: 485.2071 $(M+H)^+$. Found: 485.2071. Anal. Calcd. for $C_{29}H_{28}O_5N_2$: C, 71.88; H, 5.82; N, 5.78. Found: C, 71.60; H, 5.75; N, 5.73.

(1*S*,5*S*)-3-(2-((3-(4-Chlorophenyl)-4-oxo-4*H*-chromen-7-yl)oxy)ethyl)-3,4,5,6-tetrahydro-1*H*-1,5-methanopyrido[1,2-*a*][1,5]diazocin-8(2*H*)-one (10c). Yield: 76%; mp 146-147°C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.16 (d, 1H, J = 8.8 Hz), 7.94 (s, 1H), 7.51 (d, 2H, J = 8.6 Hz), 7.4 (d, 2H, J = 8.6 Hz), 7.3-7.18 (m, 1H), 6.9 (dd, 1H, J = 8.8, 2 Hz), 6.76 (d, 1H, J = 2 Hz), 6.43 (d, 1H, J = 8.8 Hz), 5.96 (d, 1H, J = 6.8 Hz), 4.14-3.86 (m, 4H), 3.08-2.92 (m, 3H), 2.75 (t, 2H, J = 5.6 Hz), 2.6-2.42 (m, 3H), 1.94-1.76 (m, 2H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 175.5, 163.7, 163.1, 158, 152.7, 151.4, 138.8, 134.2, 130.5, 130.3, 128.8, 127.9, 124.3, 118.4, 116.8, 115.1, 104.7, 100.9, 66.8, 61, 60.5, 56.2, 50.1, 35.6, 28.1, 25.8. Purity by HPLC-LRMS >98% (Fig. 3S, panel A). HRMS (ESI) Calcd for $C_{28}H_{26}O_4N_2{}^{35}Cl$: 489.1576 $(M+H)^+$. Found: 489.1578; Calcd for $C_{28}H_{26}O_4N_2{}^{37}Cl$: 491.1546 $(M+H)^+$. Found: 491.1555 Anal. Calcd. for $C_{28}H_{25}O_4N_2Cl$: C, 68.78; H, 5.15; N, 5.73. Found: C, 69.02; H, 5.41; N, 5.63.

(1*R*,5*S*)-3-(2-((3-(4-Chlorophenyl)-2-methyl-4-oxo-4*H*-chromen-7-yl)oxy)ethyl)-3,4,5,6-tetrahydro-1*H*-1,5-methanopyrido[1,2-*a*][1,5]diazocin-8(2*H*)-one (10d). Yield: 62%; mp 186-187°C; ^{1}H NMR (400 MHz, methanol-d_4) δ 7.96 (d, 1H, J = 8.4 Hz), 7.45 (d, 2H, J = 8.6 Hz), 7.39 (dd, 1H, J = 8.9, 6.9 Hz), 7.28 (d, 2H, J = 8.6 Hz), 6.94-6.9 (m, 1H), 6.89 (d, 1H, J = 2 Hz), 6.35 (dd, 1H, J = 8.9, 1.2 Hz), 6.25 (dd, 1H, J = 6.9, 1.2 Hz), 4.12-4.04 (m, 2H), 4 (d, 1H, J = 15.4 Hz), 3.88 (dd, 1H, J = 15.4, 6.4 Hz), 3.13-2.98 (m, 3H),

2.78-2.72 (m, 2H), 2.6-2.44 (m, 3H), 2.3 (s, 3H), 1.96-1.82 (m, 2H); ^{13}C NMR (100 MHz, methanol-d_4) δ 178, 165.8, 165.5, 165.1, 159.2, 153.8, 141.2, 134.9, 133.6, 133.2, 129.6, 128, 123.2, 117.8, 116.4, 116.2, 107.7, 101.9, 67.8, 61.9, 61.4, 57.3, 51.6, 36.9, 29.5, 26.3, 19.6. Purity by HPLC-LRMS >98% (Fig. 3S, panel B). HRMS (ESI) Calcd for $C_{29}H_{28}N_2O_4{}^{35}Cl$: 503.1732 $(M+H)^+$. Found: 503.1735; Calcd for $C_{29}H_{28}N_2O_4{}^{37}Cl$: 505.1703 $(M+H)^+$. Found: 505.1707. Anal. Calcd. for $C_{29}H_{27}O_4N_2Cl$: C, 69.25; H, 5.41; N, 5.57. Found: C, 68.98; H, 5.36; N, 5.50.

3-(4-Chlorophenyl)-7-(hex-5-en-1-yloxy)-2-methyl-4*H*-chromen-4-one (11d). To a solution of 7-hydroxyisoflavone **3d** (573 mg, 2 mmol) in DMF (10 mL) was added K_2CO_3 (690 mg, 5 mmol) and 6-bromo-1-hexene (0.6 mL, 4.5 mmol). The mixture was stirred at 80°C for 1 h under a nitrogen atmosphere. The mixture was cooled, diluted with water, and extracted with ethyl acetate. The combined organic layers were washed successively with water and brine and dried over anhydrous $MgSO_4$. The product was isolated by column chromatography using ethyl acetate-hexanes as eluent (from 5:95 to 1:5) to afford 600 mg (81%) of **11d** as a white solid: mp 104°C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 7.93 (d, 1H, J = 8.6 Hz), 7.49 (d, 2H, J = 8.4 Hz), 7.32 (d, 2H, J = 8.4 Hz), 7.14 (d, 1H, J = 2.3 Hz), 7.04 (dd, 1H, J = 8.6; 2.3 Hz), 5.9-5.76 (m,1H), 5.08-4.95 (m, 2H), 4.13 (t, 2H, J = 6.4 Hz), 2.27 (s, 3H), 2.14-2.07 (m, 2H), 1.82-1.72 (m, 2H), 1.58-1.48 (m, 2H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 174.6, 163.1 (163.13), 163.1 (163.11), 157.1, 138.5, 132.5, 132.3, 132.1, 128.1, 126.8, 121.3, 116.3, 115, 114.9, 100.8, 68.3, 32.8, 27.9, 24.6, 19.2. HRMS (ESI) Calcd for $C_{22}H_{22}O_3{}^{35}Cl$: 369.1263 $(M+H)^+$. Found: 369.1253; Calcd for $C_{22}H_{22}O_3{}^{37}Cl$: 371.1233 $(M+H)^+$. Found: 371.1227. Anal. Calcd. for $C_{22}H_{22}O_3Cl$: C, 71.64; H, 5.74. Found: C, 71.55; H, 5.59.

3-(4-Chlorophenyl)-2-methyl-7-(4-(oxiran-2-yl)butoxy)-4*H*-chromen-4-one (12d). A solution of 77% *meta*-chloroperoxybenzoic acid (896 mg, 4 mmol) in dichloromethane (5 mL) was added to a solution of **11d** (338 mg, 0.92 mmol) in dichloromethane (5 mL). The mixture was stirred for 4 h at 25°C under a nitrogen atmosphere. The product was poured into saturated $NaHCO_3$ solution and extracted with dichloromethane. The organic layers were washed with brine and dried over anhydrous $MgSO_4$. The product was purified by column chromatography using methanol-dichloromethane (ratio ranging from 2:98 to 2:48) to afford 234 mg (76%) of **12d**: mp 118-119°C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.11 (d, 1H, J = 8.8 Hz), 7.4 (d, 2H, J = 8.4 Hz), 7.23 (d, 2H, J = 8.4 Hz), 6.95 (dd, 1H, J = 8.8, 2.4 Hz), 6.82 (d, 1H, J = 2.4 Hz), 4.08 (t, 2H, J = 6.2 Hz), 3-2.92 (m, 1H), 2.79 (t, 1H, J = 4.4 Hz), 2.54-2.48 (m, 1H), 2.29 (s, 3H), 1.96-1.86 (m, 2H), 1.76-1.54 (m, 4H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 176.1, 163.5, 162.8, 157.7, 133.8, 132, 131.8, 128.7, 127.7, 122.4, 117.2, 114.7, 100.5, 68.4, 52.2, 47.1, 32.3, 28.9, 22.8, 19.5. HRMS (ESI) Calcd for $C_{22}H_{22}O_4{}^{35}Cl$: 385.1201 $(M+H)^+$. Found: 385.1212; Calcd for $C_{22}H_{22}O_4{}^{37}Cl$: 387.1172 $(M+H)^+$. Found: 387.1182. Anal. Calcd. for $C_{22}H_{21}O_4Cl$ C, 68.66; H, 5.50. Found: C, 68.39; H, 5.50.

(1*R*,2'ζ,5*S*)-3-(6-((3-(4-Chlorophenyl)-2-methyl-4-oxo-4*H*-chromen-7-yl)oxy)-2-hydroxyhexyl)-3,4,5,6-tetrahydro-1*H*-1,5-methanopyrido[1,2-*a*][1,5]diazocin-8(2*H*)-one (13d). A mixture of **12d** (327 mg, 0.8 mmol) and (1*R*,5*S*)-cytisine (194 mg, 1 mmol) in absolute ethanol (9 mL) was stirred in a pressure tube for 20 h at 90°C. The solvent was evaporated, and the product was purified by column chromatography using methanol-dichloromethane (2:48) to afford 470 mg (96%) of **13d** as a mixture of diastereoisomers epimeric at the C-2' position in the linker. The ^{1}H NMR spectrum displayed C-2' H (RC<u>H</u>(OH)R') as an unresolved multiplet at δ 3.64-3.5 (**Fig. 1S**, Panel A), but displayed the C-5 ArH of isoflavonoid as two doublets (**Fig. 1S**, Panel B) with one doublet at δ 8.08 (J = 9.2Hz) for one diastereomer and another doublet at δ 8.07 (J = 8.8 Hz) for the other diastereomer in *ca*. 1:1 ratio. Data in support of **13d**: ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.08 and 8.07 (two d, 1H, J = 9.2 Hz), 7.39 (d, 2H, J = 8.4 Hz), 7.32-7.16 (m, 3H), 6.96-6.86 (m, 1H), 6.82-6.76 (m, 1H), 6.46-6.38 (m, 1H), 6.04-5.94 (m, 1H), 4.18-3.84 (m, 3H), 3.64-3.5 (m, 1H), 3.12-2.82 (m, 3H), 2.7-2.62 (m, 1H), 2.56-2.24 (m, 4H), 2.27 (two s, 3H), 2.2-2.1 (m, 1H), 1.98-1.74 (m, 4H), 1.66-1.3 (m, 4H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 174.6, 163.2 (163.18), 163.2 (163.17), 163.1, 162.2, 157.1, 152.2 (152.23), 152.2 (152.21), 138.7, 138.6, 132.5, 132.3, 132.1, 128.1, 126.7, 121.3, 116.3, 115.1, 114.9, 103.7, 103.6, 100.8, 68.5, 67.1, 66.9, 63.05, 62.98, 61.4, 60.8, 60.4, 60, 49.7, 34.8, 34.7, 34.4, 28.64, 28.58, 27.6, 27.4, 25.2, 21.23, 21.15, 19.2. HRMS (ESI) Calcd for $C_{33}H_{36}O_5N_2{}^{35}Cl$: 575.2318 $(M+H)^+$. Found: 575.2312; Calcd for $C_{33}H_{36}O_5N_2{}^{37}Cl$: 577.2289 $(M+H)^+$. Found: 577.2295. Anal. Calcd. for $C_{33}H_{35}O_5N_2Cl{\cdot}½H_2O$: C, 67.86; H, 6.21; N, 4.80. Found: C, 68.26; H, 6.41; N, 4.72.

(1*R*,5*S*)-3-(6-((3-(4-Chlorophenyl)-2-methyl-4-oxo-4*H*-chromen-7-yl)oxy)-2-oxohexyl)-3,4,5,6-tetrahydro-1*H*-1,5-methanopyrido[1,2-*a*][1,5]diazocin-8(2*H*)-one (14d). To a suspension of Dess-Martin periodinane (520 mg, 1.2 mmol) in dichloromethane (8 mL) was added a solution of **13d** (470 mg, 0.8 mmol) in dichloromethane (5 mL). The mixture was stirred at 25°C for 2 h, diluted with dichloromethane, and washed with a 3:2 saturated solution of $Na_2S_2O_3$ and $NaHCO_3$ (20 mL. The combined organic layers were washed with brine and dried over anhydrous $MgSO_4$. The product was purified by column chromatography using methanol-dichloromethane (1:98)

to afford 380 mg (81%) of **14d** as a white foam: ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.1 (d, 1H, J = 8.8 Hz), 7.4 (d, 2H, J = 7.9 Hz), 7.32-7.2 (m, 1H), 7.23 (d, 2H, J = 7.9 Hz), 6.92 (d, 1H, J = 8.8 Hz), 6.84-6.78 (m, 1H), 6.47 (d, 1H, J = 8.8 Hz), 6 (d, 1H, J = 6.8 Hz), 4.19 (d, 1H, J = 15.6 Hz), 3.97 (t, 2H, J = 5.2 Hz), 3.9 (dd, 1H, J = 15.6, 6.8 Hz), 3.2-2.86 (m, 4H), 2.74 (d, 1H, J = 10.4 Hz), 2.62 (d, 1H, J = 10.8 Hz), 2.55 (d, 1H, J = 10.8 Hz), 2.52-2.42 (m, 1H), 2.29 (s, 3H), 2.3-2.22 (m, 1H), 2.18-2.06 (m, 1H), 1.95 (d, 1H, J = 12.7 Hz), 1.82 (d, 1H, J = 12.7 Hz), 1.7-1.52 (m, 4H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 210.5, 176.1, 163.6, 163.5, 162.9, 157.7, 151.2, 138.8, 133.8, 132, 131.8, 128.7, 127.6, 122.4, 117.1, 116.9, 114.7, 104.8, 100.5, 68.2, 67.6, 60.8 (60.83), 60.8 (60.76), 50.1, 39.1, 35.4, 28.5, 28.2, 25.4, 19.9, 19.5. HRMS (ESI) Calcd for $C_{33}H_{34}O_5N_2{}^{35}Cl$: 573.2151 $(M+H)^+$; Found: 573.2156; Calcd for $C_{33}H_{34}O_5N_2{}^{37}Cl$: 575.2121 $(M+H)^+$. Found: 575.2125. Anal. Calcd. for $C_{33}H_{33}O_5N_2Cl{\cdot}½H_2O$: C, 68.09; H, 5.89; N, 4.81. Found: C, 68.02; H, 5.95; N, 4.71.

***N*-((*E* and *Z*)-22-((3-(4-chlorophenyl)-2-methyl-4-oxo-4*H*-chromen-7-yl)oxy)-15-oxo-18-(((1*R*,5*S*)-8-oxo-1,5,6,8-tetrahydro-2*H*-1,5-methanopyrido[1,2-*a*][1,5]diazocin-3(4*H*)-yl)methyl)-3,6,9,12-tetraoxa-16,17-diazadocos-17-en-1-yl)-5-((3a*S*,4*S*,6a*R*)-2-oxohexahydro-1*H*-thieno[3,4-*d*]imidazol-4-yl)pentanamide (15d)**. A mixture of hydrazide-PEG_4-biotin (Thermo Fisher, 50 mg, 0.1 mmol), **14d** (57 mg, 0.1 mmol), and cerium trichloride (3 mg, 0.01 mmol) in methanol (3 mL) was stirred at 60°C for 4 h. The solvent was evaporated, and the product was isolated by preparative chromatography using methanol-dichloromethane (8:92) to afford 25 mg (24%) of **15d** as a mixture of *E/Z*-isomers: ^{1}H NMR (400 MHz, $CDCl_3$) δ 10.51 and 10.3 (two s, 1H), 7.98 (d, 1H, J = 8.7 Hz), 7.45 (d, 2H, J = 8.3 Hz), 7.36-7.22 (m, 1H), 7.28 (d, 2H, J = 8.3 Hz), 7.02-6.93 (m, 2H), 6.8-6.64 (m, 1H), 6.28 and 6.24 (two d, 1H, J = 9 Hz), 6.07 and 6.02 (two d, 1H, J = 6.8 Hz), 5.48-5.36 (m, 1H), 5.18-5.08 (m, 1H), 4.3-4.36 (m, 1H), 4.24-4.18 (m, 1H), 4.16-4.08 (m, 2H), 3.9-3.8 (m, 2H), 3.78-3.4 (m, 16H), 3.38-3.2 (m, 2H), 3.18-3.08 (m, 3H), 3.06-2.6 (m, 6H), 2.54-2.02 (7H), 2-1.46 (m, 10H), 1.44-1.22 (m, 4H). MS (ESI): 1060 $(M+H)^+$, 1077 $(M+NH_4)^+$, 1082 $(M+Na)^+$, 1098 $(M+K)^+$. HRMS (ESI) Calcd for $C_{54}H_{71}O_{11}N_7{}^{35}ClS$: 1060.4615 $(M+H)^+$. Found: 1060.4612; Calcd for $C_{54}H_{71}O_{11}N_7{}^{37}ClS$: 1062.4618 $(M+H)^+$. Found: 1062.4607.

(1*R*,2'ζ,5*S*)-3-(2-Hydroxy-6-phenoxyhexyl)-3,4,5,6-tetrahydro-1*H*-1,5-methanopyrido[1,2-*a*][1,5]diazocin-8(2*H*)-one (16). A mixture of phenol (2 g, 21.3 mmol), K_2CO_3 (8.8 g, 63.4 mmol), and 6-bromo-1-hexene (3.4 mL, 25.5 mmol) in DMF (15 mL) was stirred at 60°C for 5 h under a nitrogen atmosphere. The mixture was cooled, poured into water, and extracted with dichloromethane. The combined organic layers were washed successively with water and brine and dried over anhydrous $MgSO_4$. The product was purified by column chromatography using ethyl acetate-hexanes (5:95) to afford 1.8 g (96%) of (hex-5-en-1-yloxy)benzene as colorless oil: ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.32-7.24 (m, 2H), 6.96-6.86 (m, 3H), 5.9-5.76 (m, 1H), 5.08-4.94 (m, 2H), 3.96 (t, 2H, J = 6.5 Hz), 2.18-2.08 (m, 2H), 1.84-1.76 (m, 2H), 1.62-1.52 (m, 2H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 159.2, 138.7, 129.6, 120.6, 114.9, 114.6, 67.7, 33.6, 28.9, 25.5. NMR data was consistent with reported data in $CDCl_3$.[15-17] A mixture of 77% *meta*-chloroperbenzoic acid (3.4 g, 15.3 mmol) in dichloromethane (3 mL) was added dropwise to a solution of (hex-5-en-1-yloxy)benzene (1.8 g, 10.2 mmol) in dichloromethane (3 mL). The mixture was stirred at 25°C for 2 h. The mixture was poured into saturated solution of $NaHCO_3$ and extracted with dichloromethane. The combined organic layers were washed with brine, and dried over anhydrous $MgSO_4$. The product was purified by column chromatography using ethyl acetate-hexanes (5:95) to afford 1.6 g (81%) of 2-(4-phenoxybutyl)oxirane as colorless oil: ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.32-7.24 (m, 2H), 6.96-6.86 (m, 3H), 3.98 (t, 2H, J = 6.3 Hz), 2.98-2.9 (m, 1H), 2.78-2.72 (m, 1H), 2.52-2.46 (m, 1H), 1.9-1.8 (m, 2H), 1.72-1.58 (m, 4H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 159.1, 129.6, 120.7, 114.6, 67.6, 52.3, 47.2, 32.3, 29.2, 22.8. NMR data was consistent with reported data in $CDCl_3$.[15] A mixture of 2-(4-phenoxybutyl)oxirane (385 mg, 2 mmol) and cytisine (457 mg, 2.4 mmol) in methanol (8 mL) was stirred in a pressure tube for 8 h at 90°C. The solvent was evaporated, and the product was purified by column chromatography using methanol-dichloromethane (ratio ranging from 2:98 to 7:93) to afford 750 mg (98%) of **16** as a mixture of diastereoisomers. The ^{1}H NMR spectrum did not display any signals that would enable calculation of the ratio of diastereomers. The ^{1}H NMR spectrum displayed C-2' H (RC<u>H</u>(OH)R') as an unresolved multiplet at δ 3.62-3.46 (**Fig. 1S**, Panel C). Data in support of **16**: ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.32-7.22 (m, 3H), 6.92 (t, 1H, J = 7.3 Hz), 6.89-6.84 (m, 2H), 6.44-6.4 (m, 1H), 6.01-5.94 (m, 1H), 4.11 and 4.05 (two d, 1H, J = 15.5 Hz), 3.96-3.84 (m, 3H), 3.62-3.46 (m, 1H), 3.09-2.95 (m, 2H), 2.88 and 2.83 (two d, 1H, J = 11.1 and 10.7 Hz), 2.66-2.4 (m, 3H), 2.36-2.06 (m, 3H), 1.97-1.67 (m, 4H), 1.64-1.22 (m, 4H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 163.4 (163.44), 163.4 (163.42), 159.1, 150.9, 150.6, 138.9, 138.8, 129.5, 120.55, 120.53, 117.03, 116.96, 114.52, 114.5, 104.8, 104.7, 67.64, 67.63, 66.5, 65.9, 63.9, 63.5, 62.7, 62, 59.1, 58.9, 50.1, 50, 35.8, 35.2, 34.5, 34.2, 29.4, 29.3, 28.3, 27.9, 26, 25.9, 22.2, 22.1. HRMS (ESI) Calcd for $C_{23}H_{31}O_3N_2$: 383.2329 $(M+H)^+$. Found: 383.2340. Anal. Calcd. for $C_{33}H_{30}O_3N_2$: C, 72.22; H, 7.91; N, 7.32. Found: C, 71.94; H, 7.93; N, 7.28.

(1*R*,5*S*)-3-(2-oxo-6-phenoxyhexyl)-3,4,5,6-tetrahydro-1*H*-1,5-methanopyrido[1,2-*a*][1,5]diazocin-8(2*H*)-one (17). To a suspension of Dess-Martin periodinane (424 mg, 1

mmol) in dichloromethane (3 mL) was added a solution of **16** (258 mg, 0.7 mmol) in dichloromethane (3 mL). The mixture was stirred at 25°C for 2 h, and the reaction was quenched with a 2:1 mixture of saturated $Na_2S_2O_3$ and $NaHCO_3$ (10 mL). The product was extracted with dichloromethane. The combined organic layers were washed successively with saturated $NaHCO_3$ solution and brine and dried over anhydrous $MgSO_4$. The solvent was evaporated, and the product was purified by column chromatography using methanol-dichloromethane (1:24) to afford 147 mg (57%) of **17** as a colorless, viscous oil: ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.32-7.2 (m, 3H), 6.93 (t, 1H, J = 7.3 Hz), 6.9-6.84 (m, 2H), 6.45 (d, 1H, J = 9 Hz), 5.98 (d, 1H, J = 6.8 Hz), 4.17 (d, 1H, J = 15.5 Hz), 3.94-3.82 (m, 3H), 3.08-2.84 (m, 4H), 2.72 (d, 1H, J = 10.6 Hz), 2.62 (d, 1H, J = 10.9 Hz), 2.58-2.51 (m, 1H), 2.5-2.42 (m, 1H), 2.3-2.04 (m, 2H), 1.93 (d, 1H, J = 12.8 Hz), 1.8 (d, 1H, J = 12.8 Hz), 1.66-1.5 (m, 4H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 210.6, 163.6, 159.1, 151.1, 138.8, 129.5, 120.7, 116.9, 114.6, 104.8, 67.6, 67.4, 60.7 (60.73), 60.7 (60.7), 50.1, 39.3, 35.5, 28.8, 28.2, 25.5, 20. HRMS (ESI) Calcd for $C_{23}H_{29}O_3N_2$: 381.2173 $(M+H)^+$. Found: 381.2177. Removing traces of solvent from the viscous oil precluded obtaining a satisfactory combustion analysis of **17**. An oxime derivative of **17** was prepared using 110 mg of **17**, hydroxylamine hydrochloride (30 mg, 0.4 mmole), and sodium acetate (39 mg, 0.5 mmol) in ethanol to afford 83 mg (72%) of a hygroscopic solid as mixture of *syn/anti*-isomers: mp 62-70°C (recrystallized from diethyl ether-hexanes). HRMS (ESI) Calcd for $C_{23}H_{30}O_3N_3$: 396.2282 $(M+H)^+$. Found: 396.2277. Anal. Calcd. for $C_{23}H_{29}O_3N_3 \cdot H_2O$: C, 66.81; H, 7.56; N, 10.16. Found: C, 66.93; H, 7.28; N, 10.07.

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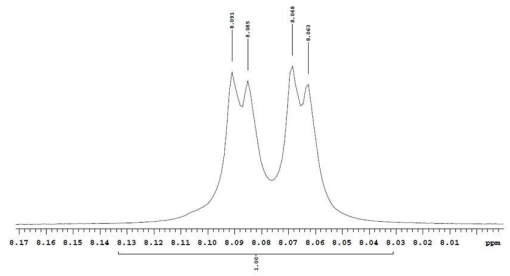
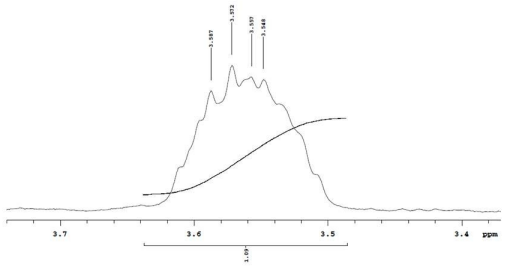
Supplemental Figures

A



B



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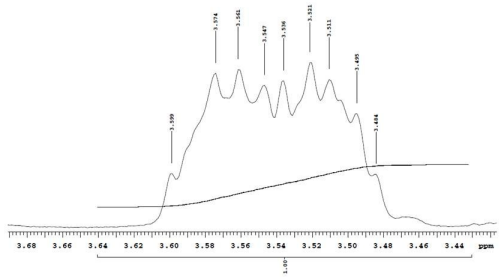

Fig. S1. A. ^{1}H NMR ($CDCl_3$) of C-2′ H (*i.e.*, RC<u>H</u>(OH)R′) in **13d**. B. ^{1}H NMR ($CDCl_3$) of C-5 ArH of isoflavonoid in **13d**. ^{1}H NMR ($CDCl_3$) of C-2′ H (*i.e.*, RC<u>H</u>(OH)R′) in **16**.

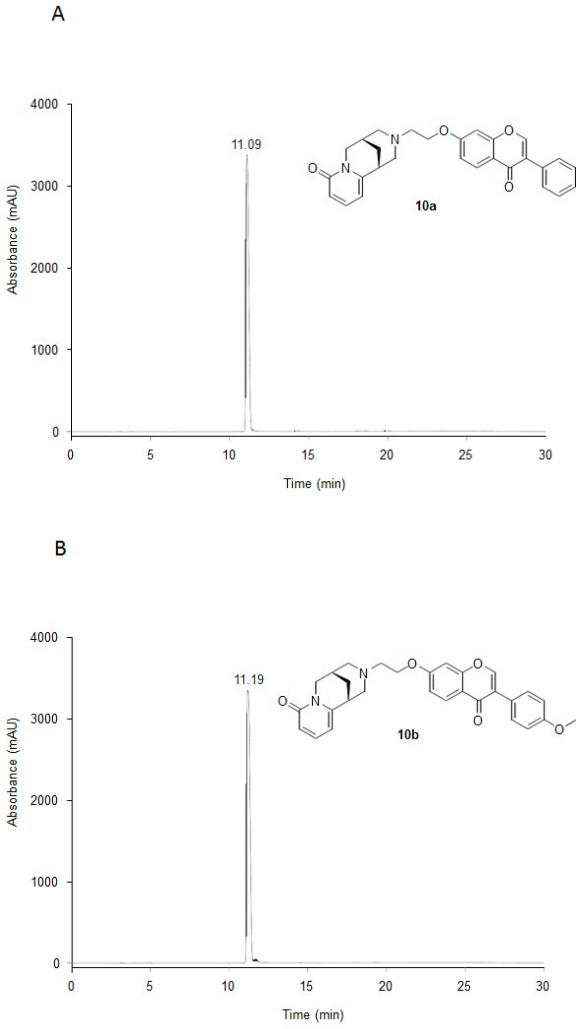

Fig. S2. A. HPLC trace for compound **10a**. R_t = 11.09 min.
B. HPLC trace for compound **10b**. R_t = 11.19 min.

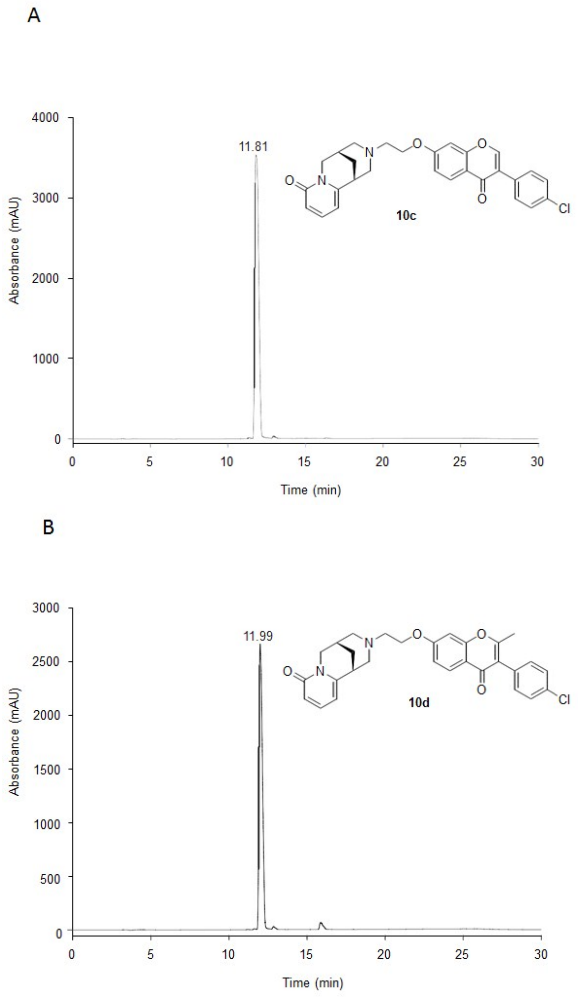

Fig. S3. A. HPLC trace for compound **10c**. R_t = 11.81 min.
B. HPLC trace for compound **10d**. R_t = 11.99 min.

		Normal Cells		Cancer Cells	
		BEAS-2B	**HEL 299**	**LS174T**	**PC-3**
		inhibition (%)	inhibition (%)	inhibition (%)	inhibition (%)
Cytinsine	10μM	0 ± 5.1	0 ± 5.6	13.9 ± 0.6	13.4 ± 7.6
10c	1μM	14 ± 4.0	15.9 ± 4.4	54.5 ± 2.7	33.9 ± 2.2
	10μM	23.3 ± 0.8	32.5 ± 1.3	88.4 ± 0.7	63.7 ± 3.7
Doxorubicin	1μM	98.2 ± 0.1	95.8 ± 1.3	98.2 ± 0.0	97.3 ± 1.2
	10μM	98.5 ± 0.1	94.3 ± 0.5	99.9 ± 0.1	99.5 ± 0.0
Erlotinib	1μM	8.5 ± 7.4	11.6 ± 1.9	33.2 ± 5.0	10.5 ± 2.7
	10μM	25.5 ± 9.2	73.5 ± 2.4	74.4 ± 2.6	51 ± 3.2
5-FU	1μM	0 ± 5.4	16.7 ± 1.8	22.4 ± 10.3	36.3 ± 11.5
	10μM	64 ± 2.1	75.3 ± 6.7	95.5 ± 1.1	70.3 ± 1.0
Sorafenib	1μM	0 ± 5.4	10.5 ± 1.9	26 ± 14.5	31.9 ± 5.1
	10μM	90 ± 1.7	97.8 ± 0.1	99.2 ± 0.0	96.6 ± 1.5

Table. S1. Effects of cytisine, **10c** and selected FDA-approved anticancer drugs on the proliferation of normal cells and cancer cells.