Supporting Information

Solid phase combinatorial synthesis of xanthone library using click chemistry and its application to embryonic stem cell probe

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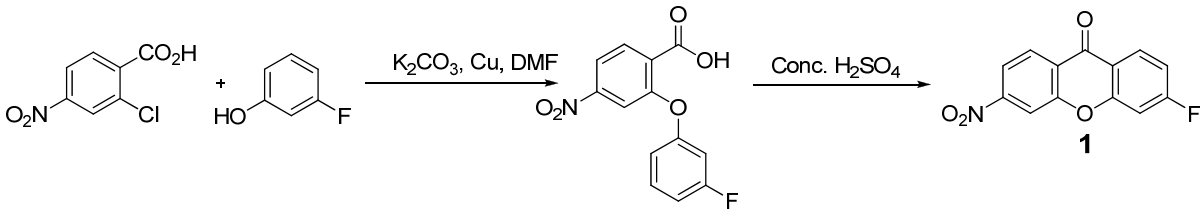
List of Information

Synthetic Materials and Methods

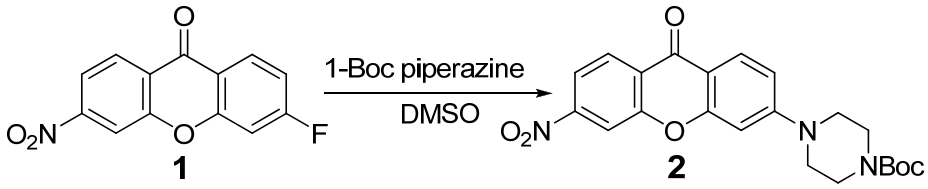
All the chemicals including entire alkyne building blocks and solvents were purchased from Sigma Aldrich, Alfa Aesar, Fluka, MERCK or Across, and used without further purification. 2-Chlorotrityl alcohol resin (1.37 mmol/g) was purchased from BeadTech Inc., Korea. All library compounds were characterised by HPLC-MS (Agilent-1200 series) with a DAD detector and a single quadrupole mass spectrometer (6130 series) with an ESI probe. Unless indicated the analytical method: eluents: A: H2O (0.1% HCOOH), B: ACN (0.1% HCOOH), gradient from 5 to 95%B in 7 min; C18(2) Luna column (4.6 x 50mm, 5um particle size). ^{1}H NMR spectra were recorded on a Bruker Avance 300 NMR spectrometer. Spectroscopic measurements were done in BioTek microplate reader or SpectraMax M2 spectrophotometer (Molecular Devices). All the spectroscopic measurements were done in DMSO solutions and coumarin1 (quantum yield=0.59) was used as a reference for quantum yield calculations.

Synthesis of 3-fluoro-6-nitro-9H-xanthen-9-one (1)



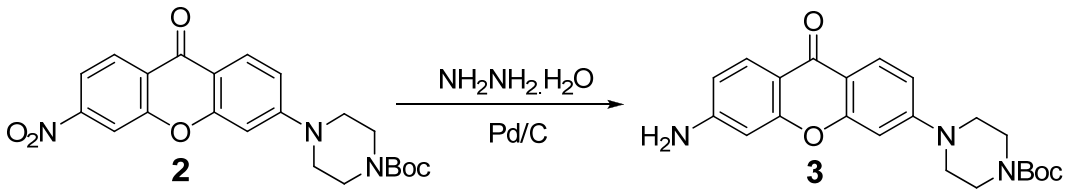
To a solution of 2-chloro-4-nitrobenzoic acid (3.0 g, 14.88 mmol) in DMF (40 mL) was added 3- fluorophenol (2.47 g, 16.38 mmol), potassium carbonate (3.08 g, 16.38 mmol) and copper Powder (102 mg, 1.61 mmol). After heating at 130 ^{o}C overnight the reaction mixture was then filtered through celite and washed with DMF. Once evaporating the DMF, 1 N HCl at 0 ^{o}C was added to it. The solution was stirred until the brown solid was formed. The solid was filtered off and washed with cold water to yield a brown solid (3.1 g). A crude solid was dissolved in concentrated sulfuric acid (20 mL) and heated at 90 ^{o}C for 1 h. After cooling to room temperature, the reaction mixture was poured to ice (350 mL volume) and stirred for one hour. Then the solution was filtered and the precipitate was dried. ^{1}H-NMR (300 MHz, $CDCl_3$): δ 8.50 (d, 1H, *J*=8.7 Hz), 8.37 (m, 2H), 8.20 (dd, 1H, *J*=8.7, 2.1 Hz), 7.21 (m, 2H) ESI *m/z* ($C_{13}H_6FNO_4$) calc: 259.03; found: 260.0

Synthesis of tert-butyl 4-(6-nitro-9-oxo-9H-xanthen-3-yl)piperazine-1 carboxylate (2)



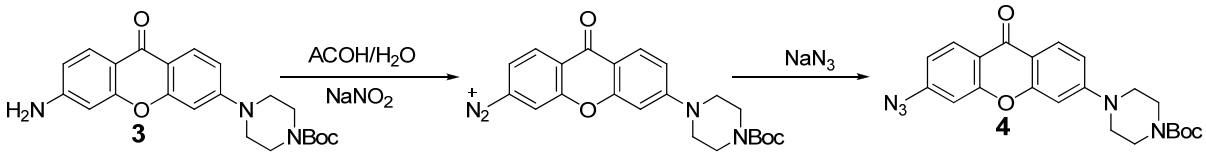
To a solution of 6-fluoro-3-Nitro-9H-Xanthone (**1**) in (2 g, 7.72 mmol) in DMSO (23 mL) 1-Boc piperazine (3.59 g, 19.3 mmol) was added. After heated at 90 ^{o}C for overnight the solution was diluted with EtOAc and washed with water and brine. The organic layer was collected and the solvents were removed under reduced pressure and the residue was purified by flash column chromatography (30 to 40% EtOAc/Hexane). ^{1}H-NMR (300 MHz, $CDCl_3$) δ 8.46 (d, 1H, *J*=8.7 Hz), 8.29 (d, 1H, *J*=2.0 Hz), 8.18 (d, 1H, *J*=9.0 Hz), 8.13 (dd, 1H, *J*=8.7, 2.1 Hz), 6.94 (dd, 1H, *J*=9.0, 2.1 Hz), 6.73 (d, 1H, *J*=2.1 Hz), 3.64 (t, 4H, *J*=4.8 Hz), 3.48 (t, 4H, *J*=4.8 Hz), 1.50 (s, 9H).^{13}C-NMR (300MHz, $CDCl_3$) δ174.1, 158.5, 155.7, 155.5, 154.5, 150.7, 128.3, 126.1, 117.8, 113.7, 113.2, 112.3, 99.5, 80.4, 46.8, 28.3. ESI *m/z* ($C_{22}H_{23}N_3O_6$) calc: 425.16; found: 426.0

Synthesis of tert-butyl 4-(6-amino-9-oxo-9H-xanthen-3-yl)piperazine-1-carboxylate (3)



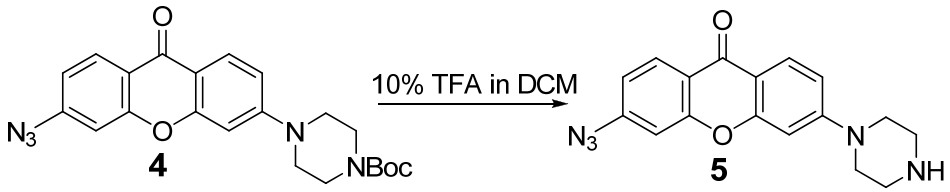
To a solution of **2** was added $NH_2NH_2.H_2O$ (1.76 g, 32.5 mmol) and Pd/C (300 mg, 20% of the compound) and heated at 90 ^oC for 4 hour .The hot solution was filtered through celite and the solvents were removed under reduced pressure. ^{1}H-NMR (300 MHz, $CDCl_3$): δ 8.12 (d, 1H, *J*=9.0 Hz), 8.07 (d, 1H, *J*=9.0 Hz), 6.83 (dd, 1H, *J*=9.0, 2.4 Hz), 6.64 (d, 1H, *J*=2.4 Hz), 6.60 (dd, 1H, *J*=9.0, 2.1 Hz), 6.51(d, 1H, *J*= 2.1 Hz), 3.59 (t, 4H, *J*=4.8 Hz), 3.35 (t, 4H, *J*=4.8 Hz), 1.48 (s, 9H). ^{13}C NMR (300 MHz, $CDCl_3$): δ 175.0, 158.2, 157.9, 154.8, 154.6, 152.3, 128.2, 127.7, 113.9, 112.0, 111.5, 100.3, 99.9, 80.2, 47.3, 28.4. ESI *m/z* ($C_{22}H_{25}N_3O_4$) calc: 395.18; found: 396.0

Synthesis of tert-butyl 4-(6-azido-9-oxo-9H-xanthen-3-yl) piperazine-1-carboxylate (4)



To a solution of tert-butyl 4-(6-amino-9-oxo-9H-xanthen-3-yl) piperazine-1-carboxylate (1 g, 2.53 mmol) in AcOH/H_2O (1:1) $NaNO_2$ (209.6 mg, 3.03 mmol) at 0 ^oC was added and stir it for 1 hour. The diazonium salt obtained was filtered and to the filtrate at 0 ^oC NaN_3 (246.7 mg, 3.8 mmol) was added and stir it for one and half hour. The solution was then neutralised with saturated $NaHCO_3$ and extracted with DCM. After evaporating the solvents the residue was subjected to flash column chromatography (30 to 40% EtOAc in Hexane). ^{1}H-NMR (300 MHz, $CDCl_3$): δ 8.24 (d, 1H, *J*=9.0 Hz), 8.12 (d, 1H, *J*=9.0 Hz), 8.96 (m, 2H), 8.87 (dd, 1H, *J*=9, 2.4), 6.65 (d, 1H, *J*=2.4 Hz), 3.60 (t, 4H, *J*=4.5 Hz), 3.40 (t, 4H, *J*=4.5 Hz), 1.48 (s, 9H). ^{13}C-NMR (300MHz, $CDCl_3$): δ 175.5, 158.7, 157.7, 156.0, 155.3, 146.5, 129.2, 128.7, 119.9, 115.7, 114.1, 112.5, 107.6, 100.56, 81.0, 47.7, 29.1 ESI *m/z* ($C_{22}H_{23}N_5O_4$) calc: 421.18; found: 422.0

Synthesis of 3-azido-6-(piperazin-1-yl)-9H-xanthen-9-one (5)

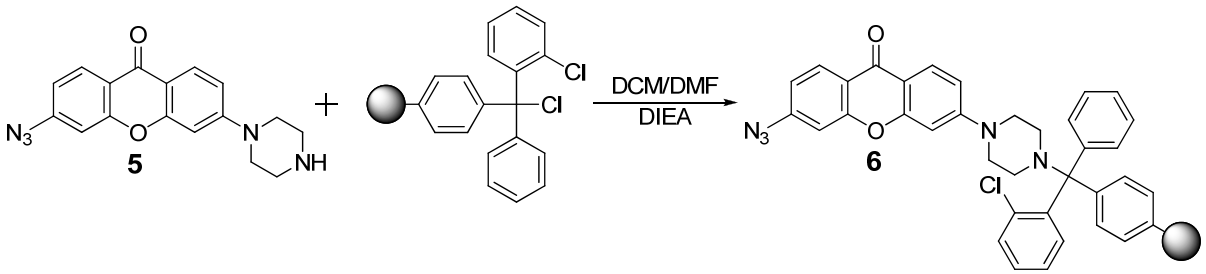


500 mg (1.2 mmol) of compound **4** was added to 25 mL of 10% TFA in DCM and stir it at room temperature for one hour. The solution was then evaporated several times with DCM. The compound used in the next step without further purifications. ESI *m/z* ($C_{17}H_{15}N_5O_2$) calc: 321.12; found: 322.0

Preparation of 2-chlorotrityl chloride from 2-chlorotrityl alcohol resin

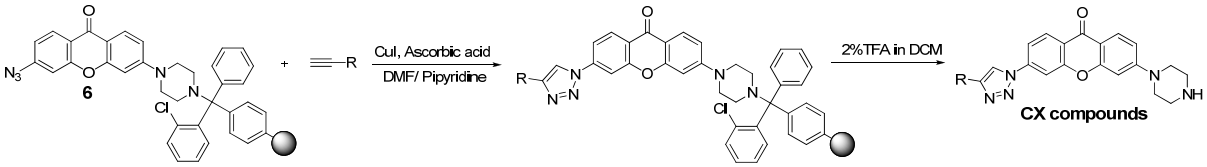
2-Chlorotrityl alcohol resin (2 g 1.37 mmol/g) was suspended in dichloromethane (20 mL) for 10 min. Thionyl chloride (600 µL, 8.24 mmol) was added and the resin solution was shaken for 5 hour at room temperature. The resin was filtered and washed with DMF and DCM then dried in high vacuum.

General procedure for loading 3-azido-6-(piperazin-1-yl)-9H-xanthen-9-one to solid resin (6)



1.2 mmol of **5** was dissolved in 10 mL DMF/DCM (9:1) and DIEA was added to it. The solution was then added to 2-chlorotrityl chloride resin (0.6 mmol) suspended in dichloromethane (2 mL). After stirring for 12 h, the resin was filtered through 10 mL cartridge and washed with DMF (X5), MeOH (X10), and DCM (X10). The resin was then shaken with 20% MeOH in DMF for 2 hour. The resin again washed with DMF (X5), methanol (X5), and dichloromethane (X5) and dried using high vacuum for 2hour. The loading was 80%.

General Procedure for CX Library Synthesis on Solid Support

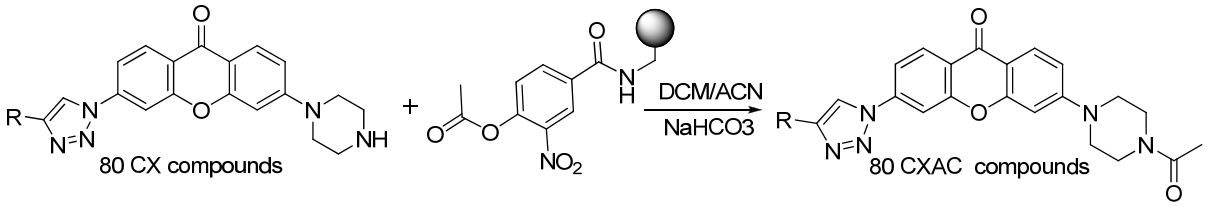


For each reaction, a resin (60 mg, 0.048 mmol) was suspended in 2 mL of DMF/ Pipyridine (4:1) in a 10 mL of syringe. 5 eq of CuI (0.24 mmol) and 5 eq (0.24 mmol) of ascorbic acid was then dissolve in 2 mL of the same solvent and added to the resin along with the 5 eq (0.24 mmol) of alkynes. The reaction mixture was shaken for overnight at room temperature and the resin was filtered through 10 mL cartridge and washed with DMF, 1% Sodium diethylthiocarbamate in DMF, 1% DIEA in DMF, 10% H_20 in DMF. Finally the resin washed with DMF (X5), MeOH (X5), and DCM (X5). The resin was dried and treated with 2% TFA in DCM (5 mL) for 10 min. The solution was drained to the 20mL vial, and dried using Speed Vacuum.

Synthesis of active ester p-nitrophenol resin

The nitrophenol resin was synthesized using previously reported procedure[1]. This nitrophenol resin was subsequently treated with acetyl chloride at room temperature with continuous shaking for two hours. The resin was then washed with DCM (X5) and dried under vacuum to obtain the active ester resin.

General procedure of synthesis of CXAC



20mg of active ester resin was suspended in 400 ul of DCM/ACN (7:1). 1 umol of each CX compound was also dissolved in 200 ul of same solvent and added to the resin shake it at room temperature. After 3 hour the solution was filtered and dried it in fume hood.

General protocol for spectral measurement of CX and CXAC compounds

CX and CXAC compounds spectral properties were measured in DMSO solvents. The CX compounds are excited at 360 nm wavelength and their emission spectrum are recorded from 390 nm to 600 nm. The CXAC compounds are excited at 365nm and their corresponding emission spectra are recorded from 410 nm to 540 nm. For quantum yield calculation we

integrated emission area of the fluorescent spectra and compared the value to the same area measured for **Coumarin 1** (φ=0.58) in DMSO. The quantum yields are calculated using the equation **1**, where *F* stands for area of fluorescent emission, η is reflective index of the solvent, and *Abs* is absorbance at excitation wavelength selected for standards and samples. Emission was integrated 390 nm to 600 nm for CX and for CXAC the integrated area was 410 nm to 540 nm.

$$\varphi_{flu}^{sample} = \varphi_{flu}^{reference}\left(\frac{F^{sample}}{F^{reference}}\right)\left(\frac{\eta^{sample}}{\eta^{reference}}\right)\left(\frac{Abs^{reference}}{Abs^{sample}}\right) \qquad \text{.....(1)}$$

Table S1. Characterization and photophysical properties of CX library

compound	M^+(calc.)	M^+ (exp.)	λ_{abs} (nm)	λ_{em} (nm)	$\varphi^{ref\ 2}$	purity
CX 1	403.48	404.0	372	489	0.03	90
CX 2	404.46	405.0	372	491	0.04	89
CX 3	417.5	418.0	372	491	0.04	90
CX 4	387.43	388.0	371	485	0.08	89
CX 5	415.49	416.0	372	489	0.03	95
CX 7	429.51	430.1	371	487	0.11	88
CX 8	443.54	444.0	372	488	0.03	90
CX 9	423.47	424.0	374	494	0.06	92
CX 10	437.49	438.0	372	497	0.06	93
CX 11	479.57	480.1.0	371	494	0.07	96
CX 12	479.57	480.0	371	489	0.05	92
CX 13	466.53	467.0	374	492	0.01	85
CX 14	483.52	484.0	372	490	0.05	91
CX 15	507.46	507.9	371	496	0.06	91
CX 16	459.45	459.9	371	493	0.03	92
CX 17	453.49	454.0	372	493	0.06	91
CX 18	480.56	481.0	372	487	0.03	92
CX 19	451.52	552.0	371	490	0.02	93
CX 20	515.56	516.0	371	491	0.05	94
CX 21	473.53	474.0	371	484	0.35	91
CX 22	503.55	504.0	366	490	0.11	92
CX 24	523.58	523.9.0	372	490	0.05	91
CX 25	451.52	452.0	370	489	0.02	91
CX 26	459.45	459.9	366	490	0.06	90
CX 27	453.49	454.0	370	491	0.05	91
CX 28	437.49	438.0	371	488	0.06	90
CX 29	493.6	494.0	367	494	0.06	91
CX 30	559.46	595.9	366	496	0.06	90
CX 31	441.46	442.0	370	496	0.06	92
CX 32	465.55	466.0	366	489	0.06	90
CX 33	467.52	468.0	368	488	0.04	90
CX 34	453.49	453.9	366	486	0.05	88
CX 35	441.46	442.0	371	492	0.09	94
CX 36	417.5	418.0	371	489	0.02	93
CX 37	427.5	427.9	372	489	0.07	92
CX 38	492.36	491.8	371	488	0.07	90
CX 39	453.48	453.9	371	492	0.06	92
CX 40	437.49	438.0	372	488	0.07	95
CX 41	417.5	418.0	372	489	0.05	90
CX 42	403.48	404.0	372	489	0.07	95
CX 43	403.48	404.1	372	484	0.03	90
CX 44	459.58	460.0	371	487	0.11	87
CX 45	437.49	438.0	372	493	0.05	90

CX 46	451.52	452.0	372	490	0.07	90
CX 47	431.53	432.1	371	487	0.09	91
CX 48	389.45	390.0	371	482	0.04	94
CX 49	487.64	488.0	371	490	0.13	91
CX 50	445.56	446.0	370	489	0.11	90
CX 51	515.69	516.0	366	489	0.14	91
CX 55	448.48	448.9	371	495	0.10	95
CX 56	427.46	428.0	371	493	0.08	96
CX 57	424.45	425.0	366	498	0.08	95
CX 58	444.53	445.1	372	488	0.08	98
CX 59	424.45	425.0	372	494	0.08	94
CX 60	468.46	469.0	338	451	0.08	88
CX 62	391.42	392.0	372	495	0.08	91
CX 66	419.55	420.0	372	487	0.08	91
CX 67	431.49	432.0	366	489	0.08	87
CX 68	453.49	453.9	371	486	0.08	90
CX 69	447.53	448.0	371	493	0.08	91
CX 70	377.4	378.0	361	483	0.13	90
CX 6	429.51	430.1	361	478	0.17	92
CX 73	433.58	434.0	371	485	0.05	91
CX 75	506.51	592.0	369	493	0.10	88
CX 76	429.51	430.1	371	490	0.04	86
CX 78	391.42	392.0	372	486	0.05	88
CX 79	433.5	434.1	371	487	0.10	91
CX 80	433.5	434.0	369	485	0.03	91
CX 82	419.48	420.0	371	494	0.02	90
CX 83	453.49	454.0	371	494	0.07	91
CX 85	467.49	468.0	372	491	0.07	94
CX 86	467.49	468.0	372	485	0.07	89
CX 88	419.48	420.1	368	482	0.06	88
CX 90	405.45	406.0	371	484	0.06	91
CX 91	449.5	450.0	370	485	0.04	86
CX 92	405.45	406.0	371	483	0.06	90
CX 95	405.45	406.0	371	485	0.10	91
CX 84	485.51	486.0	372	488	0.02	90
CX 97	491.46	491.8	370	499	0.12	90
CX 98	491.46	491.9	369	495	0.11	92

Table S2. Characterization and photophysical properties of CXAC library

compound	M^+(calc.)	M^+ (exp.)	λ_{abs}(nm)	λ_{em}(nm)	φ [ref 2]	purity
CXAC 1	445.21	446.3	369	483	0.3	98
CXAC 2	446.21	447.3	369	485	0.27	90
CXAC 3	459.23	460.2	369	483	0.3	98
CXAC 4	429.18	430.2	368	486	0.19	96
CXAC 5	457.21	458.2	369	484	0.3	98
CXAC 7	471.23	472.3	372	484	0.19	94
CXAC 8	485.24	486.2	369	483	0.31	98
CXAC 9	465.18	466.2	370	488	0.28	98
CXAC 10	479.2	480.2	369	487	0.25	97
CXAC 11	521.24	522.3	369	487	0.29	98
CXAC 12	521.24	522.3	369	487	0.3	98
CXAC 13	508.22	509.3	372	483	0.06	90
CXAC 14	525.2	526.2	369	486	0.27	98
CXAC 15	549.16	550.2	369	492	0.28	98
CXAC 16	501.16	509.2	365	468	0.21	91
CXAC 17	495.19	496.2	369	487	0.32	89
CXAC 18	522.24	523	369	484	0.34	90
CXAC 19	493.21	494.2	369	484	0.35	99
CXAC 20	557.21	558.2	369	485	0.31	97
CXAC 21	515.2	516.2	369	487	0.32	94
CXAC 22	545.21	546.1	368	467	0.27	90
CXAC 24	565.21	566.2	368	487	0.29	98
CXAC 25	493.21	494.1	368	485	0.32	96
CXAC 26	501.16	502.1	368	483	0.27	98
CXAC 27	495.19	496.2	368	487	0.33	93
CXAC 28	479.2	480.2	369	485	0.33	97
CXAC 29	535.26	536.3	368	485	0.38	94
CXAC 30	601.15	602.2	372	503	0.3	95
CXAC 31	483.17	484.2	369	488	0.31	89
CXAC 32	507.23	508.3	368	471	0.31	90
CXAC 33	509.21	510.2	368	483	0.34	93
CXAC 34	495.19	496.2	366	485	0.31	89
CXAC 35	483.17	484.2	369	492	0.3	98
CXAC 36	459.23	460.3	368	482	0.4	98
CXAC 37	469.21	470.2	368	486	0.44	96
CXAC 38	533.1	533.9	368	488	0.4	90
CXAC 39	495.19	496.2	369	486	0.44	98
CXAC 40	479.2	480.2	369	486	0.43	98
CXAC 41	459.23	460.2	369	485	0.43	98
CXAC 42	445.21	446.3	369	482	0.47	97
CXAC 43	445.21	446.2	369	482	0.42	97
CXAC 44	501.27	502.3	369	489	0.4	98

CXAC 45	479.2	480.2	372	486	0.46	97
CXAC 46	493.21	494.2	368	485	0.4	97
CXAC 47	473.24	474.3	368	485	0.47	98
CXAC 48	431.2	432.2	369	483	0.41	96
CXAC 49	529.31	530.3	368	483	0.53	97
CXAC 50	487.59	488.3	369	482	0.43	98
CXAC 51	557.34	558.3	368	485	0.46	97
CXAC 55	490.18	491.2	369	496	0.61	92
CXAC 56	469.19	470.2	369	487	0.54	90
CXAC 57	466.18	467.2	368	494	0.47	89
CXAC 58	486.24	486.9	369	488	0.41	88
CXAC 59	466.18	467.2	369	487	0.39	94
CXAC 60	510.17	511.2	338	463	0.29	87
CXAC 62	433.18	434.2	369	488	0.43	94
CXAC 66	461.19	462.2	368	484	0.54	97
CXAC 67	473.21	474.2	372	479	0.08	85
CXAC 68	495.19	496.2	372	485	0.7	90
CXAC 69	489.24	490.3	369	483	0.48	88
CXAC 70	419.16	420.3	366	469	0.17	85
CXAC 6	471.23	472.1	369	484	0.44	85
CXAC 73	475.2	476.2	369	481	0.48	90
CXAC 75	448.18	449	368	480	0.35	87
CXAC 76	471.23	472.3	368	479	0.45	90
CXAC 78	433.18	434.2	369	483	0.58	96
CXAC 79	475.22	476.3	368	484	0.56	98
CXAC 80	475.22	476.2	369	482	0.54	91
CXAC 82	461.21	462.2	369	484	0.57	96
CXAC 83	495.19	496.2	369	487	0.34	84
CXAC 85	509.19	510.2	369	487	0.38	91
CXAC 86	509.19	510.2	369	485	0.32	93
CXAC 88	461.21	462.2	368	473	0.41	89
CXAC 90	447.19	448	368	483	0.47	87
CXAC 91	491.2	492.2	368	478	0.41	92
CXAC 92	447.19	448.2	368	485	0.41	96
CXAC 95	447.19	448.2	368	482	0.49	96
CXAC 84	527.2	528.2	365	473	0.3	86
CXAC 97	533.17	534.1	369	491	0.52	91
CXAC 98	533.17	534.1	372	493	0.44	93

For HPLC-MS characterisation of CXAC Library following analytical method was used Eluents: A: H2O (0.1% HCOOH), B: ACN (0.1% HCOOH), gradient from 30 to 100% B in 2.5 min; C18(2) Luna column (4.6 x 50 mm, 5 um particle size)

General protocol for high throughput cell imaging

The 384 well plates were coated with gelatin for at least 1 hour. Then the appropriate amount of MEF (2.5 X 10^4/mL) and mESC (1 X 10^5/mL) were seeded into the wells. After one day, 1 uM concentration of compound was added to the cells and incubated for 1 hour. The image was then taken using ImageXpress machine with the DAPI filter. Transmitted light images were also taken simultaneously.

General protocol for flow cytometry measurement

1 uM concentration of compound was added to the cells and incubated for 1 hour. After which the staining pattern of the cells were checked before and after washing (3 times with PBS) using the fluorescent microscopy. The cells were trypisinized using Trypsin, 0.25% (1X) with EDTA and transferred to 5ml polystyrene round bottom tube (Falcon®). The pellet obtained were then resuspended and washed with cold PBS (3 times).

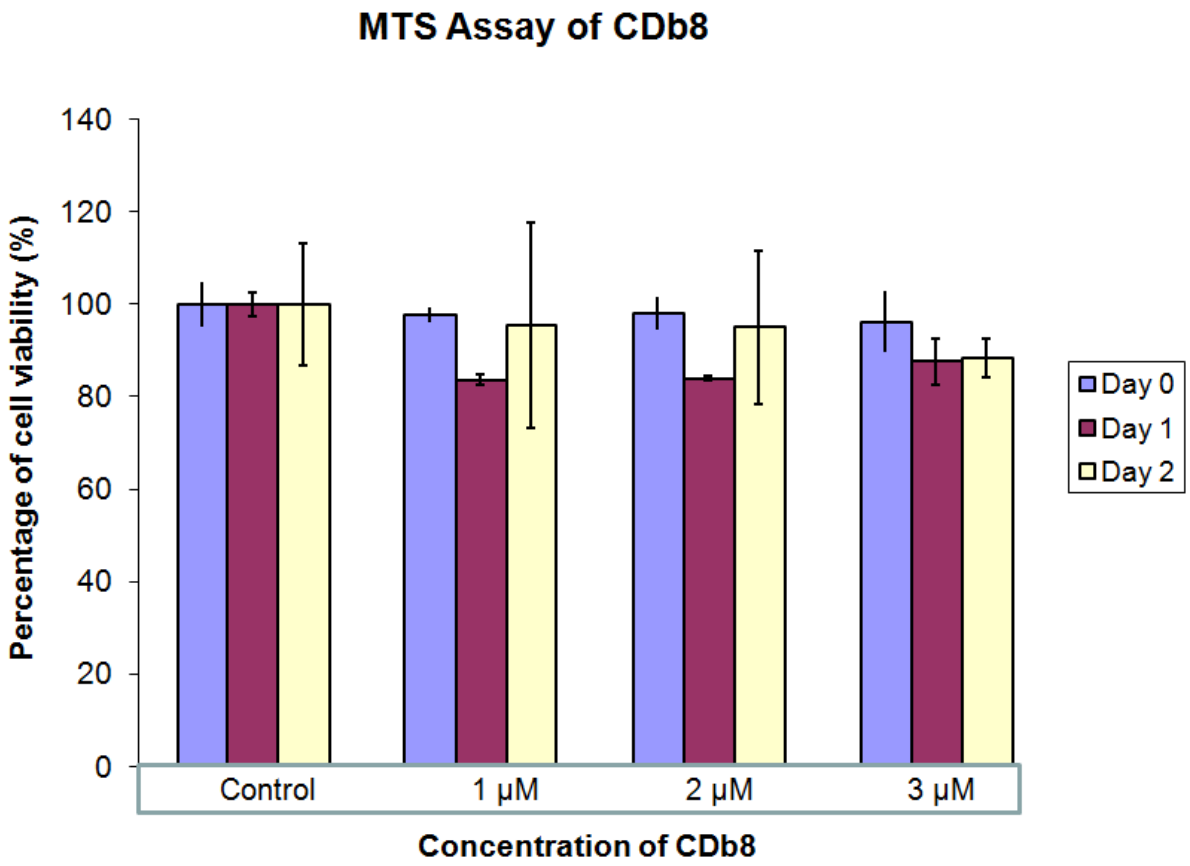


Figure S1. Cytotoxic effect of **CDb8** (CXAC-F5) in mESC**.** The cytotoxic effect of **CDb8** was tested by the **MTS** (3-(4,5-dimethylthiazol-2-yl)-5-(3-carboxymethoxyphenyl)-2-(4-sulfophenyl)-2H-tetrazolium, inner salt) assay using the CellTiter 96 nonradioactive cell proliferation colorimetric assay kit (Promega) on mESC cells. First, the mESC cells (1 X 10^4 cells in 100 µl of media) were seeded onto 96 well plate and then different concentrations (0 µM, 1 µM, 2 µM and 3 µM) of **CDb8** was added to the cells on the following day and incubated them at 37^{o}C for 3days. After one day(Day 0), 20 µl of MTS solution was added to each well and incubated for another 2 hours before the absorbance was measured at 490 nm. The same experiments were done for Day 1 and Day 2. The control cells are 100% alive without compound condition. At least 80% of the **CDb8** treated cells are alive after 3 dyas of incubation at 3 µM concentration. This indicates that the **CDb8** is nontoxic to mESC.

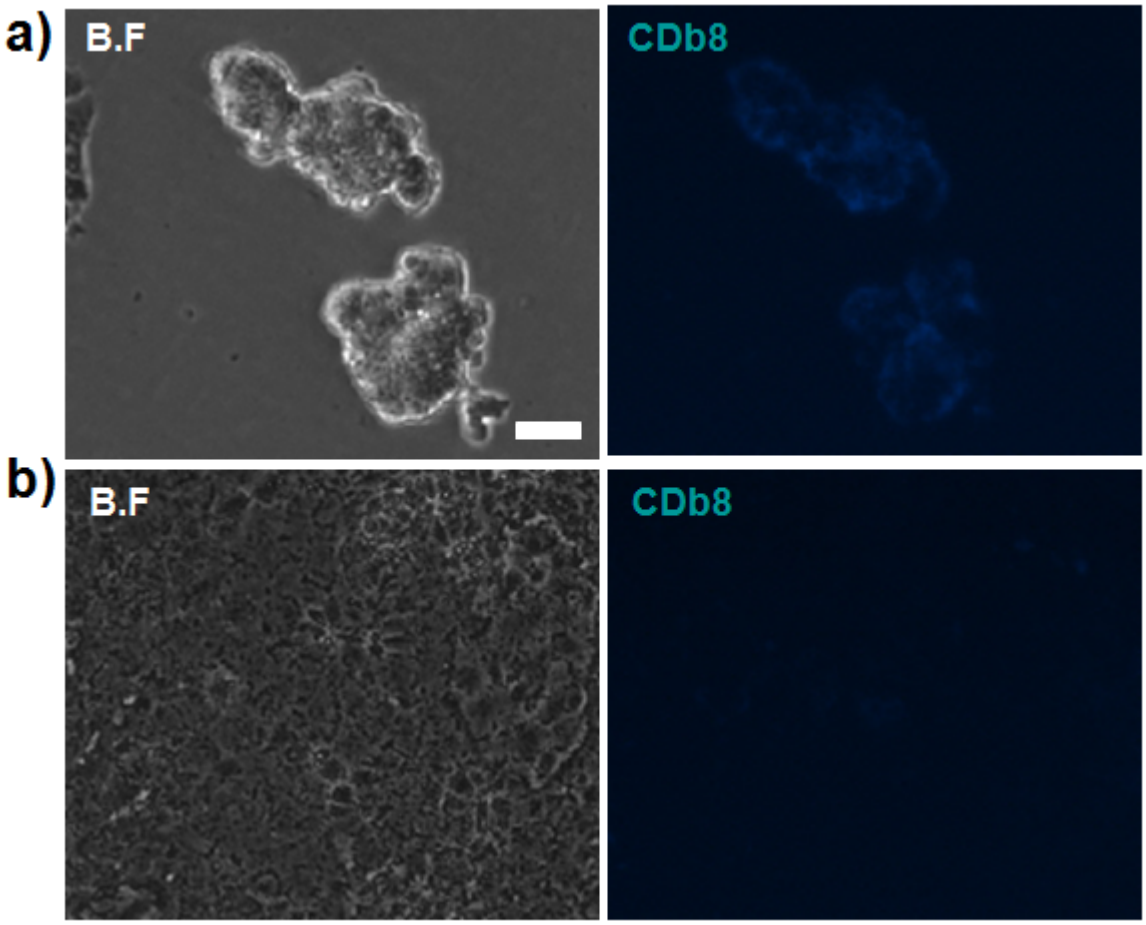


Figure S2. Differentiated mESC staining with **CDb8**. All of differentiated mESC were distinguished from mESC 14days after removing LIF culture. Almost all differentiated cells were **CDb8** negative. a) mESC selectively stained by **CDb8** b) The differentiated mESC were not stained by **CDb8.** B.F, bright field. Scale bar, 100 μm.

HPLC-MS characterisation of CDb8

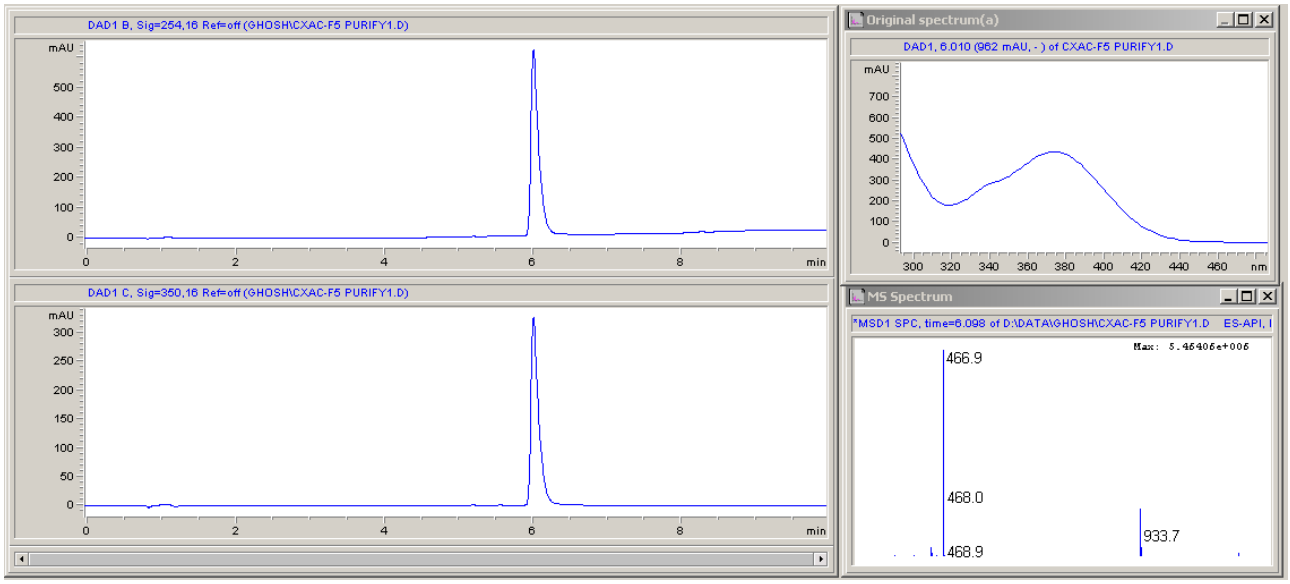
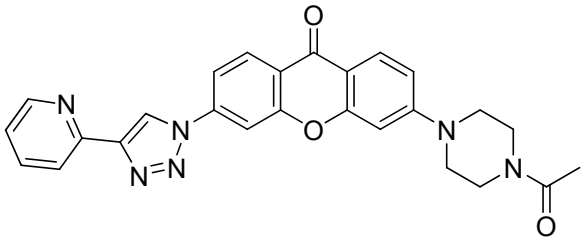


Figure S3. HPLC-MS characterization of CDb8**.** chromatograms (*descending order*) at 254 nm, 350 nm . Spectra profile (300-500 nm). ESI-MS positive spectra. HPLC conditions: A: H_2O-HCOOH: 99.9:0.1. B: ACN-HCOOH: 99.9:0.1; gradient 5% B to 95% B (7 min), isocratic 95% B (1.5 min). Reverse-phase Phenomenex C_{18} Luna column (4.6 x 50 mm^2) 3.5 µm, flow rate: 0.8 mL/min.

^{1}H-NMR (300 MHz, $CDCl_3$): δ 8.76 (1H, *s*), 8.64 (1H, dd, *J*= 2.7, 1.8 Hz), 8.47 (1H, dd, *J*= 8.7, 1.8 Hz), 8.28 (1H, d, *J*= 8.1 Hz), 8.20 (1H, dd, *J*= 9, 2.7 Hz), 7.98 (1H, dd, *J* = 2.7, 2.1 Hz), 7.85 (1H, td, *J*= 8.7, 1.8 Hz), 7.78 (1H, dd, *J*= 8.7, 2.4 Hz), 7.30 (1H, m), 6.94 (1H, dd, *J* = 9, 2.4 Hz), 6.77 (1H, d, *J*= 2.4 Hz), 3.83 (1H, m), 3.70 (1H, m), 3.50 (1H, m) & 2.17 (3H, s). ESI *m/z* ($C_{26}H_{22}N_6O_3$) calc: 466.18; found: 466.9

Structure of **CDb8**:



References:

1. J. W. Lee, Y. Q. Louie, D. P. Walsh and Y. T. Chang, *J. Comb. Chem.*, 2003, **5**, 330-335.

2. J. S. Lee, N. Y. Kang, Y. K. Kim, A. Samanta, S. Feng, H. K. Kim, M. Vendrell, J. H. Park and Y. T. Chang, *J Am Chem Soc*, 2009, **131**, 10077-10082