Supporting Information

Solid Phase Synthesis of Ultra-Photostable Cyanine NIR dye library

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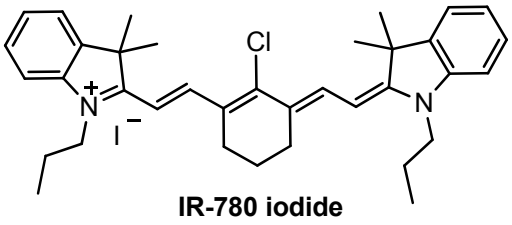
Materials and Methods

Amine building block and all other chemicals and solvents for the synthesis were purchased from the Alfa Aesar, Fluka, Acros, MERCK, and Sigma Aldrich and were used without any purification. Merck Silica Gel 60 (particle size: 0.04-0.063 mm, 230-400 mesh) was used for the normal phase column chromatographic purification. From BeadTech Inc., Korea, 2-chlorotrityl alcohol resin (1.37mmol/g) was purchased. All the, **CyN**, **CyNA** derivatives which were previously synthesized by our group were used for the comparative photostability study with **CyR** derivatives. For analytical characterization of **CyR** compounds HPLC-MS (Agilent-1200 series) with a DAD detector and a single quadrupole mass spectrometer (6130 series) with an ESI probe was routinely used. Analytical process, except specified: eluents: A: H_2O (0.1% HCOOH), B: ACN (0.1% HCOOH), gradient from 0 to 100% B in 4 min; C_{18} (2) Luna column (4.6 x 50mm^2, 5µm particle size) was used. ^{1}H-NMR and ^{13}C-NMR spectra were recorded on both Bruker Avance 300 MHz and 500 MHz NMR spectrometer, and chemical shifts are expressed in parts per million (ppm) and approximate coupling constants were calculated in Hz. Quantum yields and all other photophysical properties, photostability evaluation study of **CyN**, **CyR** and **CyNA** derivatives were performed in SpectraMax M2 spectrophotometer (Molecular Devices) instrument and the obtained data were analyzed using the Microsoft Office Excel 2007. In order to employ the strong UV irradiation to the

compounds of interest, UVP Blak-Ray® B-100AP high intensity mercury lamp (100W, 365 nm) at 2 cm distance was used.

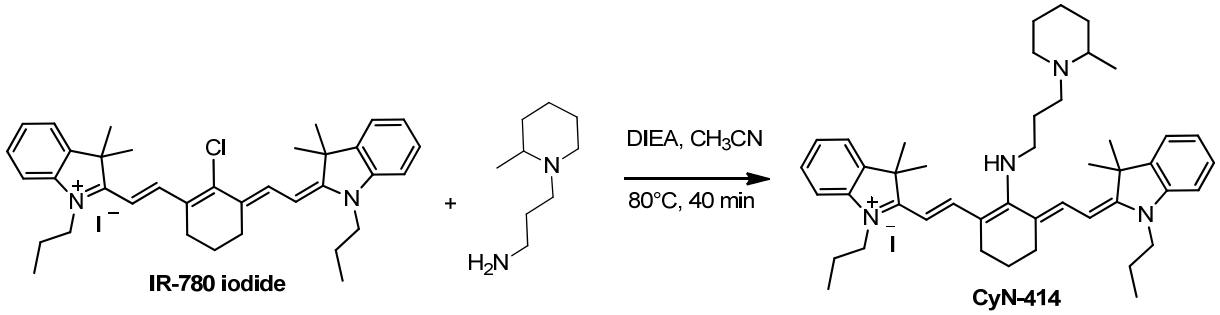
Synthesis of IR-780 iodide

For the synthesis of compound **IR-780 iodide**, we followed the reported procedure[3].



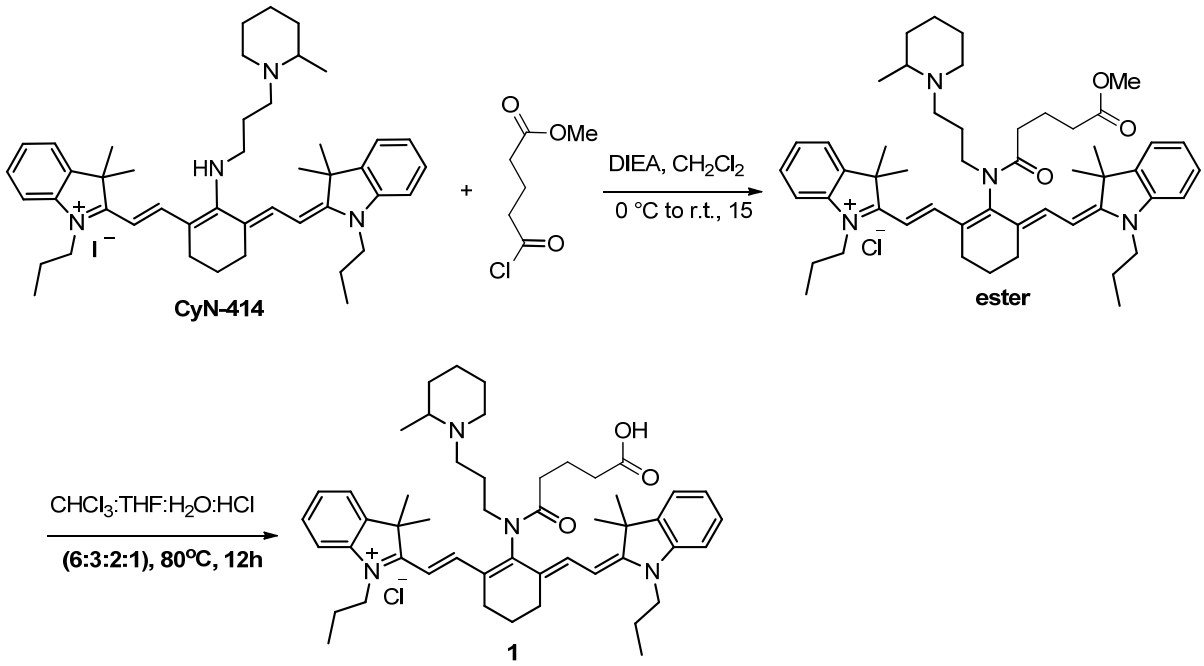
IR-780 iodide

Synthesis of CyN-414



IR-780 iodide (600 mg, 0.9 mmol, 1 eq.) and 1-(3-aminopropyl)-2-pipecoline (340 mg, 1.8 mmol, 2 eq.) were dissolved in ACN (2 mL), and N,N-diisopropylethylamine (DIEA) (174 µL, 1.34 mmol, 1.5 eq.) was added. The reaction mixture was heated to 80° C for 40 min, and the resulting blue color crude **CyN-414** was neutralized with 0.1 N HCl (aq.) and concentrated under vacuum. The crude product obtained was directly used for the next step synthesis of **1**.

Synthesis of 1



The crude compound **CyN-414** was dissolved in DCM under N_2 atmosphere, and treated with excess DIEA (1.4 mL, 10.8 mmol, 12 eq.) and methyl 4-(chloroformyl)butyrate (220 µL, 1.34 mmol, 1.5 eq.) at 0^o C for 15 min. The resulting green product (ester) was washed with 0.1 (N) HCl and brine, concentrated under vacuum. The obtained crude product (ester) was directly employed for next step synthesis.

The crude product (ester) was dissolved in solvent mixture of $CHCl_3$:THF:H_2O:HCl(conc) (6:3:2:1)(v/v/v/v) (90 mL) . The resulting mixture was stirred at 0^o C. After 5 min., the reaction mixture was refluxed at 80^o C for 12 h, and monitored by LC-MS. After complete hydrolysis of the methyl ester, $CHCl_3$ was added to the reaction mixture, and the organic layer was collected (3 x 40 mL), washed with water and purified by a normal-phase silica column using DCM-MeOH (ranging from 100:0 to 88:12) as the eluting solvent.

Characterization data for **1** (140 mg, 20% from **IR-780 iodide**)

Characterization of Compound 1

^{1}H-NMR (300 MHz, $CDCl_3$): 1.06 (t, 6H, J=7.5Hz), 1.24 (d, 3H, J=6.6 Hz), 1.39 (m, 2H), 1.61 (s, 6H), 1.62 (s, 6H), 1.79-1.95 (m, 6H), 2.22 (t, 2H, J=7.8 Hz), 2.33 (t, 2H, J=6.6 Hz), 2.52-2.56 (m, 4H), 2.82 (t, 2H, J=5.4 Hz), 2.87 (t, 2H, J=5.4 Hz), 2.96-2.98 (m, 2H), 3.09-3.12 (m, 1H), 3.36 (t, 4H), 3.53 (t, 2H, J=6.6 Hz), 3.67 (t, 2H, J=6.6 Hz), 4.06 (t, 2H, 4.2 Hz), 4.15 (t, 2H, J=4.8 Hz), 6.15 (d, 1H, J=14.1Hz), 6.20 (d, 1H, J=14.1Hz), 7.07-7.38 (m,8H), 7.51 (d, 1H, J=14.1Hz), 7.60 (d, 1H, J=14.1Hz).

^{13}C-NMR (75 MHz, CDCl3): 106.2, 110.6, 110.8, 114.9, 118.8, 122.3, 125.5, 125.6, 127.7, 128.1, 128.6, 140.6, 140.9, 141.4, 142.1, 142.2, 144.6, 153.9, 160.9, 161.4, 171.7, 172.5, 173.6, 174.3, 101.5, 101.9, 102.4, 11.6, 12.2, 19.5, 20.4, 20.6, 20.7, 22.2, 22.9, 24.8, 28.1, 28.2, 28.3, 31.3, 32.3, 41.9, 43.9, 48.3, 49.1, 49.3, 50.2, 51.8, 53.7, 60.4,

ESI (HRMS) m/z ($C_{50}H_{69}N_4O_3^+$), calc: 773.5364; found: 773.5351.

Synthesis of CyNA-414

The **CyNA-414** was synthesized according to the reported procedure.

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

Thionyl chloride (1.2 mL, 16.48 mmol, 3 eq) was added to 2-Chlorotrityl alcohol resin (4g, 1.37 mmol/g, 5.48 mmol, 1 eq) suspended in 40 mL of anhydrous DCM. Then the solution mixture was shaken for overnight at room temperature. Then the resin was filtered and washed thoroughly with DMF (3X 40 mL) followed by DCM (3X40 mL) and then the resin was dried in vacuum.

General procedure for loading of solid supported 3-bromopropylamine

(3.5 g, 15.75 mmol, 5 eq) 3-bromopropylamine was dissolved in THF(5 mL/g) and then(6 mL, 32.5 mmol, 10 eq) DIEA was added to the solution. The resulting solution was then added to 2-chlorotrityl chloride resin (2.5 g, 3.25 mmol, 1 eq, 1.3 mmol/g) suspended in dichloromethane (10 mL/g). After stirring for 12 h, the resin was filtered through 10 mL cartridge and washed with DMF (5X 40 mL), methanol (5X 40 mL), and dichloromethane (10X 40 mL). The resin was then shaken with 20% MeOH in DMF for 2 h for the capping of excess 2-chlorotrityl chloride resin. The resin obtained was again washed with DMF (5X 40 mL), methanol (5X 40 mL), and dichloromethane (5X 40 mL) and then was dried using high vacuum.

General procedure for synthesis of solid supported secondary amines

For each reaction, resin (solid supported 3-bromopropylamine) (100 mg, 0.1 mmol, 1 eq, 1 mmol/g) was suspended in 2 mL of N-Methylpyrrolidone (NMP) in a 20 mL of glass vial. 7 eq of each amine (0.7 mmol) and 14 eq of DIEA (1.4 mmol) were then added in 2 ml of the same solvent The reaction mixture was shaken for overnight at 70 ^{o}C temperature in the heat block and the resin was filtered through 10 mL cartridge and washed with DMF (5X 5 mL), methanol (5X 5mL), and dichloromethane (5X 5mL). The solid supported secondary amine resins obtained, were dried and used for next step reactions.

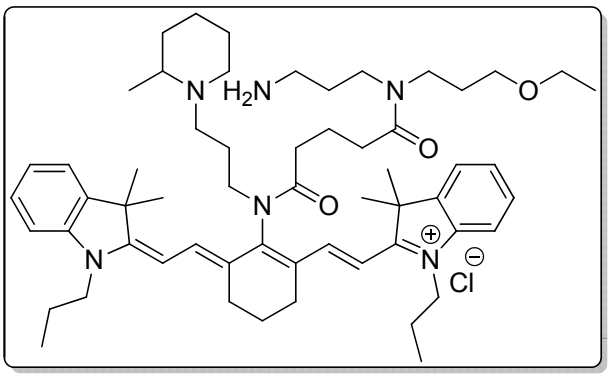
General procedure for synthesis of CyR library

To synthesize **CyR** library, each of (solid supported secondary amine) resin (50 mg, 0.035 mmol, 1eq, 0.7 mmol/g) was suspended in 3 mL of DMF in a 10 mL syringe then (28 mg, 0.07 mmol, 1 eq) **1** (30 mg, 0.77 mmol, 2.2 eq), 2-(7-Aza-1H-benzotriazole-1-yl)-1,1,3,3-tetramethyluronium hexafluorophosphate (HATU) and (30 µL, 0.168 mmol, 4.8 eq) DIEA were added. At a time 40 individual reaction mixtures were placed on orbital shaker for 24 h at room temperature and after complete the reaction the resin was filtered through 10 mL cartridge washed with DMF (5X 5mL), methanol (5X 5mL), and dichloromethane (5X 5mL). The resin was dried under high vacuum to afford solid supported compounds resin **2** and then the subsequently the dried resin of 50 mg was treated with 2% TFA in dichloromethane (5 mL) for 10 min. The solution was drained to the 20 mL vial and then organic layer was washed with the saturated $NaHCO_3$ solution, and then the organic layer was separated and dried using Speed Vacuum to afford the **CyR** library products. Each of **CyR** compound was

solid and primarily characterized by LC-MS. The representative products are characterized by LC-MS, HR-MS, ^{1}H-NMR, ^{13}CNMR.

Representative CyR compounds characterization

CyR 167 (14 mg, 60%)

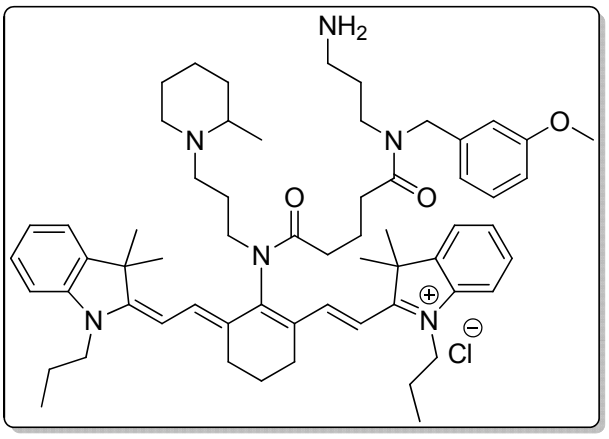


^{1}H-NMR (500 MHz, $CDCl_3$):1.03-1.64 (m, 18H), 1.61 (s, 6H), 1.64 (s, 6H), 1.70-1.89 (m, 6H), 2.05 (m, 2H), 2.19-2.34 (m, 5H), 2.62-2.87 (m, 11H), 3.38-3.60 (m, 4H), 3.67 (m, 2H), 3.69 (s, 3H), 3.8 (s, 2H), 3.99-4.06 (m, 6H), 6.15 (d, 1H, J=13.85 Hz), 6.17 (d, 1H, J=13.85 Hz), 6.6-6.89 (m, 4H), 7.07-7.37 (m, 8H), 7.54 (d, 1H, J=13.85 Hz), 7.57 (d, 1H, J=13.85)

^{13}C-NMR (75 MHz, CDCl3):11.60, 14.09, 15.02, 15.11, 20.41, 20.76, 22.66, 23.53, 24.74, 25.08, 27.70, 27.75, 27.77, 28.02, 28.70, 29.67, 31.93, 32.21, 36.87, 41.34, 44.80, 45.93, 48.61, 49.22, 49.26, 49.30, 66.11, 66.14, 66.84, 67.87, 101.76, 101.96, 110.48, 110.54, 115.64, 117.97, 122.67, 125.49, 125.58, 127.89, 128.52, 128.59, 140.87, 141.11, 141.23, 141.31, 141.94, 142.05, 142.09, 153.36, 153.39, 161.76, 162.03, 172.38, 172.63, 173.90, 175.16

ESI (HRMS) m/z ($C_{58}H_{87}N_6O_3^+$) calc: 915.6834; found: 915.6864.

CyR 387 (12 mg, 52%)

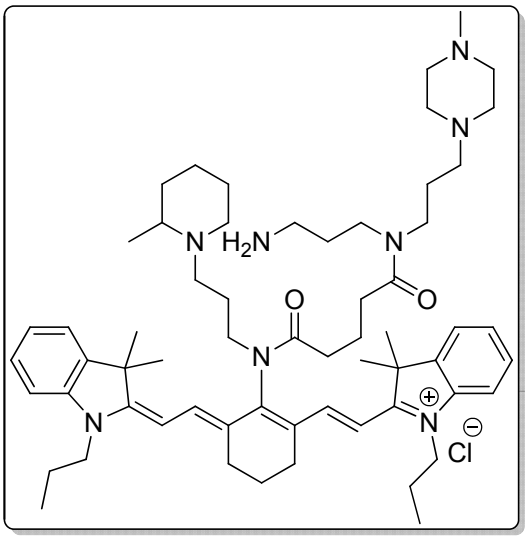


^{1}H-NMR (500 MHz, $CDCl_3$): 1.04-1.59 (m, 12H), 1.65 (s, 6H), 1.66 (s, 6H), 1.71-2.06 (m, 20H), 2.25-2.39 (m, 6H), 2.57-2.77 (m, 6H), 2.90-3.0 (m, 2H), 3.30-3.46 (m, 11H), 3.67 (m, 2H), 4.01 (m, 4H), 6.14 (d, 1H, J=13.9 Hz), 6.13 (d, 1H, J=13.85 Hz), 7.07-7.44 (m, 8H), 7.52 (d, 1H, J=13.85 Hz), 7.54 (d, 1H, J=13.25 Hz)

^{13}C-NMR (75 MHz, CDCl3): 11.51, 11.56, 20.58, 20.78, 21.02, 24.76, 28.03, 28.11, 28.13, 32.32, 32.64, 37.57, 42.33, 44.63, 44.98, 45.93, 47.97, 49.09, 49.13, 49.20, 49.26, 50.44,

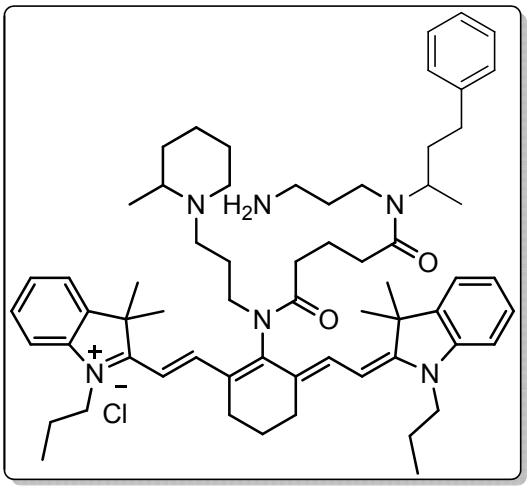
50.77, 51.46, 55.09, 55.17, 56.59, 59.79, 101.66, 101.91, 110.64, 110.74, 110.82, 11.94, 112.60, 112.79, 113.39, 114.47. 118.08, 118.26, 120.07, 121.38, 122.26, 122.45, 125.27, 125.36, 125.42, 125.51, 128.31, 128.66, 129.89, 138.07, 141.00, 141.25, 141.53, 142.22, 142.25, 150.30, 160.04, 161.69, 171.96, 172.16, 173.21, 174.38
ESI (HRMS) m/z ($C_{61}H_{85}N_6O_3^+$) calc: 949.6678; found: 915.6671.

CyR 526 (17 mg, 55%)



^{1}H-NMR (500 MHz, $CDCl_3$):0.88-1.77 (m, 16H), 1.24 (s, 6H), 1.20 (s, 6H), 2.2 (m, 7H), 2.61 (s, 3H), 2.98 (m, 10H), 3.17 (m, 5H), 3.32 (m, 8H), 3.45-3.65 (m, 11H), 3.98 (m, 2H), 4.21 (m, 1H), 5.35 (m, 2H), 6.42-7.13 (m, 12H)
^{13}C-NMR (125 MHz, CDCl3): 14.17, 14.23, 22.58, 22.60, 22.68, 26.40, 29.15, 29.20, 29.38, 29.41, 29.62, 29.69, 30.68, 31.73, 31.75, 31.92, 39.50, 39.68, 39.84, 40.01, 40.17, 40.34, 40.5, 59.00, 59.05, 59.13, 63.78, 64.06, 67.80, 67.93, 114.62, 114.83, 115.15, 123.61, 124.28, 126.35, 131.80, 132.21, 132.35, 133.68, 140.30, 140.46, 141.74, 141.83, 154.80, 155.01, 161.43, 161.72, 162.22, 162.35, 162.70, 168.13, 168.59
ESI (HRMS) m/z ($C_{61}H_{93}N_8O_2^+$) calc: 969.7416; found: 969.7410.

CyR 275 (9 mg, 61%)

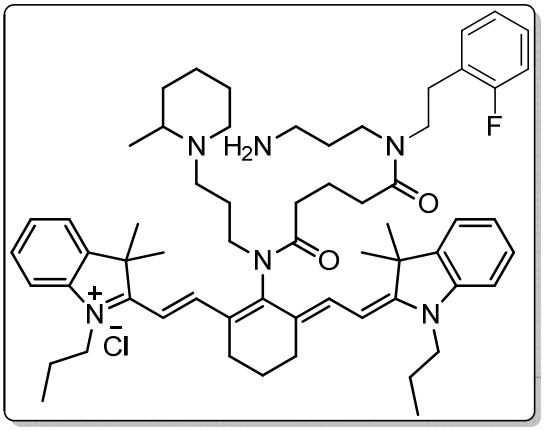


^{1}H-NMR (500 MHz, $CDCl_3$): 1.10 (t, 6H, J=4Hz), 1.18 (d, 3H, J= 5Hz), 1.2(d, 3H, J=8.5Hz), 1.33(m, 2H), 1.43-1.45(m, 2H), 1.66(s, 12H), 1.88-1.92(m, 6H), 2.02-2.04(m, 1H), 2.22(t, 2H, J=8Hz), 2.33 (t, 2H, J=5Hz), 2.51-2.55(m, 4H), 2.75 (t, 4H, J=5Hz), 3.12 (m, 2H), 3.22-

3.24 (m, 1H), 3.29 (t, 4H, J=5Hz), 3.68 (t, 2H, J=5Hz), 4.03 (t, 2H, J=4.7Hz), 4.33 (t, 2H, J=5Hz), 6.15 (d, 1H, J=14.5Hz), 6.34 (d, 1H, J=15Hz), 7.08-7.13 (m, 5H), 7.21-7.28 (m, 5H), 7.49 (d, 1H, J=14Hz). 7.55 (d, 1H, J=15 Hz).

ESI (HRMS) m/z ($C_{63}H_{89}N_6O_2^+$), calc: 961.7042; found 961.7057

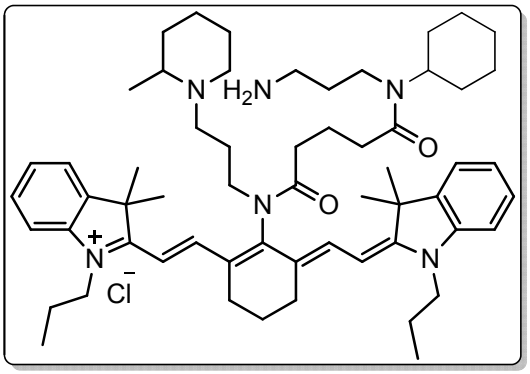
CyR 477 (13 mg, 56%)



^{1}H-NMR (500 MHz, $CDCl_3$): 0.89 (t, 6H, J=7Hz), 1.10 (d, 3H, J=5Hz), 1.28-1.34 (m, 2H), 1.64 (s, 12H), 1.828 (m, 2H), 1.91-1.94 (m, 2H), 2.04 (t, 2H, J= 6Hz), 2.22 (t, 2H, J=8Hz), 2.228 (t, 2H, J=7Hz), 2.33 (t, 2H, J=5), 2.37 (t, 2H), 2.49-2.58 (m, 4H), 2.77 (t, 2H, J=6Hz), 2.86 (t, 2H, J=7Hz), 2.89 (t, 2H, J=7.5Hz), 2.94-2.96 (m, 2H), 3.03-3.05 (m, 1H), 3.45 (t, 4H), 3.58 (t, 2H, J=7Hz), 3.64 (t, 2H, J=8Hz), 4.01 (t, 2H, J=8Hz), 4.13 (t, 2H, J=6.5Hz), 6.07 (d, 1H. J=14 Hz), 6.11 (d, 1H, J=14Hz), 6.97-7.05 (m, 4H), 7.07-7.2 (m, 8H), 7.48 (d, 1H, J=14Hz), 7.54 (d, 1H, J=14Hz)

ESI (HRMS) m/z ($C_{61}H_{84}FN_6O_2^+$), calc: 951.6634; found 951.6651

CyR 164 (15 mg, 52%)



^{1}H-NMR (300 MHz, $CDCl_3$): 0.825 (t, 6H, J=7.2Hz), 1.03 (d, 3H, J=7.2Hz), 1.25-1.4 (m, 12 H), 1.61 (s, 12H), 1.79-1.91 (m, 6H), 1.84-1.89 (m, 2H), 2.0 (t, 2H), 2.12 (t, 2H, J=3.3 Hz), 2.21 (t, 2H, J=7.8Hz), 2.31 (t, 2H, J=2.1Hz), 2.41-2.52 (m, 4H), 2.71-2.76 (m, 4H), 2.95-3.0 (m, 4H), 3.25-3.26 (m, 1H), 3.34 (t, 4H), 3.5-3.62 (m, 4H), 3.93 (t, 4H, J=3.9Hz), 6.0 (d, 1H. J=14.1 Hz), 6.17 (d, 1H, J=14.1Hz), 7.0-7.39 (m, 8H), 7.54 (d, 1H, J=14.1 Hz), 7.47 (d, 1H, J=14.1 Hz)

ESI (HRMS) m/z ($C_{61}H_{84}FN_6O_2^+$), calc: 911.6885; found 911.6882

Photostability evaluation of CyR derivatives by fluorescence spectroscopy

Procedure: 1% DMSO in10 mM HEPES buffer (pH 7.4) was used for dissolving all the **CyR** derivatives and **CyNA-414** compound. The final concentrations of all the compounds were

10 μM. The compound solution were taken in 96-well black plate and then fluorescence intensity measurements were carried out in each 10 min interval for a total period of 8 h (excitation-emission 790-820 nm for **CyR** and **CyNA** compounds). For the strong UV condition, same stock solution was used for the photostability study.

Table S1 Characterization and purity determination by HPLC-MS, photophysical properties and preliminary photostability assessment of **CyR** library compounds and **CyNA-414** by using the SpectraMax M2 spectrophotometer under the xenon lamp

Compound	M^+(calc.)	M^+(exp.)	λ_{abs}(nm)	λ_{em}(nm)	ϕ*1	purity*2	F/F_o*3	Yield*3
CyR 28	1025.8	1025.9	807	824	0.12	93	98	53
CyR 32	947.6	947.6	806	822	0.11	92	89	56
CyR 49	963.6	963.6	807	823	0.13	93	91	62
CyR 77	947.6	947.7	806	824	0.10	96	90	51
CyR 92	913.7	913.6	807	823	0.08	93	86	50
CyR 100	1023.7	1023.6	807	823	0.12	92	92	56
CyR 101	933.6	933.7	807	825	0.10	97	90	55
CyR 103	963.6	963.7	807	822	0.10	95	93	54
CyR 105	926.7	926.6	807	822	0.12	95	94	53
CyR 111	934.6	935.6	807	823	0.10	92	92	60
CyR 131	961.7	961.8	807	822	0.16	95	91	61
CyR 135	987.5	987.5	806	822	0.08	91	89	59
CyR 164	911.6	911.8	806	824	0.10	91	92	58
CyR 165	901.6	901.6	807	822	0.10	93	94	52
CyR 167	915.6	915.6	807	824	0.11	92	100	60
CyR 177	887.6	887.6	806	823	0.10	92	93	53
CyR 180	897.6	897.6	807	822	0.12	92	98	56
CyR 185	899.6	899.6	807	824	0.10	91	96	54
CyR 193	999.7	999.7	806	823	0.12	93	100	55
CyR 201	961.7	961.7	807	822	0.09	90	93	59
CyR 211	933.6	933.6	807	823	0.14	91	89	61
CyR 218	937.6	937.6	807	822	0.13	92	82	65
CyR 220	937.6	937.7	806	823	0.09	92	97	63
CyR 221	953.6	953.6	807	822	0.12	93	91	60
CyR 222	871.6	871.7	807	824	0.10	95	97	56
CyR 230	941.7	941.7	807	823	0.10	91	96	51
CyR 240	995.5	995.6	806	822	0.11	88	93	49
CyR 262	979.6	979.6	806	823	0.10	91	93	61
CyR 272	940.7	940.6	806	822	0.11	91	95	54
CyR 274	929.7	929.6	806	821	0.10	96	91	52
CyR 275	961.7	961.6	807	822	0.08	91	88	51
CyR 277	927.7	927.7	806	823	0.10	92	95	56
CyR 282	942.7	942.7	807	822	0.10	90	91	54
CyR 319	979.6	979.7	806	822	0.08	91	95	53
CyR 323	915.6	915.6	806	821	0.14	90	94	59
CyR 329	997.57	997.6	806	822	0.11	91	100	61
CyR 330	988.3	988.6	806	823	0.10	96	91	59
CyR 335	947.6	947.7	806	821	0.08	94	92	51
CyR 341	997.5	997.7	806	821	0.11	94	100	49
CyR 358	979.6	979.7	806	822	0.10	94	95	51
CyR 359	978.6	978.5	807	823	0.10	94	94	54
CyR 360	941.7	941.7	806	822	0.11	94	94	51
CyR 361	964.6	964.5	807	822	0.13	95	90	52
CyR 364	953.6	953.5	807	822	0.11	93	93	59
CyR 368	998.7	998.7	807	822	0.13	95	91	63
CyR 374	919.6	919.6	807	822	0.11	96	97	62
CyR 375	937.6	937.6	807	823	0.09	94	94	61
CyR 381	949.6	949.7	807	825	0.10	89	95	59
CyR 384	941.7	941.7	807	822	0.13	96	100	52
CyR 387	949.6	949.7	807	823	0.10	91	98	53
CyR 388	949.6	949.7	806	823	0.12	92	89	56
CyR 395	963.6	963.7	806	823	0.10	90	100	50
CyR 396	933.6	997.8	807	822	0.10	98	100	51
CyR 399	955.6	955.7	806	822	0.11	91	80	52

CyR 403	953.6	953.6	807	823	0.11	93	100	56
CyR 405	951.6	951.6	807	822	0.12	91	100	59
CyR 407	969.7	969.7	806	821	0.10	92	100	54
CyR 412	955.6	955.6	806	824	0.11	96	86	53
CyR 414	968.7	968.6	807	823	0.12	94	94	55
CyR 419	899.6	899.6	807	823	0.10	93	100	56
CyR 420	928.7	928.6	806	823	0.13	95	100	51
CyR 425	899.6	899.7	806	824	0.09	94	94	52
CyR 429	925.7	925.6	807	824	0.10	93	96	58
CyR 439	913.7	913.6	807	823	0.12	94	94	54
CyR 442	899.6	899.6	806	823	0.11	94	91	52
CyR 446	909.6	909.7	806	821	0.12	93	92	57
CyR 447	987.6	987.6	806	822	0.10	93	100	48
CyR 477	951.6	951.5	806	823	0.10	91	91	53
CyR 479	951.6	951.7	807	820	0.10	93	95	55
CyR 526	969.7	969.7	807	824	0.12	92	100	54
CyR 548	1009.7	1009.8	807	823	0.12	91	100	51
CyR 554	955.7	955.6	807	822	0.12	92	92	54
CyR 565	989.2	989.6	807	823	0.12	92	93	55
CyR 572	933.6	1013.5	807	822	0.11	96	100	56
CyR 574	899.6	899.7	806	822	0.10	92	100	52
CyR 577	997.8	933.7	807	822	0.13	93	92	61
CyR 599	1009.7	1009.7	807	823	0.12	89	90	60
CyR 602	913.7	913.6	806	822	0.11	93	93	55
CyR 677	967.6	967.6	806	824	0.10	91	100	54
CyR 686	1001.6	1001.6	807	822	0.11	92	94	53
CyNA-414[4]	701.5	701.2	804	819	0.10	96	100	51

*[1] Quantum yields were measured in DMSO, using Cardiogreen as a standard (ϕ : 0.13, in DMSO).[1]
*[3] Quotients of fluorescent intensities after 8 h vs. fluorescent intensities (after 0 h), in a time-course fluorescence measurement using 10 μM (1% DMSO) solutions in HEPES buffer (10 mM, pH 7.4).
*[2] Purities were determined according to UV absorption at 365 nm.
*[4] **CyNA-414** was used as the standard for the photostability evaluation.
*[5]Yield was calculated of the solid compound after complete drying in high vacuum. Average yield of the 80-membered CyR library was 56 %.

Table S2 Photostability evaluation of **CyR** library compounds and **CyNA-414** by using the UVP Blak-Ray® B-100AP high intensity mercury lamp

compound	F/F_o*[5]	F/F_o*[6]
CyR 28	44	34
CyR 32	55	43
CyR 49	48	37
CyR 77	44	39
CyR 92	50	40
CyR 100	56	48
CyR 101	46	44
CyR 103	52	39
CyR 105	62	53
CyR 111	51	41
CyR 131	43	36
CyR 135	54	42
CyR 164	58	45
CyR 165	60	47
CyR 167	67	**63($)**
CyR 177	59	50
CyR 180	48	40
CyR 185	55	43
CyR 193	43	36
CyR 201	57	48
CyR 211	62	55
CyR 218	51	39
CyR 220	49	46
CyR 221	41	37

CyR 222	43	34
CyR 230	63	52
CyR 240	48	33
CyR 262	43	32
CyR 272	47	37
CyR 274	48	42
CyR 275	43	35
CyR 277	45	38
CyR 282	48	43
CyR 319	42	36
CyR 323	61	58
CyR 329	51	40
CyR 330	44	39
CyR 335	51	37
CyR 341	59	50
CyR 358	45	40
CyR 359	45	38
CyR 360	57	39
CyR 361	45	37
CyR 364	56	47
CyR 368	51	40
CyR 374	62	57
CyR 375	58	49
CyR 381	45	39
CyR 384	52	46
CyR 387	70	**68($)**
CyR 388	43	39
CyR 395	62	50
CyR 396	37	35
CyR 399	49	41
CyR 403	48	39
CyR 405	45	36
CyR 407	47	37
CyR 412	47	36
CyR 414	49	39
CyR 419	42	36
CyR 420	48	34
CyR 425	61	57
CyR 429	47	38
CyR 439	48	39
CyR 442	45	38
CyR 446	55	49
CyR 447	41	32
CyR 477	48	38
CyR 479	49	41
CyR 526	69	**60 ($)**
CyR 548	62	60
CyR 554	48	38
CyR 565	57	44
CyR 572	61	54
CyR 574	48	46
CyR 577	44	35
CyR 599	49	38
CyR 602	53	40
CyR 677	48	43
CyR 686	51	36
CyNA-414	51	47

($) Best selected three compounds

*[5] Quotients of fluorescent intensities after 1 h vs. fluorescent intensities (after 0 h), in a time-course fluorescence measurement under the strong Hg-lamp (high intensity UV radiation) using 10 μM (1% DMSO) solutions in HEPES buffer (10 mM, pH 7.4).

*[6] Quotients of fluorescent intensities after 2 h vs. fluorescent intensities (after 0 h), in a time-course fluorescence measurement under the strong Hg-lamp(high intensity UV radiation) using 10 μM (1% DMSO) solutions in HEPES buffer (10 mM, pH 7.4).

Photodecomposition of Study

Procedure: 10 µM **CyR 526, CyR 387, CyR 167** and **CyNA-414** solutions in 10 mM HEPES buffer (pH 7.4) containing 1% DMSO were placed in a 96-well black plate under the strong UV light, and fluorescence intensity measurements were performed every 10 min for a total period of 2 h (excitation-emission: 790-820 nm).Values are fitted to a non-linear regression one-phase exponential decay. Rate constants of dye photodecomposition were acquired from the plots of $-\ln(F/F_0)$ vs. time, considering $-\ln(F/F_0) = k*t$ as a pseudo-first order rate equation[2].

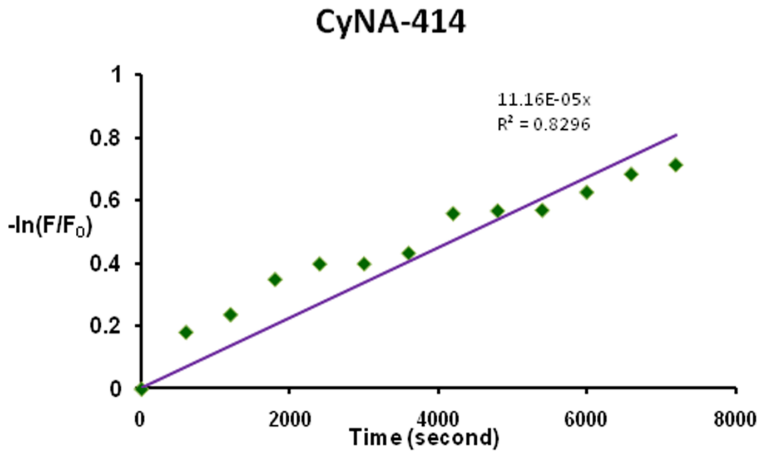


Fig. S1 Photodecomposition of **CyNA-414**

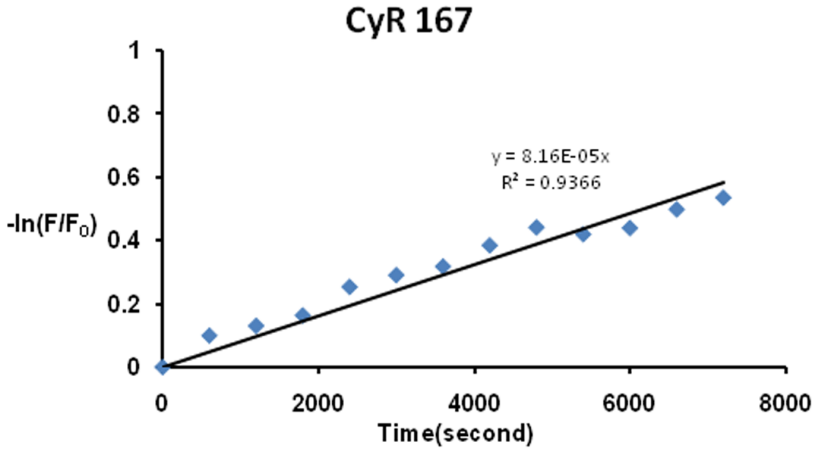


Fig. S2 Photodecomposition of **CyR 167**

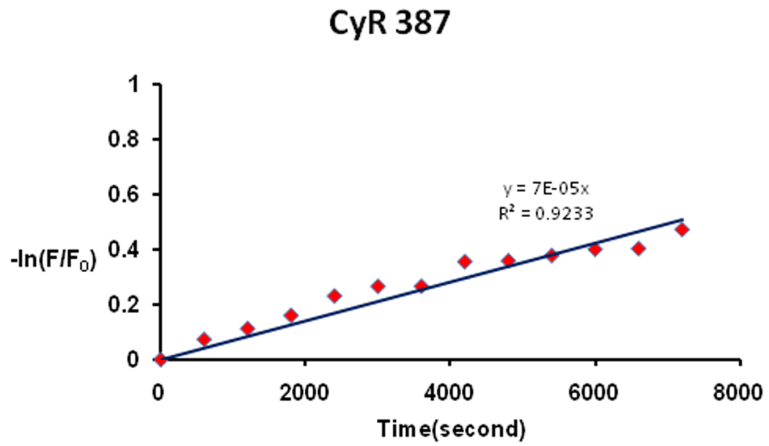


Fig. S3 Photodecomposition of **CyR 387**

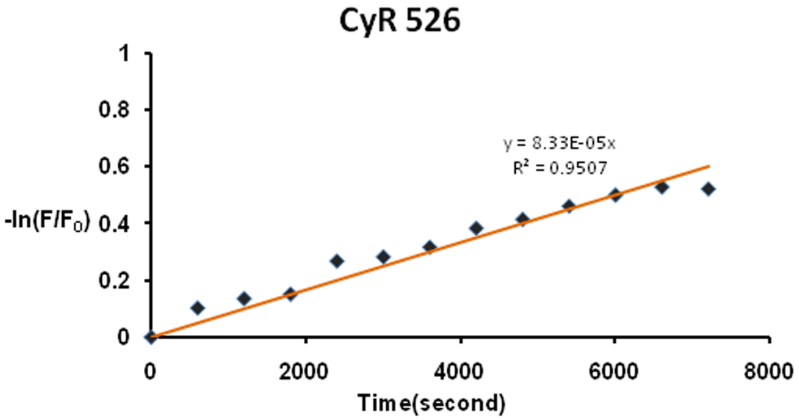


Fig. S4 Photodecomposition of **CyR 526**

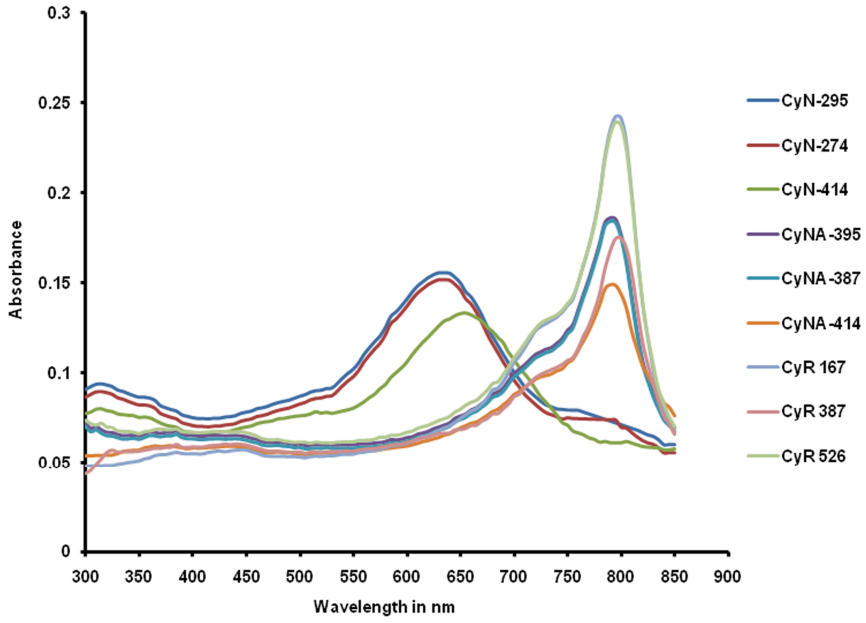


Fig. S5 Absorption spectra of selected **CyN, CyNA** and **CyR** compounds in 1% DMSO in 10 mM HEPES buffer (pH 7.4) at 10 µM concentration

Table S3 Structures of best selected photostable **CyR** compounds compared with **CyNA-414** under high intensity UV radiation

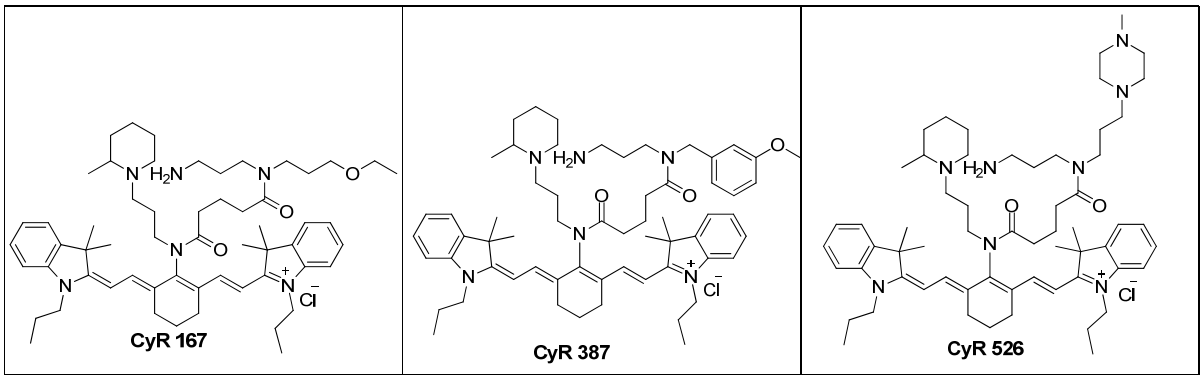


Table S4 structures of the **CyNA** compounds used in high intensity UV condition

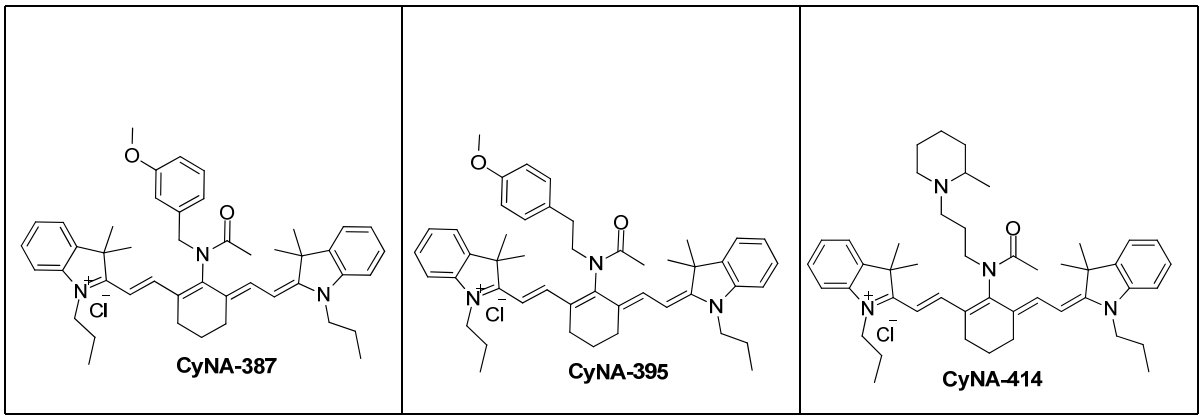
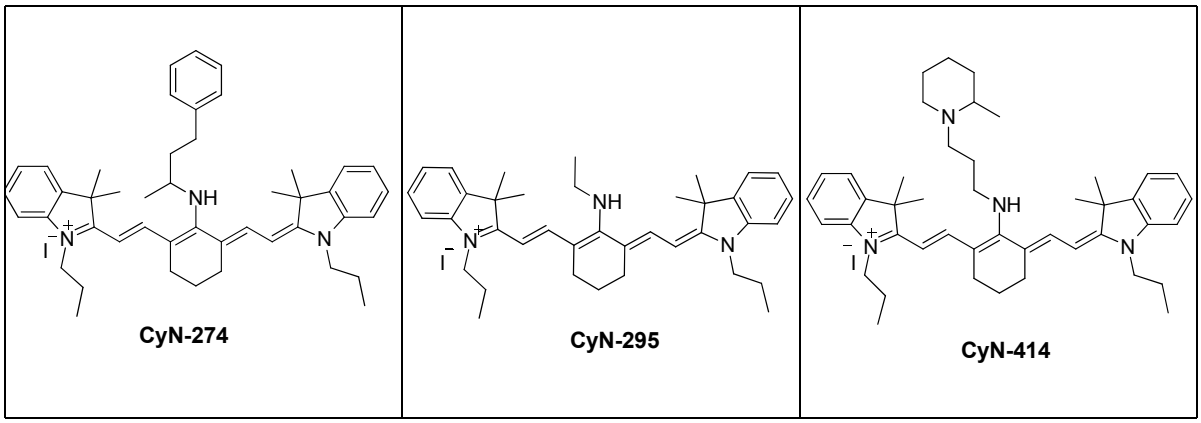


Table S5 Structure of **CyN** used in high intensity UV condition



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2. A. Toutchkine, D.V. Nguyen, K.M. Hahn. *Org. Lett*. 2007, **9**, 2775-2777.
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