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Supporting Information

Efficient Photosensitizers with Aggregation-Induced Emission Characteristics for Lysosome- and Gram-Positive Bacteria-Targeted Photodynamic Therapy

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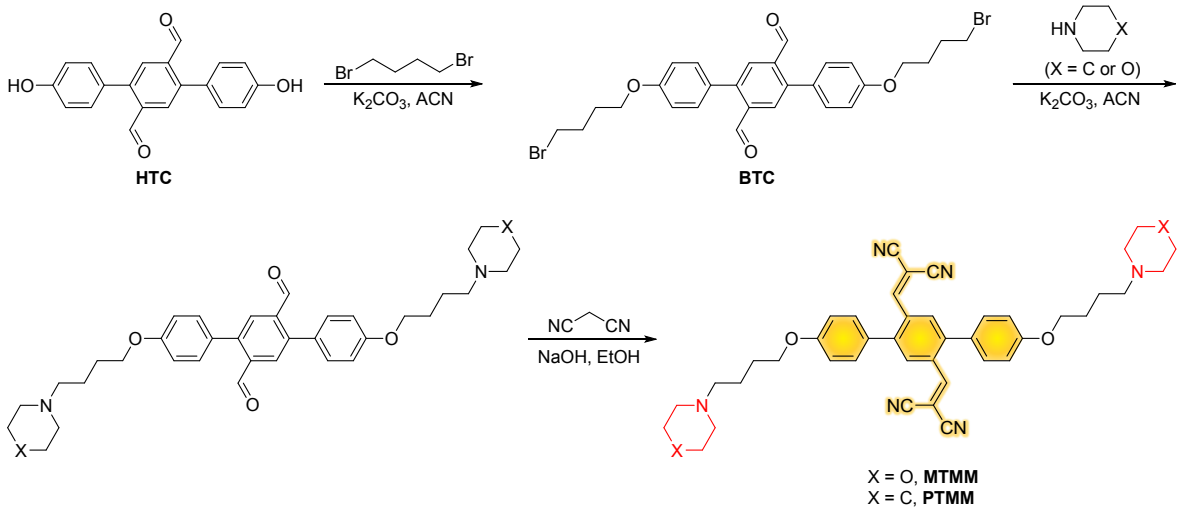
1. General Information

Petroleum ether and ethyl acetate for chromatography were distilled before used. All other reagents and solvents were used directly from the corresponding supplier without further purification. All starting materials were purchased from Sigma-Aldrich, Aladdin, Energy, Tansoole and use directly. HeLa cells were purchased from the Type Culture Collection of the Chinese Academy of Sciences, Shanghai, China. Nuclear magnetic resonance spectra (^{1}H, ^{13}C NMR) were recorded on a Bruker Avancf 300 (^{1}H at 300 MHz, ^{13}C at 75 MHz) or a Bruker Ascend 600 (^{1}H at 600 MHz, ^{13}C at 151 MHz). The chemical shifts are reported as ppm and solvent residual peaks were shown as following: $CDCl_3$ δ H (7.26 ppm) and δ C (77.16 ppm); d_6-DMSO δ H (2.50 ppm) and δ C (39.52 ppm). UV-visible absorption spectra were measured on Purkinje TU-1950 spectrometer. Fluorescence spectra were recorded on a Hitachi F-7000 spectrometer. Single crystal was collected on Oxford diffraction Eos CCD detector. The fluorescence imaging was collected on Olympus FV1200. Dynamic Light Scattering (DLS) was carried out on Malvern Zetasizer Nano ZS90. High-resolution Mass spectra (HRMS) were obtained on a Bruker Maxis and Microflex and reported as m/z (relative intensity). Accurate masses are presented as molecular ion $[M+Na]^+$ or $[M+H]^+$, respectively. All theoretical calculations reported were performed using the Gaussian 09 code.

2. Experimental Procedure and Characterization Data

2.1 Reaction procedures



Scheme S1. Synthetic routes of target compounds.

4,4''-dihydroxy-[1,1':4',1''-terphenyl]-2',5'-dicarbaldehyde (HTC) was synthesized according to the previous literature.[1] Yield: 47%. ^{1}H NMR (600 MHz, d_6-DMSO) δ 9.98 (s, 2H, CHO), 9.83 (s, 2H, OH), 7.87 (s, 2H, Ar H), 7.33 (d, J = 8.5, 4H, Ar H), 6.93 (d, J = 8.5, 4H, Ar H); ^{13}C NMR (151 MHz, d_6-DMSO) δ 191.69, 157.97, 142.95, 135.99, 131.24, 129.66, 126.69, 115.63.

Synthesis of 4,4''-bis(4-bromobutoxy)-[1,1':4',1''-terphenyl]-2',5'-dicarbaldehyde (BTC). 4,4''-dihydroxy-[1,1':4',1''-terphenyl]-2',5'- dicarbaldehyde (382 mg, 1.2 mmol), potassium carbonate(249 mg, 1.8mmol) and 1,4-dibromobutane (440 uL, 3.6 mmol) was dissolved in acetonitrile (10 mL). The reaction was refluxed at 80 °C for 8 h. After the reaction was completed based on TLC, the acetonitrile was removed under reduced pressure and purified by column chromatography on silica gel (DCM) to afford desired product as yellow solid. Yield: 47%. ^{1}H NMR (600 MHz, $CDCl_3$) δ 10.08 (s, 2H, CHO), 8.06 (s, 2H, Ar H), 7.35 (d, J = 8.5 Hz, 4H, Ar H), 7.02 (d, J = 8.5 Hz, 4H, Ar H), 4.08 (t, J = 6.0 Hz, 4H, CH_2), 3.52 (t, J = 6.6 Hz, 4H, CH_2), 2.14-2.09 (m, 4H, CH_2), 2.03-1.98 (m, 4H, CH_2); ^{13}C NMR (151 MHz,

$CDCl_3$) δ 192.22, 159.54, 143.93, 136.68, 131.47, 130.29, 128.98, 114.90, 67.23, 33.47, 29.60, 28.03.

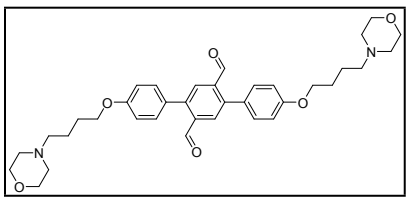
Synthesis of 4,4''-bis(4-morpholinobutoxy)-[1,1':4',1''-terphenyl]-2',5'-dicarbaldehyde (MTC). 4,4''-bis(4-bromobutoxy)-[1,1':4',1''-terphenyl]- 2',5'-dicarbaldehyde (200 mg, 0.34 mmol), potassium carbonate(188 mg, 1.36mmol) and morpholine(133 μL, 1.53mmol) were dissolved in 6 mL of acetonitrile. The reaction was refluxed at 80 °C for 6 h. After the reaction was completed based on TLC analysis, the acetonitrile was removed under reduced pressure and the mixture was purified by column chromatography on silica gel (DCM/MeOH = 30/1) to afford desired compound as yellow solid with 80% yield.

Synthesis of 4,4''-bis(4-(piperidin-1-yl)butoxy)-[1,1':4',1''-terphenyl]-2',5'-dicarbaldehyde (PTC). 4,4''-bis(4-bromobutoxy)-[1,1':4',1''-terphenyl]- 2',5'-dicarbaldehyde (200 mg, 0.34 mmol), potassium carbonate(188 mg, 1.36mmol) and piperidine(140 μL, 1.53mmol) were dissolved in 6 mL of acetonitrile. The reaction was refluxed at 80 °C for 6 h. After the reaction was completed based on TLC analysis, the acetonitrile was removed under reduced pressure and the mixture was purified by column chromatography on silica gel (DCM/MeOH = 30/1) to afford desired compound as yellow solid with 90% yield.

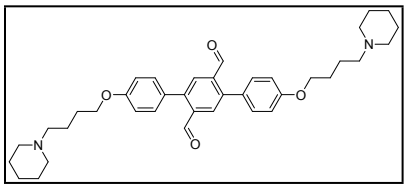
Synthesis of 2,2'-((4,4''-bis(4-morpholinobutoxy)-[1,1':4',1''-terphenyl]- 2',5'-diyl)bis(methanylylidene)) dimalononitrile (MTMM). 4,4''-bis (4-morpholinobutoxy)-[1,1':4',1''-terphenyl]-2',5'-dicarbaldehyde (189.6 mg, 0.31 mmol) and malononitrile (62.65 mg, 0.93 mmol) were dissolved in 8 mL of ethanol, Then 68 μL of 1M NaOH solution was added. The reaction was stirred at room temperature for 3 h and filtered to afford yellow solid of **MTMM** with 58% yield.

The synthetic procedure of 2,2'-((4,4''-bis(4-(piperidin-1-yl)butoxy)- [1,1':4',1''-terphenyl]-2',5'-diyl)bis(methanylylidene))dimalononitrile **(PTMM)** was similar to that of **MTMM**.

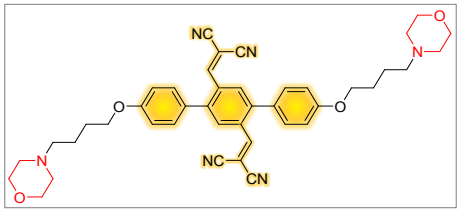
2.2 Compound data



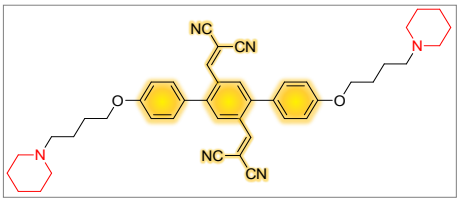
4,4''-bis(4-morpholinobutoxy)-[1,1':4',1''-terphenyl]-2',5'-dicarbaldehyde (MTC). Yield: 80%. ^{1}H NMR (600 MHz, $CDCl_3$) δ 10.07 (s, 2H, CHO), 8.05 (s, 2H, Ar H), 7.33 (d, *J* = 8.6 Hz, 4H, Ar H), 7.01 (d, *J* = 8.6 Hz, 4H, Ar H), 4.06 (t, *J* = 6.3 Hz, 4H, CH_2), 3.73 (t, *J* = 4.6 Hz, 8H, CH_2), 2.48-2.43 (m, 12H, CH_2), 1.89-1.84 (m, 4H, CH_2), 1.74-1.69 (m, 4H, CH_2); ^{13}C NMR (151 MHz, $CDCl_3$) δ 192.25, 159.68, 143.94, 136.66, 131.43, 130.26, 128.78, 114.89, 68.02, 67.08, 58.71, 53.84, 27.30, 23.19.



4,4''-bis(4-(piperidin-1-yl)butoxy)-[1,1':4',1''-terphenyl]-2',5'-dicarbaldehyde (PTC). Yield: 90%. ^{1}H NMR (600 MHz, $CDCl_3$) δ 10.08 (s, 2H, CHO), 8.05 (s, 2H, Ar H), 7.34 (d, *J* = 8.5 Hz, 4H, Ar H), 7.02 (d, *J* = 8.5 Hz, 4H, Ar H), 4.05 (t, *J* = 6.3 Hz, 4H, CH_2), 2.45-2.43 (m, 12H, CH_2), 1.87-1.83 (m, 4H, CH_2), 1.77-1.72 (m, 4H, CH_2), 1.65-1.62 (m, 8H, CH_2), 1.46 (brs, 4H, CH_2); ^{13}C NMR (151 MHz, $CDCl_3$) δ 192.24, 159.55, 143.91, 136.63, 131.43, 130.25, 128.84, 114.88, 67.84, 58.59, 54.31, 27.28, 25.02, 23.87, 22.78.



2,2'-((4,4''-bis(4-morpholinobutoxy)-[1,1':4',1''-terphenyl]-2',5'-diyl)bis (methanylylidene)) dimalononitrile (MTMM). Yield: 58%. ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.21 (s, 2H, CH), 7.83 (s, 2H, Ar H), 7.25 (d, *J* = 8.5 Hz, 4H, Ar H), 7.05 (d, *J* = 8.7 Hz, 4H, Ar H), 4.08 (t, *J* = 6.2 Hz, 4H, CH_2), 3.73 (t, *J* = 4.5 Hz, 8H, CH_2), 2.48-2.41 (m, 12H, CH_2), 1.93-1.84 (m, 4H, CH_2), 1.76-1.67 (m, 4H, CH_2); ^{13}C NMR (151 MHz, $CDCl_3$) δ 160.43, 158.77, 142.92, 132.76, 131.47, 130.71, 128.80, 115.48, 113.09, 112.28, 86.46, 68.16, 67.15, 58.70, 53.89, 27.26, 23.23. HRMS (ESI-TOF) m/z: $[M+H]^+$ calcd for $C_{42}H_{45}N_6O_4^+$, 697.3502, found, 697.3490.



2,2'-((4,4''-bis(4-(piperidin-1-yl)butoxy)-[1,1':4',1''-terphenyl]-2',5'-diyl)bis (methanylylidene)) dimalononitrile (PTMM). Yield: 72%. ^{1}H NMR (600 MHz, $CDCl_3$) δ 8.20 (s, 2H), 7.83 (s, 2H), 7.24(d, *J* = 8.5 Hz, 4H, Ar H), 7.05 (d, *J* = 8.6 Hz, 4H, Ar H), 4.06 (t, *J* = 6.3 Hz, 4H), 2.41-2.37 (m, 12H), 1.86-1.82 (m, 4H), 1.74-1.69 (m, 4H), 1.62-1.58 (m, 8H), 1.44 (brs, 4H); ^{13}C NMR (151 MHz, $CDCl_3$) δ 160.43, 158.81, 142.90, 132.74, 131.45, 130.70, 128.74, 115.48, 113.08, 112.28, 86.41, 68.23, 59.07, 54.72, 27.44, 26.02, 24.53, 23.51. HRMS (ESI-TOF) m/z: $[M+H]^+$ calcd for $C_{44}H_{49}N_6O_2^+$, 693.3917, found, 693.3905.

Reference

1. J. N. Zhang, H. Kang, N. Li, S. M. Zhou, H. M. Sun, S. W. Yin, N. Zhao and B. Z. Tang, *Chem. Sci.,* 2017, **8**, 577-582.

3. Optical Properties

Aggregation-induced emission properties of target compounds. Nanoaggregates preparation: 5 mM stock solution of target compound in THF was firstly prepared. Then aliquots of above stock solution was transferred into 5 mL volumetric flasks and appropriate amounts of water were added to obtain 50 μM solution with different volume fractions of water (0%, 10%, 30%, 50%, 70%, 80%, 90%, 95%, 99%). After that, the PL measurements of the resulting solutions were performed immediately.

Stabilities of MTMM and PTMM. The fluorescent spectra of **MTMM** and **PTMM** solution (20 μM in DMEM medium without phenol red, f_w = 99%) were recorded from 0 min to 480 min. Then the highest of their emission peaks (I/I_0) at different time spots were plotted to evaluate their aqueous stabilities.

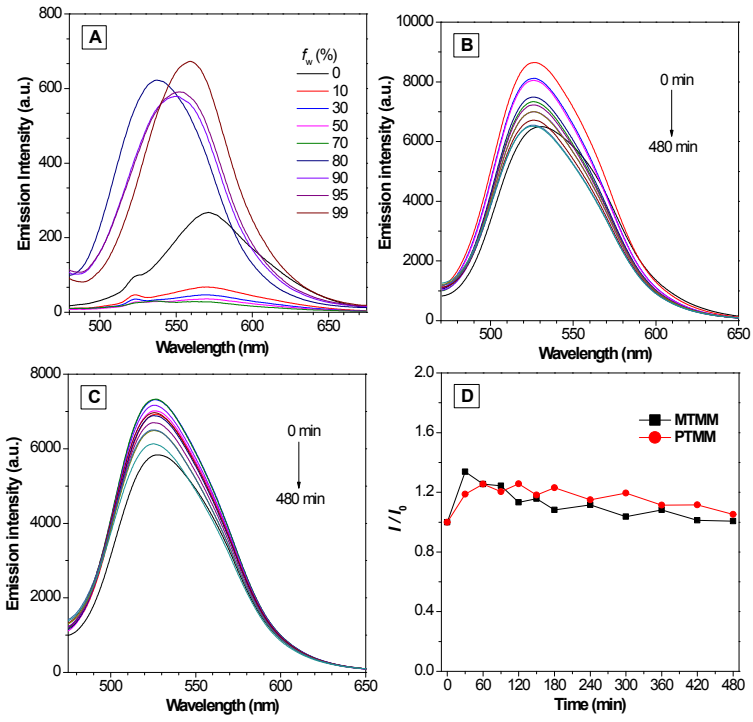


Fig. S1 (A) The emission spectra of **MTMM** (50 μM) in THF/water mixtures with varied water volume fractions (f_w). (B) The emission spectra change of **MTMM** in DMSO/DMEM (1/99) mixtures with different standing times. (C) The emission spectra change of **PTMM** in DMSO/DMEM (1/99) mixtures with different standing times. (D) Corresponding plots of I/I_0 at the highest emission peak in B and C with different standing times.

Table S1. Optical properties of **MTMM** and **PTMM**.

Compd.	Solution state[a]				Aggregation State[b]	
	λ_{abs} [nm]	ε [$M^{-1}cm^{-1}$]	λ_{em} [nm]	$Ø_f$[b]	λ_{em} [nm]	$Ø_f$[b]
MTMM	420	10900	570	0.6	559	2.1
PTMM	421	9840	571	0.3	552	1.0

[a]THF solution; [b]In THF/water mixtures with f_w of 80%; [c]Absolute fluorescence quantum yield measured using the calibrated integrating sphere system.

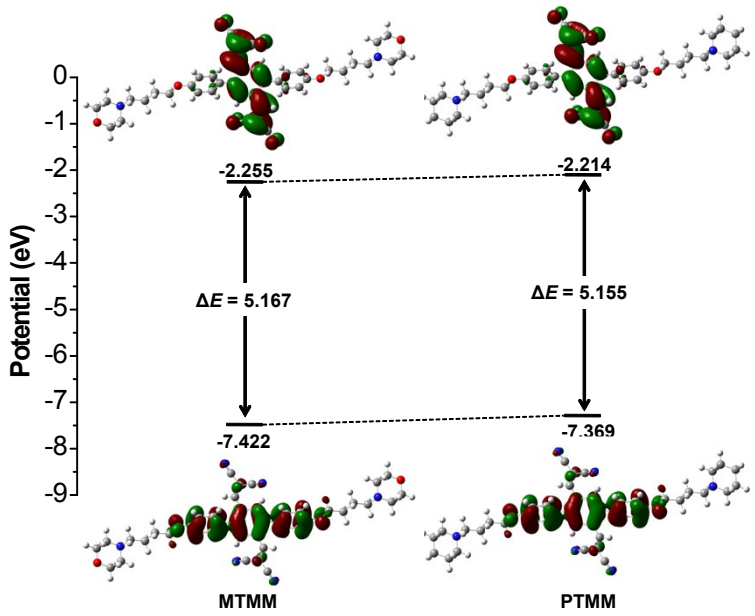


Fig. S2 Molecular frontier orbital amplitude plots of **MTMM** and **PTMM** calculated by CAM-B3LYP/6-31G (d, p) basis set.

Table S2. Cartesian Coordinates of Optimized **MTMM** Structures

Symbol	X	Y	Z
C	-0.91506	-0.9933	0.34767
C	0.435525	-1.33554	0.243458
C	1.382945	-0.3363	-0.07337
C	0.916013	0.958067	-0.27764
C	-0.43445	1.300423	-0.17287
C	-1.38191	0.301008	0.143197
C	2.832375	-0.60494	-0.21632
C	3.762303	0.261901	0.361251
C	3.320071	-1.68996	-0.95955
C	5.129251	0.056397	0.225856
H	3.415259	1.112714	0.939781
C	4.676955	-1.90548	-1.10328

C	5.594396	-1.03489	-0.50846
H	5.056728	-2.73756	-1.68475
C	-2.8316	0.569743	0.283738
C	-3.32063	1.652025	1.030013
C	-3.76031	-0.29345	-0.30107
C	-4.67778	1.868856	1.16924
H	-2.62718	2.316796	1.535052
C	-5.12753	-0.08618	-0.17085
H	-3.41199	-1.14246	-0.88148
C	-5.59404	1.002804	0.565989
H	-5.05874	2.699137	1.752511
H	-5.814	-0.77337	-0.64766
O	6.899707	-1.32858	-0.70508
O	-6.89973	1.298503	0.757194
C	7.888843	-0.46799	-0.15719
H	7.756749	0.549558	-0.54875
H	7.785279	-0.42485	0.935152
C	-7.88777	0.452639	0.185013
H	-7.76983	-0.57074	0.565697
H	-7.76858	0.422158	-0.90618
C	9.244929	-1.0214	-0.54552
H	9.327809	-2.04343	-0.16002
H	9.291816	-1.09206	-1.63769
C	-9.24486	1.012691	0.560068
H	-9.31223	1.06078	1.652408
H	-9.30886	2.043491	0.194901
C	10.3918	-0.16136	-0.01997
H	10.2776	0.859842	-0.40089
H	10.35155	-0.09484	1.072928
C	-10.3907	0.177792	-0.00669
H	-10.2931	-0.85423	0.348943
H	-10.3322	0.140016	-1.10014
C	11.75344	-0.72541	-0.41508
H	11.81597	-1.75799	-0.05365
H	11.82905	-0.77672	-1.51857
C	-11.7535	0.745167	0.379543
H	-11.8539	0.754493	1.48223
H	-11.7941	1.791813	0.057591
H	2.625737	-2.35764	-1.45946
H	5.81665	0.746668	0.69685
C	0.881709	-2.69278	0.549951
H	1.868017	-2.78354	0.994398
C	-0.88077	2.657879	-0.47814
H	-1.86769	2.749023	-0.92113
H	-1.63162	-1.74531	0.650796
H	1.632368	1.710004	-0.58134
C	0.216407	-3.85199	0.339313

C	-0.21508	3.816919	-0.26775
C	0.797768	-5.09768	0.752762
C	-1.05387	-3.93968	-0.3237
C	1.056274	3.904194	0.39324
C	-0.797	5.062883	-0.6796
N	1.272329	-6.09788	1.086683
N	-2.07594	-4.01472	-0.86
N	2.079288	3.97889	0.927786
N	-1.27199	6.0633	-1.01224
C	14.10995	-0.7605	0.093621
C	13.07221	1.314669	-0.44855
C	15.23567	0.009775	0.764032
H	14.39808	-0.9781	-0.95183
H	13.96672	-1.71758	0.606195
C	14.22682	2.033532	0.231346
H	13.28937	1.224101	-1.52971
H	12.16654	1.917602	-0.34013
H	16.18248	-0.52632	0.66472
H	15.00743	0.128744	1.83432
H	14.42998	2.9865	-0.26291
H	13.96338	2.230117	1.281977
C	-13.0954	-1.27911	0.294755
C	-14.0984	0.830544	-0.17428
C	-14.2426	-1.9527	-0.44159
H	-13.3349	-1.2349	1.374089
H	-12.1941	-1.88715	0.178199
C	-15.2177	0.104937	-0.90286
H	-14.4067	1.003583	0.873741
H	-13.9336	1.80824	-0.63927
H	-14.4667	-2.92466	0.00424
H	-13.9584	-2.10436	-1.49423
H	-16.1605	0.647116	-0.79928
H	-14.9675	0.031963	-1.97238
N	12.87118	0.005687	0.161872
N	-12.867	0.053563	-0.25145
O	-15.4256	-1.18445	-0.36264
O	15.41677	1.275289	0.161469

Table S3. Cartesian Coordinates of Optimized **PTMM** Structures

Symbol	X	Y	Z
C	-0.91504	-0.97487	0.315369
C	0.435337	-1.31722	0.20887
C	1.382226	-0.31838	-0.1109
C	0.915069	0.975724	-0.31611
C	-0.43532	1.318065	-0.20969
C	-1.3822	0.319237	0.110135

C	2.831272	-0.58791	-0.25575
C	3.762683	0.276195	0.323459
C	3.317024	-1.6716	-1.00218
C	5.129259	0.069026	0.187044
H	3.416958	1.126034	0.904218
C	4.673511	-1.88824	-1.14774
C	5.592722	-1.02064	-0.55099
H	5.051855	-2.71925	-1.73168
C	-2.83125	0.588675	0.255012
C	-3.31707	1.672335	1.001461
C	-3.76258	-0.27549	-0.32422
C	-4.67357	1.888852	1.147034
H	-2.62136	2.338121	1.502067
C	-5.12919	-0.06844	-0.18777
H	-3.41679	-1.12527	-0.90503
C	-5.59271	1.021145	0.550309
H	-5.052	2.719832	1.730959
H	-5.81783	-0.7562	-0.66056
O	6.897151	-1.31516	-0.74953
O	-6.89717	1.315571	0.748912
C	7.8881	-0.46417	-0.18887
H	7.759588	0.558579	-0.56767
H	7.781538	-0.43486	0.903653
C	-7.88807	0.464384	0.188434
H	-7.75916	-0.55838	0.567034
H	-7.78185	0.435274	-0.90413
C	9.243918	-1.01533	-0.58076
H	9.318802	-2.04656	-0.21881
H	9.298598	-1.06063	-1.67399
C	-9.24391	1.015119	0.580833
H	-9.2982	1.060342	1.674088
H	-9.31923	2.046364	0.218987
C	10.39094	-0.17399	-0.02595
H	10.27755	0.859657	-0.37158
H	10.34947	-0.14473	1.068477
C	-10.3909	0.173455	0.026428
H	-10.2772	-0.8601	0.37222
H	-10.3497	0.143998	-1.06801
C	11.75359	-0.72601	-0.43691
H	11.80504	-1.77416	-0.12211
H	11.83429	-0.7304	-1.54166
C	-11.7536	0.725271	0.437607
H	-11.8339	0.730129	1.542384
H	-11.8054	1.773274	0.122364
H	2.621296	-2.33733	-1.50283
H	5.817957	0.756796	0.659746
C	0.882679	-2.67368	0.516853

H	1.870583	-2.76308	0.958033
C	-0.88276	2.674477	-0.51769
H	-1.87077	2.763728	-0.95867
H	-1.63102	-1.72633	0.621223
H	1.631055	1.727212	-0.62187
C	0.216938	-3.83361	0.311492
C	-0.21712	3.834532	-0.3127
C	0.79985	-5.07804	0.726558
C	-1.05542	-3.92364	-0.34714
C	1.055451	3.92492	0.345484
C	-0.80034	5.078797	-0.72783
N	1.275322	-6.07734	1.061904
N	-2.07907	-4.00107	-0.88009
N	2.079293	4.002607	0.878023
N	-1.27608	6.077949	-1.06321
C	-13.0891	-1.29724	0.393881
C	-14.095	0.821729	-0.12268
C	-14.1785	-2.05359	-0.35981
H	-13.367	-1.22136	1.464968
H	-12.155	-1.86248	0.348442
C	-15.2241	0.152867	-0.8992
H	-14.4189	0.985717	0.924954
H	-13.8833	1.807888	-0.54792
C	-15.482	-1.2582	-0.37471
H	-14.3261	-3.03556	0.101332
H	-13.8387	-2.22294	-1.38762
H	-16.1283	0.766239	-0.83019
H	-14.9401	0.10579	-1.95645
H	-16.2398	-1.76533	-0.98019
H	-15.8808	-1.19779	0.646374
C	14.0949	-0.82322	0.123886
C	13.08946	1.296246	-0.39168
C	15.22394	-0.155	0.901027
H	14.41896	-0.98666	-0.92377
H	13.88296	-1.80958	0.54852
C	14.17891	2.05194	0.362689
H	13.36762	1.220928	-1.46275
H	12.15551	1.861632	-0.34611
C	15.48226	1.256312	0.377385
H	16.12808	-0.76851	0.83188
H	14.93977	-0.10846	1.95824
H	14.32672	3.034167	-0.09782
H	13.83892	2.220718	1.390534
H	16.24001	1.762952	0.9833
H	15.8812	1.196407	-0.64366
N	-12.8721	0.027552	-0.17798
N	12.87216	-0.02885	0.179354

4. X-Ray Single Crystal Data and Packing Mode

4.1 Single crystal data summary

Table S4. Crystallographic data.

Crystal	**MTMM**
formula	$C_{42}H_{44}N_6O_4$
crystal system	triclinic
space group	P -1
a [Å]	6.5347 (3)
b[Å]	11.4519 (5)
c[Å]	14.2937 (6)
β [deg]	95.468 (4)
V [Å^3]	954.35 (8)
Z	2
μ [mm^{-1}]	0.635
T [K]	293
θ_{min}-θ_{max} [deg]	4.2230-71.3480
R	0.0469
wR_2	0.1434
GOOF	1.047
grow condition	CH_2Cl_2/*n*-Hexane
diffraction measurement device	four-circle diffractometer
diffraction detector	Eos CCD plate
diffraction radiation type	CuK\a
diffraction measurement method	w scans
CCDC number	1959346

Table S5. The selected bond lengths (Å) and torsion angles (°) of **MTMM**.

Crystal	**MTMM**	
Bond length (Å)	O2-C9	1.3626(17)
	O2-C8	1.426(2)
	C16-C15	1.3919(17)
	C16-C12	1.4795(17)
	C12-C11	1.3899(19)
	C18-C17	1.4643(17)
	C18-C19	1.339(2)
	N1-C5	1.459(2)
	N1-C2	1.451(2)
	O1-C1	1.420(2)
	C19-C21	1.435(2)
	C21-N3	1.129(2)
	C5-C6	1.518(2)
	C2-C1	1.509(3)
Torsion angles (°)	O2-C9-C10-C11	177.32(15)
	C16-C12-C11-C10	-179.55(13)
	C15-C16-C12-C11	136.96(15)
	C15-C16-C12-C13	-41.90(19)
	C15-C16-C17-C18	171.98(13)
	C12-C16-C15-C17	-178.61(12)
	C12-C16-C17-C18	-9.0(2)
	N1-C5-C6-C7	-171.42(16)
	N1-C2-C1-O1	57.9(2)
	C17-C18-C19-C20	169.23(14)
	C19-C18-C17-C16	149.24(15)
	C19-C18-C17-C15	-38.3(2)
	C5-N1-C2-C1	-177.03(14)
	C2-N1-C5-C6	-65.1(2)
	C2-N1-C3-C4	56.74(19)
	C1-O1-C4-C3	59.4(2)

4.2 X-ray single crystallographic packing of MTMM

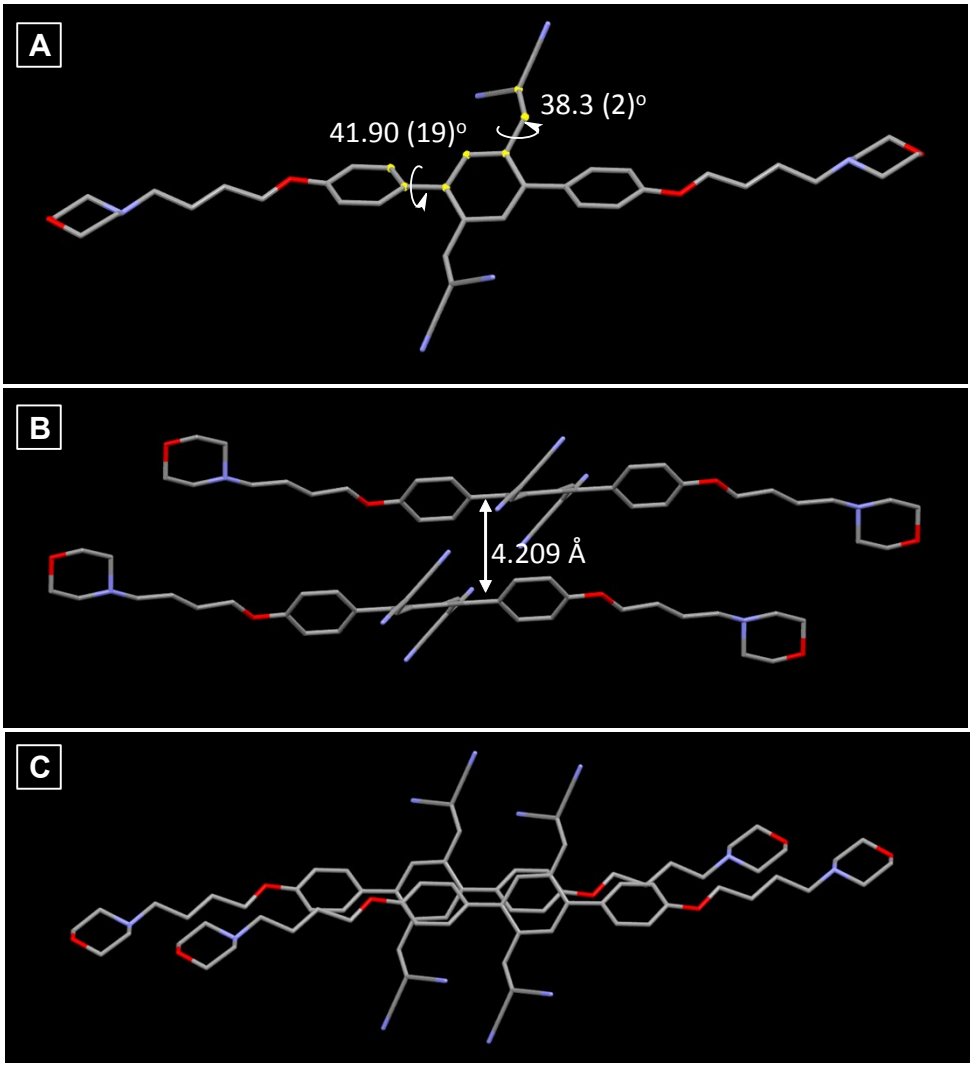


Fig. S3 (A) Single crystal structure and torsion angles of **MTMM**. (B) Side view and the distances between neighboring molecules in the crystal lattice of **MTMM**. (C) Top view of the crystal of **MTMM**. Carbon, oxygen, and nitrogen atoms are shown in gray, red, and blue, respectively. Hydrogen atoms are omitted for clarity.

5. Cell Imaging and Photodynamic Therapy of Cancer Cells

Cytotoxicity study. To determine the cytotoxicity in the dark, a MTT-based cell viability assay was performed in 96-well plates. HeLa cells were seeded at a density of 10000 cells per well and then incubated for 24 h. The AIEgens were dissolved in DMSO solution to give 50 mM stock solution. After 24 h, adding specific amount of above stock solution into cell culture medium to gave desired concentration and incubated for 1 h. Then cells were treated with fresh DMEM and further cultured for 4 h in the dark. After that, replaced with fresh DMEM medium and the cells were incubated with MTT for another 4 h. The formed formazan crystals were solubilized in 100 μL of lysate buffer. Absorbance at 570 nm of each well was measured on a Spectra Max M384 (Molecular Devices) and the data was recorded using Softmax Pro 6.4 software.

Cell culture and imaging. HeLa cells were cultured in DMEM containing 10% FBS, and incubated at 37 °C in 5% CO_2 atmosphere. After the cells reached to 80% confluence, the medium was removed and the adherent cells were washed once with PBS buffer to remove the remnant growth medium. Then **MTMM** (or **PTMM**) stock solution (2 mM in DMSO) was added into the cell plates to give final concentration of 20 μM (for **MTMM** or **PTMM**). The cells were incubated for 60 min and washed two times with PBS buffer. Then the resulted cells were used for bio-imaging under confocal laser scanning microscope (CLSM). **MTMM** and **PTMM** were excited at 405 nm and the emission was collected at range of 500-555 nm. No background fluorescence of cells was detected under the setting condition.

Co-localization experiments. The HeLa cells were first incubated with **MTMM** or **PTMM** (20 μM) for 60 min. Then the cells were washed once with PBS buffer and incubated with LysoTracker Red (50 nM) or MitoTracker Deep Red (100 nM) for 30 min. After that, the cells were further washed two times with PBS buffer and used for bio-imaging under CLSM. **MTMM** or **PTMM** were excited at 405 nm and the emission was collected at range of 500-

555 nm, LysoTracker Red was excited at 559 nm and the emission was collected at range of 570-670 nm, MitoTracker Deep Red was excited at 635 nm and the emission was collected at range of 655-755 nm. No background fluorescence of cells was detected under the setting condition.

Wash-free staining experiments. HeLa cells were incubated with **PTMM** (20 μM) for 60 min (or LysoTracker Red (50 nM) for 30 minutes). After that, the cells were imaged directly using CLSM without washing.

Extracellular ROS detection. The ROS generation was measured using DCF-DA as an indicator since the emission of DCF-DA increases upon reaction with ROS. For ROS detection, the DCF-DA (5 μM) was mixed with the **MTMM** (or **PTMM**) (20 μM) in DMSO/PBS buffer (1:99, v/v) and exposed to LED white light irradiation (25 mW cm^{-2}). The oxidation of DCF-DA was monitored by the emission increase at 525 nm.

Extracellular singlet oxygen detection. The singlet oxygen generation was measured using 9,10-anthracenediyl bis-(methylene)-dimalonic acid (ABDA) as an indicator since the absorbance of ABDA decreases upon reaction with singlet oxygen. For singlet oxygen detection, the ABDA (100 μM) was mixed with the **MTMM** (or **PTMM**) (20 μM) in DMSO/PBS buffer (1:99, v/v) and exposed to white light irradiation (25 mW cm^{-2}). The decomposition of ABDA was monitored by the absorbance decrease at 378 nm.

$^1O_2$ quantum yield measurements. The ABDA was used as $^1O_2$-sensitive indicator, and Rose Bengal (RB) was employed as the standard photosensitizer. In general, the ABDA solution was added into sample solution (100 μM), and the white light (25 mW cm^{-2}) was used as the irradiation source. The absorbance of ABDA at 378 nm was recorded at different irradiation times to obtain the decay rate of the photosensitizing process. The $^1O_2$ quantum yield of the desired PS in water was calculated using the following formula (**Eq. 1**):

$$\Phi_{PS} = \Phi_{RB}\frac{K_{PS}A_{RB}}{K_{RB}A_{PS}} \qquad \textbf{Eq. 1}$$

where K_{PS} and K_{RB} represent the decomposition rate constants of ABDA by the **MTMM** (or PTMM) and RB, respectively. A_{PS} and A_{RB} represent the light absorbed by the **MTMM** (or PTMM) and RB, respectively, and were determined by integrating of the areas under the absorption bands in the wavelength range of 400-600 nm. Φ_{RB} is the $^1O_2$ quantum yield of RB, which is 0.75 in water.

Intracellular ROS detection. Intracellular ROS generation under white light irradiation was detected using DCF-DA as an indicator. HeLa cells were cultured in DMEM with 10% FBS at 37 °C under 5% CO_2 atmosphere. After cells reached 80% confluence, the culture medium was removed and washed twice with PBS buffer and the cells were treated with DCF-DA (200 μM) for 30 min under dark and washed once with PBS buffer. The cells were then incubated with **MTMM** or **PTMM** (20 μM) for 60 min under dark. Then the cells were washed twice with PBS buffer and exposed to white light irradiation (25 mW cm^{-2}) for different time. For control experiment, the cells were only incubated with DCF-DA (200 μM) for 30 min, then cells were washed once with PBS buffer and exposed to white light irradiation (25 mW cm^{-2}) for different time. Under CLSM, DCF-DA was excited at 488 nm and the emission was collected at range of 500-550 nm. No background fluorescence of cells was detected under the setting condition.

Cytotoxicity of PDT process. To determine the cytotoxicity induced by PDT process, a MTT-based cell viability assay was performed in 96-well plates. HeLa cells were seeded at a density of 10000 cells per well and then incubated for 24 h. The AIEgens were dissolved in DMSO solution to give 50 mM stock solution. After 24 h, adding specific amount of above stock solution into cell culture medium to gave desired concentration and incubated for 1 h. Then replaced by fresh DMEM and selected wells were exposed to white light irradiation (25 mW cm^{-2}, 60 min) and further cultured for 3 h under dark. After that, replaced with fresh DMEM medium and the cells were incubated with MTT for another 4 h. The formed formazan crystals were solubilized in 100 μL of lysate buffer. Absorbance at 570 nm of each

well was measured on a Spectra Max M384 (Molecular Devices) and the data was recorded using Softmax Pro 6.4 software.

Cell apoptosis detection. Adherent HeLa cells were pre-incubated with **MTMM** (20 μM) or **PTMM** (20 μM) for 60 min at 37°C. The cells were further stained with annexin V-FITC and propidium iodide following the protocols of the manufacturer (Life Technologies) and imaged with the elapse of time after white light irradiation (25 mW cm^{-2}, 60 min). For control experiment, adherent HeLa cells were pre-incubated with 20 μM **MTMM** or **PTMM** for 60 min at 37°C and then stained with annexin V-FITC and propidium iodide following the protocols of the manufacturer (Life Technologies) and imaged without white light irradiation. Annexin V-FITC was excited at 488 nm and the emission was collected at range of 500-550 nm. PI was excited at 559 nm and the emission was collected at range of 570-670 nm.

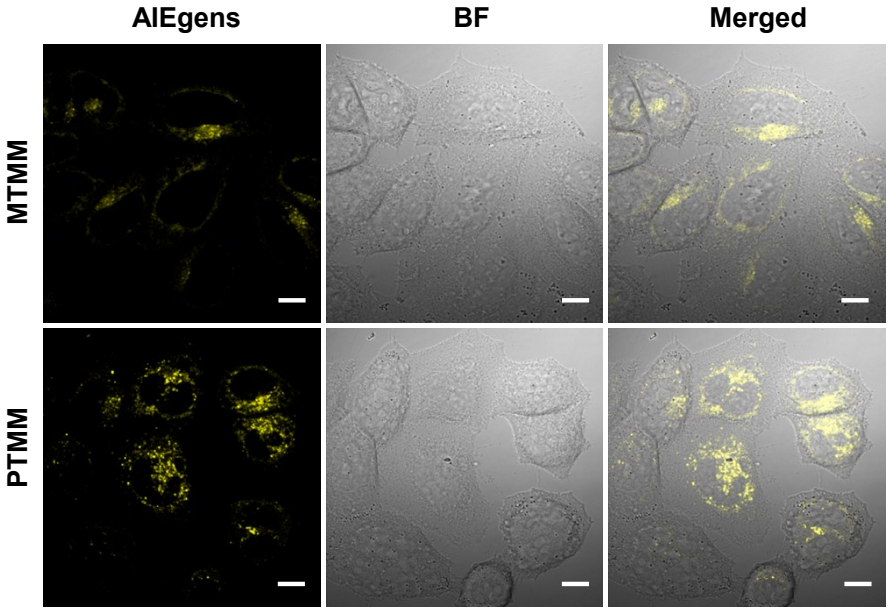


Fig. S4 CLSM images of HeLa cells after stained with **MTMM** (20 μM) and **PTMM** (20 μM). Scale bars: 10 μm.

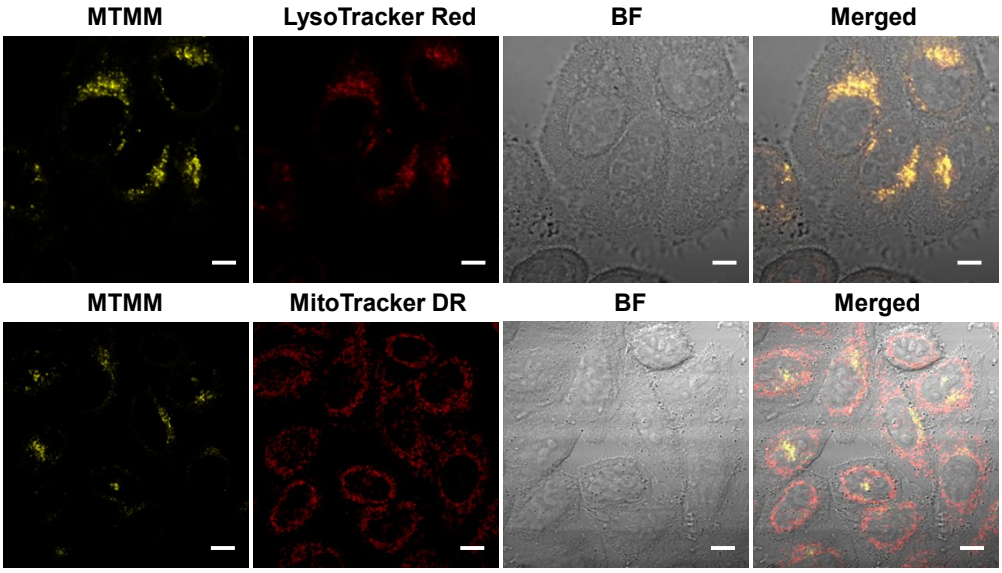


Fig. S5 Co-localization CLSM images of HeLa cells stained with **MTMM** (20 μM) and LysoTracker Red (50 nM) or MitoTracker Deep Red (100 nM). Scale bar: 10 μm.

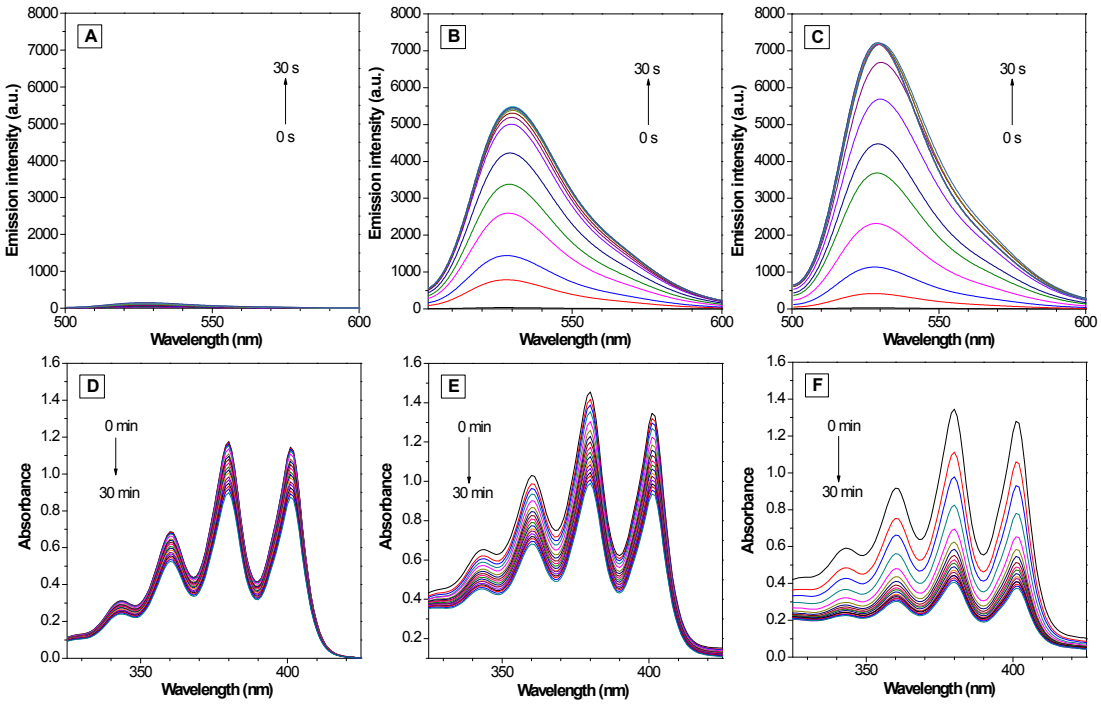


Fig. S6 Change of emission spectra for DCF-DA (5 μM) (A) without AIEgens (B) in the presence of **MTMM** (20 μM) and (C) in the presence of **PTMM** (20 μM) in PBS buffer after different durations under white light irradiation. Change of absorption spectra for ABDA (100 μM) (D) without AIEgens (E) in the presence of **MTMM** (20 μM) and (F) in the presence of **PTMM** (20 μM) in PBS buffer after different durations under white light irradiation.

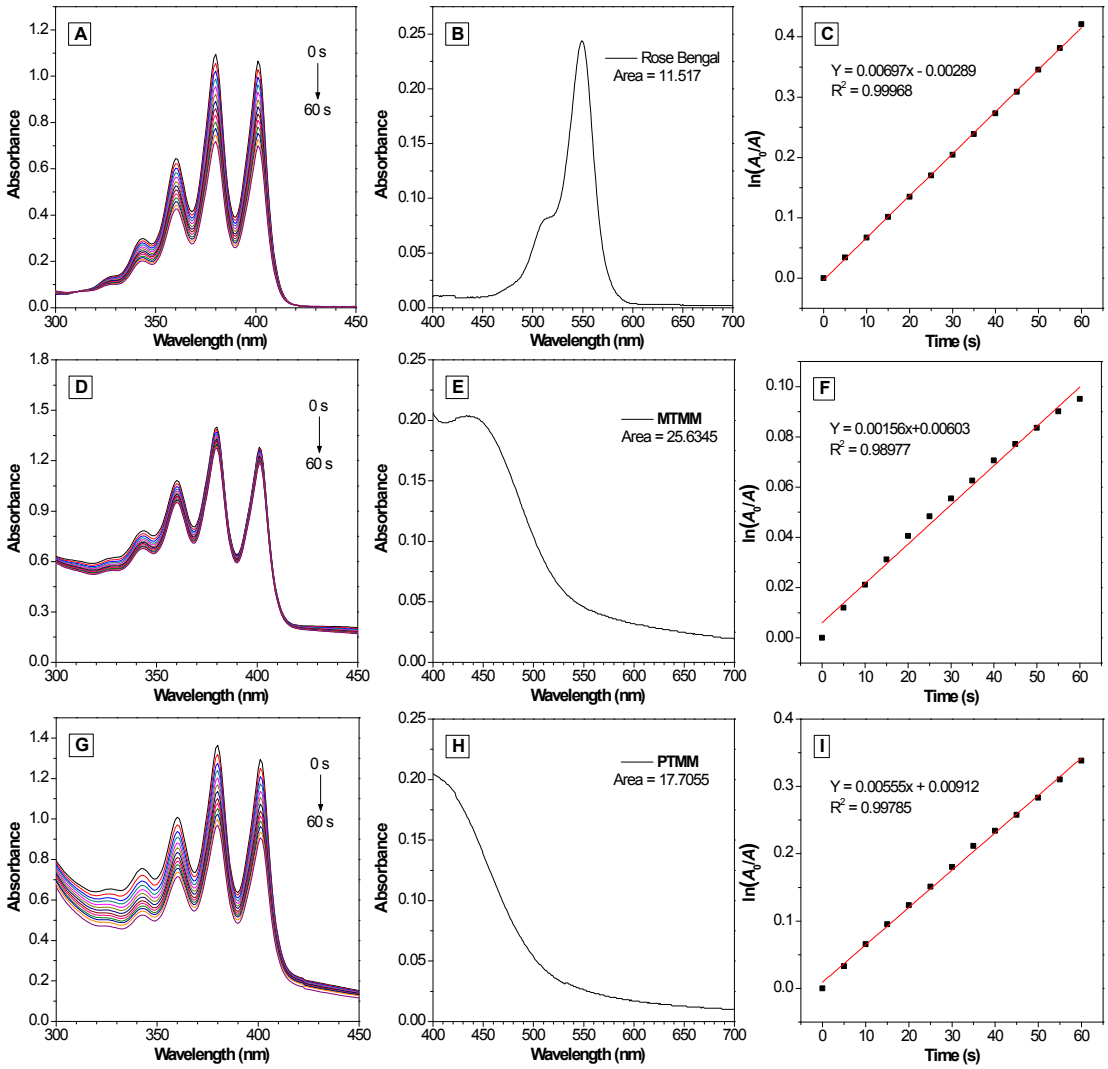


Fig. S7 Photodegradation of ABDA with RB (A), **MTMM** (D), and **PTMM** (G). The absorption peak area of RB (B), **MTMM** (E), and **PTMM** (H); The decomposition rate constants of RB (C), **MTMM** (F), and **PTMM** (I). To eliminate the inner-filter effect, the absorption maxima were adjusted to ~0.2 OD.

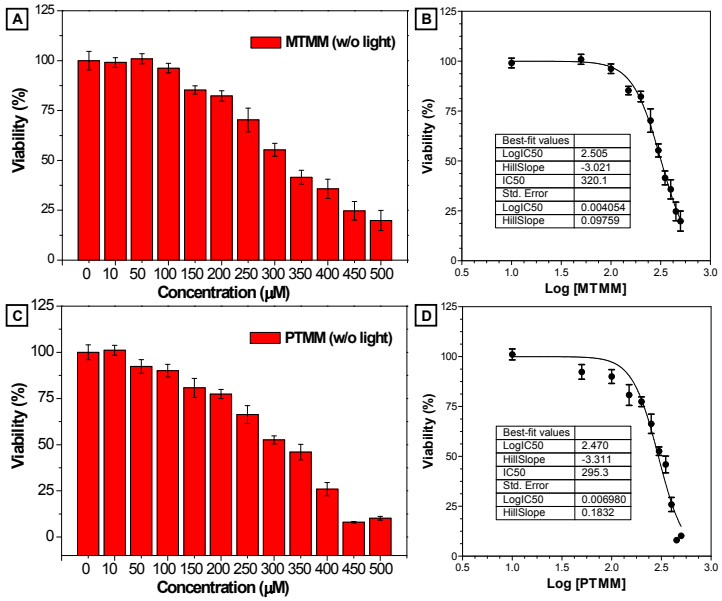


Fig. S8 (A) The dark-toxicity of **MTMM** towards HeLa cells. (B) The curve of IC_{50} of **MTMM** in the dark. (C) The dark-toxicity of **PTMM** towards HeLa cells. (D) The curve of IC_{50} of **PTMM** in the dark. Error bars: mean ± SD (n = 7). Inset in B and D: IC_{50} values of **MTMM** and **PTMM**.

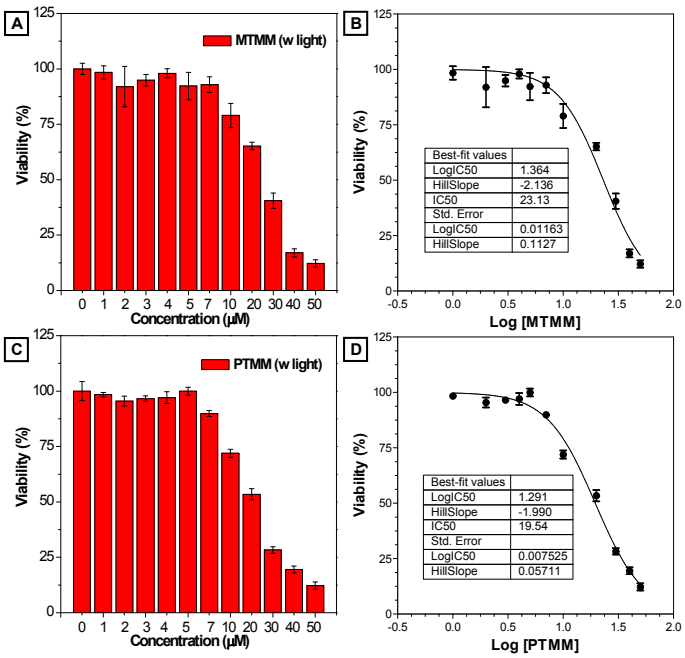


Fig. S9 (A) The photo-toxicity of **MTMM** towards HeLa cells. (B) The curve of IC_{50} of **MTMM** upon white light irradiation (25 mW cm^{-2}, 60 min). (C) The photo-toxicity of **PTMM** towards HeLa cells. (D) The curve of IC_{50} of **PTMM** upon white light irradiation (25 mW cm^{-2}, 60 min). Error bars: mean ± SD (n = 7). Inset in B and D: IC_{50} values of **MTMM** and **PTMM**.

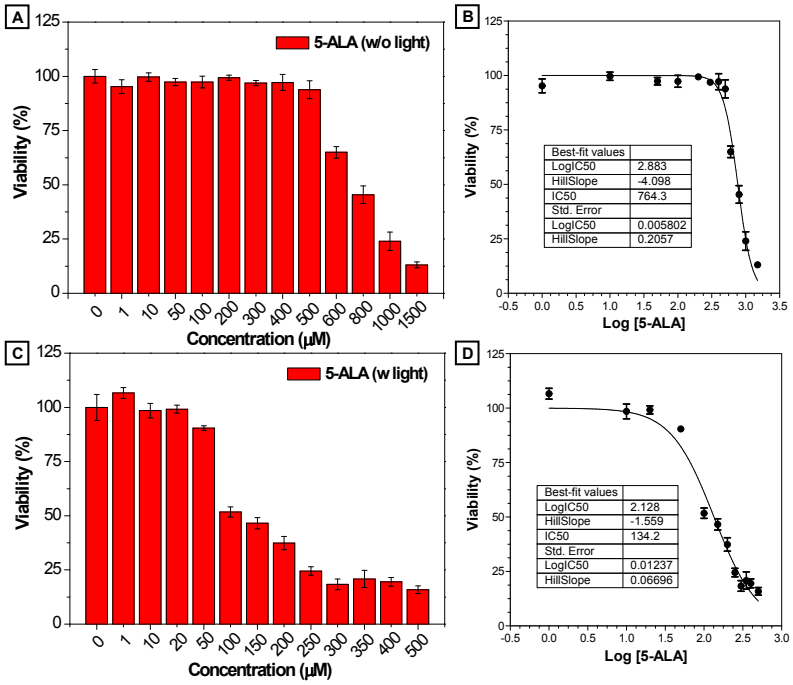


Fig. S10 (A) The dark-toxicity of 5-ALA towards HeLa cells. (B) The curve of IC_{50} of 5-ALA in the dark. (C) The photo-toxicity of 5-ALA towards HeLa cells. (D) The curve of IC_{50} of 5-ALA upon white light irradiation (25 mW cm^{-2}, 60 min). Error bars: mean ± SD (n = 6). Inset in B and D: IC_{50} values of 5-ALA in the dark and upon white light irradiation, respectively.

Table S6. Dark- and photo-toxicity of **MTMM**, **PTMM** and 5-ALA towards HeLa cells.

Compd.	HeLa cells		
	Dark[a]	Light[a]	PI[b]
MTMM	320.1	23.13	13.8
PTMM	295.3	19.54	15.1
5-ALA[c]	191.1	33.6	5.7

[a]IC_{50} values were an average of seven measurements. [b]PI refers to the phototoxicity index, which means the ratio between the IC_{50} values in the dark and upon white light irradiation (25 mW cm^{-2}, 60 min). [c]The measured IC_{50} values were adjusted since four 5-ALA molecules are needed to form the active species of protoporphytin IX.

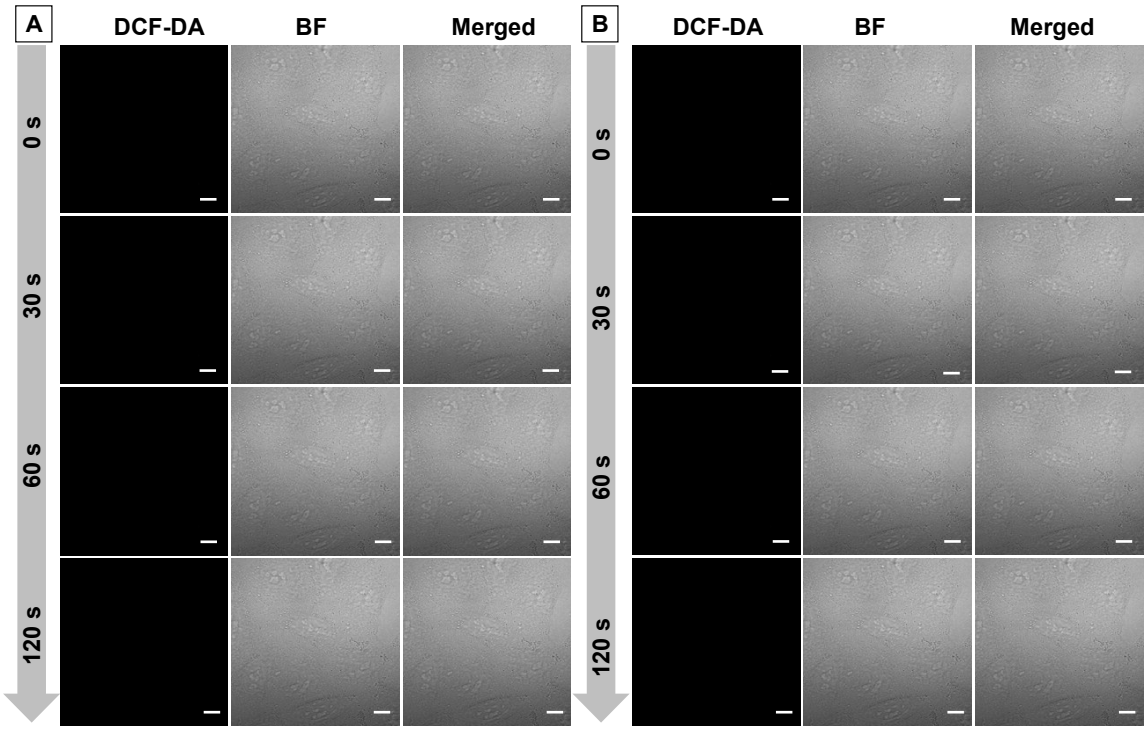


Fig. S11 Intracellular ROS detection by CLSM after HeLa cells were stained with DCF-DA (200 μM) in the absence of (A) **MTMM** and (B) **PTMM** under white light illumination (25 mW cm^{-2}) for different time. Scale bar: 10 μm.

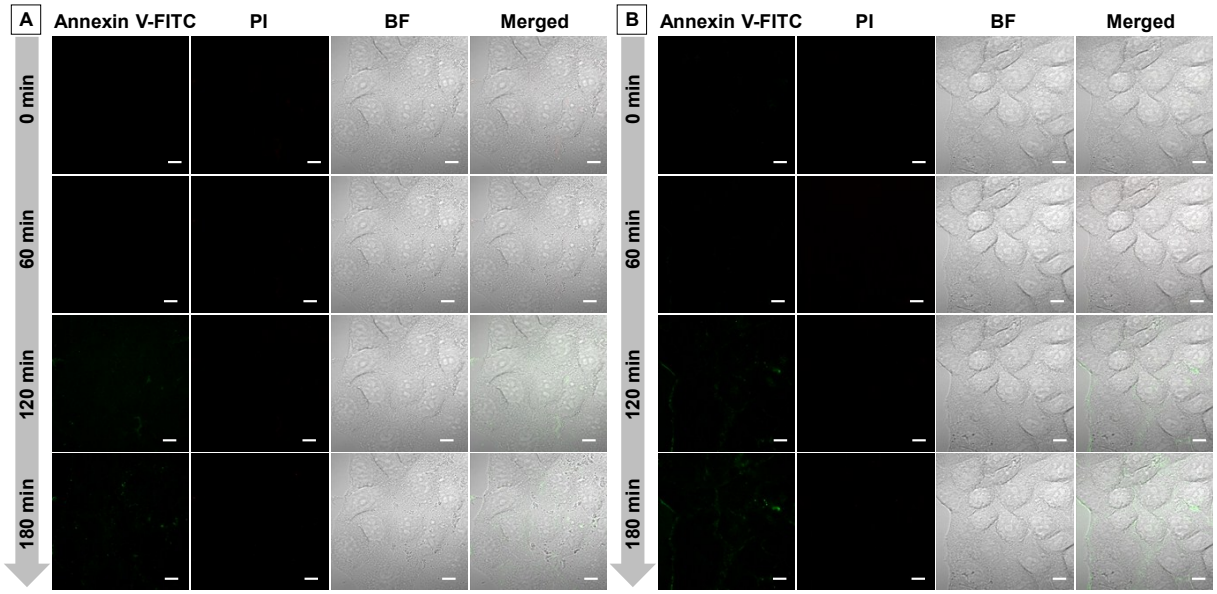


Fig. S12 HeLa cell apoptosis observed by CLSM images after pre-stained with Annexin V-FITC and PI in the absence of (A) **MTMM** and (B) **PTMM** under white light illumination (25 mW cm^{-2}). Scale bar: 10 μm.

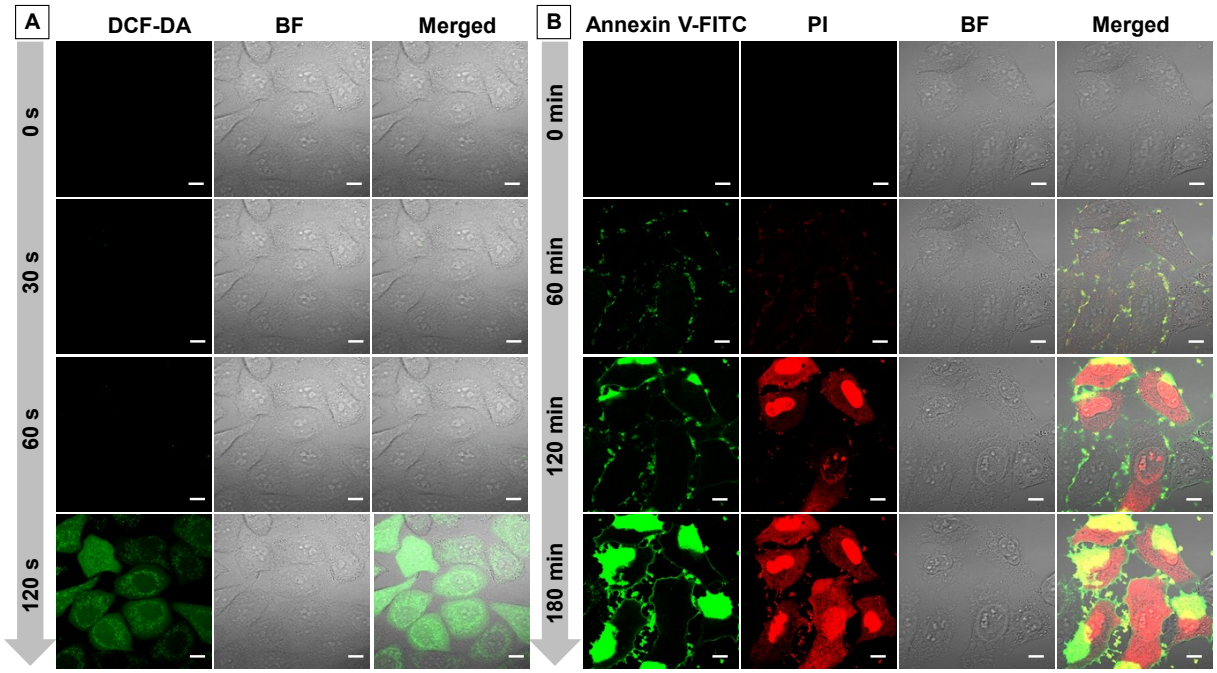


Fig. S13 HeLa cell apoptosis observed by CLSM images after treated with **MTMM** (20 μM) under white light illumination (25 mW cm^{-2}). Scale bar: 10 μm.

6. Bacterial Imaging and Killing of Bacteria

Bacterial staining experiment. A single colony of *S. aureus* or *E. coli* that picked from LB or BHI plates was incubated in 2YT or BHI medium overnight with shaking at 37 ^{o}C. After that, the bacterial culture was centrifuged at 4000 rpm at 4 ^{o}C for 10 min and the cell pellet was re-suspended in 0.9% NaCl solution. This washing procedure was repeated twice. The final concentration of bacteria was around 2×10^{7}/mL and then the bacteria were transferred into a 1.5 mL EP tube. Then bacteria were harvested by centrifuging at 4000 rpm for 5 min. After removal of the supernatant, 1 mL of the **MTMM** or **PTMM** (5 μM) solution was added into EP tube that contained bacteria and allowed to inbubate at room temperature for 10 min. To take CLSM images, about 10 μL of above stained bacteria solution was transferred on to a glass slide and then covered by a coverslip. **MTMM** and **PTMM** were excited at 405 nm and the emission was collected at range of 500-555 nm. No background fluorescence of bacteria cell was detected under the setting condition.

Bacterial killing experiments. The antibacterial activity of **MTMM** and **PTMM** were determined by incubating 0.9% NaCl solution of *E. coli* (~1.5×10^{5} CFU mL^{-1}) or S. aureus (~5×10^{5} CFU mL^{-1}) suspensions with **MTMM** or **PTMM** (5 μM) at room temperature for 60 min under dark. Then the bacteria were either exposed to white light irradiation (25 mW cm^{-2}, 60 min) or incubated under dark for 60 min. After that, the bacteria suspensions were serially diluted by 100-fold with 0.9% NaCl solution, Then picked 100 μL diluted bacterial and spread on the agar plate (1.2% agar + brain heart infusion for *S. aureus* and 1.2 % agar + Lysogeny Broth for *E. coli*) and incubated at 37 ^{o}C overnight.

The antibacterial activities of **MTMM** or **PTMM** were evaluated based on the reduced ratio of colonies. The bacteria colonies on the agar plates were counted and the reduced ratio was calculated based on the equation [(A - B)/A] × 100%, where A is the mean number of bacteria colonies in the control sample (without AIE gens)，and B is the mean number of bacteria

after incubated with the AIEgens. The results were repeated for three times.

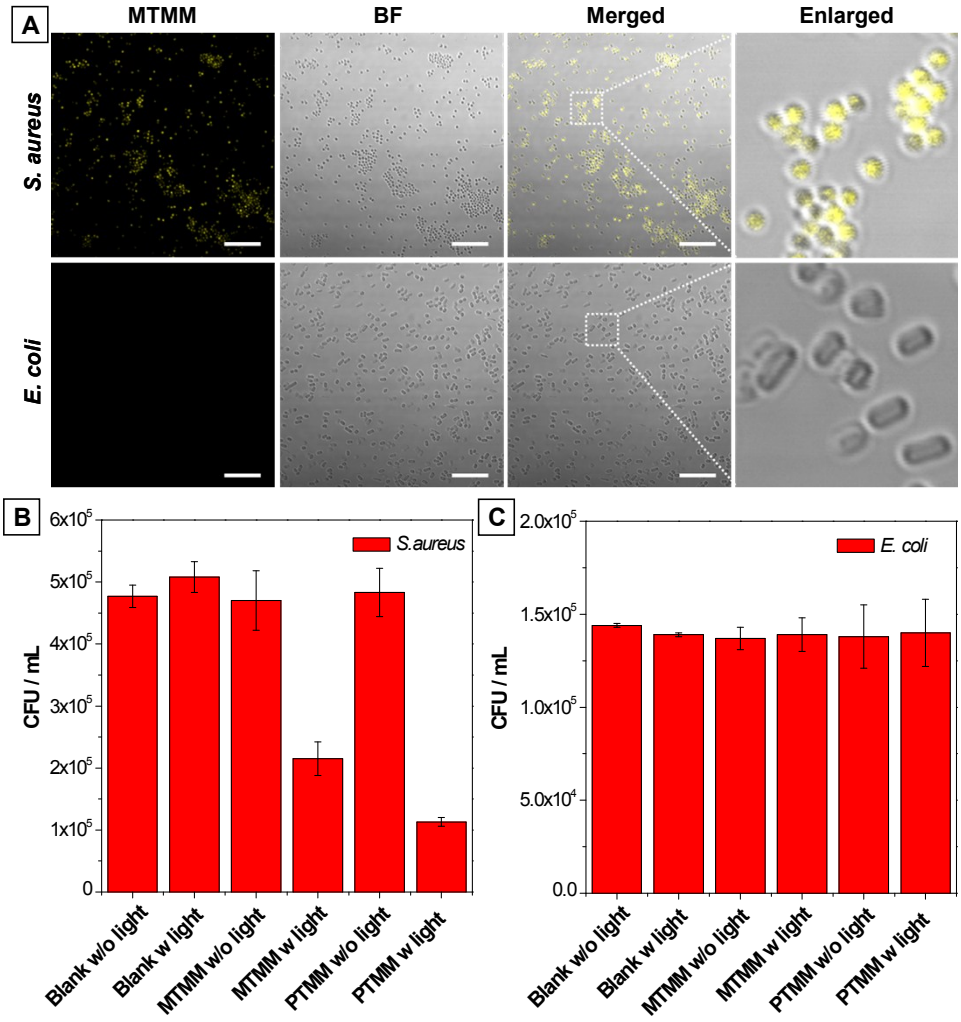


Fig. S14 (A) CLSM images of *S. aureus* and *E. coli* stained with **MTMM** (5 μM). (B) CFU of *S. aureus* treated with **MTMM** (5 μM) and **PTMM** (5 μM) in the dark or upon white light irradiation (25 mW cm^{-2}, 60 min). (C) CFU of *E. coli* treated with **MTMM** (5 μM) and **PTMM** (5 μM) in the dark or upon white light irradiation (25 mW cm^{-2}, 60 min). Scale bar in A: 10 μm. Error bars in B and C: mean ± SD (n = 3).

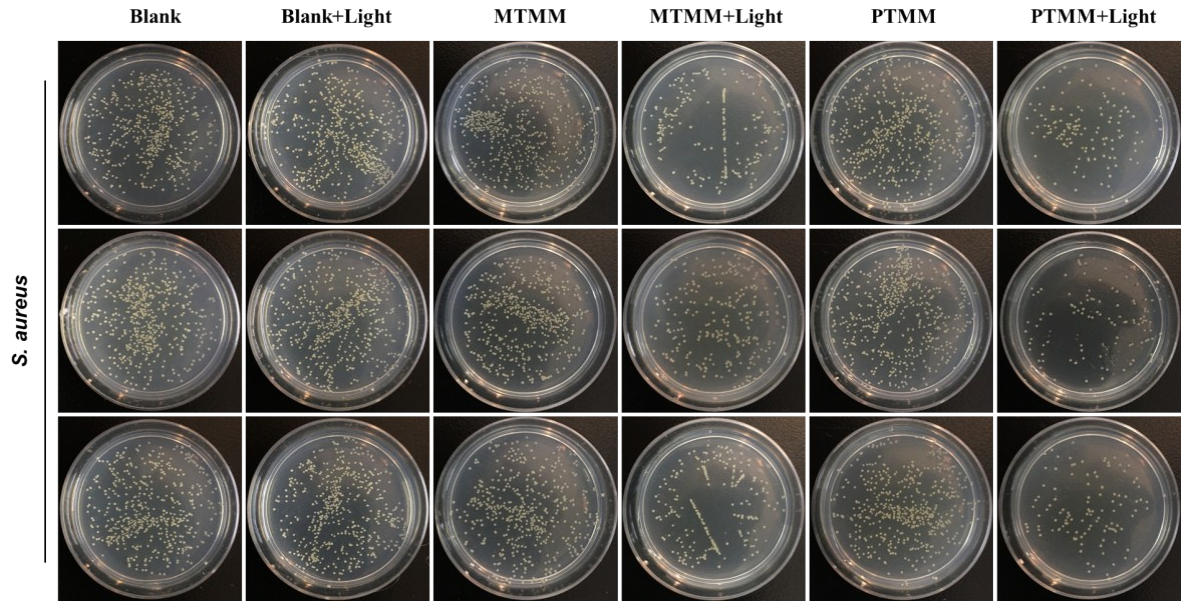


Fig. S15 Plates of *S. aureus* treated with/without **MTMM** (5 μM) and **PTMM** (5 μM) in the dark or upon white light irradiation (25 mW cm^{-2}, 60 min).

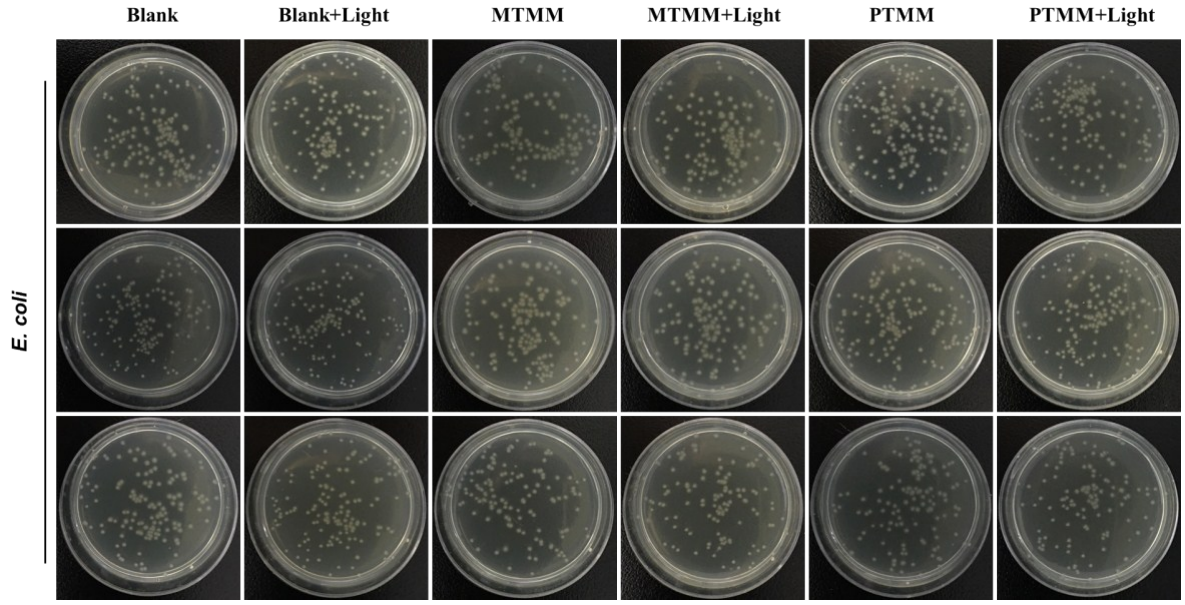


Fig. S16 Plates of *E. coli* treated with/without **MTMM** (5 μM) and **PTMM** (5 μM) in the dark or upon white light irradiation (25 mW cm^{-2}, 60 min).

7. NMR and Mass Spectra

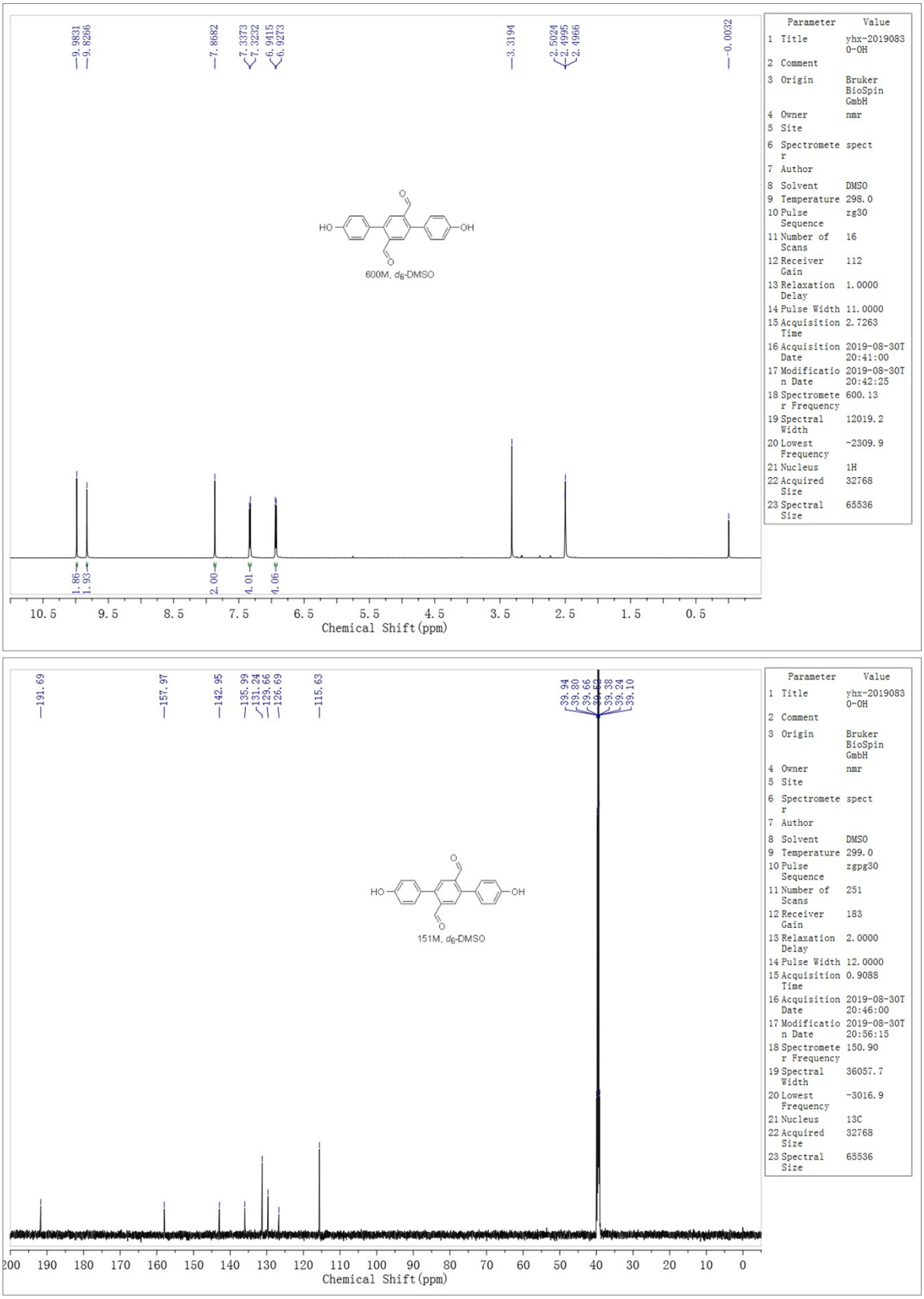
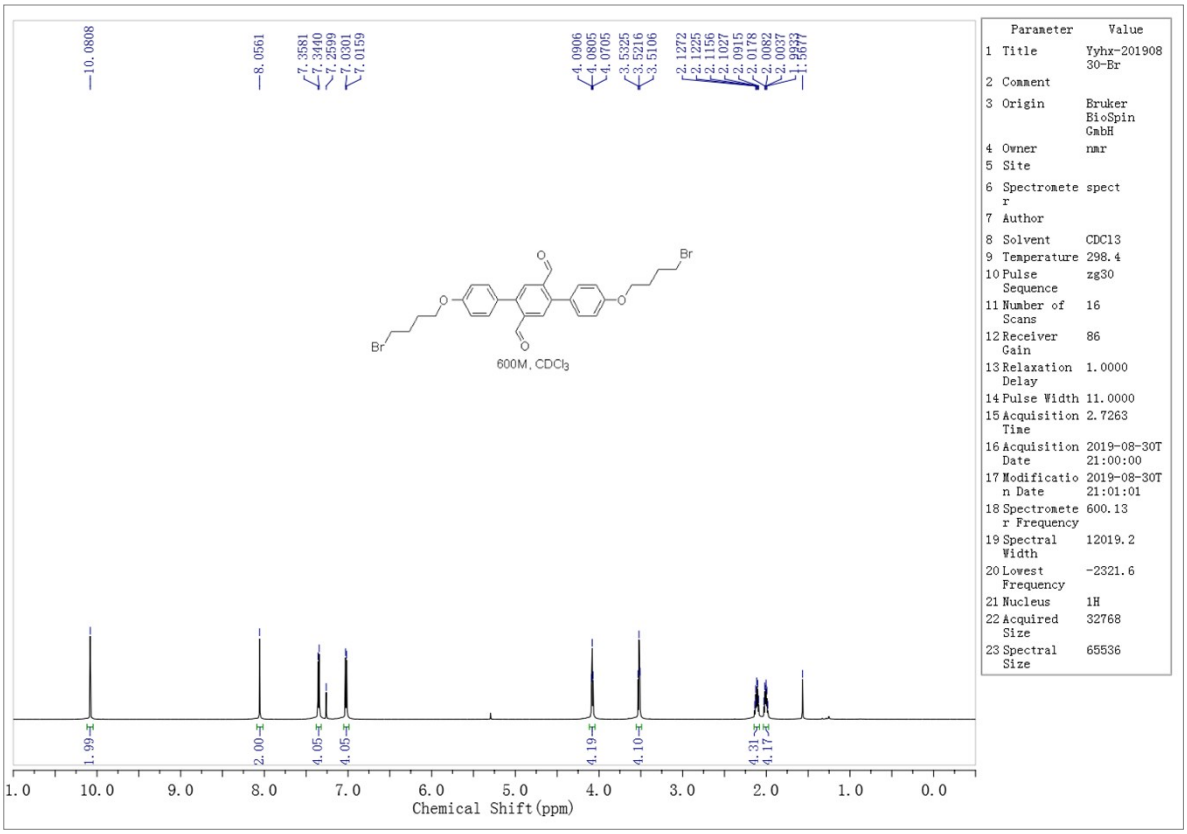


Fig. S17 ^{1}H NMR (d_6-DMSO, 600 MHz) and ^{13}C NMR (d_6-DMSO, 151 MHz) spectra of **HTC**.



	Parameter	Value
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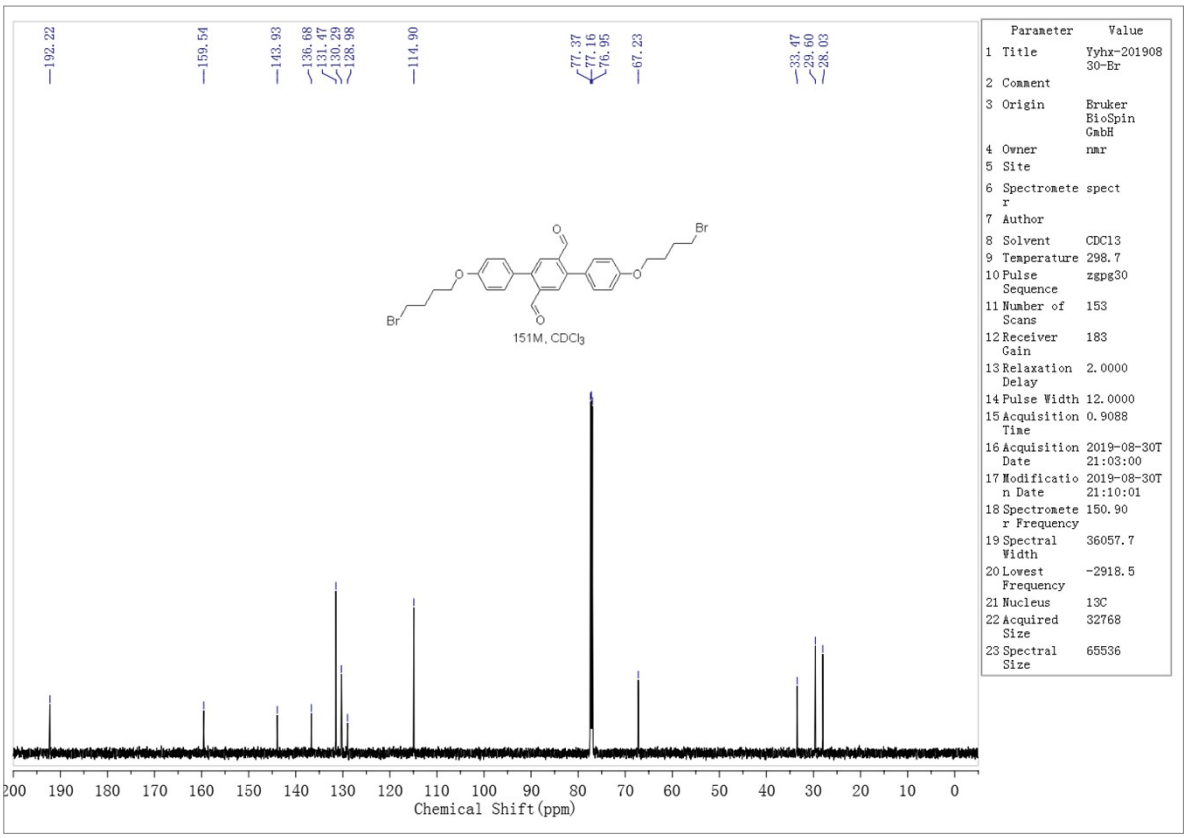
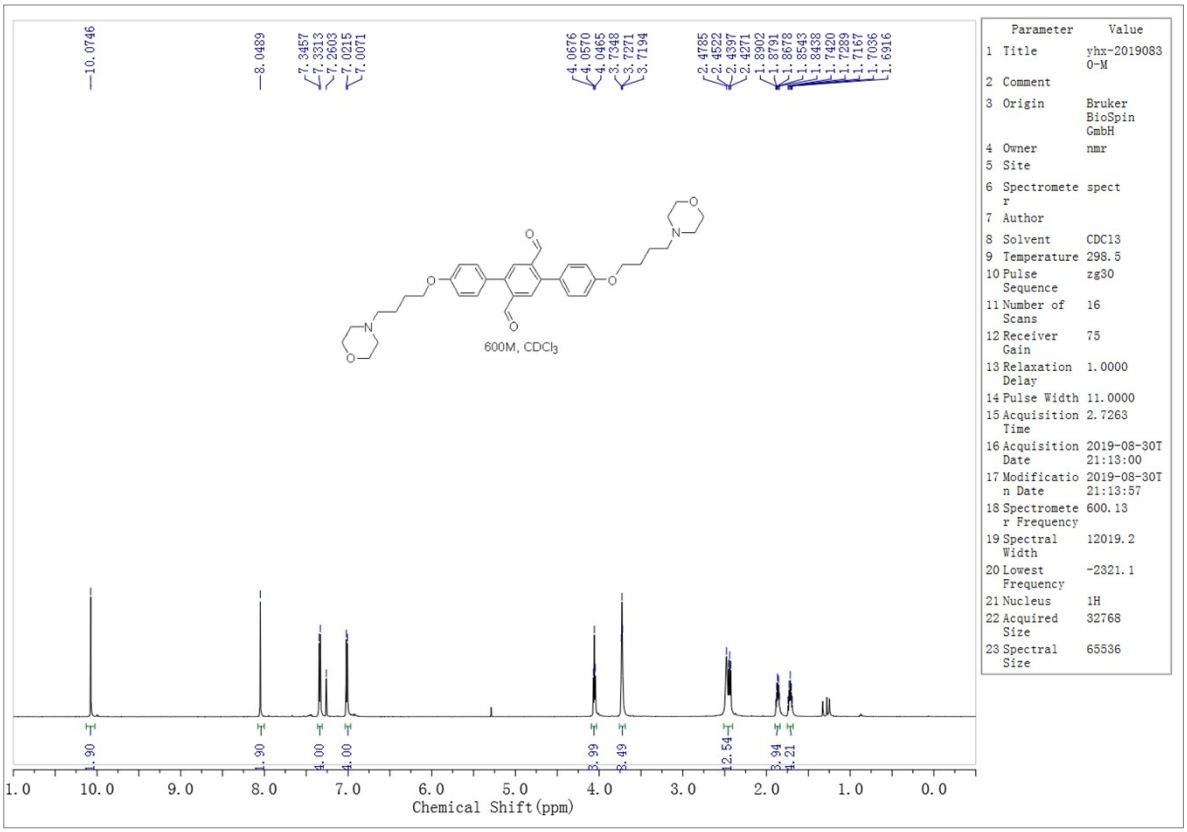
2	Comment	
3	Origin	Bruker BioSpin GmbH
4	Owner	nmr
5	Site	
6	Spectrometer	spect
7	Author	
8	Solvent	CDCl3
9	Temperature	298.4
10	Pulse Sequence	zg30
11	Number of Scans	16
12	Receiver Gain	86
13	Relaxation Delay	1.0000
14	Pulse Width	11.0000
15	Acquisition Time	2.7263
16	Acquisition Date	2019-08-30T21:00:00
17	Modification Date	2019-08-30T21:01:01
18	Spectrometer Frequency	600.13
19	Spectral Width	12019.2
20	Lowest Frequency	-2321.6
21	Nucleus	1H
22	Acquired Size	32768
23	Spectral Size	65536



	Parameter	Value
1	Title	Yyhx-20190830-Br
2	Comment	
3	Origin	Bruker BioSpin GmbH
4	Owner	nmr
5	Site	
6	Spectrometer	spect
7	Author	
8	Solvent	CDCl3
9	Temperature	298.7
10	Pulse Sequence	zgpg30
11	Number of Scans	153
12	Receiver Gain	183
13	Relaxation Delay	2.0000
14	Pulse Width	12.0000
15	Acquisition Time	0.9088
16	Acquisition Date	2019-08-30T21:03:00
17	Modification Date	2019-08-30T21:10:01
18	Spectrometer Frequency	150.90
19	Spectral Width	36057.7
20	Lowest Frequency	-2918.5
21	Nucleus	13C
22	Acquired Size	32768
23	Spectral Size	65536

Fig. S18 ^{1}H NMR ($CDCl_3$, 600 MHz) and ^{13}C NMR ($CDCl_3$, 151 MHz) spectra of **BTC**.



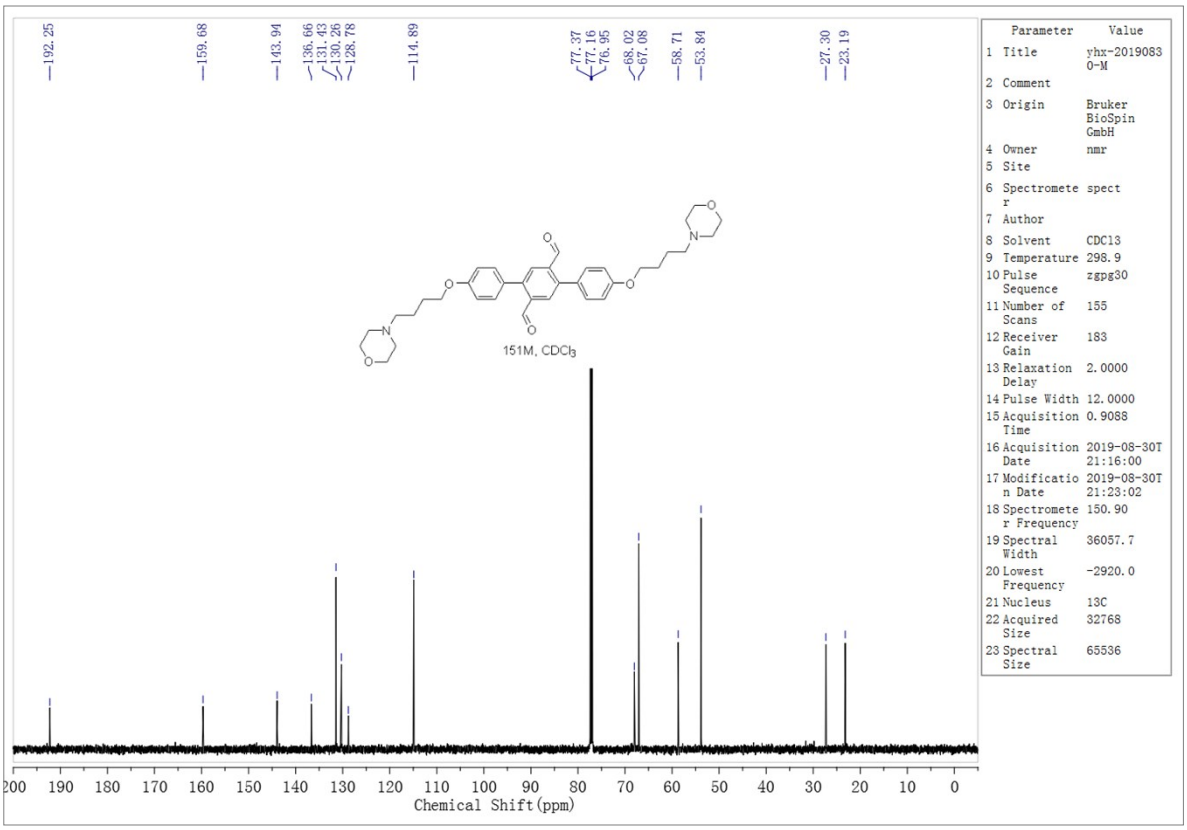
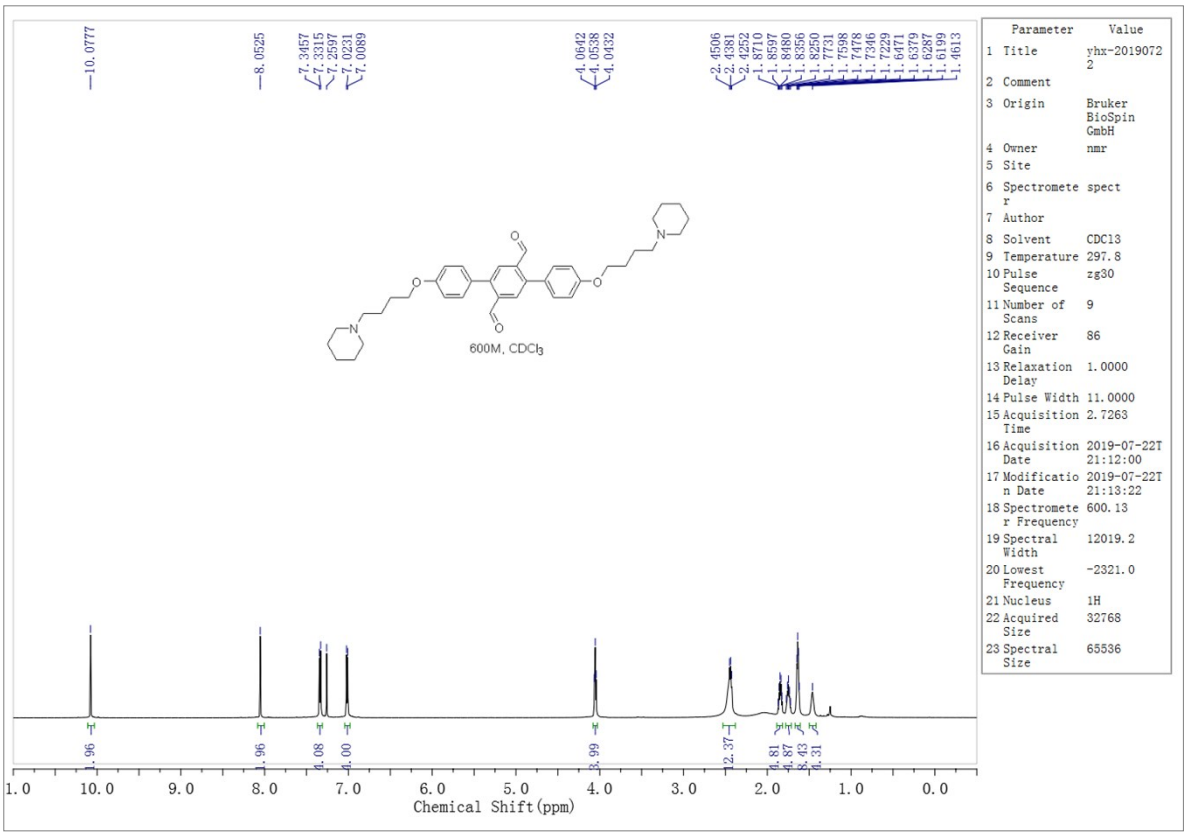
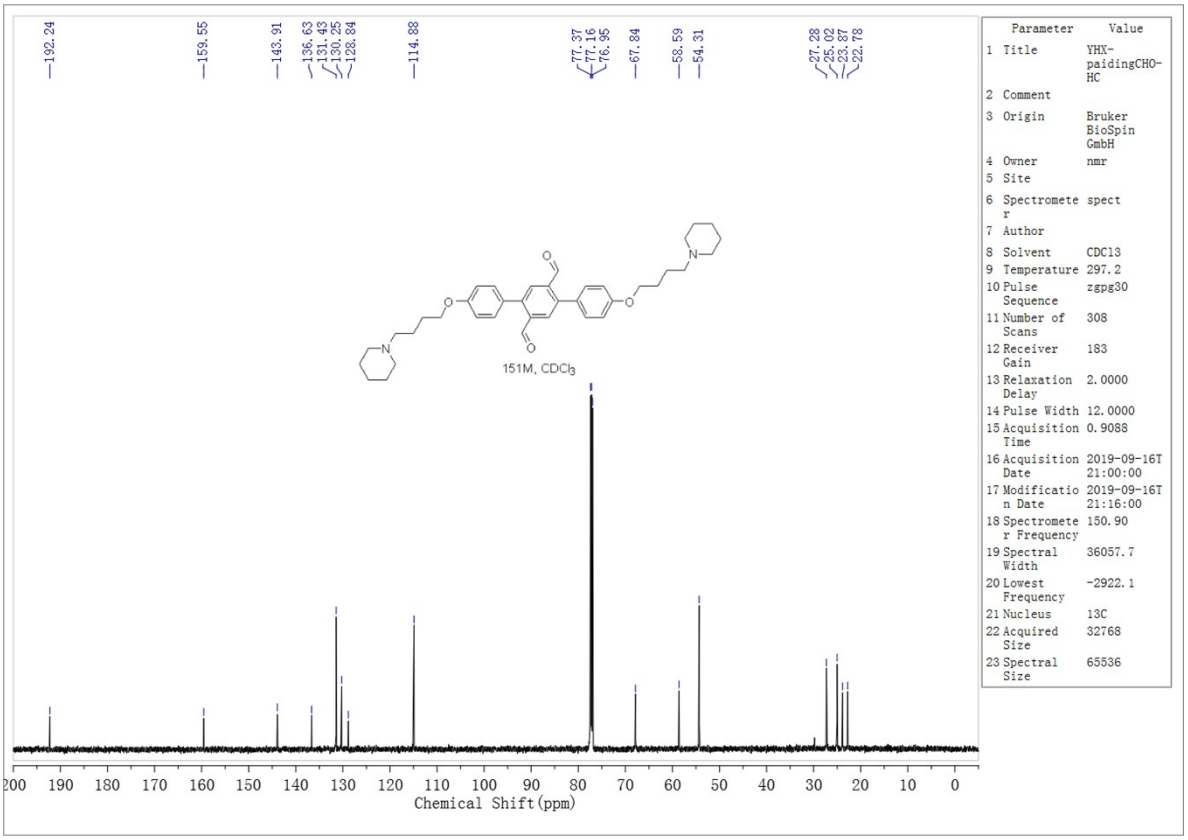


Fig. S19 ^{1}H NMR ($CDCl_3$, 600 MHz) and ^{13}C NMR ($CDCl_3$, 151 MHz) spectra of **MTC**.



Parameter	Value
1 Title	yhx-20190722
2 Comment	
3 Origin	Bruker BioSpin GmbH
4 Owner	nmr
5 Site	
6 Spectrometer	spect
7 Author	
8 Solvent	CDCl3
9 Temperature	297.8
10 Pulse Sequence	zg30
11 Number of Scans	9
12 Receiver Gain	86
13 Relaxation Delay	1.0000
14 Pulse Width	11.0000
15 Acquisition Time	2.7263
16 Acquisition Date	2019-07-22T 21:12:00
17 Modification Date	2019-07-22T 21:13:22
18 Spectrometer Frequency	600.13
19 Spectral Width	12019.2
20 Lowest Frequency	-2321.0
21 Nucleus	1H
22 Acquired Size	32768
23 Spectral Size	65536



Parameter	Value
1 Title	YHX-paidingCHO-HC
2 Comment	
3 Origin	Bruker BioSpin GmbH
4 Owner	nmr
5 Site	
6 Spectrometer	spect
7 Author	
8 Solvent	CDCl3
9 Temperature	297.2
10 Pulse Sequence	zgpg30
11 Number of Scans	308
12 Receiver Gain	183
13 Relaxation Delay	2.0000
14 Pulse Width	12.0000
15 Acquisition Time	0.9088
16 Acquisition Date	2019-09-16T 21:00:00
17 Modification Date	2019-09-16T 21:16:00
18 Spectrometer Frequency	150.90
19 Spectral Width	36057.7
20 Lowest Frequency	-2922.1
21 Nucleus	13C
22 Acquired Size	32768
23 Spectral Size	65536

Fig. S20 ^{1}H NMR ($CDCl_3$, 600 MHz) and ^{13}C NMR ($CDCl_3$, 151 MHz) spectra of **PTC**.

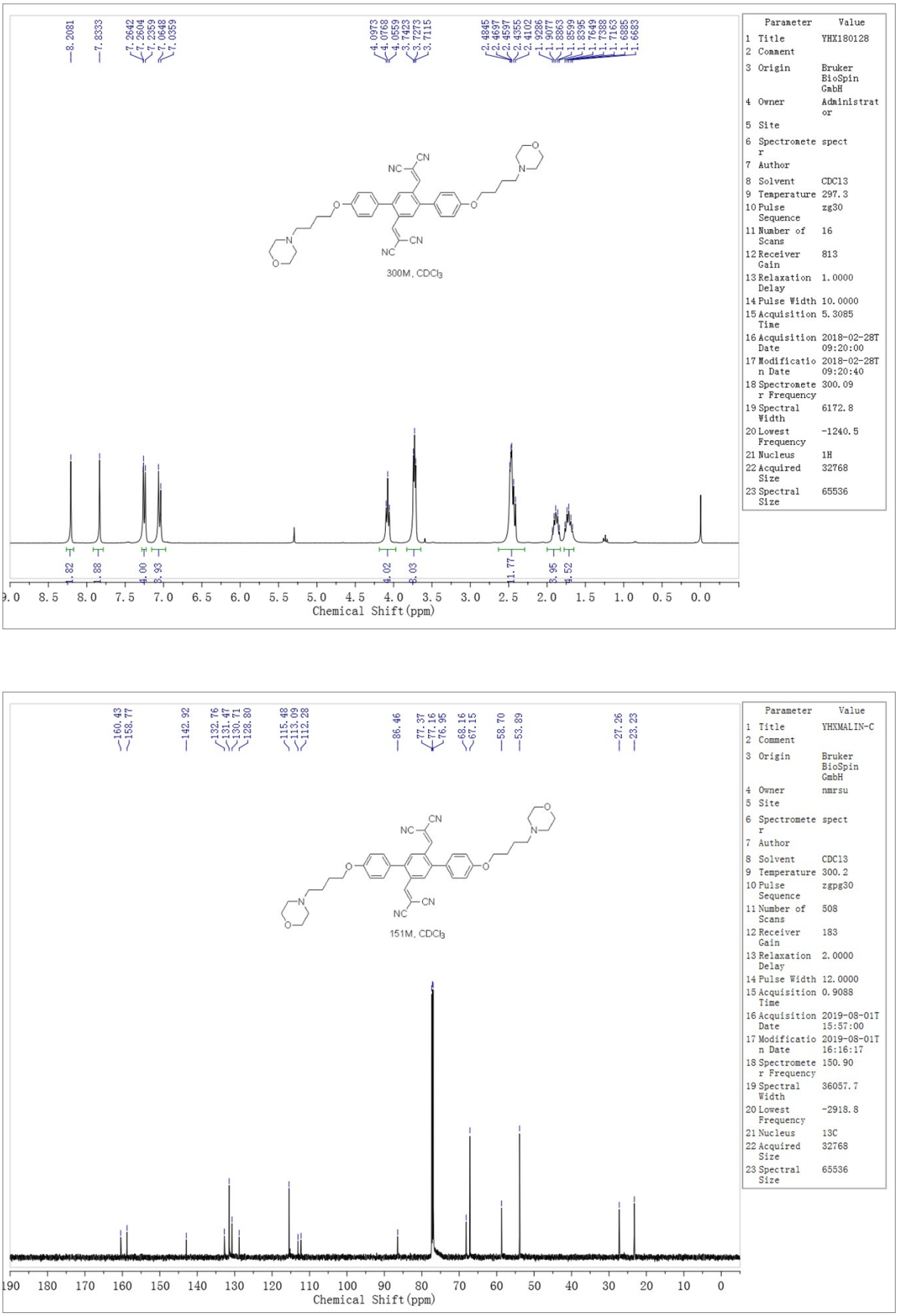


Fig. S21 ^{1}H NMR ($CDCl_3$, 300 MHz) and ^{13}C NMR ($CDCl_3$, 151 MHz) spectra of **MTMM**.

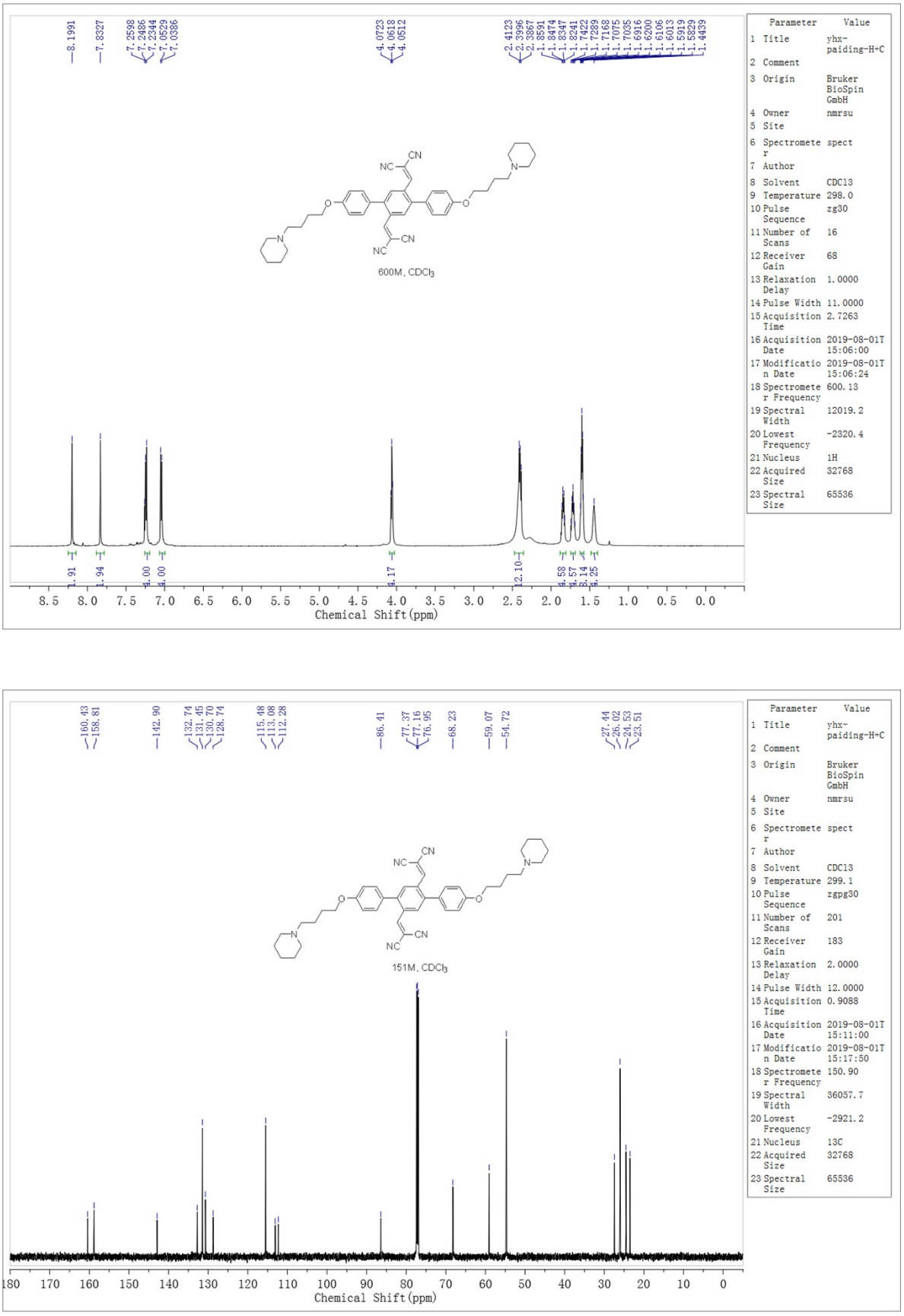


Fig. S22 ^{1}H NMR ($CDCl_3$, 600 MHz) and ^{13}C NMR ($CDCl_3$, 151 MHz) spectra of **PTMM**.

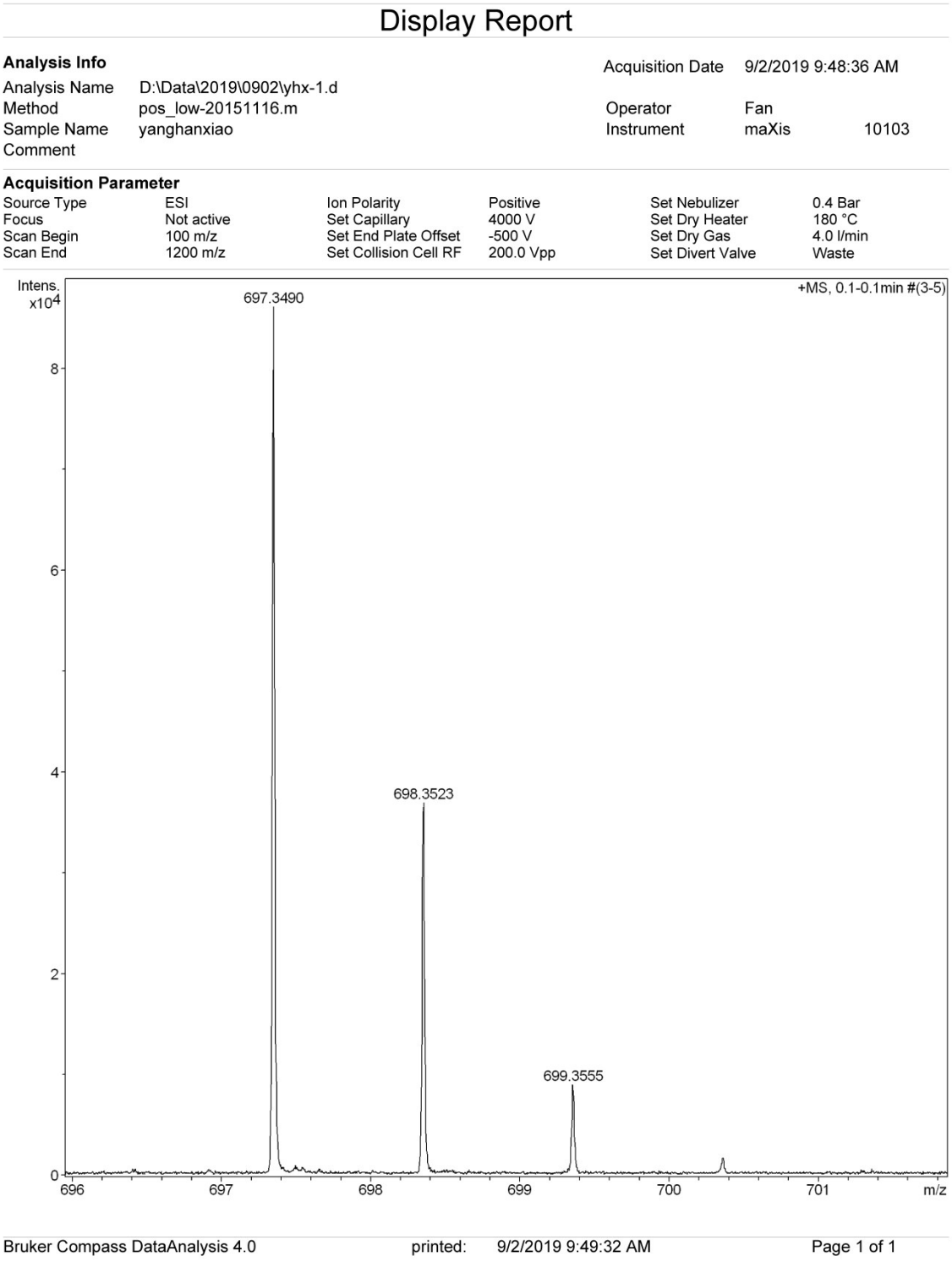


Fig. S23 High-resolution mass spectra of **MTMM**.

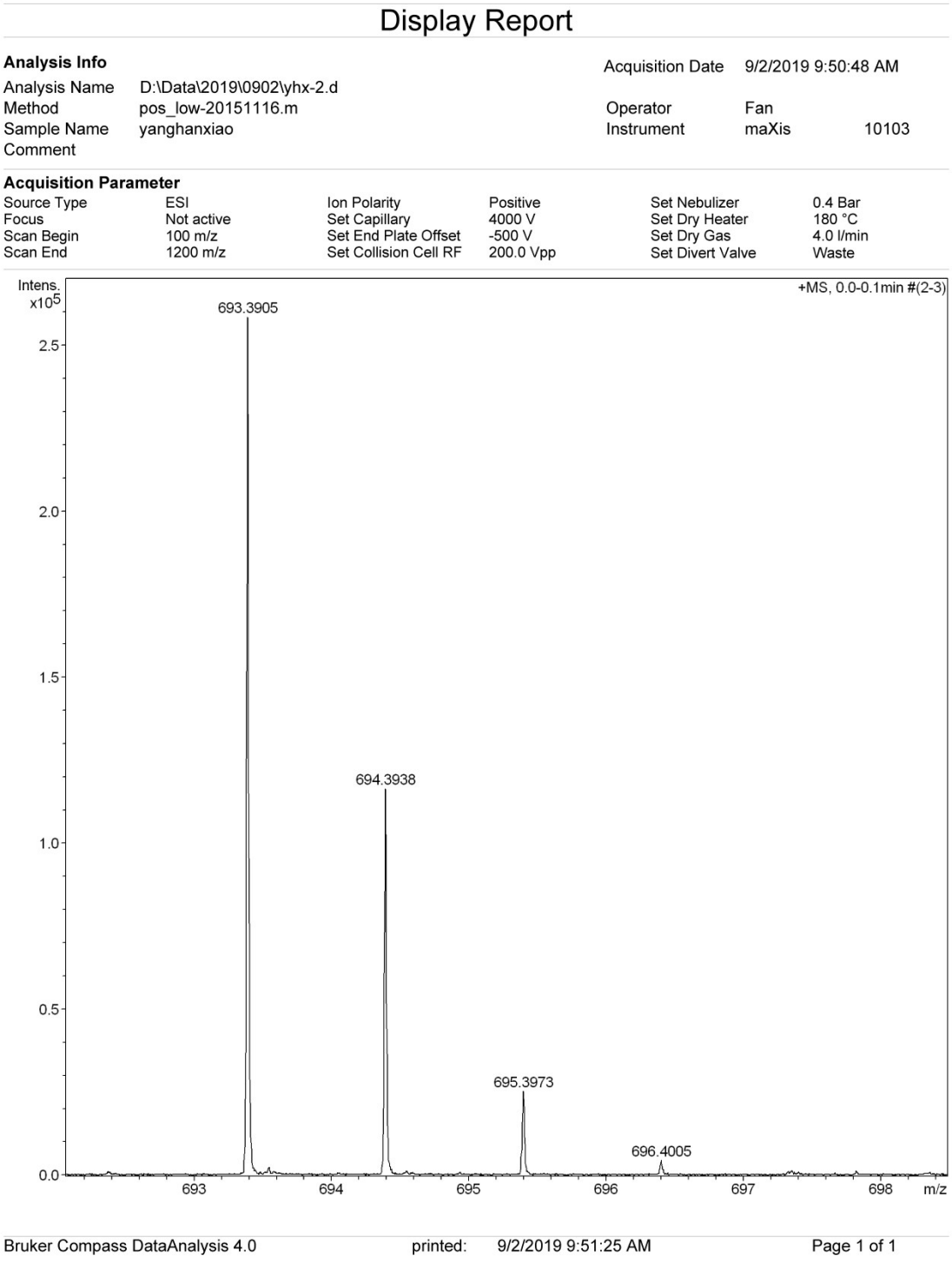


Fig. S24 High-resolution mass spectra of **PTMM**.