Supporting Information

Diketopyrrolopyrrole-based ratiometric fluorescent probe for the sensitive and selective detection of cysteine over homocysteine and glutathione in living cells

Xiao Zhang, Yandi Hang, Weisong Qu, Yongchao Yan, Ping Zhao*, Jianli Hua*

Key Laboratory for Advanced Materials, Institute of Fine Chemicals, East China University of Science and Technology, 200237, Shanghai, China

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Experimental section

Instrumentation and materials

^{1}H NMR and ^{13}C NMR spectra were recorded on a Bruker AM 400 MHz spectrometer, and tetramethyl silane (TMS) were used as an internal standard. Electrospray ionization and time-of-flight analyzer (ESI-TOF) mass spectra were determinated by the Waters Micromass LCT mass spectrometer. Absorption spectra were recorded on a Varian Cary 500 UV-vis spectraphotometer. Fluorescence spectra were measured on a Horiba Fluoromax-4 Fluorescence spectrometer.

N, *N*-Dimethylformamide (DMF) was refluxed with calcium hydride and distilled before use. Tetrahydrofuran (THF) was pre-dried over 4 Å molecular sieves and distilled under an argon atmosphere from sodium sodium benzophenone ketyl immediately before use. All other reagents and reactants were purchased as commercial products from Sigma-Aldrich and used as received without further purification.

Absorption and fluorescence spectra were measured in DMSO-buffer solution (PBS: DMSO = 6: 4, v/v, pH = 7.4) at 37 ℃.

Compound **1** and 4-(bis(4-methoxyphenyl)amino)phenyl boronic acid were prepared according to previous literature protocols [1, 2].

Intracellular imaging

Hela cells were cultured at 37 ºC and 5% CO_2 air condition and maintained 24 h before imaging by the confocal laser scanning miroscopy. The cells was seeded in 14 mm glass culture flasks and allowed to adhere for 12 h.

Time-dependent fluorescent spectra of DPP-AC towards Cys

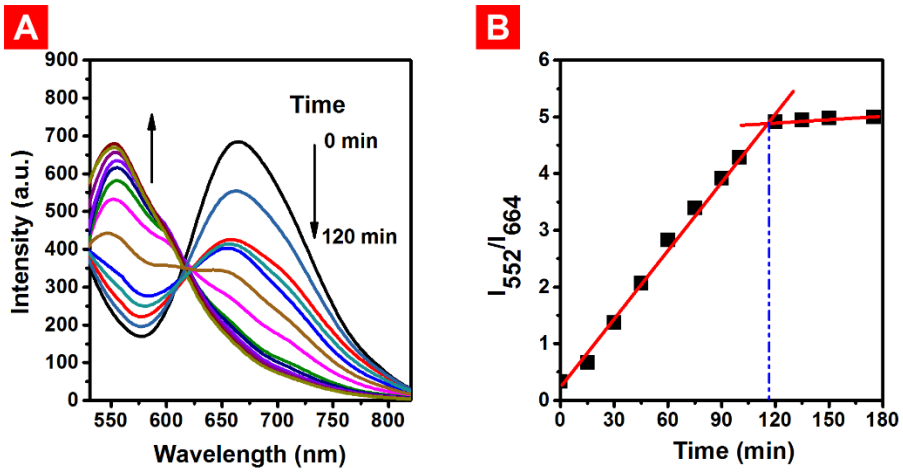


Figure S1. (A) Time-dependent fluorescent spectra of **DPP-AC** (10 μM) towards Cys (90 μM) with λ_{ex} = 510 nm in DMSO-buffer solution (PBS: DMSO = 6: 4, v/v, pH = 7.4) at 37 ℃. (B) Linear correction between the fluorescence ratio (I_{552}/I_{664}) and reaction time (0-180 min) with λ_{ex} = 505 nm in DMSO-buffer solution (PBS: DMSO = 6: 4, v/v, pH = 7.4) at 37 ℃.

MS titration of DPP-AC with Cys

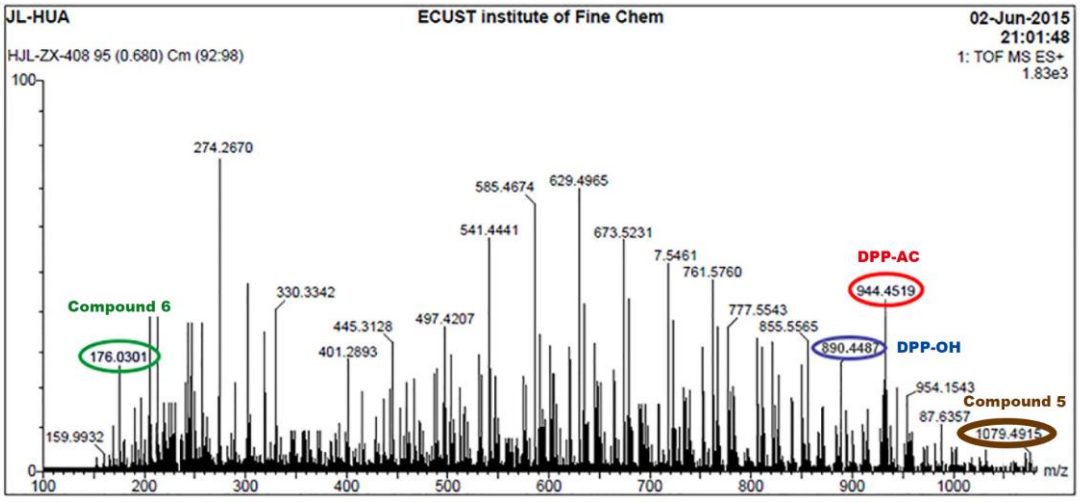
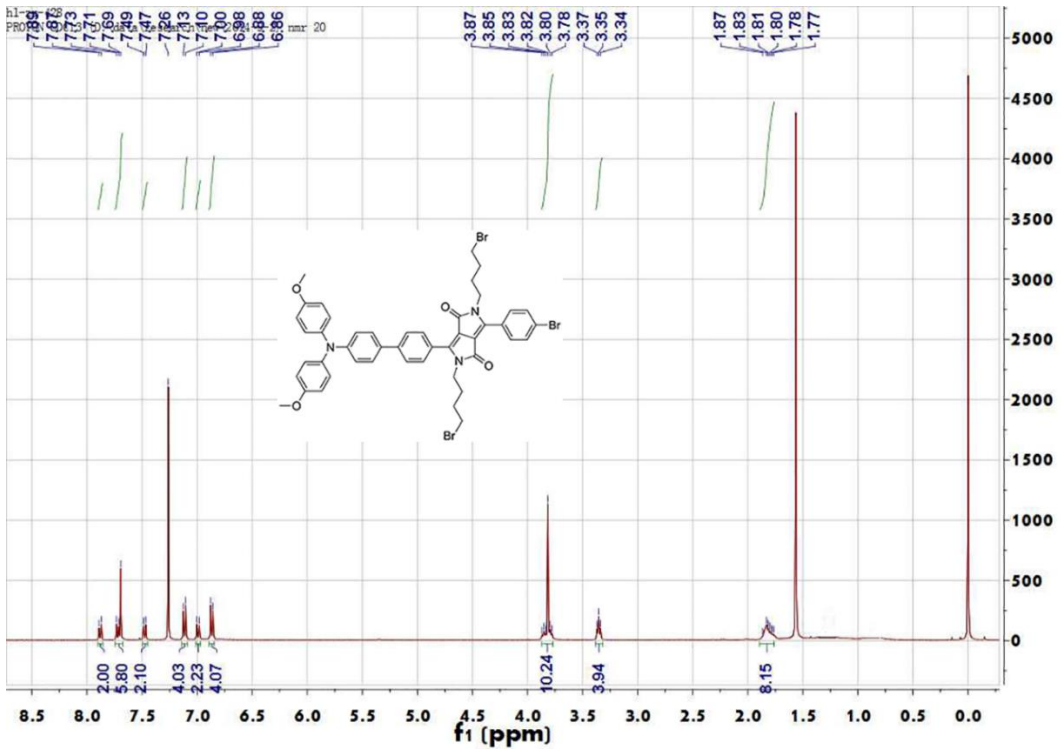


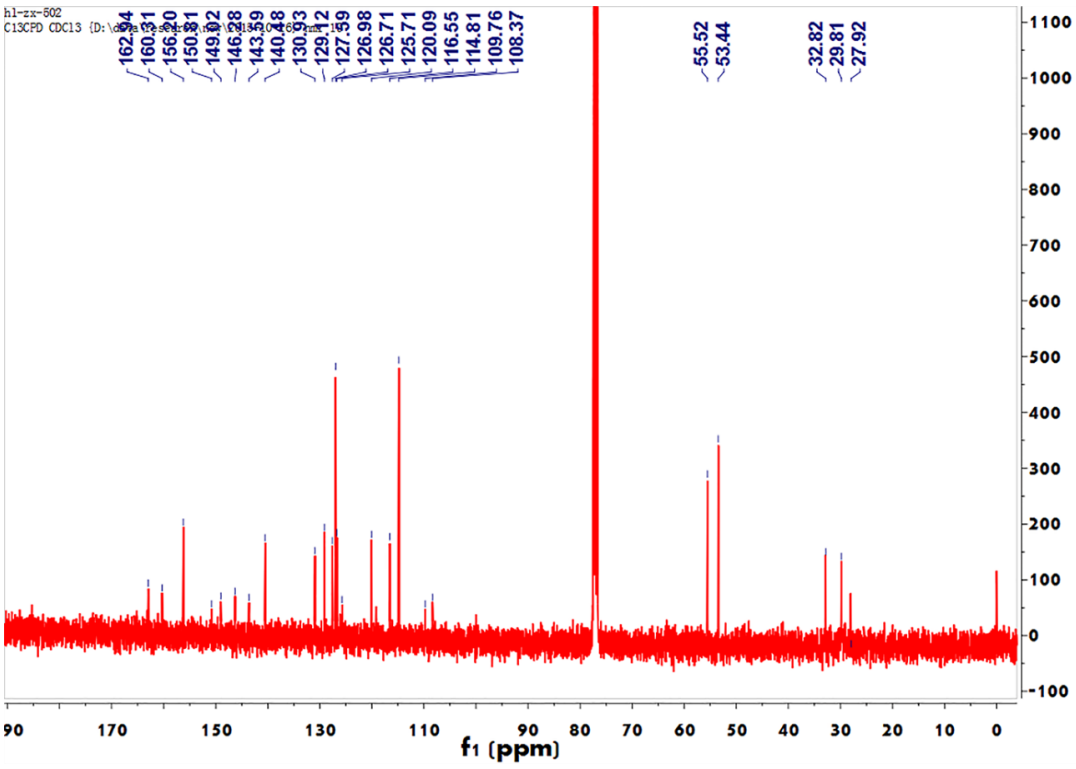
Figure S2. The MS titration of **DPP-AC** (10 μM) after the reaction with Cys (10 μM) for 30 min.

Original spectral copy of new compounds

^{1}H NMR of compound 2



^{13}C NMR of compound 2



MS of compound 2

Elemental Composition Report **Page 1**

Single Mass Analysis
Tolerance = 30.0 mDa / DBE: min = -1.5, max = 100.0
Element prediction: Off
Number of isotope peaks used for i-FIT = 2

Monoisotopic Mass, Even Electron Ions
189 formula(e) evaluated with 8 results within limits (up to 1 best isotopic matches for each mass)
Elements Used:
C: 0-60 H: 0-80 N: 0-5 O: 0-10

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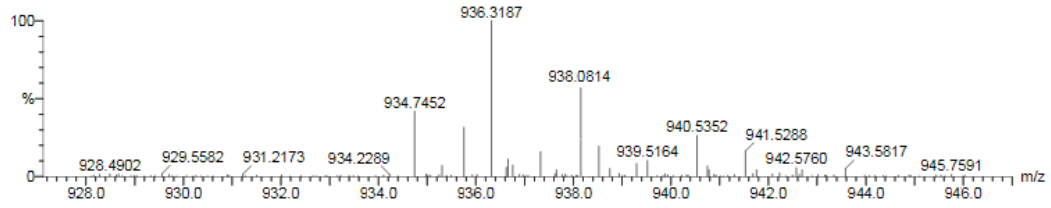
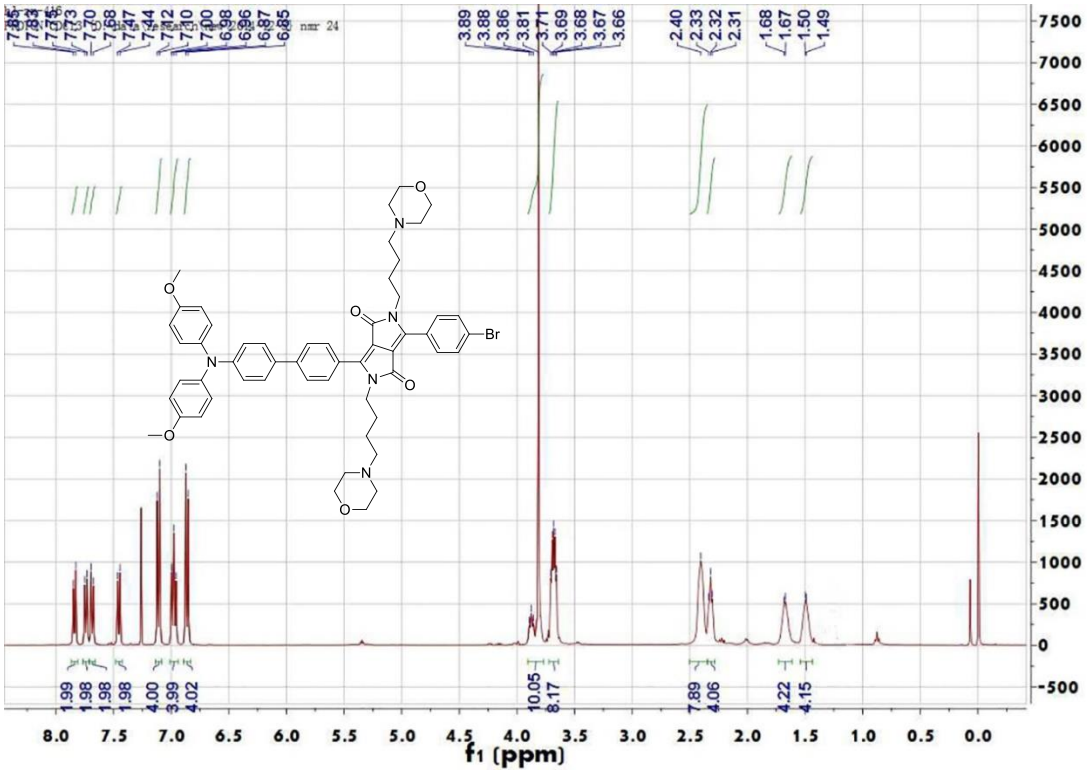
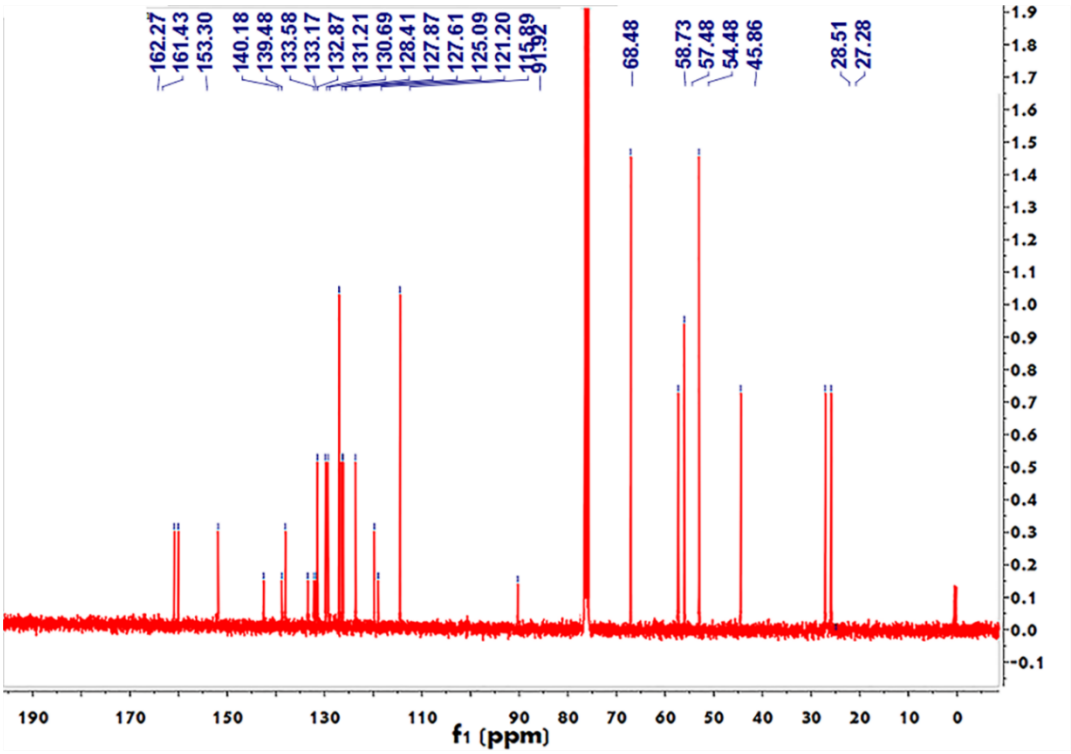


Minimum:				-1.5			
Maximum:		30.0	50.0	100.0			
Mass	Calc. Mass	mDa	PPM	DBE	i-FIT	i-FIT (Norm)	Formula
938.0814	938.0804	1.4	1.4	35.5	59.8	0.0	C46 H42 N3 O4 Br3

^{1}H NMR of compound 3



^{13}C NMR of compound 3



MS of compound 3

Elemental Composition Report **Page 1**

Single Mass Analysis
Tolerance = 30.0 mDa / DBE: min = -1.5, max = 100.0
Element prediction: Off
Number of isotope peaks used for i-FIT = 2

Monoisotopic Mass, Even Electron Ions
530 formula(e) evaluated with 1 results within limits (up to 1 best isotopic matches for each mass)
Elements Used:
C: 0-54 H: 0-59 N: 0-5 O: 0-6 Br: 0-1

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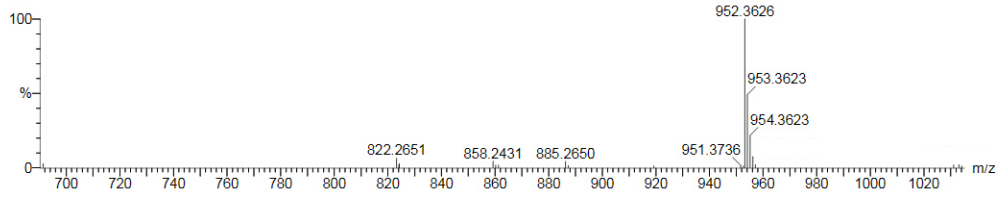
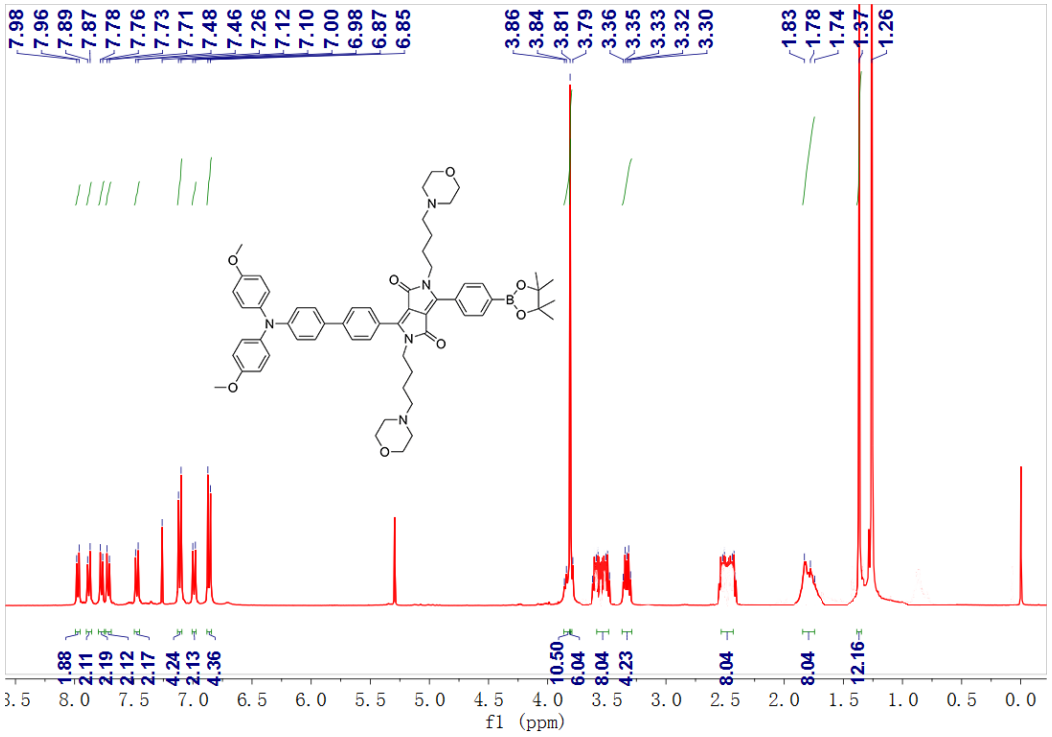
HJL-ZX-408 35 (0.840) Cm (34:36) 1: TOF MS ES- 4.56e+004

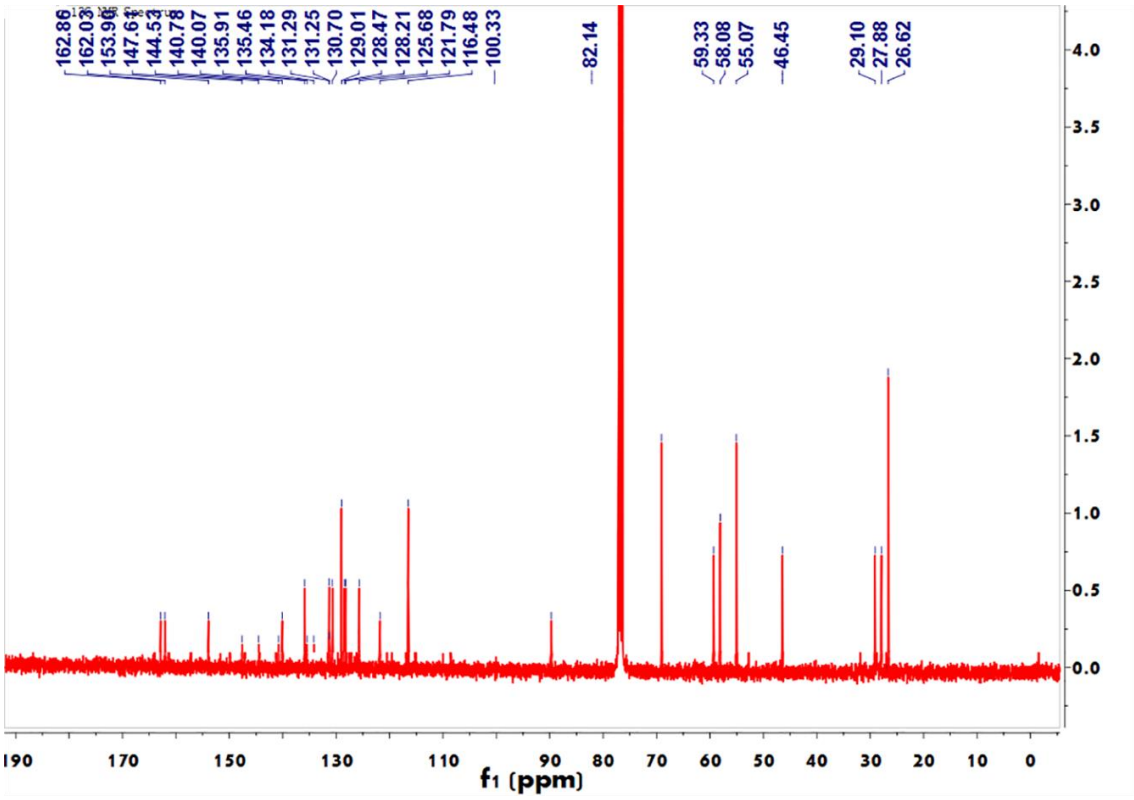


Minimum:				-1.5			
Maximum:		30.0	50.0	100.0			
Mass	Calc. Mass	mDa	PPM	DBE	i-FIT	i-FIT (Norm)	Formula
952.3626	952.3649	0.0	0.0	32.5	21.3	0.0	C54 H59 N5 O6 Br1

^{1}H NMR of compound 4



^{13}C NMR of compound 4



MS of compound 4

Elemental Composition Report **Page 1**

Single Mass Analysis
Tolerance = 50.0 PPM / DBE: min = -1.5, max = 100.0
Element prediction: Off
Number of isotope peaks used for i-FIT = 2

Monoisotopic Mass, Even Electron Ions
44 formula(e) evaluated with 1 results within limits (up to 1 closest results for each mass)
Elements Used:
C: 0-60 H: 0-73 N:0-5 O: 0-8 B: 0-1

JL-HUA ECUST institute of Fine Chem 10-Jun-2015 21:41:41
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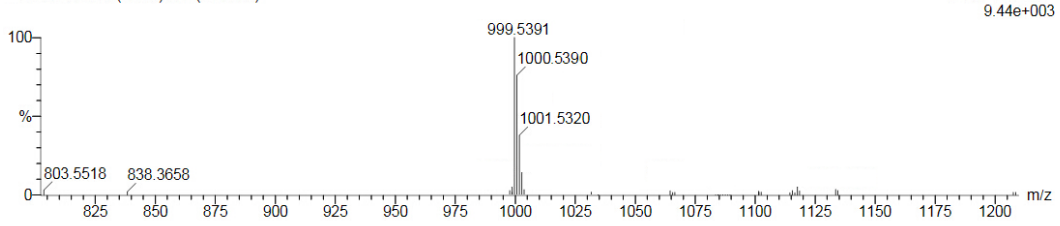
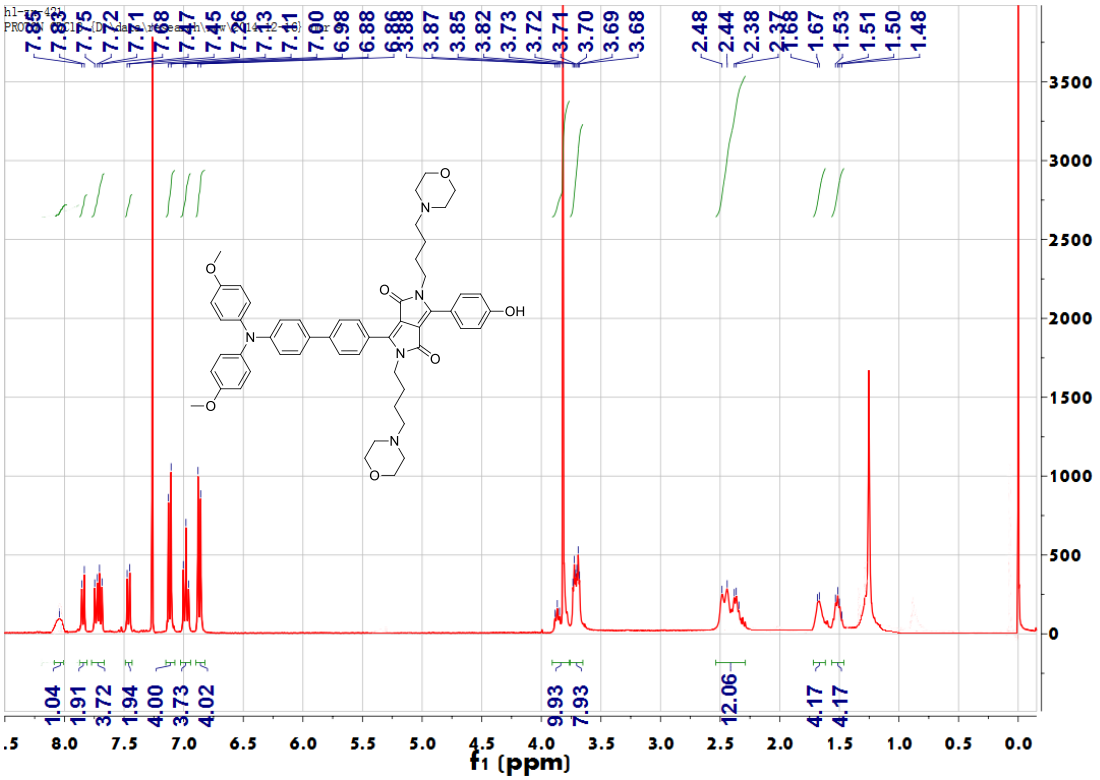
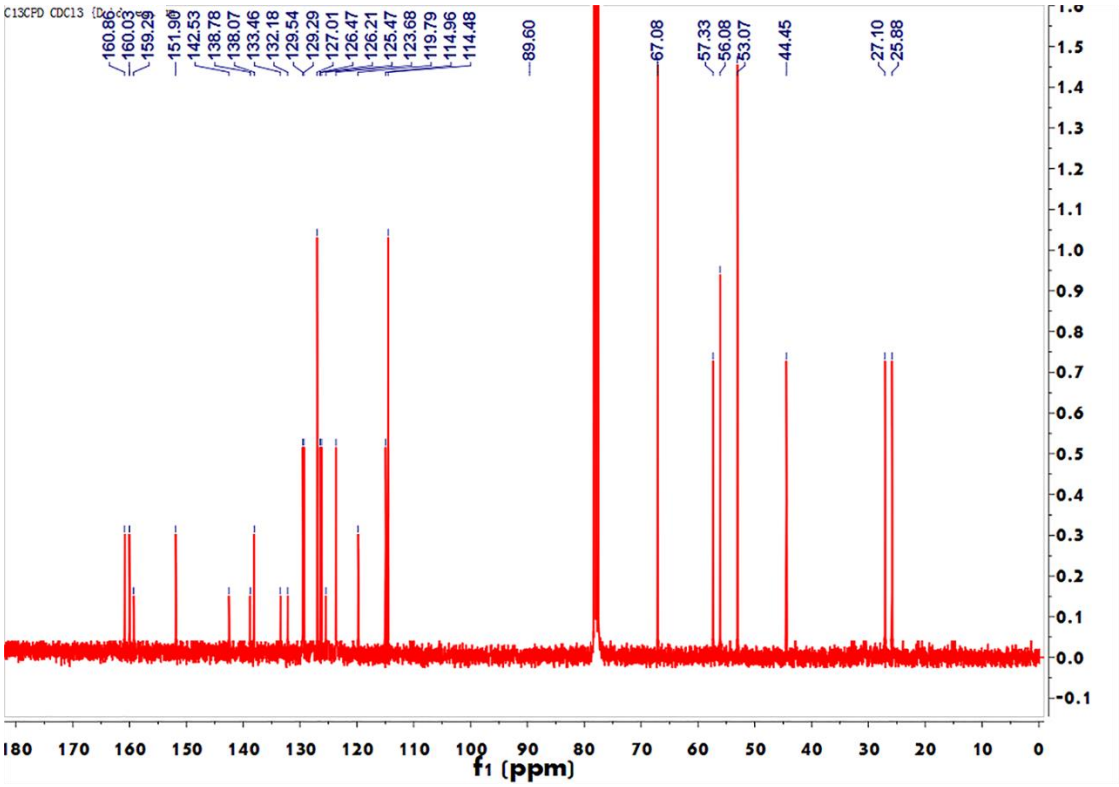


Minimum:				-1.5			
Maximum:		300.0	50.0	100.0			
Mass	Calc. Mass	mDa	PPM	DBE	i-FIT	i-FIT (Norm)	Formula
1000.5390	1000.5396	0.8	0.7	35.5	12.4	0.0	C60 H73 N5 O8 B1

^{1}H NMR of compound DPP-OH



^{13}C NMR of compound DPP-OH



MS of compound DPP-OH

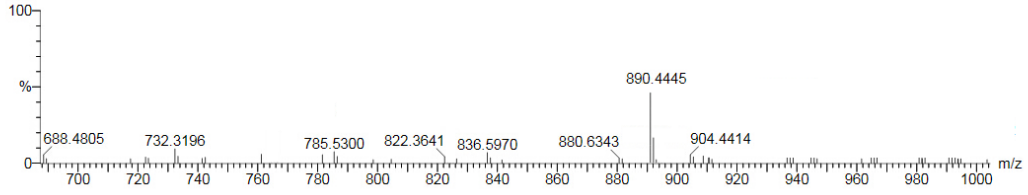
Elemental Composition Report **Page 1**

Single Mass Analysis
Tolerance = 50.0 PPM / DBE: min = -1.5, max = 100.0
Element prediction: Off
Number of isotope peaks used for i-FIT = 2

Monoisotopic Mass, Even Electron Ions
1 formula(e) evaluated with 1 results within limits (up to 1 closest results for each mass)
Elements Used:
C: 0-54 H: 0-60 N: 0-5 O: 0-7

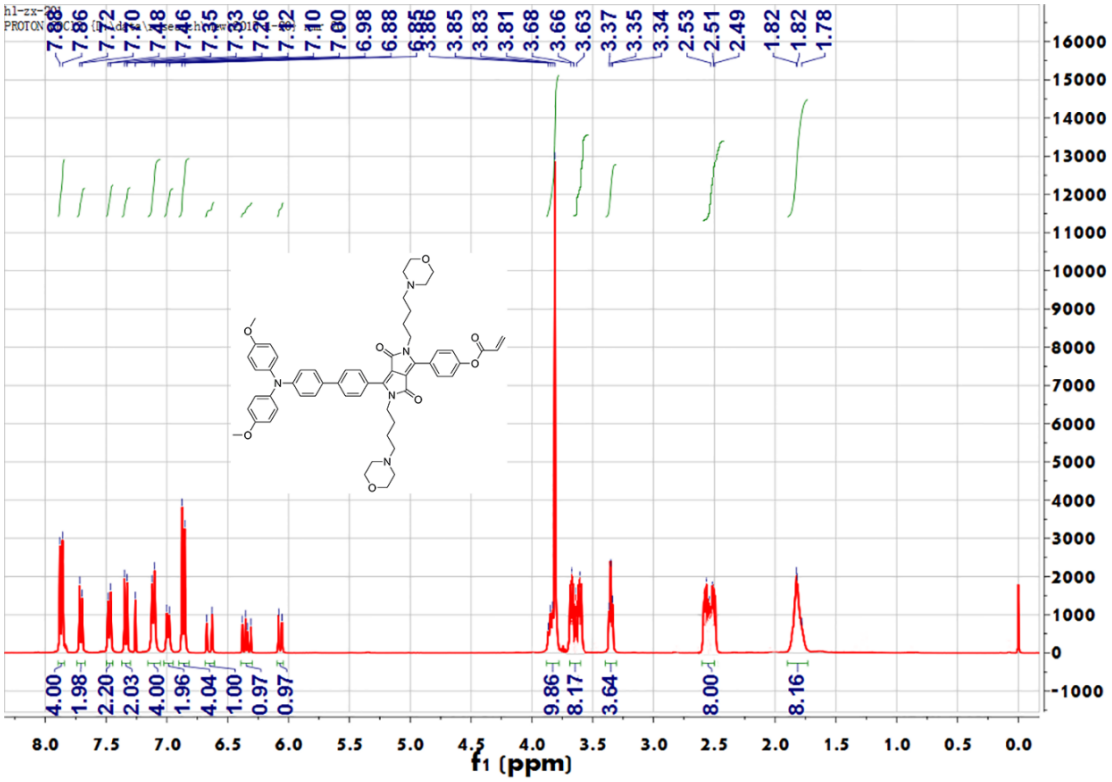
JL-HUA ECUST institute of Fine Chem 10-Jun-2015 21:22:35

HL-ZX-016 103 (0.729) Cm (101:104) 1: TOF MS ES+ 2.63e+003

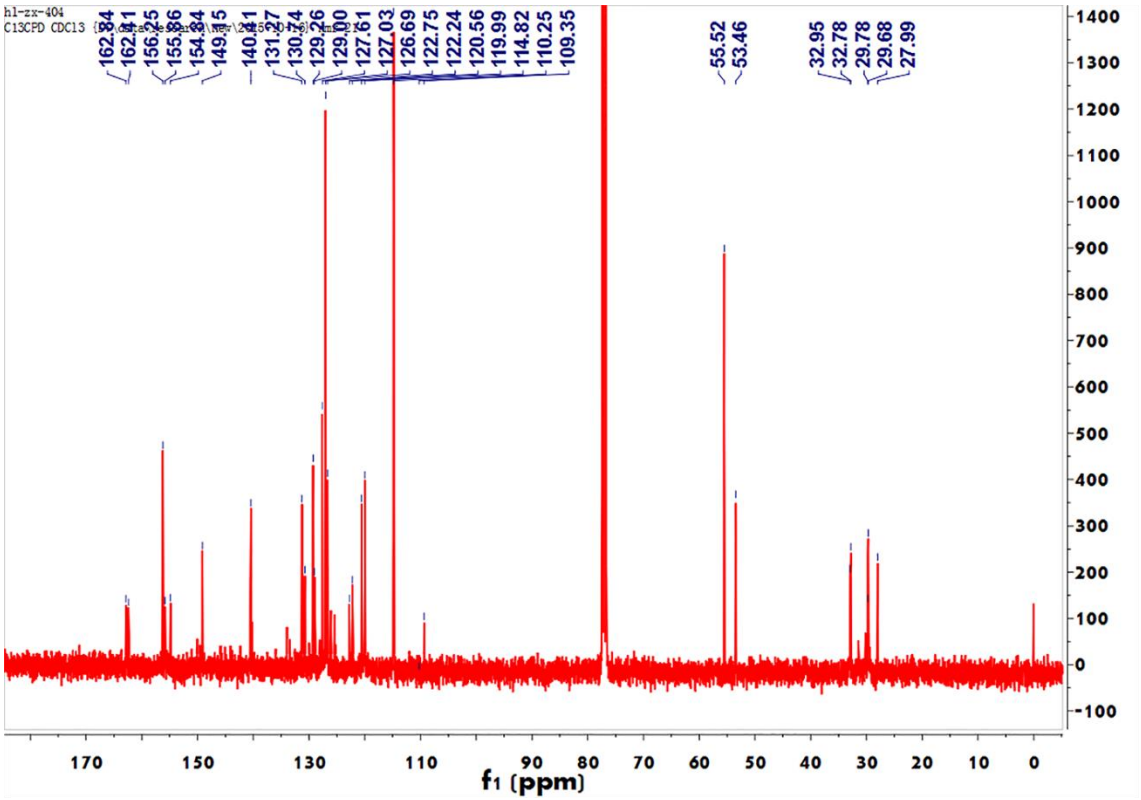


Minimum:				-1.5			
Maximum:		300.0	50.0	100.0			
Mass	Calc. Mass	mDa	PPM	DBE	i-FIT	i-FIT (Norm)	Formula
890.4445	890.4493	-0.9	-1.1	39.5	19.3	0.0	C54 H60 N5 O7

^{1}H NMR of compound DPP-AC



^{13}C NMR of compound DPP-AC



MS of compound DPP-AC

Single Mass Analysis
Tolerance = 50.0 PPM / DBE: min = -1.5, max = 100.0
Element prediction: Off
Number of isotope peaks used for i-FIT = 2

Monoisotopic Mass, Even Electron Ions
171 formula(e) evaluated with 6 results within limits (up to 1 closest results for each mass)
Elements Used:
C: 0-57 H: 0-100 N: 0-5 O: 0-8

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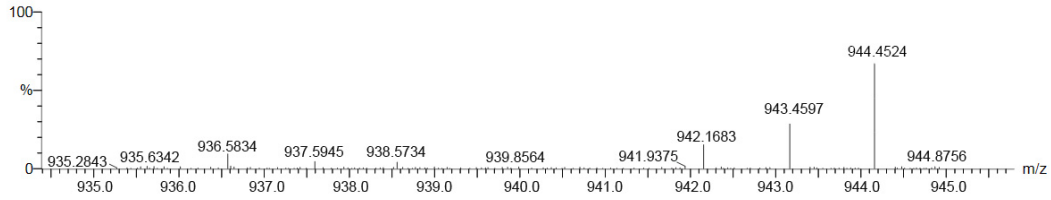

Minimum:			-1.5	
Maximum:		300.0	50.0	100.0

References and notes

[1] S. Y. Qu, W. J. Wu, J. L. Hua, C. Kong, Y. T. Long and H. Tian, J. *Phys. Chem. C*, 2010, **114**, 1343-1349.

[2] Y. D. Hang, L. Yang, Y. Qu and J. L. Hua, *Tetrahedron Letter*, 2014, **55**, 6998-7001.