Electronic Supplementary Material (ESI) for RSC Advances

Supporting Information

A novel pH "off-on" fluorescent probes for lysosome imaging

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1. Co-staining of PC-3 cells with LTP and LT Red

PC-3 cells were grown in RPMI-1640 medium supplemented with 10% (v/v) fetal bovine serum in an atmosphere of 5% CO_2, 95% air at 37 °C. Cells were adjusted to the density of 80,000/cm^2 and incubated for 32 h, then washed with cell culture medium twice. The solution of LTP (7 μM) and LT Red (70 nM) in cell culture medium were added and incubated at 37 °C for 1h. Then the samples were readily to be detected by confocal microscopy (Zeiss LSM 700). The colocalization coefficient was calculated by using imageJ software.

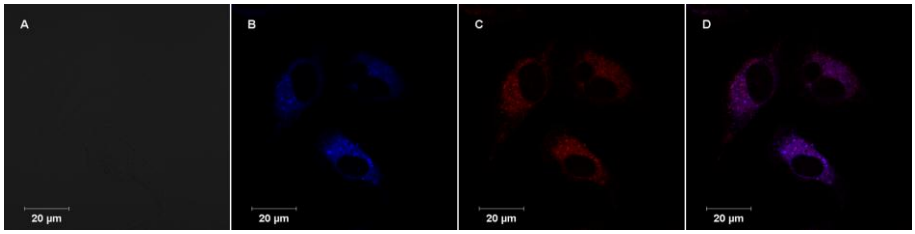


Figure S1 Co-staining of PC-3 cells with LTP(7μM) and LT Red (70 nM). A, bright-field image. B, cells stained by LTP, Ex 405nm; C, cells stained by LT Red, Ex 555nm; D, merged images. All images were taken under 63× oil immersion objective.

2. ^{1}H NMR, ^{13}C NMR and HR-MS of LTP

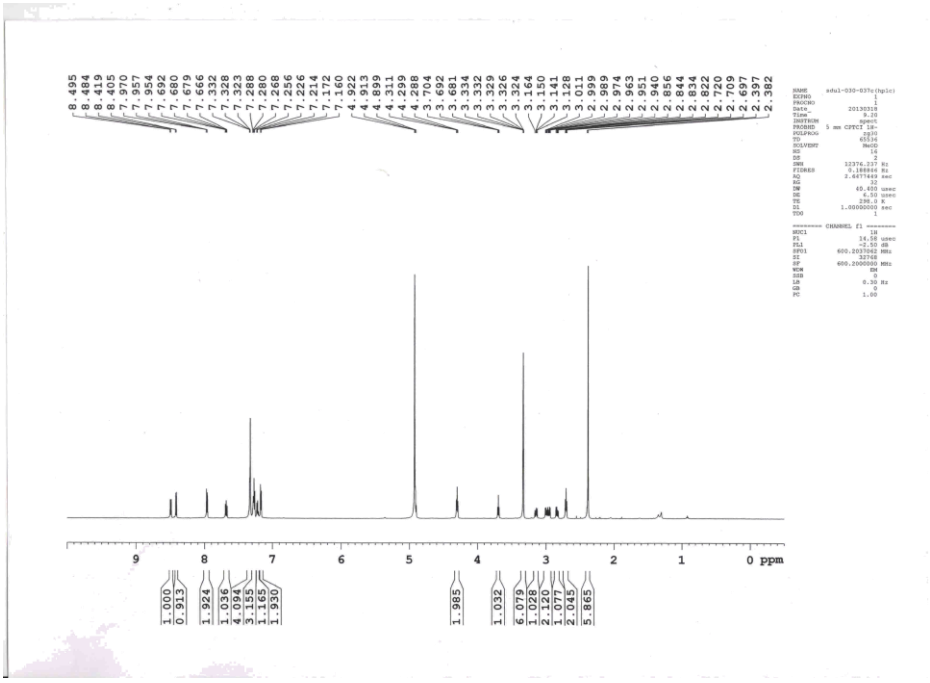


Figure S2 ^{1}H NMR (300 MHz) spectrum of **LTP**.

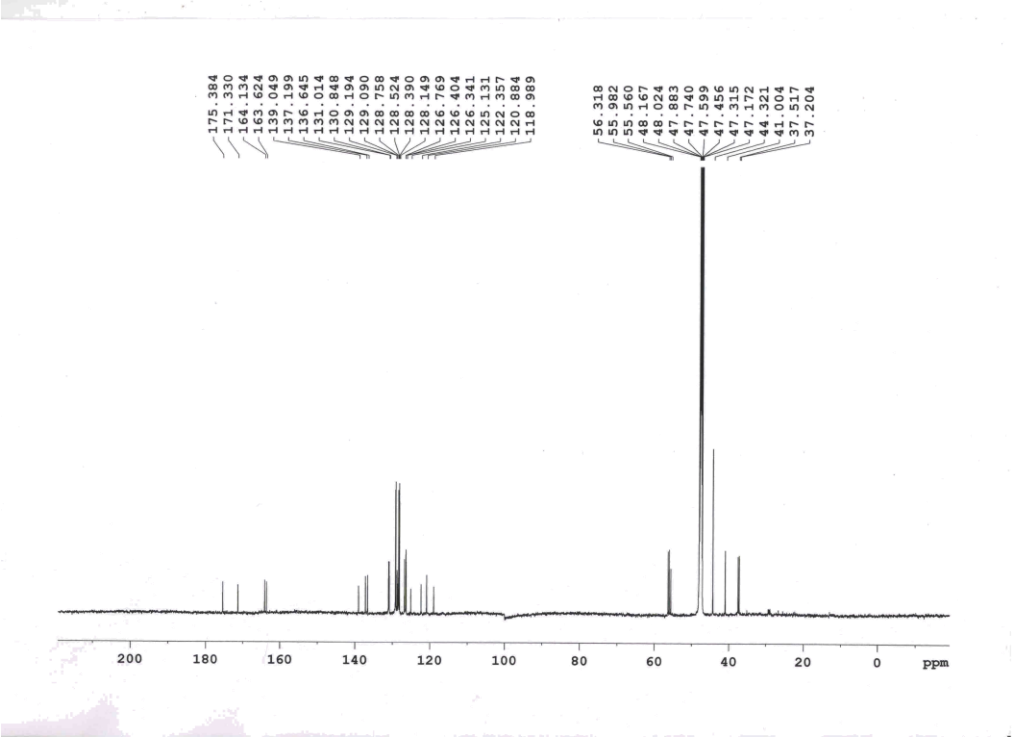
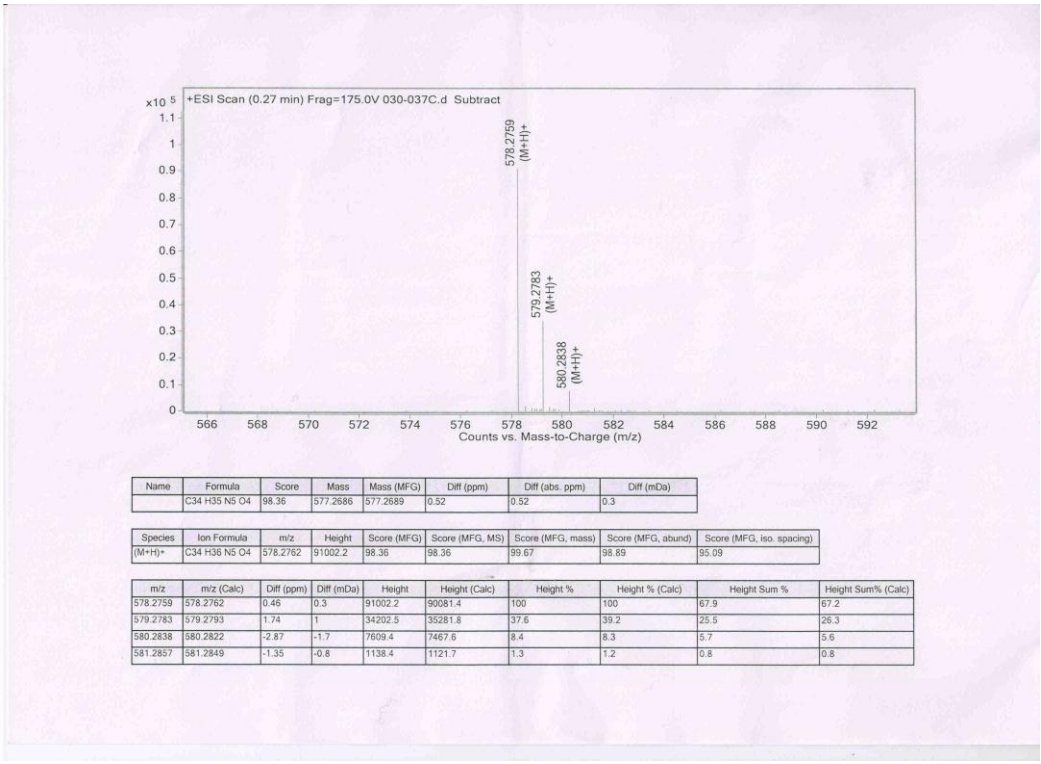


Figure S3 ^{13}C NMR (150 MHz) spectrum of **LTP**.



Name	Formula	Score	Mass	Mass (MFG)	Diff (ppm)	Diff (abs. ppm)	Diff (mDa)
	C34 H35 N5 O4	98.36	577.2686	577.2689	0.52	0.52	0.3

Species	Ion Formula	m/z	Height	Score (MFG)	Score (MFG, MS)	Score (MFG, mass)	Score (MFG, abund)	Score (MFG, iso. spacing)
(M+H)+	C34 H36 N5 O4	578.2762	91002.2	98.36	98.36	99.67	98.89	95.09

m/z	m/z (Calc)	Diff (ppm)	Diff (mDa)	Height	Height (Calc)	Height %	Height % (Calc)	Height Sum %	Height Sum% (Calc)
578.2759	578.2762	0.46	0.3	91002.2	90081.4	100	100	67.9	67.2
579.2783	579.2793	1.74	1	34202.5	35281.8	37.6	39.2	25.5	26.3
580.2838	580.2822	-2.87	-1.7	7609.4	7467.6	8.4	8.3	5.7	5.6
581.2857	581.2849	-1.35	-0.8	1138.4	1121.7	1.3	1.2	0.8	0.8

Figure S4 HR-MS spectrum of **LTP**.

3. HPLC spectrum of LTP

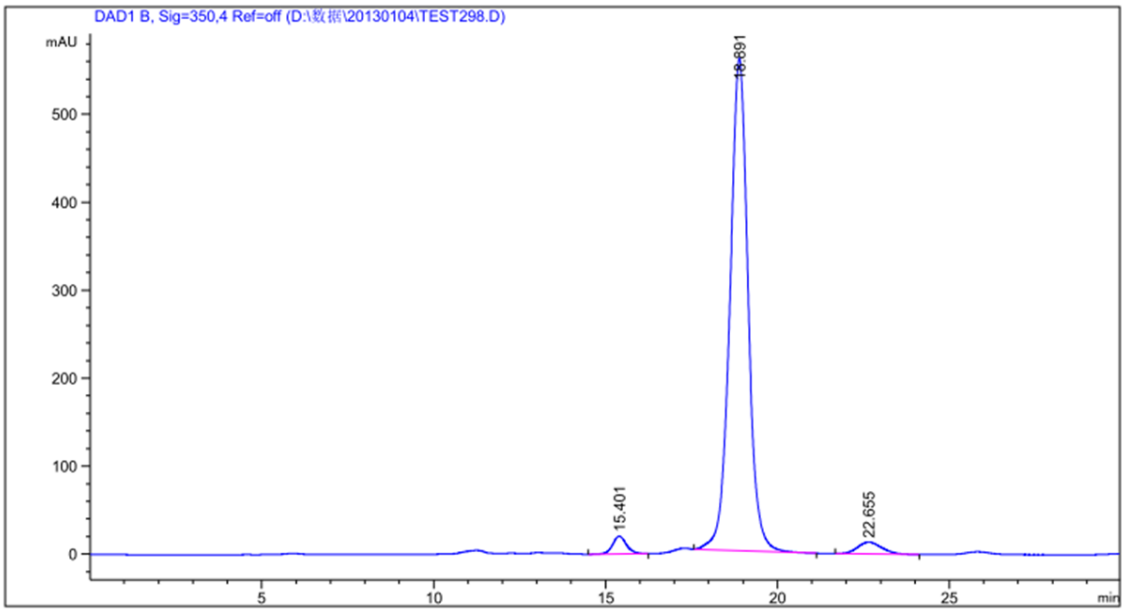


Figure S5 HPLC spectrum of **LTP**. Purity of **LTP** was determined by RP-HPLC method with Agilent 1260, using the Luna C18 (250×10.0 mm, 5μm) as the solid phase, methanol-water-diethylamine (81.85:18:0.15) as the mobile phase. The detection wavelength was 350 nm and the flow rate was 1.5 mL/min.