Supporting Information

Discriminating normal and inflammatory models by viscosity

changes with a mitochondria-targetable fluorescent probe

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Cytotoxicity test

Firstly, living cells were cultivated with 96-well plate in a 5% CO₂ incubator at 37 °C. Secondly, after 12 h, living cells fresh were cultivated using fresh medium containing different concentrations **EIMV** (1, 5, 10, 20, 30 μ M). Thirdly, after 24 h, the excess medium and probes were cleared up, and then 10 μ L (4,5-dimethyl-2-thiazolyl)-2, 5-diphenyltetrazolium bromide (MTT) (5 mg/mL in phosphate buffer solution (PBS)) was dropped into the above plate. Subsequently, the culture medium was cleared up, and 100 μ L DMSO was dropped into the dishes to dissolve the formazan crystal product. Fourthly, 96-well plate was shaken up for a moment. Absorbance data was obtained using multiscan spectrum. Finally, cell viability was obtained with the equation: The cell viability (%) = (OD_{490 sample} - OD_{490 blank})/(OD_{490 control} - OD_{490 blank}) × 100%. OD_{490 control} was cells untreated with probes of different concentrations, OD_{490 control} was cells untreated with probe, OD_{490 blank} was cell containing culture medium.

Synthesis



Scheme. S1. The synthetic route to the probe EIMV.



Fig. S1 (A) Absorption spectra and (B) fluorescence responses of EIMV (10 μ M.) in various solvents.

Solvents	λª /λʰ (nm)	Stokes shifts	Φc
DMSO	511/597	86	0.41
DMF	510/590	80	0.32
EtOH	520/610	90	0.20
H ₂ O	485/602	117	0.25
MeCN	500/605	105	0.32
MeOH	520/610	90	0.18
PBS	490/601	111	0.31
Glycerol	510/602	92	19.4

^aMaximum absorption wavelength (nm). ^bMaximum emission wavelength (nm). ^c Φ is fluorescence quantum yield (error limit: 8%) determined by using Rhodamine 6G (Φ =0.95) in Water as the standard.



Fig. S2 The linear relationship between log (I_{600}) and log (viscosity) in the PBS-glycerol solvent.

Table S2 Cytotoxicity Data of EIMV in HeLa cells ^a.

Incubate	0	1	5	10	20	30
concentration(µM)						
(% cell survival)	100±4	97±4	92±4	90±4	85±4	82±4

^a Cell viability was quantified by the MTT assays (mean ± SD).



Fig. S3 Confocal fluorescence images of the Hela cells incubated with 10 μ M **EIMV** for 30 min; (A): Bright-field images; (B): Merged pictures; (C): Red channel: $\lambda_{ex} = 561$ nm, $\lambda_{em} = 570-620$ nm.



Fig. S4 (A) Fluorescence images of Hela cells incubated with **EIMV** (10 μ M) acquired at different times under successive excitation.($\lambda_{ex} = 561 \text{ nm}$, $\lambda_{em} = 570-620 \text{ nm}$); (C) Fluorescence intensities of Hela cells incubated with **EIMV** (10 μ M) under successive excitation at different times.

Characterization.



Fig. S5 ¹H NMR spectrum of EIMV







Figure S7 HRMS spectrum of EIMV