Early diagnosis of heterotopic ossification with a NIR fluorescent probe by targeting type II collagen

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Supporting Information

Isolation and characterization of tissue-resident MPCs: Mouse tissue-resident mesenchymal progenitor cells (MPCs) were harvested from the insertion of the tendon into the calcaneus to the distal gastrocnemius including encompassing areas of tendon and surrounding soft tissues. All collected tissues were mechanically dissociated using a sterile scissors for all murine samples. Tissues were digested for 120 minutes in a solution of 300 U/mL of collagenase type II (Sigma-Aldrich, St. Louis, MO) at 37℃ under sustained agitation. Samples were filtered using consecutive 70-μm and 40-μm sterile strainers, and digestion was quenched using equal parts standard growth medium (Dulbecco's modified Eagle's medium [DMEM] supplemented with 10% fetal bovine serum [FBS] and 1% penicillin/streptomycin) (Procell, Wuhan, China) to generate a single-cell suspension. Cells were centrifuged at 1,000 rpm for 5 minutes before removing the supernatant and washing it in PBS. The cell pellet was resuspended in standard growth media and subsequently plated at a density of 1×10^6 cells/well in 6-well tissue culture plates. To characterize tMSC cells, we used flow cytometry for several stromal cell markers using Mouse Mesenchymal Stem Cell Characterization Kit

(Cyagen, Suzhou, China). This kit contains a panel of positive and negative selection markers for the characterization of the mesenchymal stromal cell population. The MPCs in passages 3–5 were used in the following experiments.

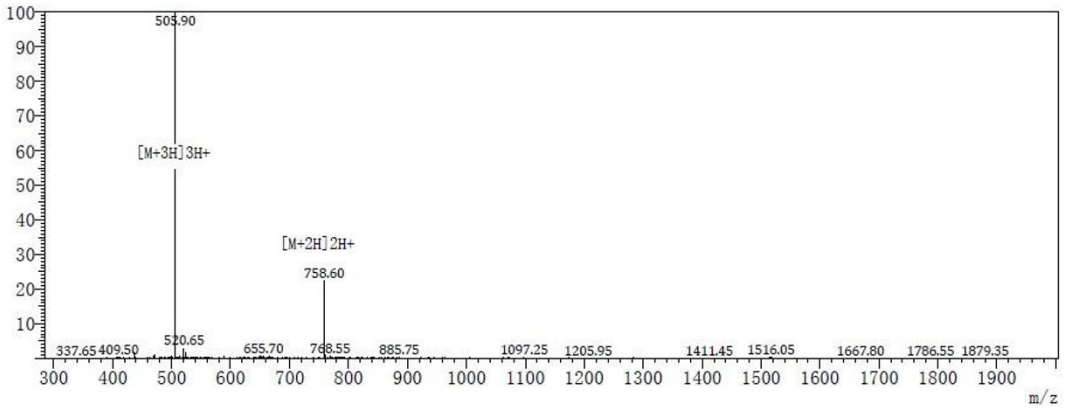

Fig. S1 The MS Spectra of WL-808.

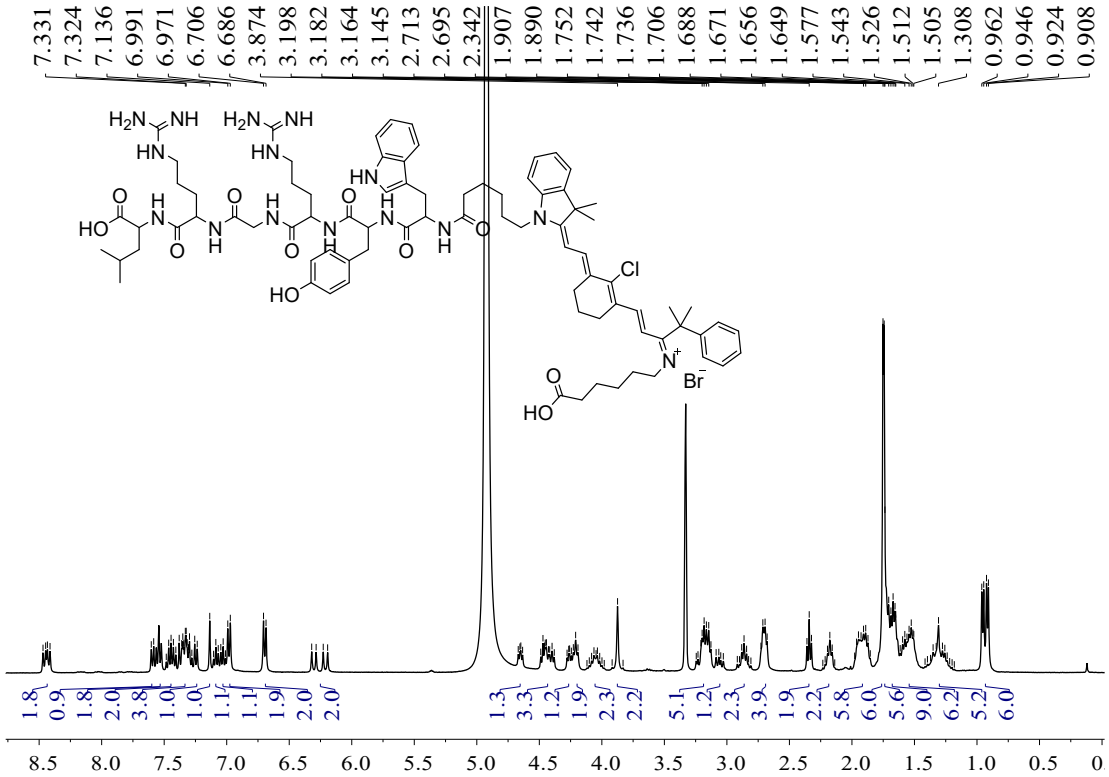

Fig. S2 The ^{1}H NMR spectrum of WL-808.

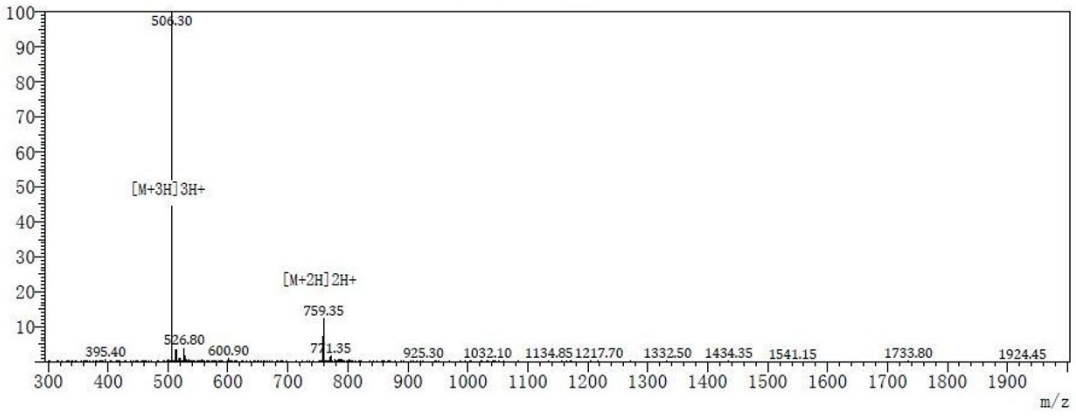

Fig. S3 The MS Spectra of YW-808.

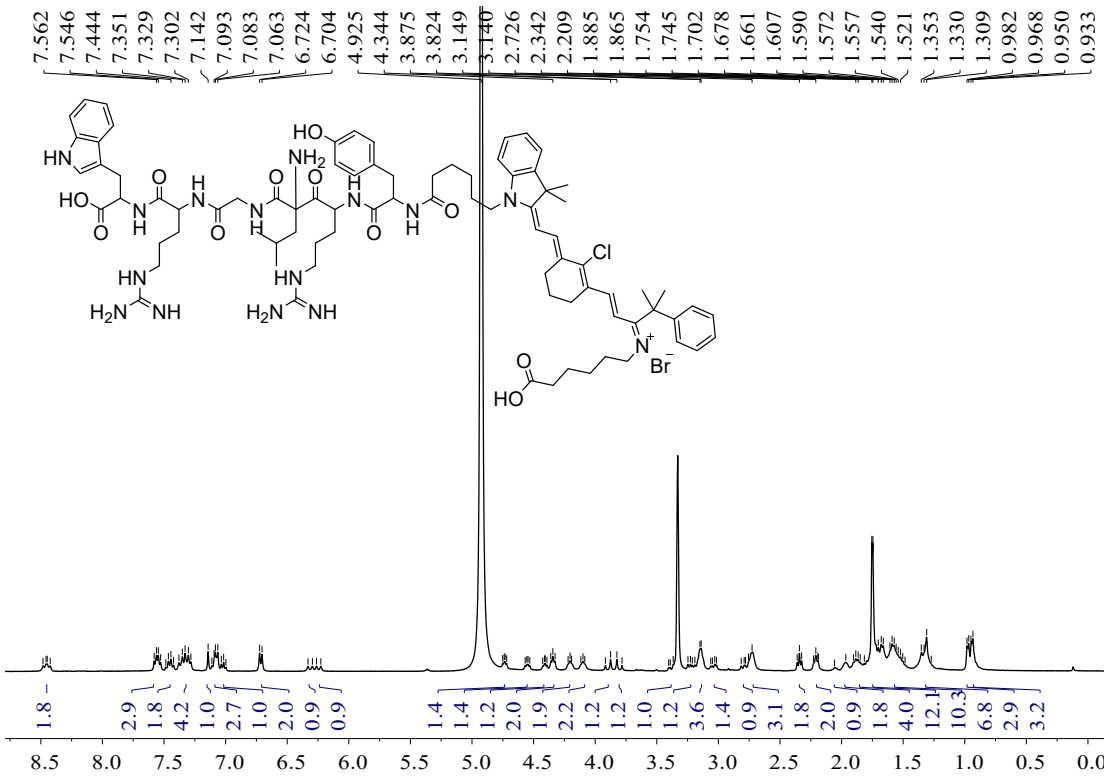

Fig. S4 The ^{1}H NMR spectrum of YW-808.

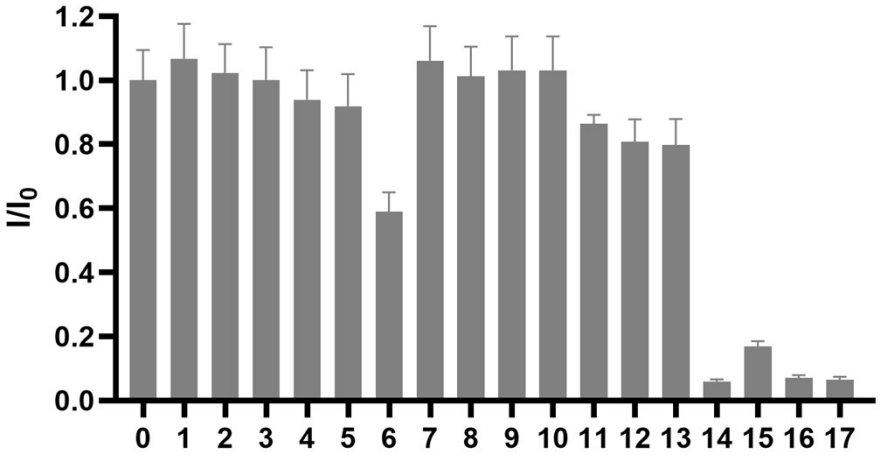

Fig. S5 Anti-interference evaluation of WL-808 (1 μM) in the presence of 17 interfering components at 10 mM (0-17: control, Glu, Pro, Ala, Arg, Asp, Cys, NaCl, KCl, $NaNO_3$, $FeCl_2$, Na_2CO_3, H_2O_2, Na_2SO_3, Na_2S, $FeCl_3$, $ZnCl_3$)

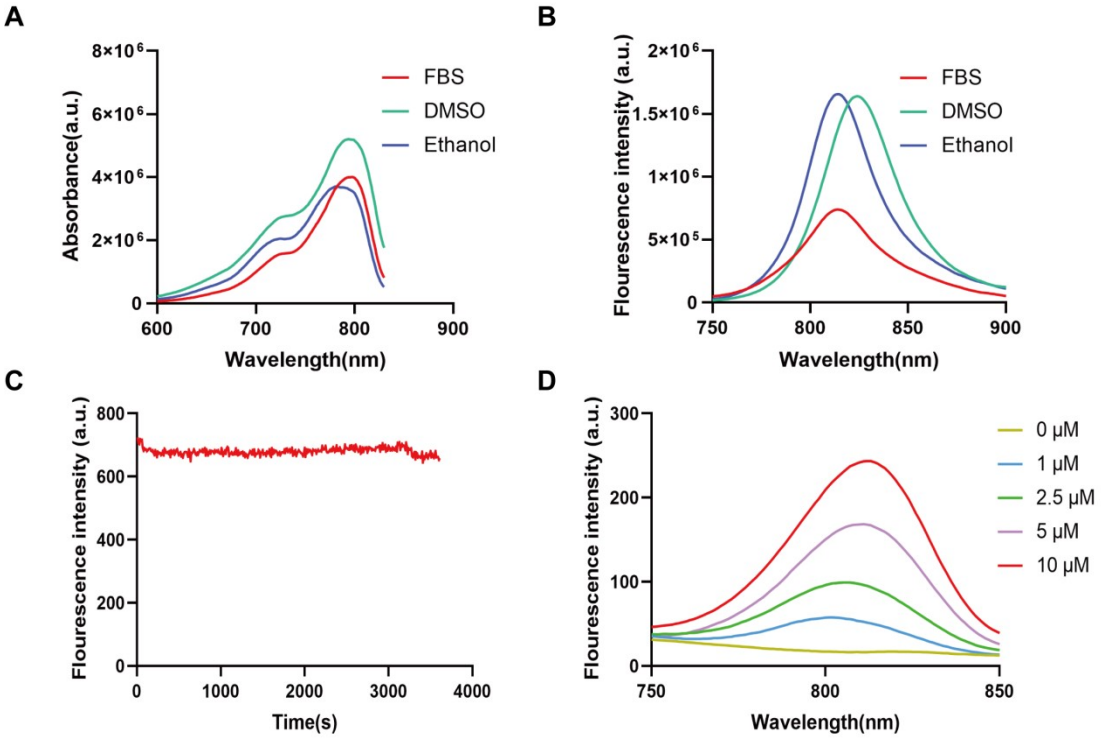

Fig. S6 The optical properties of YW-808. (A) Emission spectra of YW-808 (2.5 μM) in different solvents. (B) Absorption spectra of YW-808 (2.5 μM) in different solvents. (C) Photostability of YW-808 under continuous excitation (excitation wavelength = 750 nm, scan wavelength = 790 nm). (D) Emission spectra of YW-808 at different concentrations (1-10 μM) in FBS. (DMSO: dimethyl sulfoxide; FBS: fetal bovine

serum).