

Electronic Supplementary Information

Near-infrared fluorescence imaging for vascular visualization and fungal detection in plants

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1. Experimental section

1.1. Reagents and Materials.

IR-820 dye was purchased from Sigma-Aldrich (St. Louis, Missouri, USA).

1.2. Optical characterization of IR-820 dyes.

The absorption spectra of IR-820 in different solvents were measured by a UV/vis absorption spectrophotometer (Lambda 25, Perkin-Elmer, USA) ¹. Fluorescence emission spectra of IR-820 in different solvents were measured by a fluorescence spectrometer equipped with R928P and G8605-23 photodetectors, respectively (F920, Edinburgh Instruments, Ltd. UK).

1.3. NIR-Ia and NIR-Ib fluorescence imaging.

NIR-Ia images were acquired through an IVIS spectrum system (Caliper Life Sciences, Hopkinton, MA, USA) equipped with a silicon camera (detection range 350–900 nm) and a 700–900 nm bandpass filter. The excited light was 680 nm and NIR-Ia images were taken at a fixed exposure time of 1s. The images were processed by Maestro software. NIR-Ib images were captured using a 640 × 512 pixel two-dimensional InGaAs camera (detection range 900–1,700 nm) (Photonic Science, UK) equipped with a 900–1,000 nm bandpass filter (Thorlabs FEL, Newton, NJ, USA). A NIR lens pair SWIR-35 (Navitar, Rochester, NY, USA) was used to focus the image onto the photodetector. An 808 nm diode laser (laser glow technologies, Canada) offered excitation light. NIR-Ib images were captured at a fixed exposure time of 200 ms and Matlab 7 software was applied to process the images for any necessary flat-field correction.

1.4. Imaging experiment for *R. rugosa*.

R. rugosa purchased from a local florist was hydroponically grown in culture solution containing 50 µg/mL IR-820 under long-day conditions (16 h light, 8 h dark) at 25 °C and 40% humidity. Leaves, flowers and stems were fixed and imaged every 6 h without extraction.

1.5. Imaging experiment for *G. sinensis*, *C. esculenta* and *C. interruptus*.

G. sinensis and *C. esculenta* were obtained from the campus garden of Shenzhen Institute of Advanced Technology. *C. interruptus* was obtained from the hills of Liangtian Town, Chenzhou in Hunan Province. All plants were cultured in solution containing 50 µg/mL IR-820 under long-day conditions (16 h light, 8 h dark) at 25 °C and 40% humidity. Leaves and stems were cut, fixed and imaged after incubation for 24 h.

1.6. Study of water transpiration in *G. sinensis*.

G. sinensis was incubated in culture solution containing 50 µg/mL IR-820 under long-day conditions (16 h light, 8 h dark) at 25 °C and 40% humidity. One of the plant's side branches was either incubated inside an incubation chamber at 35 °C or irradiated by white light from a standard 15-W fluorescent light bulb placed 0.5 m away. Leaves and stems were extracted, fixed and imaged after incubation/irradiation for 6 h.

1.7. Detection and identification of fungal pathogens.

A naturally diseased *K. senegalensis* was obtained from the campus garden of Shenzhen Institute of Advanced Technology. The plant was cut, and its stem was cultured in solution containing 50 µg/mL IR-820 under long-day conditions (16 h light, 8 h dark) at 25 °C and 40% humidity. The diseased part of the stem was further cut, fixed and imaged after incubation for 24 h. For the inoculation experiments, *C. gleosporoides* and *A. argyroxiphii* were purchased from ATCC (Manassas, Virginia, USA). A 1 mL drop of inoculate containing 10 mg of either *C. gleosporoides* or *A. argyroxiphii* by dry weight was added to the healthy leaves of *K. senegalensis* and then left to grow at 25 °C and 90% humidity for 3–5 days. The plant was cut, and its stem was cultured in solution containing 50 µg/mL IR-820 under long-day conditions (16 h light, 8 h dark) at 25 °C and 40% humidity. Diseased leaves were extracted, fixed and imaged.

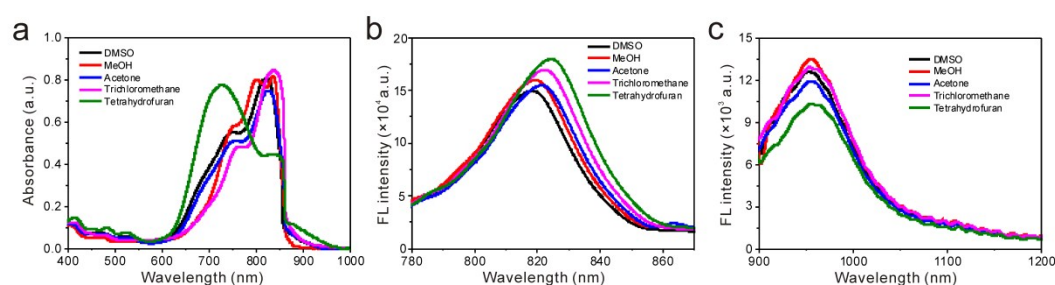


Fig.S1 Optical properties of IR-820 dye in different solvents. (a) UV/Vis absorption spectra in different solvents. (b) NIR-Ia emission spectra ($\lambda_{\text{ex}} = 680 \text{ nm}$) and (d) NIR-Ib emission spectra ($\lambda_{\text{ex}} = 808 \text{ nm}$) of IR-820 dye in different solvents.

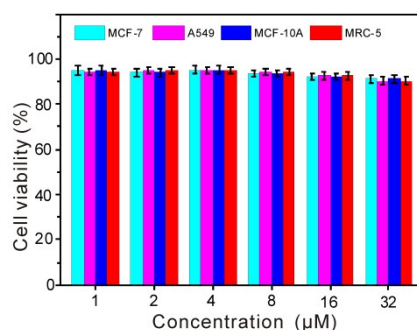


Fig.S2 Toxicity analysis of IR-820 dye.

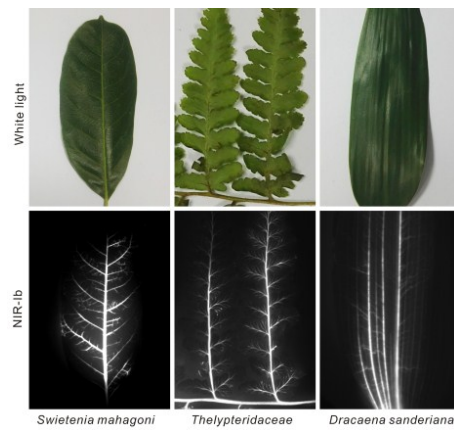


Fig.S3 White-light and NIR-Ib images of the leaf of *Swietenia mahagoni* (L.) Jacq, *Thelypteridaceae* and *Dracaena sanderiana*.

References

1. M. J. Jiang, X. G. Gu, J. W. Y. Lam, Y. L. Zhang, R. T. K. Kwok, K. S. Wong and B. Z. Tang. *Chem. Sci.*, 2017, **8**, 5440–5446.