GORYO CHEMICAL

For research use only

Updated: Mar. 2, 2021

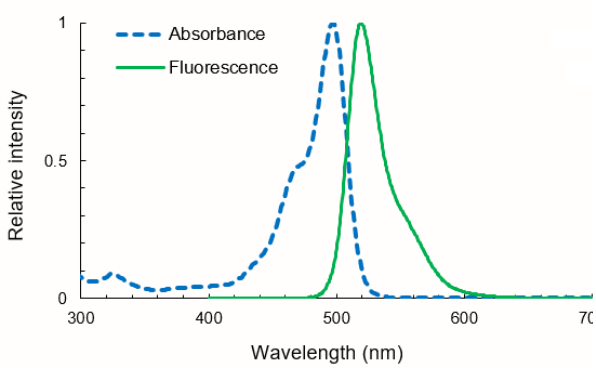
SaraFluor™ 497 actin probe

Product information

Catalog no.	Product name	Amount	Storage upon receipt	Stability
AR5601-N5	SaraFluor 497 actin probe	30 nmol × 5	≤–20°C, keep desiccated and protected from light.	1 year (when unopened and stored as described.)

1. About SaraFluor 497 actin probe

The SaraFluor 497 actin probe is a fluorescent probe that specifically binds to actin filaments (F-actin) and can visualize F-actin with green fluorescence. It is applicable to both living and fixed cells. Since it is an organic fluorescent small molecule with high membrane permeability, it is possible to visualize intracellular actin filaments by adding to the cell culture medium or extracellular fluid without a rinsing step. This product is thought to bind to F-actin through the common binding site for phalloidin and jasplakinolide.



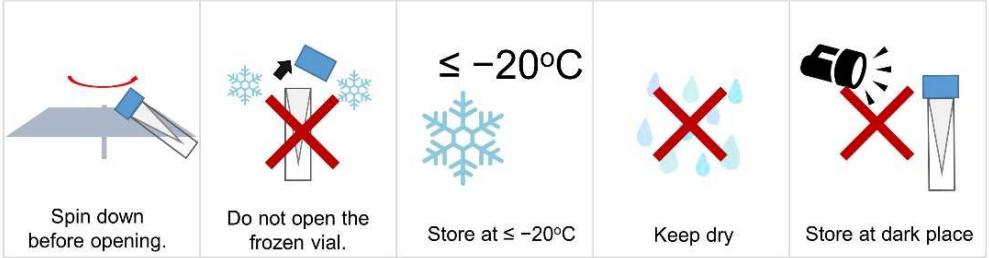
Spectra of SaraFluor 497 actin probe

■ **Storage**

This product is shipped as a dried solid in a nitrogen gas-filled vial. Upon receipt, store at ≤–20°C, keep desiccated and protected from light.

■ Preparation of the reagent

1. SaraFluor 497 actin probe is orange solid. Before opening the cap, warm the vial to room temperature. Then, use a micro-centrifuge to spin down the solid that might adhere to the cap.
2. Add 30 µL of DMSO to one vial to prepare 1 mM solution. Finally, dissolve the solid entirely by pipetting ~5 times. The solution will become a pale-yellow liquid.
3. After dissolving to DMSO, store as small aliquots at ≤–20°C. Avoid freeze-thaw cycles and use them as soon as possible.



4. An example of live cell imaging

1. Prepare cells on a glass-bottom dish.
2. Add SaraFluor 497 actin probe solution to the culture medium at the final concentration of 10-100 nM*. To add 100 nM probe, prepare 10 µM solution by diluting the 1 mM solution with PBS, then add the 1/100 volume of the 10 µM solution to the culture medium. The final concentration of DMSO should be ≦0.1%. Gently mix the medium and incubate the dish in a CO_2 incubator for >10 minutes.
3. Observe the cells using a fluorescence microscope. Do not replace the medium. We do not recommend rinsing the cells with a fresh medium before the observation. Use of microscope stage top incubator might be necessary.

* Higher concentration of the reagent may affect the cell shape or show cytotoxicity. The effect differs on the cell types. Therefore, we recommend using the lowest possible concentration if the target cells are sensitive to this reagent and the shape and dynamics are essential for your purpose.

5. An example of fixed cell imaging

1. Prepare cells on a glass-bottom dish.
2. Fix cells by adding PBS(+) supplemented with 4% paraformaldehyde and incubating at 37°C for 15 minutes.
3. Remove the fixation buffer from the cell culture dish, add PBS supplemented with 100 nM – 1 of SaraFluor 497 actin probe, and incubate for 15 minutes.
4. Observe the cells using a fluorescence microscope.

■ Fluorescent observation

The SaraFluor 497 actin probe's fluorescence can be observed with a commonly used Blue excitation and green fluorescence filter set for GFP/FITC. For live-cell imaging, use lower concentrations of SaraFluor 497 actin probe, weaken the excitation light intensity and use high-sensitivity cameras to reduce the exposure time. Consider the use of antifade reagent for time-lapse or continuous imaging.

For the observation of the detailed actin structure, users may consider the use of a confocal microscope, a total internal reflection (TIRF) microscope, a structured illumination microscope (SIM), or superresolution resolution radial fluctuation (SRRF). Stimulated emission depletion (STED) microscopy is also applicable. Use of 488 nm excitation laser with 592 nm depletion laser is appropriate.

Adjust the image intensity offset to reduce the fluorescence background signal derived from the unbound probe in the medium. We do not recommend exchanging the medium to reduce the background because the probe concentration in the medium and the concentration of actin-bound probes form an equilibrium.

Related Products

Catalog no.	Product name	Major applications
GC301	AcidiFluor ORANGE	Acidic-environment sensitive fluorescent probe for lysosome staining
A201-01	HaloTag® SaraFluor 650B Ligand	Spontaneously blinking fluorophore with Halo-tag ligand for superresolution imaging. Excited by a red laser.
A209-01	SaraFluor 650B-NHS	Spontaneously blinking fluorophore for superresolution imaging. Excited by a red laser.
A218-01	SaraFluor 488B-NHS	Spontaneously blinking fluorophore for superresolution imaging. Excited by a blue laser.
A308-01	HaloTag® SaraFluor 650T Ligand	Halo-tag binding deep-red fluorophore. Applicable for STED imaging.