

DFHBI

3'5'-Difluoro-4-Hydroxybenzylidene
Imidazolinone

Cat. No. 400-1mg/5mg/10mg



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Product

3'5'-difluoro-4-hydroxybenzylidene imidazolinone (DFHBI) is a non-fluorescent small molecule derived from the GFP chromophore. Upon binding to the "Spinach"[™] aptamer, DFHBI is converted to a highly fluorescent state that can be detected at the emission wavelength of 501 nm. DFHBI is cell permeable with negligible toxicity in living cells. DFHBI can be used to label any genetically encoded Spinach[™] family of RNA tags that include Spinach[™], Spinach2[™], and Broccoli[™]. Live-cell imaging of tagged RNA can be performed using a standard fluorescence microscopy. Further, DFHBI can also selectively detect tagged RNA in total RNA gel electrophoresis. Thus, bypassing the need for Northern blot analysis.

Presentation

Each vial contains lyophilized DFHBI dyes. Resuspension in DMSO at 20-40 mM concentration is recommended before transferring to the desired experimental buffer. DFHBI can also be resuspended in water [pH >9.0] at 100 μ M. Once all the dyes are in solution, titrate back to neutral pH to ensure stability.

Storage

Store at -20 °C. Stable for 2 years at -20 °C from the date of shipment. Non-hazardous. No MSDS needed.

Specifications

Excitation maximum: 469 nm
Emission maximum: 501 nm
Extinction coefficient ($M^{-1} cm^{-1}$)^a: 24,271
Quantum yield: 0.72
 K_D : 537 nM
Brightness^b: 80

^a Extinction coefficient of Spinach[™]/DFHBI complex was measured at 7.4, where all species were in the phenolate form.

^b Brightness is reported relative to Aequorea GFP.

Data

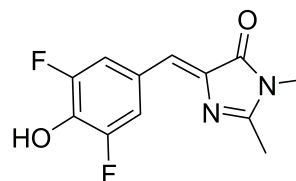


Figure 1. Structure of DFHBI. MW = 252.22

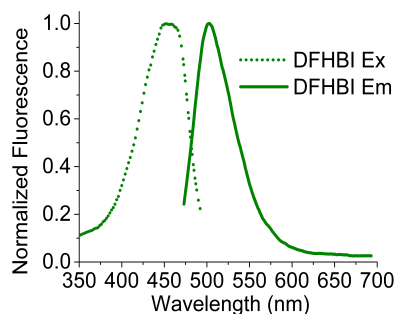


Figure 2. Excitation and emission spectra of Spinach[™]/DFHBI complex.

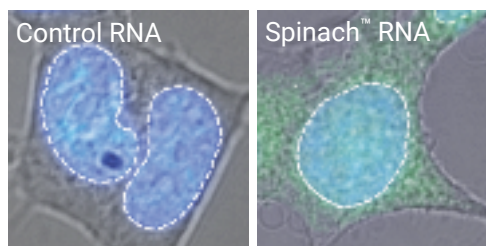


Figure 3. HEK293T cells were transfected with either 5S-control aptamer or 5S-Spinach[™] plasmids and incubated with 20 μ M DFHBI. Images are phase overlay with Hoechst-stained nuclei (blue) and Spinach[™] fluorescence (green).

References

- Paige JS, et al. 2011. RNA mimics of green fluorescent protein. *Science* 333(6042), 642-646.
- Paige JS, et al. 2012. Fluorescence imaging of cellular metabolites with RNA. *Science* 335(6073), 1194.