

## OBI

3,5-Difluoro-4-Hydroxybenzylidene-  
Imidazolinone-2-Oxime-1-Benzimidazole

Cat. No. 610-1mg



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### Product

OBI is a DFHBI derivative that binds either Broccoli™ or Red Broccoli™ and generates red fluorescence. Conversely, Red Broccoli™ also binds BI to emit green fluorescence. This “plug-and-play” capability allows the spectral properties of fluorogenic aptamers and fluorophores to be altered based on the specific experimental needs of the users. In the cells, OBI binds Red Broccoli™ with higher affinity and makes it resistant to thermal unfolding. This enables the Red Broccoli™/OBI complex to be readily detectable in live mammalian cells. This is particularly useful because green fluorescent probes suffer from the cellular green background fluorescence derived from endogenous vitamins and co-factors. Thus, red fluorescent RNA-based tags and probes would improve the accuracy of intracellular imaging.

### Presentation

Each vial contains lyophilized OBI dyes. Resuspension in DMSO at >10 mM concentration is recommended before transferring to the desired experimental buffer.

### 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: 541 nm  
Emission maximum: 590 nm  
Extinction coefficient ( $M^{-1} cm^{-1}$ )<sup>a</sup> : 57,800  
Quantum yield: 0.67  
 $K_D$  : 23 nM  
Brightness<sup>b</sup> : 266

<sup>a</sup> Extinction coefficient of OBI was measured in buffer containing 40 mM HEPES [7.4], 100 mM KCl, 5 mM  $MgCl_2$ .

<sup>b</sup> Brightness is relative to Red Broccoli™/DFHO.

### Data

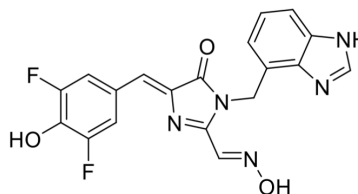


Figure 1. Structure of OBI. MW = 397.34

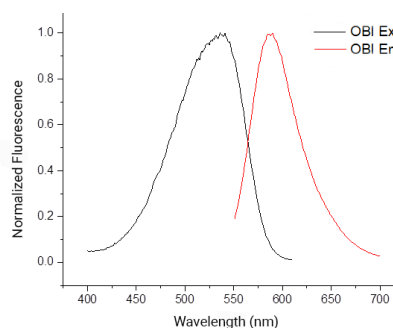


Figure 2. Excitation and emission spectra of Red Broccoli™/OBI complex.

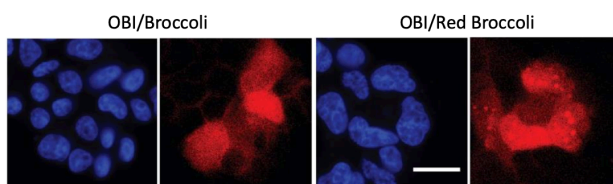


Figure 3. HEK293T cells expressing Red Broccoli, Broccoli, or control RNA were incubated with 10  $\mu M$  OBI. Red fluorescence were seen in both Red Broccoli- and Broccoli-expressing cells, but not in control RNA-expressing cells. Cell nucleus is labeled with Hoechst stain (blue) and imaged with DAPI and TRITC filters.

### References

Li X, *et al.* 2020. Imaging intracellular S-adenosyl methionine dynamics in live mammalian cells with a genetically encoded red fluorescent RNA-based sensor. *J Am Chem Soc*, 142(33):14117-14124.