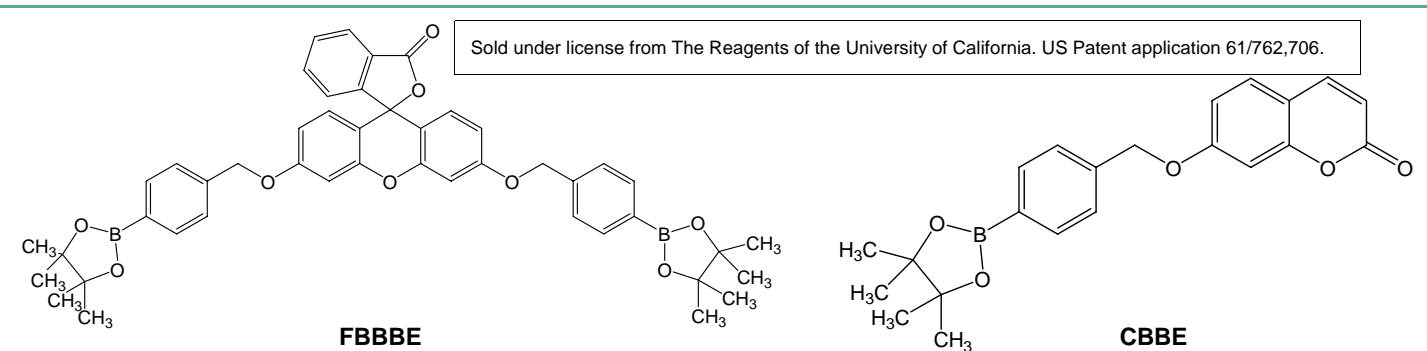


Kit Catalog number: 96-0350

Active Ingredients: Catalog number 05-0058 Fluorescein bis(benzyl boronic ester) **FBBBE**
 Catalog number 05-0054 Coumarin benzyl boronic ester **CBBE**



Introduction: **FBBBE** and **CBBE** are robust fluorescent probes, effective for imaging hydrogen peroxide in biological settings. In the supplied form, the fluorescence signal is effectively quenched by the addition of the benzyl ether boronic ester protecting groups to the fluorophores. In the presence of biologically relevant levels of hydrogen peroxide (10-200 μM), the protecting groups cleave and a strong fluorescent signal is observed (FBBBE: $I_{\text{exc}} = 480 \text{ nm}$, $I_{\text{em}} = 512 \text{ nm}$; CBBE: $I_{\text{exc}} = 370 \text{ nm}$, $I_{\text{em}} = 450 \text{ nm}$)

1. *ChemBioChem*, **2013**, *14*, 593.

Contents: **FBBBE:** 3 x 5mg **CBBE:** 3 x 5mg
 Dimethylsulfoxide (ACS spectrophotometric grade): 6 x 1.0ml

MSDS: The Material Safety Data Sheets for the three products contained in this kit can be downloaded from the Strem Chemicals Web Site at www.strem.com. Locate the MSDS using the following catalog numbers:
FBBBE: 05-0058 **CBBE:** 05-0054
 Dimethylsulfoxide (ACS spectrophotometric grade): 97-4940

Storage conditions: Both compounds are stable at ambient conditions for several days. For long term storage, the kit should be stored at -20°C and protected from light.

Preparation of FBBBE:

Step 1: Add 654 μL of DMSO to 5.0 mg FBBBE (MW: 764.3 g/mol) (resulting concentration = 10 mM). FBBBE is readily soluble in DMSO. The solution can be portioned into 50 μL aliquots as needed. These solutions must be stored in the freezer at $< -20^{\circ}\text{C}$.

Step 2: Add 50 μL of 10 mM stock to 50 μL DMSO (resulting concentration = 5 mM). This stock solution can be stored in the freezer at $< -20^{\circ}\text{C}$.

Step 3: This solution can be further diluted to the desired concentration in buffer. For example add 10 μL of the 5 mM stock solution to 990 μL 1X PBS, resulting in a 50 μM solution (this solution should be used the same day in which it was prepared). This will result in a solution containing only 1% DMSO (v/v). This solution can then be used in the biological setting to image H_2O_2

Preparation of CBBE:

Step 1: Add 661 μL of DMSO to 5.0 mg CBBE (MW: 378.2 g/mol) (resulting concentration = 20 mM). Vortex until all solid has been thoroughly dissolved. The solution can be portioned into 50 μL aliquots as needed. These solutions must be stored in the freezer at $< -20^{\circ}\text{C}$.

Step 2: Add 50 μL of 20 mM stock to 150 μL DMSO (resulting concentration = 5 mM). This stock solution can be stored in the freezer at $< -20^{\circ}\text{C}$.

Step 3: This solution can be further diluted to the desired concentration in buffer. For example, add 10 μL of the 5 mM stock solution to 990 μL 1X PBS, resulting in a 50 μM solution (this solution should be used the same day in which it was prepared). This will result in a solution containing only 1% DMSO (v/v). This solution can then be used to image H_2O_2 in the biological setting.

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