

EP-HMRG

Table 1. Product information

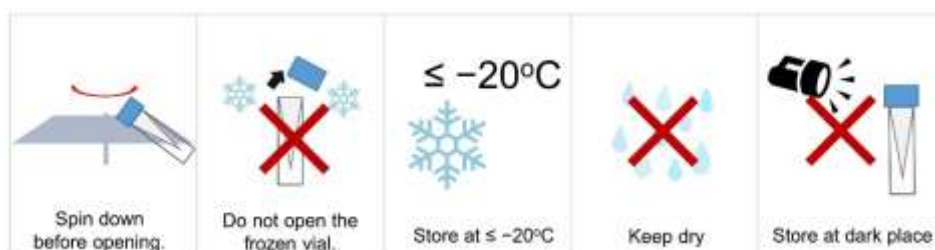
Catalog no.	Product name	Amount	Storage upon receipt	Stability
GC801	EP-HMRG	30 nmol × 5	≤-20°C, keep desiccated and protected from light.	1 year (when unopened and stored as described.)

1. About EP-HMRG

EP-HMRG is almost nonfluorescent material at pH 7 or higher. It fluoresces by the reaction with proteases that can digest the peptide sequence of EP (glutamic acid, proline). A green fluorescent product has high membrane permeability and localizes inside the lysosomes

■ Storage

Upon receipt, store the product desiccated and protected from light at ≤ -20°C. Please use up the solution after dissolving the product. Storing as a solution is not recommended.



■ Precautions

Under an acidic environment, EP-HMRG emits fluorescent by the mechanism different from proteases hydrolysis. Therefore, please dilute the probe with a neutral or weak basic solution (pH > 7). The probe also degrades and generates fluorescent products at high temperature, so use the solution promptly after dissolving. To store the solution temporarily, keep the aqueous solution on ice or freeze DMSO solution. Please use the probe rapidly as possible.

2. An example of cell imaging

■ Preparation of reagent

1. EP-HMRG is an orange solid. Before opening the vial, spin down the solid to the bottom by a microcentrifuge or by a desktop centrifuge.
2. Warm the vial to the room temperature and add 30 μL of DMSO or PBS to one vial to make 1 mM solution. Dilute the stock solution with a neutral buffer or culture medium and use the diluted solution immediately.

■ Reaction with cells

1. Dilute 1 mM EP-HMRG with HBSS or RPMI-1640 to 1-2 μ M.
2. Replace the cell culture medium with the cell staining solution and incubate at 37°C, 5% CO₂. for 5-30 minutes. A sufficient increase in fluorescent intensity is observed in the cells which highly expressed proteases that can digest the peptide sequence of EP at 5-15 minutes.
3. Since the background is negligible, it is possible to observe fluorescence without any changes. The fluorescent intensity might increase when enzyme activity proceeds. To prevent such an increase, replace EP-HMRG solution with observation buffer (such as HBSS) and then observe the cells. However, repeated washing would decrease the fluorescent intensity.

■ Fluorescent observation

The generated fluorophore can be excited by 488 nm laser and emits fluorescence at 525 nm. For fluorescence microscopy, blue excitation filter cubes for FITC or GFP are suitable.

Table 2. Related Products

Catalog no.	Product name	Major applications
GC6000	HMRG	Hydroxymethyl rhodamine green (green fluorescent probe).
A101-01	MAR	Detection of cellular hypoxia.
GC301	AcidiFluor ORANGE	Detection of lysosomes with orange fluorescence.
SK3001-01	HPF	Detection of hydroxyl radical (\cdot OH) and peroxynitrite (ONOO ⁻).
SK3002-01	APF	Detection of hydroxyl radical (\cdot OH), peroxynitrite (ONOO ⁻) and hypochlorous acid (HOCl).
GC3004-01	OxiORANGE	Detection of hydroxyl radical (\cdot OH) and hypochlorous acid (HOCl).