BioActs
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Certificate of Analysis

Flamma™ P Series

FPR-648

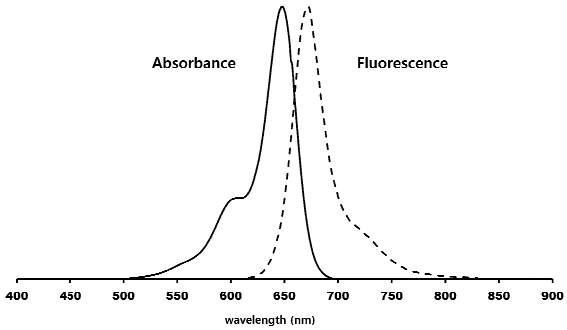
1. Contents and storage Information

Material	Amount	Lot. No.	Storage
FPR-648	1 vial (1 mg)	ZA1122FPA03	2 ~ 8 ℃ Protect from light
Carbonate buffer (pH 9.0) (Lyophilized powder)	1 vial	AL0630FBA01	

2. Product Information

Product Name	FPR-648
Code No.	PWA1215
Lot. No.	ZA1122FPA03
Formula	$C_{39}H_{51}N_3O_9S_3$
Molecular Weight	802.03
Absorption Wavelength (Maximum Peak, Room Temperature)	648 nm (1 x PBS buffer, 3.12 x 10^{-6} M) 647 nm (Distilled water, 3.12 x 10^{-6} M) 649 nm (Methanol, 3.12 x 10^{-6} M)
Molar Extinction Coefficient (Maximum Peak, Room Temperature)	2.02 × 10^5 $M^{-1}cm^{-1}$ (1 x PBS buffer, 3.12 x 10^{-6} M) 2.04 × 10^5 $M^{-1}cm^{-1}$ (Distilled water, 3.12 x 10^{-6} M) 2.32 × 10^5 $M^{-1}cm^{-1}$ (Methanol, 3.12 x 10^{-6} M)
Emission wavelength (Maximum Peak, Room Temperature)	672 nm (1 x PBS buffer, 3.12 x 10^{-6} M, ex. at 648 nm) 671 nm (Distilled water, 3.12 x 10^{-6} M, ex. at 647 nm) 676 nm (Methanol, 3.12 x 10^{-6} M, ex. at 649 nm)
Usable pH range	5 ~ 12 (8.5 ~ 9.5 recommended labeling pH)
Usable Temperature	~ 60 ℃ (25 ~ 37 ℃ recommended labeling temperature)
Usable Solvent for Stock Solution	Distilled water, DMF, DMSO (Distilled water recommended solvent)
Storage Conditions (recommended)	Store refrigerated at 2 ~ 8 ℃ in the dark. (before use) Store refrigerated at -20 ℃ in the dark. (stock solution)
Date of Manufacture	
Date of Expiration	

UV/PL Spectrum



3. Product Feature

- This product can be dissolved in distilled water, DMF and DMSO for preparing stock solution.
- Stock solutions can be diluted in water and various buffers that are used in chemical and biological experiments such as PBS buffer. (It is recommended to keep for up to 2 weeks at -20 ℃ if use buffers.)
- Conjugation with protein, peptide, antibody, biopolymer, organic compound, and nanoparticle including amino group is possible.
- Long-term storage is possible at room temperature when dissolved in water, DMF or DMSO, but it is recommended to store at -20 ℃.

4. Labeling Protocol for Protein

- Dye solution
  ① Dissolve dye (1 mg Flamma™ dye / 50 μl distilled water).
  ② Dilute the dye solution (+50 μl distilled water).
  ③ Store in refrigerated at -20 ℃.

  ※ PBS can be used for stock solution, but it is recommended to use distilled water

- Example for albumin protein (from bovine serum)
  ① Prepare protein solution with 0.1M carbonate buffer (pH 9) (2 mg protein / 1 ml buffer).
  ② Mix dye solution 1 μl and protein solution 11 μl. (or protein 1eq. : dye 4~50 eq.)
  ③ Incubate more than 0.5 hour at room temperature.
  ④ Filter the labeled protein using the separation gel or HPLC.

※ This procedure has been optimized for albumin (from bovine serum) protein. However, most of water soluble proteins are also labeled by this procedure.
※ You can select a proper filter or a suitable gel separation column for the purpose of separating the excess dyes which did not label.
※ If you want to label protein/antibody product in its stable buffer or unstable protein/antibody in basic condition, we recommend the next protocol as follow. (protein/antibody concentration 1mg/ml)
  ① Mix dye solution 2 μl and protein solution 100 μl (or protein 1eq. : dye 4~50 eq.)
  ② Add 20 μl (~100 μl) 0.1M carbonate buffer (pH 9) to the reaction solution
  ③ Mix vigorously and incubate 0.5~2 hour at room temperature.
  ④ Filter the labeled protein using the membrane filter.

- Protocol for Peptide
  ① Dissolve 1 mol peptide. (1 mg peptide / 0.1 ml DMF or DMSO or water or stable buffer)
  ② Mix 1 mol dye solution (or dye powder) and vortex. (Total Volume X ml)
  ③ Double the total volume with 0.1M carbonate buffer (pH 9).
  ④ Incubate for 4 hours at room temperature.
  ⑤ Separate the labeled peptide through HPLC.

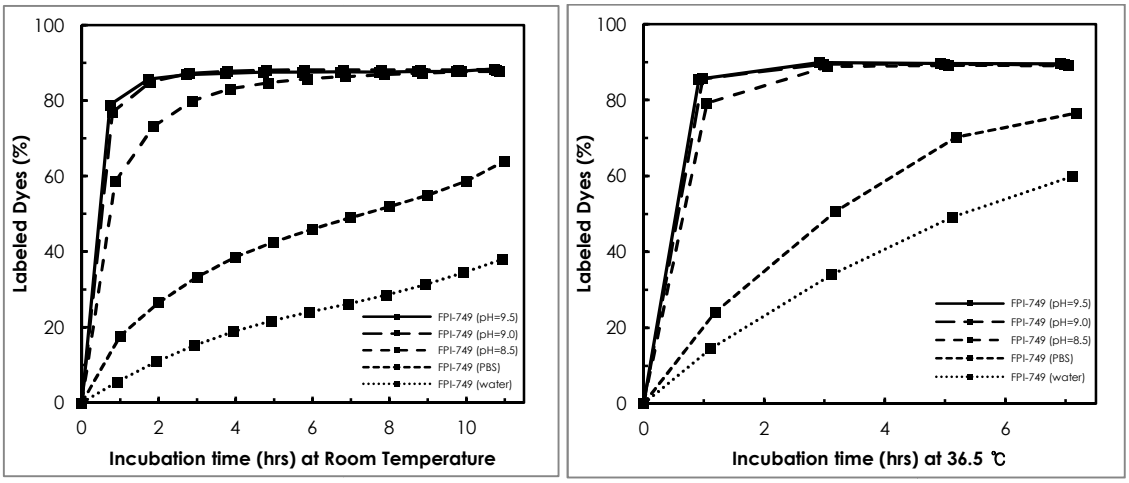
※ When your peptide doesn't dissolve perfectly in DMF (or DMSO), it is exchanged the dissolving solvent such as methanol, ethanol and dioxane.

※ If your peptide is unstable in basic condition, it is reacted in pH 7.5 0.1M phosphate buffer (or DMF or DMSO) for more than 24 hrs.

※ In case your peptide is water-soluble and stable in basic condition, it can be labeled in pH 9.0 0.1M carbonate (or phosphate) buffer) in 0.5~1 hour.

5. Labeling Information (Flamma™ P series) for Researchers

- Reaction temperature range : 4 ~ 60 °C (the best reaction rate - 37 °C)
- Reaction pH range : pH 7.5 ~ 11 (the best reaction rate - pH 9.5)



- Reaction solvent : 0.1M carbonate or phosphate buffer
- Usable reaction solvent : DMF, DMSO, Alcohol (Methanol, Ethanol), distilled water, neutral pH carbonate/phosphate/Tris buffer and most of neutral/basic biological buffer
- Reaction Time : 30 min ~ 24 hours (recommended more than 2 hours)
- The recommended usage of Flamma™ P series dye
  ① Organic Compound/ Peptide : equivalent
  ② Protein/Antibody : equivalent ~ 0.25 mg dye per 1 mg protein/antibody
  ③ Polymer : 1 mg (or what you want) dye per 1 mg polymer

6. Safety Warnings and Precautions

① Warning: For research only. Not recommended or intended for diagnosis of disease in humans or animals. Do not use internally or externally in humans or animals.

② We recommend that this products and components are handled only by those persons who have been trained in laboratory techniques and that it is used in accordance with the principles of good laboratory practice. As all chemicals should be considered as potentially hazardous, it is advisable when handling chemical reagents to wear suitable protective clothing, such as laboratory overalls, safety glasses and gloves. Care should be taken to avoid contact with skin or eyes. In case of contact with skin or eyes, wash immediately with water.

③ Caution: This dye is intensely colored and very reactive. Care should be exercised when handling the dye vial to avoid staining clothing, skin, and other items.

Michael Chung, QC Manager
August 1, 2011