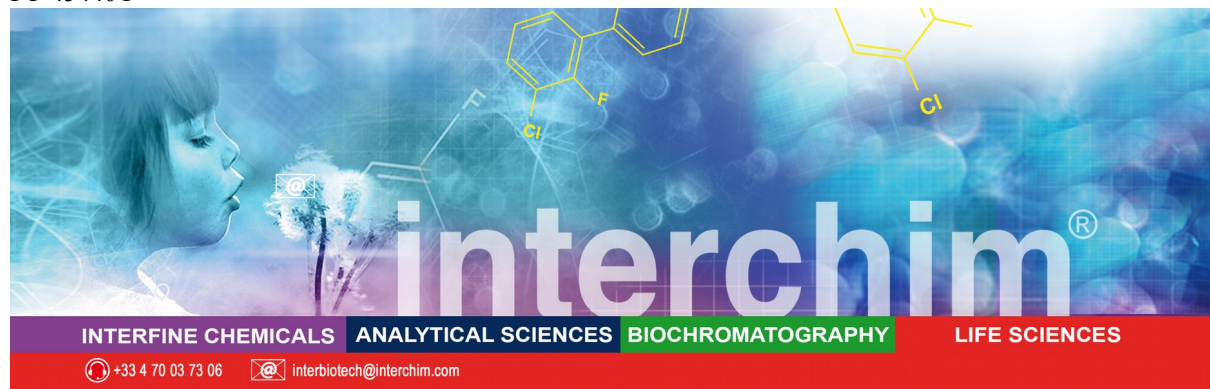


FT-45440C



BCECF

Product Information

Name :	BCECF, AM ester 2',7'-bis-(carboxyethyl)-5-(and-6)-carboxyfluorescein) acetoxymethyl ester
Catalog Number :	FP-45440A , 1 mg FP-454402 , 10x100 µg FP-45440C , 20x50 µg FP-72629A, 1 ml (1 mg/ml)
Molecular Weight :	615/821 (mean)
Soluble in:	DMF or DMSO
Absorption / Emission :	$\lambda_{exc} \lambda_{em}$ (BCECF acid, pH9 after hydrolysis) = 508 nm / 530 nm
Storage:	-20°C >1 year. (M). Protect from light and moisture

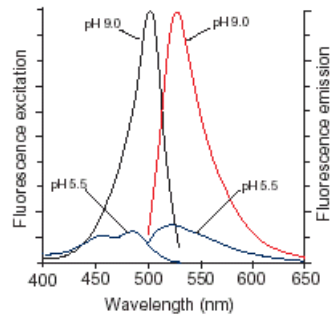
Name :	BCECF, free acid
Catalog Number :	FP-45441A , 1 mg FP-45441B, 5 mg
Structure :	C ₂₇ H ₂₀ O ₁₁
Molecular Weight :	520.45
Soluble in:	Water pH>6
Absorption / Emission :	$\lambda_{exc} \lambda_{em}$ = 508 nm / 530 nm
Extinction Coefficient :	ϵ = 90 000 M ⁻¹ m ⁻¹
Storage:	+4°C (or -20°C for long term) (K)

Introduction

BCECF is the most widely used fluorescent pH indicator. BCECF shows excellent retention in cells, and can detect small pH changes above pH 7 with higher sensitivity than fluorescein derivatives because its (ideal) pKa (6.97) is close to physiological cytosolic pH of most cells. At low pH, the dye is weakly fluorescent but becomes more fluorescent with increasing pH. The excitation spectrum of the dye undergoes a slight shift during pH change, while the wavelength of the emission maximum remains unchanged. The pH is determined ratiometrically by the relative fluorescent intensities at 535 nm when the dye is excited at 439 nm and 505 nm respectively.

FT-45440C

The applications are widely documented in the literature, including study of ionic exchanges like $\text{Cl}^-/\text{HCO}_3^-$, K^+/H^+ , Na^+/H^+ , $\text{Na}^+/\text{Ca}^{2+}$, lactate transport and metabolism, NH_4^+ transport, apoptosis, cytotoxicity, multidrug resistance, cell volume changes and cytosolic pH regulation in osteoblasts and osteoclasts, and phagocytosis.



Fluorescence excitation (detected at 535 nm) and emission (excited at 490 nm) spectra of BCECF in pH 9.0 and 5.5 buffers.

BCECF AM ester is used in intracellular pH changes monitoring in mammalian fibroblasts, gastric cells, lymphocytes, myocytes, and distal convoluted tubules. Cells in suspension or adherent are easily loaded by simple incubation in a 1–10 μM solution.

BCECF AM ester is a mixture of three components. It is a membrane permeant form of BCECF that can be loaded into cells via incubation. Once in the cells, BCECF AM ester is readily hydrolyzed by cytosolic esterases to BCECF acid.

BCECF acid is membrane-impermeant and is used for pH measurements in intercellular spaces of epithelial cell monolayers, interstitial spaces of normal and neoplastic tissue and isolated cell fractions. It can be loaded into cells via microinjection, electroporation, or scrape loading. The net anionic charge is also higher, resulting in a slower leakage from cells. Useful in cell proliferation assays.

Directions for use

Handling and Storage

Indicator salts can be stored desiccated and protected from light at room temperature, +4°C.

Stock solutions of the salts may be prepared in distilled water or aqueous buffers (pH>6) and stored at +4°C protected from light.

AM esters can be stored desiccated and protected from light at -20°C > 6 months. It should be reconstituted in anhydrous dimethylsulfoxide (DMSO) at 1–10mM. The stock solution should be stored desiccated at -20°C for at least 6 months. The working solution (diluted in aqueous media) are used as soon as possible without storage.

Guidelines for use

1- Prepare 2–100 μM BCECF solution with PBS or an appropriate buffer.

Note : It is often more convenient and effective to add the non-ionic detergent Pluronic® F127 to get further a better dissolution of AM indicator: mix the AM ester stock solution in DMSO with an equal volume of 20% (w/v) Pluronic® F127 in DMSO before dilution in the loading medium, making the final Pluronic® F127 concentration about 0.02%.

2- Add BCECF AM solution with 1/10 of the volume of cell culture medium to the cell culture. Incubate the cell at +37 °C for 15–30 min.

3- Wash cells twice with PBS or an appropriate buffer.

4- Observe the cells using a fluorescence microscope with 500 nm excitation and 530 nm emission filters.

Related products

- BAPTA AM ester, FP-486103
- SBFI, FP-82902A
- Pluronic® F127, FP-37361
- SPQ, FP-46922A
- Fura-2, FP-42776A
- CCCP, 091640

References

BCECF AM :

- **Foy R.A., et al.**, « The Effects of Hypoxia on pH_i in Porcine Coronary Artery Endothelium and Smooth Muscle A Novel Method for Measurements in Endothelial Cells In Situ », *Circulation Research.*, **80**, 21 (1997) [Article](#)
- **Futsaetner, C.M., et al.** *Can. J. Microbiol.* **39**, 180 (1993)
- **Hayashi, H., et al.**, « Intracellular Ca²⁺ concentration and pH_i during metabolic inhibition », *Am. J. Physiol.* **262**, C628 (1992) [Abstract](#)
- **Ho, A.K., et al.**, « Intracellular pH on adrenergic-stimulated cAMP and cGMP production in rat pinealocytes », *Am. J. Physiol.* **261**, C642 (1991) [Abstract](#)
- **Marechal X., et al.**, « In Vivo Application of Intestinal pH Measurement Using 2',7'-Bis(carboxyethyl)-5,6-carboxyfluorescein (BCECF) Fluorescence Imaging. », *Photochem Photobiol* **70**, 813 (1999) .
- **Rink, T.J., et al.**, « Cytoplasmic pH and Free Magnesium in Lymphocytes ». *Journal of Cell Biology* **95**, 189-196 (1982)
- **Shepherd, R.M., et al.**, « Regulation of intracellular pH in single rat zona glomerulosa cells », *Am. J. Physiol.* **262**, C182 (1992) [Abstract](#)
- **Slayman CL, et al.**, « Endosomal accumulation of pH indicator dyes delivered as acetoxymethyl esters. », *J Exp Biol.*, **196**,419 (1994)
- **Weiner, I.D. et al.**, « Apical proton secretion by the inner stripe of the outer medullary collecting duct », *Am. J. Physiol.* **256**, F957 (1989). [Abstract](#)
- **Weinlich M., et al.**, « BCECF in Single Cultured Cells: Inhomogeneous Distribution but Homogeneous Response » , *J Exp Biol* **201**, 57 (1998) [Article](#)

BCECF free acid :

- **Kolber M.A., et al.** *J. Immunol. Meth.* **108**, 255 (1988)
- **Carpenter L., et al.**, « The kinetics, substrate and inhibitor specificity of the lactate transporter of Ehrlich-Lettre tumour cells studied with the intracellular pH indicator BCECF. », *Biochem. J.* **304**, 751 (1994) [Abstract](#)
- **Seller J.R. et al.** *J. Immunol. Meth.* **172**, 255 (1994)

Ordering information

Catalog size quantities and prices may be found at <http://www.fluoprobes.com>
Please inquire for higher quantities (availability, shipment conditions).

For any information, please ask : FluoProbes® / Interchim; Hotline : +33(0)4 70 03 73 06

Disclaimer : Materials from FluoProbes® are sold **for research use only**, and are not intended for food, drug, household, or cosmetic use. FluoProbes® is not liable for any damage resulting from handling or contact with this product.

Pluronic® F127 is a trademark from BASF.