PRODUCT INFORMATION SHEET

Calcein-AM order #: MFP-C430

Product Name: **Calcein-AM**
Chemical Name: 3',6'-Di(O-acetyl)-2',7'-bis[N,N bis(carboxymethyl)aminomethyl] fluoresceintetra-acetoxymethyl ester
CAS Number: 148504-34-1
$C_{46}H_{46}N_2O_{23}$ = 994.86

Unit	Product Code
1 mg	**MFP-C430**

Product Description:

Storage: -20ºC
Shipping Condition: RT

Appearance: white or slightly yellow crystals
Purity: >90.0 % (HPLC)
λex: 490 nm, λem: 515 nm

Calcein-AM readily passes through the cell membrane of viable cells because of its enhanced hydrophobicity as compared to Calcein. After Calcein-AM permeates into the cytoplasm, it is hydrolyzed by esterases to Calcein, which remains inside the cell. Among other reagents, including BCECF-AM and Carboxy-fluorescein diacetate, Calcein-AM is the most suitable fluorescent probe for staining viable cells because of its low cytotoxicity. Calcein does not inhibit any cellular functions such as proliferation or chemotaxis of lymophocytes. In addition, viability assays using Calcein are reliable and correlate well with the standard ^{51}Cr-release assay. The excitation and emission wavelengths of calcein are 490 nm and 515 nm, respectively.

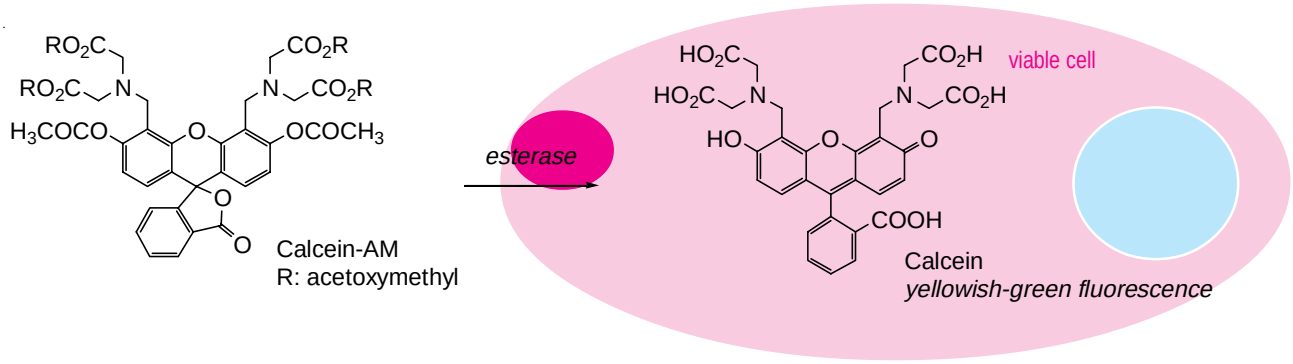
General Protocol:

1. Prepare 1 mM Calcein-AM solution with anhydrous DMSO. This stock solution should be stored aliquoted at -20°C. Prior to the experiment dilute 1-50 µM Calcein-AM solution with PBS.[a)]
2. Add Calcein-AM solution with 1/10 of the volume of cell culture medium to the cell culture.[b)]
3. Incubate the cells at 37ºC for 15-30 minutes.
4. Wash cells twice with PBS or an appropriate buffer.
5. Observe the cells using a fluorescence microscope with 490 nm excitation and 515 nm emission filters.

a) If the Calcein-AM loading into cells is difficult, use a detergent such as Pluronic F127.
b) Or you may replace the culture medium with 1/10 concentration of Calcein-AM buffer solution.

MoBiTec GmbH, Lotzestraße 22a, D-37083 Göttingen, Tel.: ++49 551-70 722- 0, Fax: ++49 551-70 722-22
E-mail: info@mobitec.com or order@mobitec.com, INTERNET: http://www.mobitec.com

PRODUCT INFORMATION SHEET



Mechanism of Viable Cell Staining

References:

1. Weston, S. A., et al., *J. Immunol. Methods*, **133**, 87 (1990).
2. X. M. Wang, et al., *Human Immunol.*, **37**, 264 (1993).
3. Lichtenfels, R., et al., *J. Immunol. Methods*, **172**, 227 (1994).
4. Rat, P., et al., *Cell. Biol. Toxicol.*, **10**, 329 (1994).
5. Braut-Boucher, F., et al., *J. Immunol. Methods*, **178**, 41 (1995).
6. Liminga, G., et al., *Anti Cancer Drugs*, **6**, 578 (1995).
7. DenGendt, C. M., et al., *Clin. Chim. Acta.*, **249**, 189 (1996).
8. Jonsson, B., et al., *Eur. J. Cancer*, **32A**, 883 (1996).
9. Sunder-Plassmann, G., et al., *Immunol. Invest.*, **25**, 49 (1996).
10. Tiberghien, F., et al., *Anticancer Drugs*, **7**, 568 (1996).
11. Decherchi, P., et al., *J. Neurosci. Methods*, **71**, 205 (1997).
12. Imbert, D., et al., *Cornea*, **16**, 666 (1997).
13. Dhar, S., et al., *Eur. J. Pharmacol.*, **346**, 315 (1998).
14. Essodaigui, M., et al., *Biochemistry*, **37**, 2243 (1998).
15. Gatti, R., et al., *J. Histochem. Cytochem.*, **46**, 895 (1998).
16. Monette, R., et al., *Brain Res. Brain Res. Protoc.*, **2**, 99 (1998).
17. Oral, H. B., et al., *Endothelium*, **6**, 143 (1998).
18. Adler, M., et al., *Neurotoxicology*, **20**, 571 (1999).
19. Giacomello, E., et al., *Biotechniques*, **26**, 758 (1999).
20. Jarvis,C. R., et al., *Neuroimage*, **10**, 357 (1999).
21. Legrand, O., et al., *Adv. Exp. Med. Biol.*, **457**, 161 (1999).
22. Petersen, T. K., et al., *Vet. Immunol. Immunopathol.*, **68**, 283 (1999).
23. Petronilli, V., et al., *Biophys. J.*, **76**, 725 (1999).
24. Roden, M. M., et al., *J. Immunol. Methods*, **226**, 29 (1999).

MoBiTec GmbH, Lotzestraße 22a, D-37083 Göttingen, Tel.: ++49 551-70 722- 0, Fax: ++49 551-70 722-22
E-mail: info@mobitec.com or order@mobitec.com, INTERNET: http://www.mobitec.com