

FT-AS2DHA



FluoProbes 365XL-Phosphate

Product Description

Name :	FluoProbes® 365XL-Phosphate
Catalog Number :	4(1H)-Quinazolinone, 6-chloro-2-[5-chloro-2-(phosphonoxy)phenyl]-,disodium salt FP-AS2DHA 5 mg FP-AS2DHB 25 mg
Structure & Properties:	CAS: [146508-65-8]
physical	MW= 431,08 g/mol
optical	Soluble: Water
	Absorption / Emission: $\lambda_{exc}\lambda_{em} = 365 / 530 \text{ nm}$
Storage:	-20°C Protect from light and moisture



Colonies of *S. aureus* grown on solid media supplemented with FluoProbes® 365XL – Phosphate, disodium salt under UV light.

Introduction

Fluorogenic and chromogenic enzyme substrates are widely used in microbial growth media to detect enzyme activities indicative for target organisms. Although more sensitive than chromogenic substrates, common fluorogenic substrates are scarcely used in plating media because their fluorescent products are diffusible and hamper localization of enzyme activity on the single colony level. Therefore, mainly indoxyl-based chromogenic substrates are employed in plating media. Substrates based on the FluoProbes® 365XL fluorogen combines the advantage of more sensitive signals of fluorogenic substrates with the precise localized staining of indoxyl substrates.

FluoProbes® 365XL - Phosphate, disodium salt is a fluorogenic water soluble substrate for phosphatase enzymes. After cleavage of the substrate by phosphatase activity an insoluble product is formed that is highly fluorescent in the solid state. Upon UV irradiation this very stable fluorophore emits green light in the 530 nm range for an extended period of time.

FluoProbes® 365XL - Phosphate in Microbiological Assays:

FluoProbes® 365XL - Phosphate is a perfectly suitable substrate for the detection of phosphatase activity in solid matrices such as agar media, tissue sections or microscopic preparations. FluoProbes® 365XL - Phosphate, disodium salt is used in microbiology as a substrate for bacterial phosphatases. The substrate is suitable to detect the specific phosphatase activity of *S. aureus* (including MRSA) or *C. perfringens* in food samples or clinical samples. Colonies

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growing on plating media supplemented with FluoProbes® 365XL emit green light upon UV light excitation (Excitation max: 365 nm; emission max: 530 nm). The enzyme product formed from FluoProbes® 365XL in agar plating media enables high precision for the localization of the relevant colonies on the plate. As a result small bacteria colonies of phosphatase-positive bacteria can be detected already after a short incubation time.

An additional advantage of FluoProbes® 365XL substrates is their very low toxicity. In contrast to most fluorogenic substrates the growth of the strains of interest is not inhibited by the substrate. FluoProbes® 365XL fluorescence does not require oxygen and therefore allows the incubation under anaerobic culture conditions.

FluoProbes® 365XL - Phosphate in Microscopy/Histology:

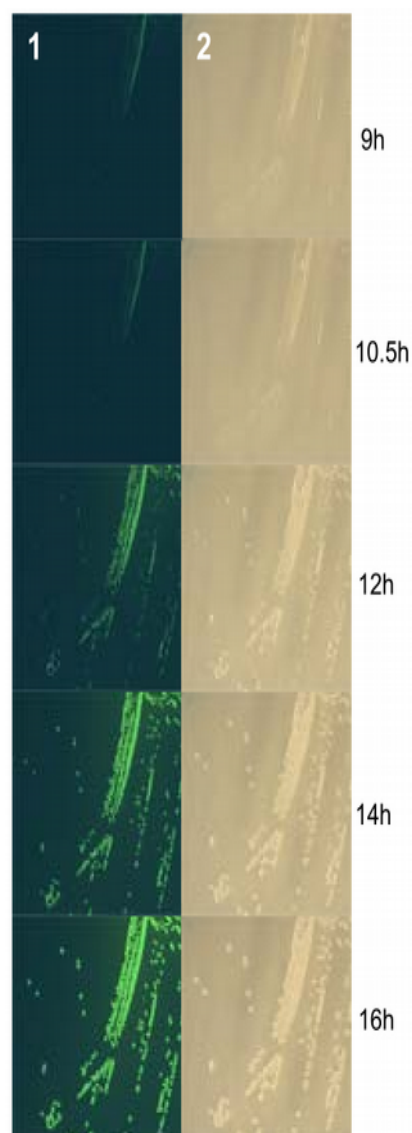
In cytochemical and histochemical imaging FluoProbes® 365XL - Phosphate, disodium salt can be used as a substrate for Alkaline Phosphatase (AP)-coupled antibodies. The fluorophore that is released from FluoProbes® 365XL - Phosphate by phosphatase activity will precipitate. This insoluble fluorophore enables a precise localization of the antibody-bound epitope in cells or tissues under a fluorescence microscope or fluorescence imaging system. The green fluorescence of FluoProbes® 365XL can be clearly differentiated from autofluorescence or counterstainings such as blue DAPI or red FluoProbes fluorescence.

Application example

MRSA RKI 02/02756 was plated on selective solid culture media (containing cefoxitin, sulbactam and polymyxin B) supplemented with FluoProbes® 365XL - Phosphate, disodium salt. Plates were incubated at 37°C for 16 hours.

Figure: A Sector of an MRSA selective medium agar plate containing FluoProbes® 365XL - Phosphate, disodium salt as seen under (1) UV light (366 nm) and (2) white light after the incubation times given on the right.

Results: Media with FluoProbes® 365XL - Phosphate proved more sensitive than media containing usual chromogenic substrates. With FluoProbes® 365XL - Phosphate the time to detection was mainly limited by the time the colonies needed to grow to a visible size, i.e. around 12 hours on the medium tested.



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Technical and scientific information

Instructions for use :

Add FluoProbes® 365XL - Phosphate, disodium salt as concentrated, filter-sterilized stock solution after autoclaving.

Stock solution:

Dissolve FluoProbes® 365XL - Phosphate, disodium salt at 10 mg/mL in deionized water. The stock solution may be turbid. Sterilize the solution using a 0.2 µm filter. This step also clarifies the solution.

Recommended working concentration:

Use FluoProbes® 365XL - Phosphate, disodium salt at a final concentration of 0.2 – 0.4 g/L in microbiological media.

Typical media composition for the detection of *S. aureus* / MRSA:

Compound:	g/L:	Supplements:
Pepton/Tryptone	11.0	FluoProbes® 365XL-Phosphate,
Yeast Extract	3.0	disodium salt 0.2 – 0.4 g/L
Meat Extract	2.0	MgCl ₂ 1 mM
NaCl	5.0	
LiCl	5.0	Optional:
Sodium pyruvate	2.0	Desferal 0.2 g/L
Cycloheximide	0.2	Mannitol 4 – 9 g/L
Agar	14.0	
pH	7.2 – 7.3	Antibiotics to adjust selectivity

Reference strains

Positive for phosphatase activity after 14 hours:

<i>S. sciurii</i>	MRSA RKI 1682/06
<i>S. intermedius</i>	MRSA RKI 1793/06
<i>S. arletta</i>	MRSA ATCC 33592
MSSA RKI 1807/06	MRSA RKI 1682/06
MSSA RKI 1823/06	MRSA RKI 1793/06
MSSA RKI 1822/06	MRSA ATCC 33592
MSSA ATCC 29213	

Legals & Disclaimer

This product is sold for Research & Development in vitro use only.

Ordering information

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

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

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