FluoProbes[®]

NBD

Product Information

Amine-reactive fluorescent probe

Name :	4-Chloro-7-nitrobenzofurazan (NBD Cl) 4-chloro-7-nitrobenz-2-oxa-1,3-diazole	CI I
Catalog Number :	FP-465409 25 mg Ultra Pure Grade FP-T3226A 1 g (\geq 98%)	
Molecular Weight :	MW= 199.55	
Solubility:	DMSO, DMF, CH ₃ OH, CH ₃ CN and CHCl ₃	Ň
Absorption / Emission :	$\lambda_{\text{exc}} \setminus \lambda_{\text{em}} (CH_3OH) = 328 / \text{none}$	
	$\lambda_{exc} \setminus \lambda_{em}$ (derivatized with n-butylamine) = 465 / 535nm	NOz
	λ_{exc} (coupled) = 464-470/512-530 nm (depends on molecule and environment)	
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Name :	4-Fluoro-7-nitrobenzofurazan (NBD F)	
Catalog Number :	FP-U0573A 5 mg Ultra Pure Grade	N,
Molecular Weight :	MW= 183.1	
Solubility:	10 mg/ml ethanol, 10 mg/ml acetonitrile	N,
Absorption / Emission :	$\lambda_{\text{exc}} \setminus \lambda_{\text{em}} (CH_3OH) = 328 / \text{none}$	I NO ₂
	λ_{exc} (coupled) = 464-470/512-530 nm (depends on molecule and environment)	

Storage:

 $+4^{\circ}C$ (or $-20^{\circ}C$ for long term) (L) Protect from light and moisture

Introduction

NBD-Cl

NBD-Cl is widely used to label peptides, proteins, drugs and other biomolecules. It is also a popular derivatizing reagent for HPLC analysis. NBD-Cl is non-fluorescent until it reacts with amino groups such as amines, amino acids, peptides, and proteins to form highly fluorescent compounds. NBD-Cl also reacts with thiol group to form fluorescent adducts.

The fluorescence spectra of NBD-amine adducts are highly environment-sensitive, the fluorescence intensity decreases significantly in aqueous solutions. NBD-labeled compounds are orange. The NBD-amine adducts have λ ex around 464 nm and λ em around 512 nm in aqueous solutions.

NBD-Cl is a pre-labeling compound for HPLC analysis of small molecules, especially amine compounds. It has i.e. be useful for agricultural chemicals that have unstable carbamate structures.

NBD-F

NBD-F has properties and applications similar to those of NBD-Cl. Upon reacting with amines and thiol compounds it generates the same fluorescent adducts as those of NBD-Cl. Compared with NBD-Cl, NBD-F is much more reactive, and should be more carefully stored. For example, the reaction of NBD fluoride with glycine is reported to be 500 times faster than the reaction of NBD chloride with glycine. Both NBD chloride and NBD fluoride are extensively used as derivatization reagents for chromatographic analysis of amino acids and other low molecular weight amines.

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FT-46540A

Directions for use

Guidelines for use

NBD-F and can label primary and secondary amines under mild conditions (1 min reaction at 60 °C in a weak basic solution).

NBD-Cl has similar properties and applications to NBD-F. It is however more reactive, and should be more carefull stored and handled.

NBD Labeling Protocol

- 1. To prepare sample solution, mix or dissolve a sample with 50 mM borate buffer (pH 8.0) with 20 mM EDTA.
- 2. Mix 300 µl of the sample solution and 100 µl of 100 mM NBD-F/acetonitrile solution in a reaction vial.
- 3. Heat the vial at 60 °C for 1 min and then cool it on an ice bath.
- 4. Add 400 µl of 50 mM HCl aqueous solution to the reaction mixture.
- 5. Use this mixture for HPLC analysis to determine NBD-labeled compounds.

Other protocols may be found in the literature.

References – NBD Cl

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- Charles R. et al., Protein Sulfenation as a Redox Sensor: Proteomics Studies Using a Novel Biotinylated Dimedone Analogue, *Mol. Cell. Proteomics*, 6: 1473 - 1484 (2007) <u>Article</u> (Analysis and Detection of Protein Sulfenic Acids)
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- **Tani M**, *et al.* (1998). Enzymatic synthesis of omega-amino-ceramide: preparation of a sensitive fluorescent substrate for ceramidase. *Anal Biochem* 263, 183-8.

Related products

See <u>BioSciences Innovations catalogue</u> and <u>e-search tool</u>. Other reagent for the detection of Amines

- ABD-F <u>FP-57564A</u> (reacts faster with thiol & amines)
- SBF-Cl <u>FP-AM858A</u> (amine reactive)
- SBF-F <u>FP-AM859A</u>

Ordering information

Ordering information

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

Info@fluoprobes.com Technicalsupport@fluoprobes.com Order-online@fluoprobes.com FluoProbes[®], powered by **interchim**^{213 Avenue J.F. Kennedy - BP 1140 ^{213 Avenue J.F. Kennedy - BP 1140} ^{213 Avenue J.F. Kennedy - BP 1140</sub> ^{213 Avenue J.F. Kennedy - BP 1140 ^{213 Avenu}}}}</sup></sup></sup></sup></sup></sup></sup></sup></sup></sup></sup></sup></sup></sup></sup></sup></sup></sup></sup></sup></sup></sup></sup></sup></sup></sup></sup>

- IANBD amide, <u>FP-79659A</u>
- OPA, <u>02727A</u>
- Fluorescamine, <u>FP-12631E</u>



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