



Fluoprobes® -dUTP, DNA labeling by Nick Translation

Product Information

Name :	Fluoprobes® 425A -dUTP,
Catalog Number :	FP-CI7950 , 10 µl of a 1 mM solution FP-CI7951 , 50 µl of a 1 mM solution
Soluble:	Soluble in clear aqueous solution pH 7.5
Absorption / Emission :	$\lambda_{exc} \lambda_{em} = 436 / 484 \text{ nm}$

Storage: For long term at -20°C (up to 12 months after date of delivery)
Protected from light.

Introduction

Fluorescent nucleotides are used for direct labeling of DNA Nick Translation using DNA Polymerase I/Dnase I. The fluorophore should not affect the polymerase activity, while eliciting excellent brightness in downstream processes.

* NTPs are available labeled by FluoProbes® dyes at various positions using different lengths of linkers, and covering the whole UV/VIS spectrum. They are supplied ready to use in aqueous solutions, at 1 mM concentration.

FluoProbes® dyes results in nucleotides conjugates that have superior properties compared to most of other commercially available dye-labeled NTPs :

- Excellent solubility in water
- Higher signal intensity
- Better photostability
- Lower molecular weight of dye resulting in minimal steric hindrance

Instructions for use

Prepare the following reaction mixture in a sterile vial.

Recommended NT assay

Sample Material can be supercoiled or linearized plasmid DNA, cosmid or BAC DNA, whole or partial chromosomes or purified PCR products.

1. Vortex the mix gently to assure homogeneity and centrifuge briefly to collect the reaction mixture at the bottom of the tube.
2. Place the tube in a precooled thermomixer at 15°C. The incubation time strongly depends on the Polymerase I / DNase I activities. A well balanced enzyme ratio is required to generate labeled fragments in the desired size range. An individual optimization of the enzyme concentrations is recommended to generate DNA fragments in the range between 300 and 800 bp at an incubation time of 60 minutes.
3. To control the length of the fragments load 2 µl of the assay on an agarose gel. Place the reaction tube at -20°C while running the gel.
4. To get smaller fragments add again Polymerase I and Dnase I and extend the incubation at 15°C.
5. For final stopping the reaction add 5µl EDTA (0.5 M, pH 8). Proceed to purification of the probe or store at -20°C.

Related products

20 µl nick translation labeling assay		
amount	final conc.	component
2 µl	1x	10x Reaction buffer
1 µl	50 µM	dATP (1 mM)
1 µl	50 µM	dCTP (1 mM)
1 µl	50 µM	dGTP (1 mM)
0.5 µl	25 µM	dTTP (1 mM)
0.5 µl	25 µM	Atto425-dUTP-NT (1 mM)
1-1.5 µg	50-75 ng/µl	Template DNA
	0.2 u/µl	DNA Polymerase I
	0.002 u/µl	DNase I
fill up to 20 µl		PCR-grade water

- Nick Translation Labeling Kits
- PCR Labeling Kits
- Fluorescent Labeled Aminoallyl-dUTP for PCR
- Standard PCR / Thermophilic Polymerases
- Deoxynucleotides (dNTPs)
- Primers and Oligonucleotides
- DNA Ladders
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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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