Data Sheet

Jena Bioscience

Atto488-dUTP-PCR

Fluorescent labeled aminoallyl-dUTP

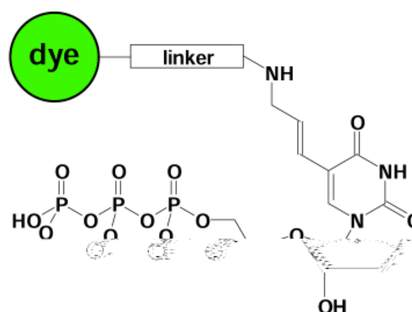
DNA labeling by PCR

Cat.-No.	Amount	Conc.
PP-302S-488	10 µl	1 mM
PP-302L-488	50 µl	1 mM

For *in vitro* use only
Quality guaranteed for 12 months
Store at -20°C in the dark
Avoid frequent thawing and freezing

Description
Atto488-dUTP-PCR is recommended for direct enzymatic labeling of DNA. The dye-dUTP is specially optimized for incorporation into DNA by PCR using Taq polymerase. The excellent stability and quantum yield of the fluorophore combined with a high incorporation rate of the dye-dUTP complex makes it the ideal choice for a broad range of fluorescence applications.
In PCR labeling, repeated cycles of denaturation, annealing and extension allow the amplification of a specific DNA fragment. The target DNA is denatured by heating followed by annealing of primers. Extension of the annealed primers with Taq polymerase results in a duplication of the DNA fragment in each cycle. When dTTP is partially substituted by dye-dUTP a fluorescent labeled double-stranded DNA is generated.
The resultant DNA is suited for application in FISH, microarray gene expression profiling and other nucleic acid hybridization assays.
Protect fluorescent labeled dUTP from light and carry out experimental procedures in low light conditions.

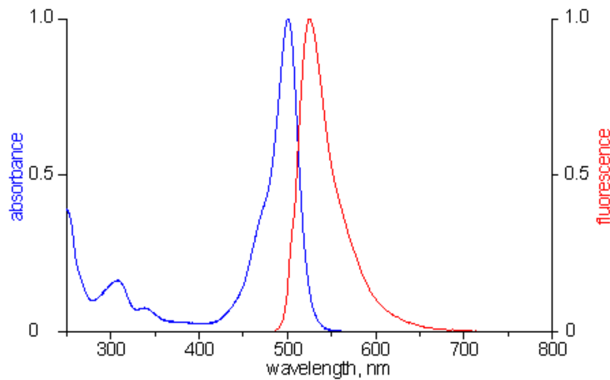
Structure



Atto488-dUTP, the dye is attached via an optimized linker to aminoallyl-dUTP

Atto488-dUTP-PCR
1 mM 5-(3-aminoallyl)-2'-deoxy-uridine-5'-triphosphate labeled with Atto488, triethylammonium salt, pH 7.5, purity >95%

Spectroscopic data
Excitation maximum: λ_{Ex} = 501 nm
Emission maximum: λ_{Em} = 523 nm
Extinction coefficient: ε_{max} = 90,000 cm^{-1} M^{-1}



Atto488 excitation and emission spectra

Recommended PCR assay

20 µl PCR labeling assay			
Component	**Stock conc.**	**Amount**	**Final conc.**
High yield buffer without $MgCl_2$ (Cat.-No. PCR-201)	10x	2 µl	1x
$MgCl_2$ stock solution	25 mM	1.6 µl	2 mM
dATP	1 mM	2 µl	100 µM
dCTP	1 mM	2 µl	100 µM
dGTP	1 mM	2 µl	100 µM
dTTP	1 mM	1 µl	50 µM
Atto488-dUTP-PCR	1 mM	1 µl [1)]	50 µM [1)]
forward Primer	10 µM	1 µl	500 nM
reverse Primer	10 µM	1 µl	500 nM
Template DNA		0.1-10 ng	5-500 pg/µl
Taq Pol (Cat.-No. PCR-201)	5 units/µl	0.2 µl (1 unit)	0.05 units/µl
PCR grade H_2O		Fill up to 20 µl	

1) The optimal final concentration of the labeled nucleotide may vary depending on the application.

Recommended cycling conditions

Initial denaturation	94°C	2 min	1x
Denaturation	94°C	30 sec	25-30x
Annealing [1)]	50-60°C	30 sec	
Elongation [2)]	72°C	1 min	
Final elongation	72°C	5 min	1x

1) The annealing temperature depends on the melting temperature of the primers used.
2) The elongation time depends on the length of the fragments to be amplified. A time of 2 min/kbp is recommended.

For optimal amplification results and high incorporation rates an individual optimization of the recommended PCR assay and cycling conditions may be necessary for each new primer-template pair.

Related products
Kits for DNA labeling
Standard PCR / Thermophilic Polymerases
Deoxynucleotides (dNTPs)
Primers and Oligonucleotides
DNA Ladders

For detailed information please visit
www.jenabioscience.com/pcr