

Product Information

CF™ Dye dCTP Conjugates

Catalog number	Size	Product	Molecular weight	Ex/Em (nm)
40067-T	5 nmol	CF™488A-dCTP	~1428	490/515
40067	25 nmol			
40057-T	5 nmol	CF™532-dCTP	~1447	527/558
40057	25 nmol			
40058-T	5 nmol	CF™543-dCTP	~1647	541/560
40058	25 nmol			
40027-T	5 nmol	CF™555-dCTP	~1553	555/565
40027	25 nmol			
40055-T	5 nmol	CF™568-dCTP	~1652	562/583
40055	25 nmol			
40056-T	5 nmol	CF™594-dCTP	~1667	593/614
40056	25 nmol			
40066-T	5 nmol	CF™640R-dCTP	~1593	642/662
40066	25 nmol			
40028-T	5 nmol	CF™647-dCTP	~1579	650/665
40028	25 nmol			
40068-T	5 nmol	CF™660R-dCTP	~1826	663/682
40068	25 nmol			

Storage and Handling

Store desiccated at $\leq -20^{\circ}\text{C}$. When stored as directed, product should be stable for at least 6 months from the date of receipt. For aqueous solutions, prepare single use aliquots and store protected from light at -20°C for up to 6 months. Avoid freeze-thaw cycles. We recommend preparing a 1 mM stock solution in 10 mM Tris pH 7.4.

Product Application

CF™ dyes are Biotium's next-generation fluorescent dyes, with combined advantages in brightness, photostability, and water solubility compared to other dyes like Alexa Fluor®, DyLight®, Cy® Dye, and IRDye®. CF dye conjugates of dCTP can be used in standard DNA labeling and synthesis protocols to generate fluorescent dsDNA and oligonucleotide probes.

Protocols

DNA labeling by PCR

1. Materials Required but not Provided

- Taq DNA polymerase (see note under product application)
- 10X Taq reaction buffer
- 25 mM MgCl_2
- dATP, dTTP, dCTP, dGTP (separate solutions), 1 mM each
- DNA template
- Forward and reverse primers, 10 μM each
- PCR clean-up kit (optional)

2. PCR reaction

2.1 For each labeling reaction, set up the PCR reaction mix as shown below:

Component	Volume per reaction	Final concentration (after addition of dUTP)
10X Taq reaction buffer	2 μL	1X
25 mM MgCl_2	2 μL	5 mM
1 mM dATP	2 μL	100 μM
1 mM dCTP	1 μL	50 μM
1 mM dGTP	2 μL	100 μM
1 mM dTTP	2 μL	100 μM
10 μM forward primer	1 μL	500 nM
10 μM reverse primer	1 μL	500 nM
Template	1 ng	50 pg/ μL
Taq	1 U	0.05 U/ μL
Molecular grade dH_2O	to 19 μL total	

2.2 Add 1 μL of 1 mM CF dye dCTP to the reaction tube.

Optional: for an unlabeled control reaction, add 1 μL of 1 mM dTTP (unlabeled) instead of CF dye dCTP.

2.3 Perform PCR according to the following cycling protocol:

Denaturing/hot-start Taq activation 94°C, 2 min. (see note 1)	Hold
Denaturing 94°C 30 sec.	Cycle 30X
Annealing (see note 2) 30 sec.	
Extension 72°C 1 min. (see note 3)	
Final extension 72°C 5 min.	Hold

Notes:

1. This protocol was optimized for Cheetah™ Hot Start Taq polymerase (see related products). Other hot-start Taq polymerases may require longer activation times.
2. Set the annealing temperature 5°C below the melting temperature (T_m) of your primers.
3. This cycling protocol was optimized for 200-300 bp amplicons. Longer amplicons may require longer extension times.

2.4 Optional: use a PCR clean-up kit to remove unincorporated nucleotides.

2.5 Run 10% of the labeled product on an agarose gel without DNA dye added to analyze the efficiency and specificity of the PCR reaction. CF dye fluorescence can be imaged on a UV light box or laser-based gel scanner. Note: Far-red fluorescence emission (650 nm or longer) is not visible to the human eye.

2.6 Post-stain the gel with DNA gel stain to image the total PCR product or optional unlabeled control PCR product.

Related Products

Catalog number	Product
30063	CF™488A TUNEL Assay Apoptosis Detection Kit
30064	CF™594 TUNEL Assay Apoptosis Detection Kit
30074	CF™640R TUNEL Assay Apoptosis Detection Kit
40004	CF™405S-dUTP
40008	CF™488A-dUTP
40002	CF™543-dUTP
40005	CF™568-dUTP
40006	CF™594-dUTP
40007	CF™640R-dUTP
40003	CF™680R-dUTP
40031	CF™555 ddCTP
40032	CF640R UTP
40001	5-Tetramethylrhodamine-dUTP
40063	Fluorescein-12-dUTP
40059	DEAC-dUTP
40029	Biotin-11-dUTP
40022	Biotin-16-dUTP
40030	Biotin-20-dUTP
40035	Biotin-11-CTP
40036	Biotin-11-dCTP
40033	Biotin-11-UTP
40023	Biotin-16-UTP
40034	Biotin-20-UTP
40078	Digoxigenin-dUTP, alkali stable
40020	5-Aminoallyl-dUTP
40021	5-Aminoallyl-UTP
40052	dNTP Set, 100 mM each
29050	Cheeta™ Hot Start Taq DNA Polymerase
41003	GelRed™ Nucleic Acid Gel Stain, 10,000X in water
41004	GelGreen™ Nucleic Acid Gel Stain, 10,000X in water

Please visit our website at www.biotium.com to view our full selection of CF™ dye bioconjugates, including antibodies, antibody labeling kits, phalloidin, Annexin V and α -bungarotoxin, as well as fluorescent reagents and kits for genomics and cell biology research.

CF dye technology is covered by pending US and international patents.

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