

# ReadiLink™ iFluor™ 555 Oligo and ssDNA Labeling Kit

Catalog number: 17482, 17483 Unit size: 10 Reactions, 20 Reactions

Component	Storage	Amount (Cat No. 17482)	Amount (Cat No. 17483)
Component A: iFluor™555-dUTP	Freeze (< -15 °C), Minimize light exposure	1 vial (20 μL)	2 vials (20 µL/vial)
Component B: TdT enzyme	Freeze (< -15 °C), Minimize light exposure	1 vial (5 μL)	2 vials (5 µL/vial)
Component C: CoCl2 solution	Freeze (< -15 °C), Minimize light exposure	1 vial (50 μL)	2 vials (50 µL/vial)
Component D: TdT Reaction Buffer	Freeze (< -15 °C), Minimize light exposure	1 vial (500 μL)	2 vials (500 μL/vial)

### OVERVIEW

ReadiLink<sup>™</sup> iFluor<sup>™</sup> 555 Oligo and ssDNA Labelling Kit enables simple and uniform tagging of single-stranded DNA or oligos with iFluor<sup>™</sup> 555, our bright, photostable and green-fluorescent fluorophore. The labelling kit uses our proprietary TAQuest<sup>™</sup> terminal deoxynucleotidyl transferase (TdT) to catalyze non-template directed nucleotide incorporation onto the 3'- end of single-stranded DNAs or oligos. The kit is optimized for efficient labelling and contains all the essential reagents required for efficient labelling of ssDNA or oligos. The resulting iFluor<sup>™</sup> 555-labelled DNA probes are ideally suited for biological applications, e.g., electrophoretic mobility shift assays (EMSA), Northern and Southern blots, colony or in situ hybridizations.

#### AT A GLANCE

#### Protocol summary

- 1. Prepare oligo or ssDNA samples
- 2. Add reagents to tube
- 3. Mix and centrifuge briefly
- 4. Incubate at 37 °C for 60 minutes
- 5. Place on ice for 5 minutes
- 6. Purify the labeled DNA

**Note:** Thaw all the kit components on ice before starting the experiment. Briefly centrifuge all the reagents to the bottom before starting the labeling process.

#### **KEY PARAMETERS**

#### **Thermal Cycler**

Instrument specification(s)

0.5 mL microcentrifuge or 0.2 mL PCR tube

## SAMPLE EXPERIMENTAL PROTOCOL

The following protocol can be used as a guideline.

- To a clean (Nuclease-free) 0.5 mL microcentrifuge tube or 0.2 mL PCR tube, prepare a reaction mix by adding the reagents in the order indicated in Table 1.
- 2. Carefully mix the reagents by a brief vortex, followed by a brief centrifuge.
- 3. Incubate the reaction at 37 °C for 60 minutes.
- 4. After incubation, place the reaction on ice for 5 minutes.
- 5. Purify the labeled DNA.

Table 1. Reagents composition per tube for each reaction

Components	Amount
Oligo or ssDNA sample	1 µg DNA diluted in Nuclease-free water to a
	final volume of 5 µL
TdT Peaction Buffer	40 ul

iFluor™ 555-dUTP	1-2 µL	
CoCl <sub>2</sub>	5 µL	
TdT enzyme	0.5 µL	
Total Volume	52 μ L (Approx.)	

Note: The amount of iFluor<sup>™</sup> 555-dUTP can be optimized to achieve the best labeling conditions.

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