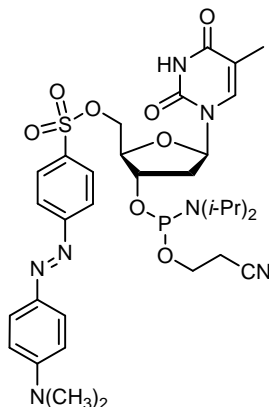


5'-O-Dabsyl-T CEP (BA 0283) Product Information



For Quenched Autoligation (QUAL) Probes

Improved methods for the detection of nucleic acids continues to be an active area of investigation. Non-enzymatic approaches involving fluorescence changes are attractive alternatives to enzyme-based methods such as PCR. Autoligation probes involve two short oligonucleotides, each of which bears a reactive functional group on one end. The two probes are designed such that they hybridize to the appropriate target sequence in an end-to-end fashion to place the two reactive functional groups in close proximity, thus promoting the formation of a covalent bond. Recent work by Kool and co-workers describes an imaginative autoligation strategy that results in the appearance of a fluorescence signal upon template-promoted ligation (Figure 1).¹ Two probes are used, one bearing a 3'-phosphorothioate (Probe 1) and the other a 5'-dabsylate and an internal fluorophore (Probe 2). The fluorophore of Probe 2 is quenched by the nearby dabsyl quencher and is thus dark. The two probes bear additional nucleotides and may bind to the correct sequence (if present) to place the sulfur nucleophile close to the 5'-O-dabsylate. A substitution reaction then occurs, displacing the dabsylate quencher and thus unquenching the fluorophore, resulting in a fluorescence signal. These "quenched autoligation probes" (QUAL probes) are more sensitive to single-nucleotide differences than most hybridization-based approaches. Further, the fluorescence change is permanent and is not subject to buffer or temperature. We now offer 5'-O-Dabsyl-T CEP (BA 0283) for the synthesis of 5'-O-dabsylate QUAL probes.

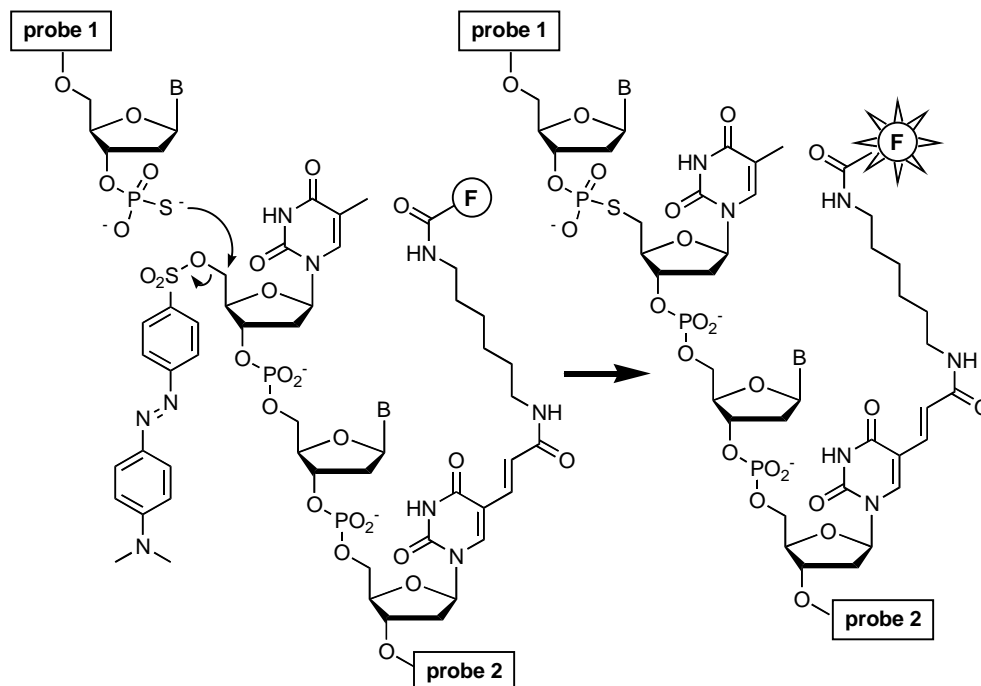


Figure 1. Quenched autoligation probes form a covalent bond in the presence of an appropriate target sequence, displacing dabsylate and thus unquenching Probe 2, resulting in a fluorescence signal. Probe 2 is made with 5'-*O*-Dabsyl-T CEP (BA 0283).

Coupling: Use standard coupling protocols to achieve >95% coupling efficiency. Extended coupling is not required, though it is not detrimental. For other monomers, use UltraMILD phosphoramidites (Pac-protected dA, *i*-PrPac-protected dG, and acetyl-protected dC). See the Supporting Information of reference 1a for details on the synthesis of dabsyl- and fluorescein-labeled oligonucleotides.

Displacement, cleavage, and nucleobase deprotection: Use 50 mM potassium carbonate in methanol for 12 h. See the Supporting Information of reference 1a for details on purification and characterization.

References:

- (1) (a) Sando, S.; Kool, E. T. *J. Am. Chem. Soc.* **2002**, *124*, 2096-2097. (b) Review: Silverman, A. P.; Kool, E. T. *Trends in Biochem.* **2005**, *23*, 225-230. (c) Review: Silverman, A. P.; Kool, E. T. *Chem. Rev.* **2006**, *106*, 3775-3789.