

A Superior New Disulfide Support for Oligonucleotide Synthesis

**Joseph T. Repine, Dennis J. McNamara,
Brian D. Gibson, Nancy S. Barta,
David A. Berry, and John C. Hodges***

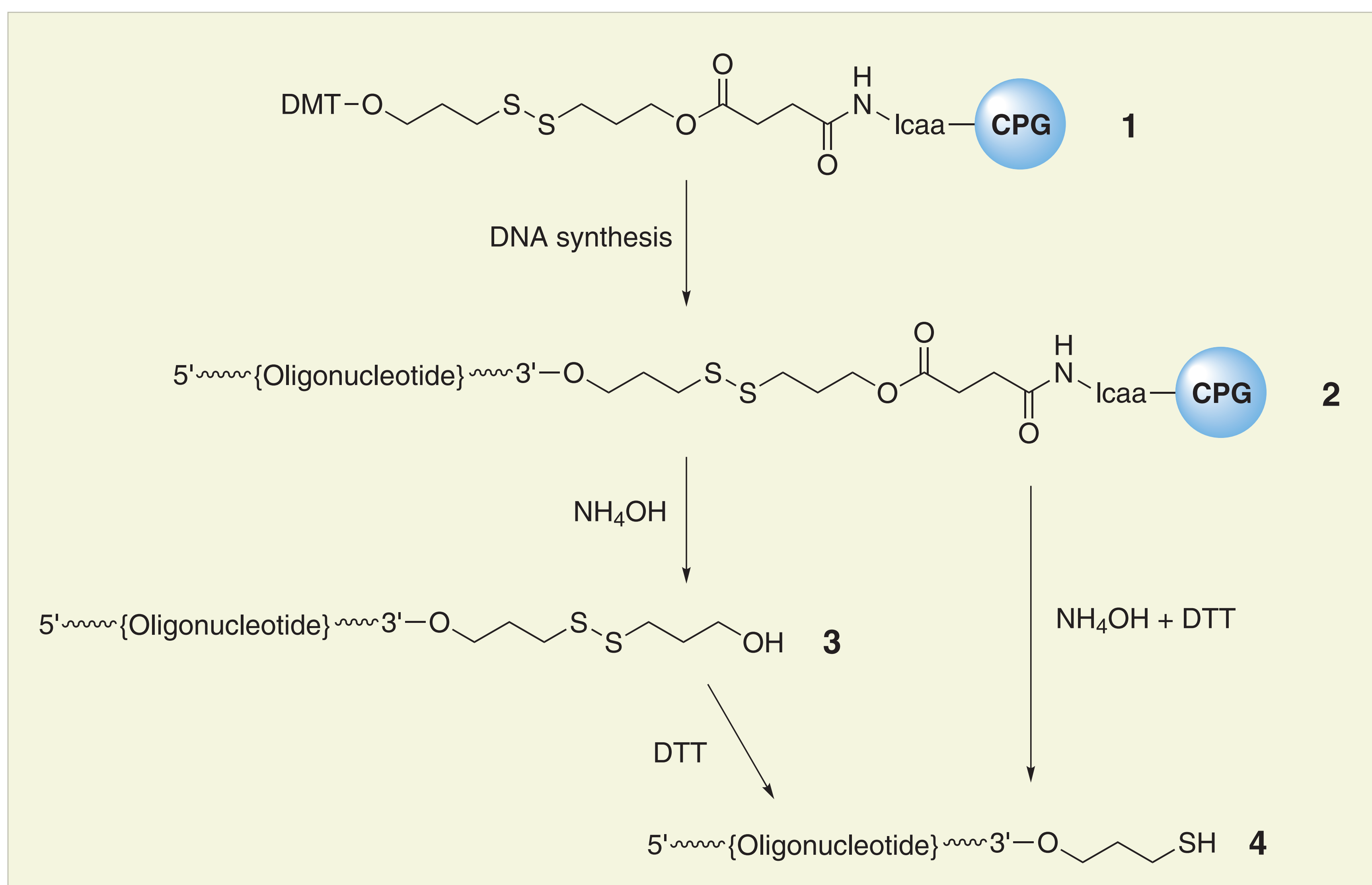
**Berry & Associates, Inc.
2434 Bishop Circle East
Dexter, MI 48130, USA**

**E-MAIL: jhodges@berryassoc.com*

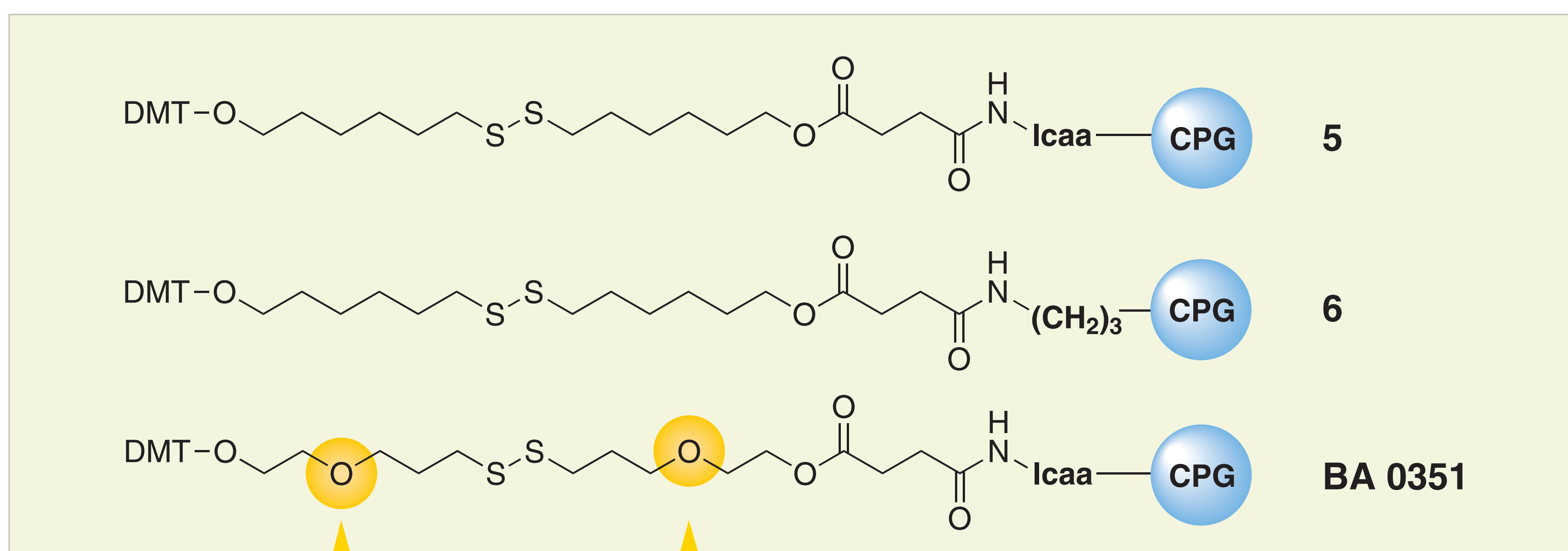
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Disulfide Supports for DNA & RNA Synthesis

In synthesizing oligonucleotides, it can be useful to include a thiol or a disulfide as a modification. These sulfur containing groups enable attachment of the oligo to surfaces or to other molecules of interest. When the modification is to occur at the 3'-terminus, one common strategy is to link one end of a symmetrical alkyl-disulfide-diol to the solid support (**1**). The oligonucleotide chain is then synthesized on the other end of the disulfide (**2**). By choosing the appropriate cleavage conditions, either a disulfide (**3**) or a free thiol (**4**) can be provided at the oligo's 3'-terminus.

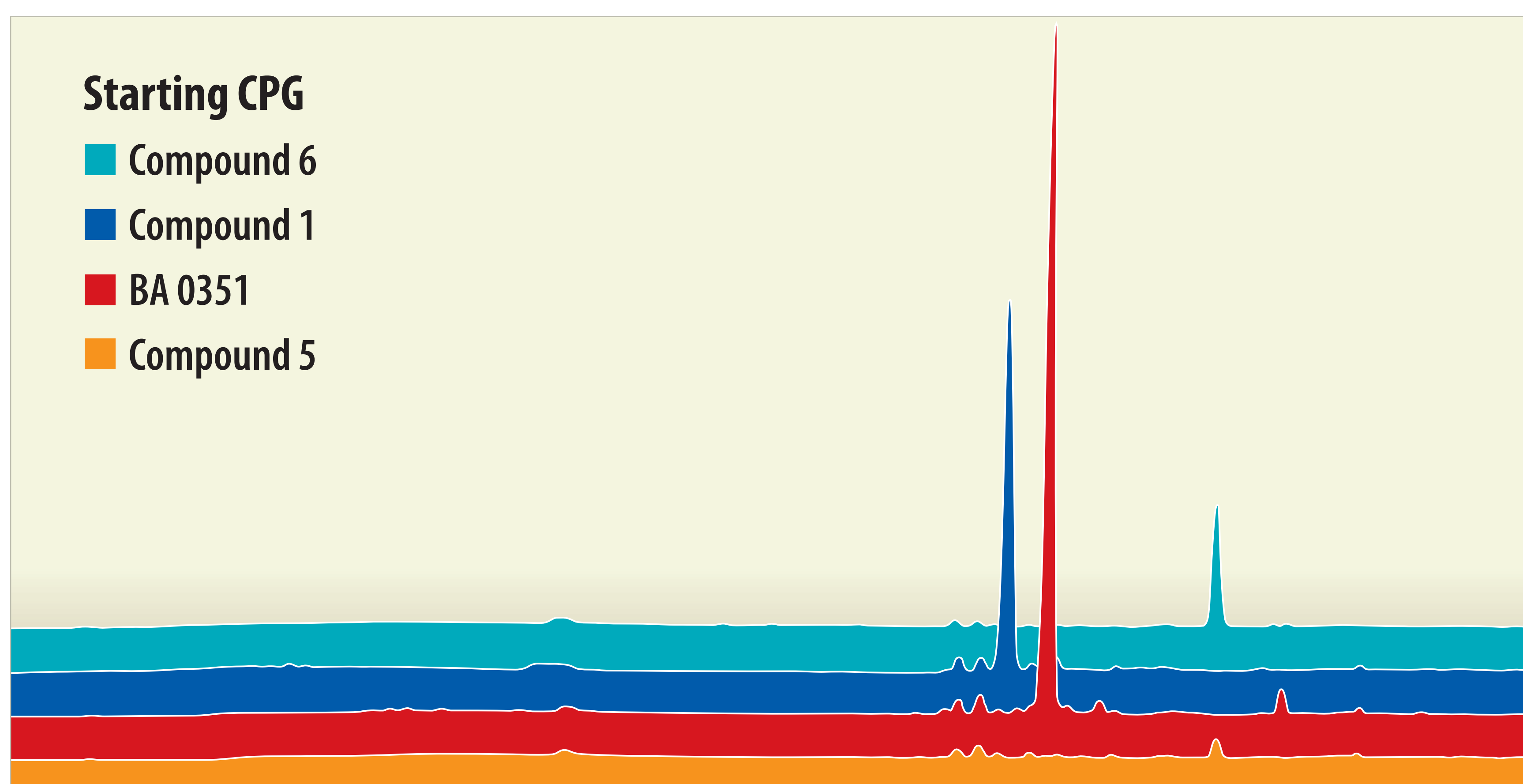


For many years, C3-disulfide-Icaa-CPG (**1**) has been the principal 3'-thiol-modifier CPG for oligonucleotide syntheses. The short alkyl chain of this product poses limitations for certain applications, but lengthening the alkyl chain from three to six carbon atoms gives a support that is dysfunctional (**5**). Switching the support to aminopropyl-CPG (**6**) gives somewhat better results. However, **6** still does not measure up to the performance of **1**.



We have discovered that including ether functionality near the middle of the C6-disulfide chains markedly enhances DNA synthesis. Compared to **5** and **6**, **BA 0351** affords vastly superior oligo yield and maximum synthesis length, while maintaining the same disulfide chain length (see data on right).

Comparative HPLC of Crude T6-Oligos from Disulfide-CPGs



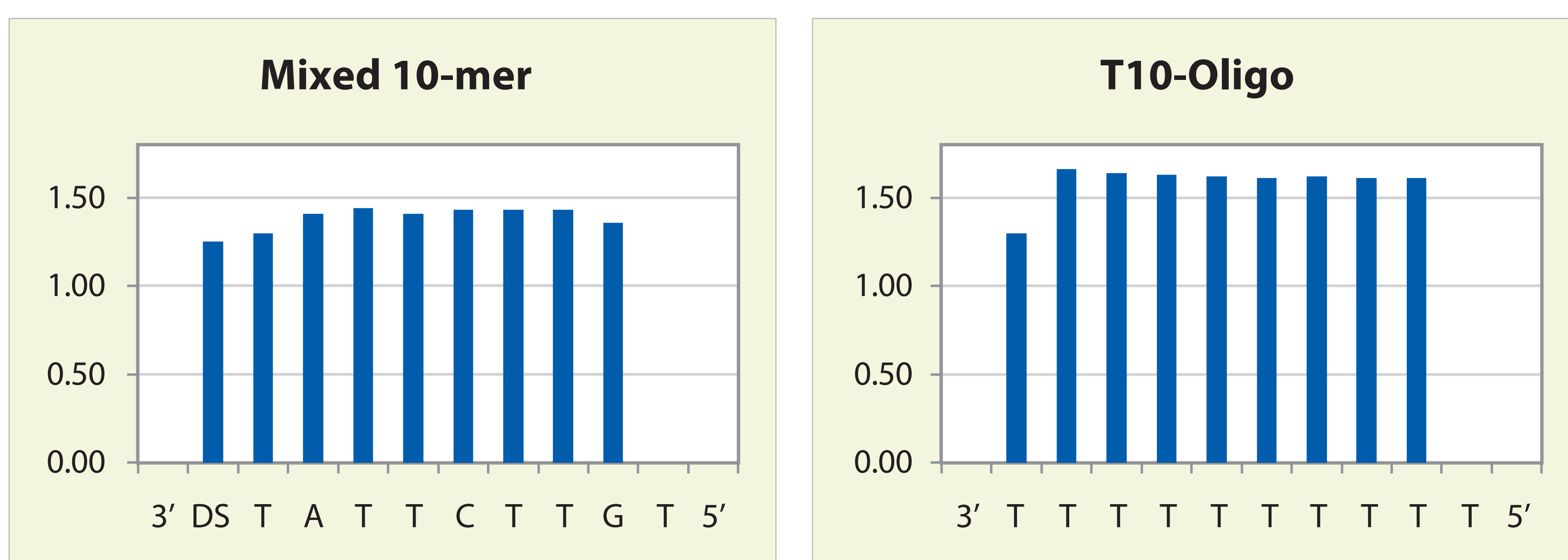
Shown above are HPLC chromatograms of crude 5'-DMT-T6-(3'-disulfide) products from 200 nM synthesis columns at equivalent dilution.

Synthesis Performance of Disulfide CPGs

Compound Number	HPLC Color	X	Support ^a	Maximum Length ^b	Relative Yield ^c
6	Teal	(CH ₂) ₆	aminopropyl-CPG	48	0.31
1	Blue	(CH ₂) ₃	Icaa-CPG	100	1.00
BA 0351	Red	(CH ₂) ₃ O(CH ₂) ₂	Icaa-CPG	104	1.68
5	Orange	(CH ₂) ₆	Icaa-CPG	9	0.05

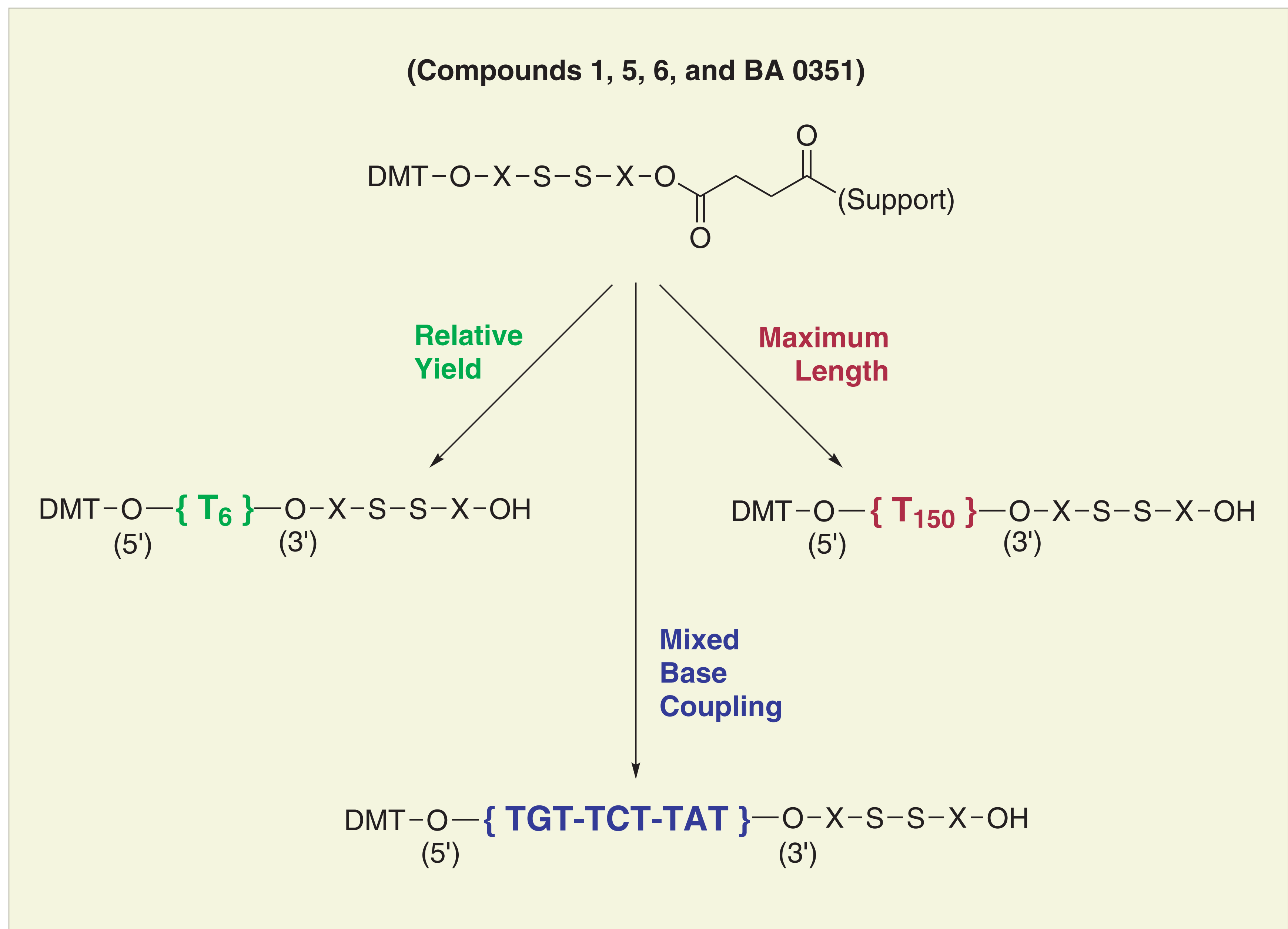
(a) All CPGs have 1000 Angstrom pores. (b) Length at which the DMT histogram drops below 70% of initial maximum. (c) Relative HPLC integration (AUC) for each 5'-DMT-T6-(3'-disulfide) peak at equivalent dilution.

Mixed Base Coupling Test



Shown above are raw integrated values for DMT-cation absorbance (498 nm) as measured by the synthesizer during deprotection. The histogram from a mixed-base oligo synthesized on **BA 0351** is comparable to that from a control T10 on Icaa-CPG, qualitatively indicating equal synthesis efficiency.

Oligo Synthesis Performance Tests for Disulfide-CPGs



Synthetic Route to BA 0351

