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BIOTIN-LABELED SYNTHETIC OLIGODEOXYRIBONUCLEOTIDES: CHEMICAL SYNTHESIS AND USES AS HYBRIDIZATION PROBES.

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Several biotinylated synthetic oligodeoxyribonucleotides have been selectively prepared by a simple and efficient chemical method. This procedure allows the specific and covalent coupling of one biotin moiety to any 5'-kinased unprotected oligodeoxyribonucleotide through an aminoalkylphosphoramide linker arm¹. The reactions are performed in aqueous solutions on unprotected oligonucleotides and proceed cleanly with good yields. This method is insensitive of the length of the polynucleotide, of the nucleotide sequence and of the nature of the 5'-terminal nucleotide.

The amount of biotin and the site of its attachment to DNA are strictly controlled. There is no noticeable disturbance of the hybridization pattern (strength and selectivity) of the biotin-labeled probe comparing to non-modified DNA. Biotin-labeled oligomers (20-mer and 23-mer) complementary to a plasmid of known base sequence have been prepared and purified to homogeneity either by affinity chromatography on avidin-agarose or by denaturing polyacrylamide gel electrophoresis. These probes were hybridized to dot and Southern blots of target plasmid DNA immobilized on nitrocellulose filters. Detection of as little as 2-6 fmol DNA was done by incubating the filters with streptavidin and biotinylated polymers of alkaline phosphatase and visualizing the complex formed.

Biotinylated oligonucleotides should prove to be very useful as stable and non-radioactive probes to identify and locate specific DNA or RNA sequences.

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