



In Situ DNA Hybridization Products

The detection of DNA sequences of infectious viral agents by complementary hybridization, using biotin-labeled DNA fragments, is a well established technology. Avidin, streptavidin or anti-biotin antibodies are generally used to introduce the tracing enzyme or detection system. The use of synthetic oligonucleotides instead of the viral DNA fragments, provides several advantages: the hybridization is more discrete and targeted to a specific domain or to repeated sequences within the global viral DNA. This offers the ability to predict the possible cross-reaction of the selected oligonucleotide with other related viruses, and diminishes the possibility of incomplete or partial hybridizations, which larger polynucleotides will be more likely to promote. In addition, the conditions or requirements to achieve proper hybridization are simple to study, establish and perform.

In situ DNA hybridization using chromogenic substrates or their binary combinations, was first reported for research applications by Ward and coworkers, 1981, and for clinical use by Brigati et al., 1983. This approach facilitates the visualization of the results by light microscopy, and simultaneously permits the determination of the morphological correlations within the tissue. This technique has evolved to become the main course in the in situ detection of viral genomic information. The adequacy or validity of the test strongly depends on the quality of the specimens. False negative results are a major concern in such applications. In order to resolve the question of the adequacy of the specimen for the test, Brigati and coworkers have advanced the use of a human genome oligonucleotide coded Alu 2, which is found repeated over 300,000 times in the human genomic structure. The biotin labeled Alu 2, as proposed by these authors, will facilitate the determination of the fitness of a specimen for in situ DNA hybridization. Damaged tissue samples producing negative results with this probe will be poor candidates for such a procedure.

Biomed now offers two biotin-labeled DNA probes under our tradename Olig-Ø-Probes. These probes are labeled with multiple biotin at a distal site of the functional oligonucleotide. The attachment site of the biotin residues away from the annealing bases which provides increased access and more efficient recognition of these residues by the tracing reagent.

References:

1. Langer, P. R. , Waldrop, A. A. , Ward, D. , Proc. Nat'l. Acad. Sci. (USA) 18:6633-6637, 1981.
2. Brigati, D. J. , Myerson, D. , Leary, J. J. , Spaholz, B. , Ward, D. C. Virology 126:32-50, 1983.
3. Montone, K. T. , Brigati, D. J. , Budgeon, L. R. Yale Journ. Biol. and Med. 62:141-158, 1989.

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