HETEROBIFUNCTIONAL LINKER BETWEEN ANTIBODIES AND REPORTER GENES FOR IMMUNOASSAY DEVELOPMENT

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The amplification inherent in transcription and translation of DNA has already been exploited for the development of highly sensitive immunoassays by using a reporter gene as a label that, upon \textit{in vitro} expression, generates multiple enzyme molecules in solution (expression immunoassay). The most challenging task in the development of an expression immunoassay is to link the antibody to a reporter gene that also contains control elements for transcription/translation. In this work, we prepare heterobifunctional linkers that consist of a modified avidin or streptavidin covalently attached to an oligonucleotide (dA)\(_{40}\). (Strept)avidin interacts with a biotinylated detection antibody whereas the oligonucleotide hybridizes with a complementary poly(dT) tail added enzymically to the 3' end of the reporter gene. The linker is evaluated in a model two-site (sandwich-type) immunoassay performed in microtiter wells. A 4.3-kilobase plasmid containing the firefly luciferase cDNA is used as a reporter.