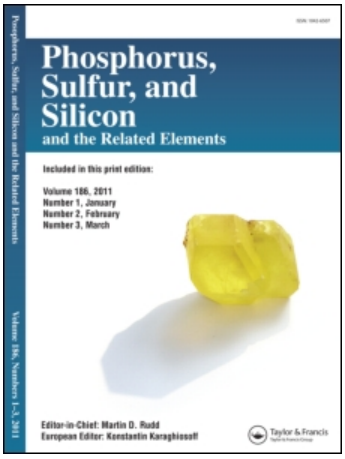


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# Simple Methods for Preparing Oligonucleotides Containing 1) Thiol Groups at their 5'-Termini, 2) 5'-Phosphates

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# Simple Methods for Preparing Oligonucleotides Containing 1) Thiol Groups at their 5′-Termini, 2) 5′-Phosphates

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A simple method has been developed for synthesising oligonucleotides containing a thiol group at their 5'-termini. The sequence required is prepared using standard solid phase phosphoramidite methods and an extra round of synthesis is then performed with S-triphenylmethyl O-methoxymorpholinophosphinyl 3-mercaptopropan (1) ol. After normal deblocking this gives an oligonucleotide containing a tritylthiol group attached to the 5'-phosphate of an oligonucleotide via a 3-carbon spacer arm. The trityl group can be removed with $AgNO_3$ at pH 5 to give the free thiol. This compound is stable at pH 8 and reacts cleanly and rapidly with sulphydryl specific probes (eg fluorescent iodoacetates) at this pH value. This method can be used to prepare a wide variety of usefully labelled oligonucleotides and it is envisaged that fluorescent oligonucleotides will be useful in the study of protein nucleic acid interactions and to replace $^{32}P$ labelled hybridisation probes.

The reagent S-triphenylmethyl O-methoxymorpholinophosphinyl 2-mercaptoethanol can be similarly used to prepare oligonucleotides containing a tritylthiol group attached to the 5'-phosphate via a 2-carbon spacer arm. Removal of the trityl group with $AgNO_3$ at pH 8 ($t_{\frac{1}{2}} \simeq 20$ min) gives the 5'-phosphate. This provides a simple method for preparing oligonucleotides containing a 5'-phosphate which are of use in total gene synthesis