A Publication
of Reliable Methods
for the Preparation
of Organic Compounds

Working with Hazardous Chemicals

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September 2014: The paragraphs above replace the section "Handling and Disposal of Hazardous Chemicals" in the originally published version of this article. The statements above do not supersede any specific hazard caution notes and safety instructions included in the procedure.

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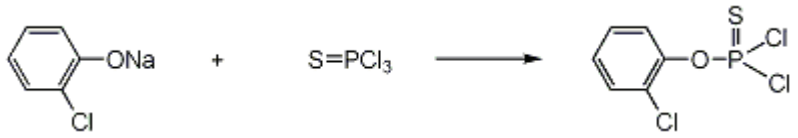
2-CHLOROPHENYL PHOSPHORODICHLORIDOTHIOATE

[Phosphorodichloroidothioic acid, O-(2-chlorophenyl)ester]



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Checked by Adam R. Renslo and Rick L. Danheiser.

1. Procedure

A 2-L, three-necked, round-bottomed flask equipped with a 250-mL addition funnel, glass stoppers, and a magnetic stirring bar is charged with thiophosphoryl chloride (271 g, 1.60 mol) (Note 1), tetrabutylammonium bromide (3.2 g; 0.01 mol) (Note 1), and 400 mL of dichloromethane (Note 2), and the resulting solution is cooled in an ice bath at 0-5°C. 2-Chlorophenol (51.4 g, 0.400 mol) (Note 1) is added to a magnetically stirred solution of sodium hydroxide (25%, 350 mL, (Note 1)) in a 500-mL Erlenmeyer flask cooled in an ice bath at 0-5°C, and the resulting solution is transferred to the addition funnel and added slowly over a period of 30 min to the reaction mixture. The resulting two-phase mixture is stirred for 8 hr while the bath is allowed to warm to room temperature, and the aqueous layer is then separated and extracted with 100 mL of dichloromethane . The combined organic layers are washed with 100 mL of brine , dried over magnesium sulfate , and filtered. The dichloromethane is removed by rotary evaporation, and the residue is distilled under reduced pressure through a short-path still head (Note 3) to give 93.2-98.4 g (89-94%) of 2-chlorophenyl phosphorodichloridothioate as a colorless oil (Note 4).

2. Notes

1. Reagent grade 2-chlorophenol, sodium hydroxide, thiophosphoryl chloride, and tetrabutylammonium bromide were purchased from Aldrich Chemical Company, Inc. , and used without purification.
2. HPLC-grade dichloromethane was purchased from Mallinckrodt Inc. and used without further purification.
3. Two traps cooled in liquid nitrogen are connected between the distillation apparatus and the vacuum pump to trap excess thiophosphoryl chloride.
4. 2-Chlorophenyl phosphorodichloridothioate has the following properties: bp 90-93°C (0.2 mm); IR (neat) cm^{-1}: 3160, 1580, 1475, 1450, 1260, 1210, 1060, 1040, 940, 780, 760, 720 ; ^{1}H NMR ($CDCl_3$, 300 MHz) δ: 7.22-7.34 (m, 2 H), 7.43-7.51 (m, 2 H) ; ^{13}C NMR ($CDCl_3$, 75 MHz) δ: 122.5, 122.6, 126.7, 126.8, 127.7, 127.8, 127.91, 127.95, 131.09, 131.12, 146.7, 146.9 ; ^{31}P NMR ($CDCl_3$, 202 MHz) δ: 54.4 . Anal. Calcd for $C_6H_4Cl_3OPS$: C, 27.56; H, 1.54. Found: C, 27.57; H, 1.42.

Waste Disposal Information

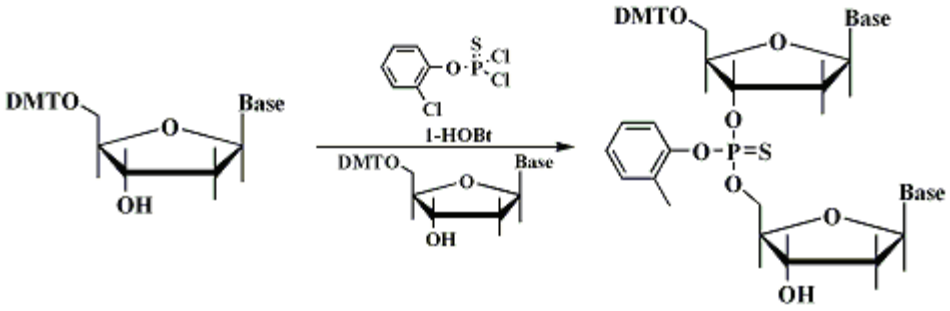
All toxic materials were disposed of in accordance with "Prudent Practices in the Laboratory"; National Academy Press; Washington, DC, 1995.

3. Discussion

Deoxyribonucleotides, deoxyribonucleotide phosphorothioates, modified DNA, and analogs have wide applications in molecular biology, antisense applications, antigene therapy, etc. Three methods are available for the synthesis of oligonucleoside phosphorothioates: phosphoramidite, H-phosphonate, and phosphotriester approaches. Different protecting groups, most of which are base labile, have been used for the phosphoramidite approach. In the H-phosphonate approach, no protecting group is involved. In the phosphotriester approach, aryl groups are used as O-protecting groups, and deprotection occurs via a

nucleophilic attack on the phosphorus center with the aryloxy group being the leaving group. Because of their base labile nature, most of the groups used in the phosphoramidite approach are not suitable for the synthesis of aryl phosphorodichloridothioates, which are used as the starting material in the phospho triester approach. The previously reported[2,3] routes for the synthesis of aryl phosphorodichloridothioates involve drastic conditions such as refluxing or the use of liquid sulfur dioxide, and are not amenable to very large scale synthesis. A much simpler alternative method involving phase transfer reaction is described here.

Phase transfer catalysis[4] is a valuable tool in organic synthesis. The process is exemplified by the convenient synthesis of 2-chlorophenyl phosphorodichloridothioate. Using this phase transfer reaction, a number of dichloridothioates of substituted phenyl, benzyl, thiophenyl, and thiobenzyl alcohols are accessible. The phosphorodichloridothioate reacts with various coupling reagents to form activated species that are useful in the synthesis of oligonucleotide phosphorothioates via the phosphotriester approach as illustrated below.[5,6]



The procedure shown here describes the preparation of a fully protected phosphorothioate triester dimer. The dimer can then be coupled subsequently in a similar way to form elongated oligomers.

References and Notes

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Appendix
Chemical Abstracts Nomenclature (Collective Index Number); (Registry Number)

2-Chlorophenyl phosphorodichloridothioate:
Phosphorodichlorodiothioic acid, O-(2-chlorophenyl)ester (10); (68591-34-4)

Thiophosphoryl chloride: HIGHLY TOXIC (8,9); (3982-91-0)

Tetrabutylammonium bromide (8);
1-Butanaminium, N,N,N-tributyl-, bromide (9); (1643-19-2)

2-Chlorophenol:
Phenol, o-chloro- (8);
Phenol, 2-chloro- (9): (95-57-8)

Sodium hydroxide (8,9); (1310-73-2)