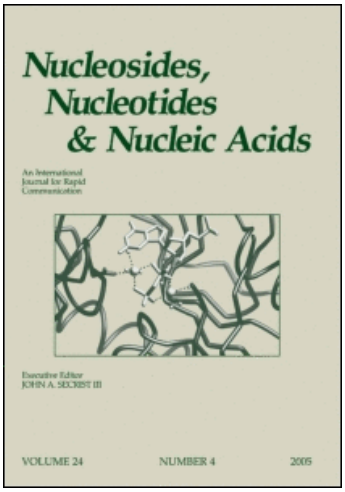

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SYNTHESIS OF CHLORODIDEOXYNUCLEOSIDES USING TRIS-(2,4,6-TRIBROMOPHENOXY)DICHLOROPHOSPHORANE (BDCP)

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ABSTRACT: 5'-Chloro-5'-deoxy-N^3,3'-*O*-dibenzoylthymidine (3a), 5'-chloro-5'-deoxy-N^4,3'-*O*-dibenzoyldeoxycytidine(3b), 5'-chloro-5'-deoxy-N^6,3'-*O*-dibenzoyldeoxyadenosine(3c), N^3-benzoyl-1-(3-chloro-2,3-dideoxy-5-*O*-trityl-ß-D-xylofuranosyl)thymine (5a) and N^6-benzoyl-9-(3-chloro-2,3-dideoxy-5-*O*-trityl-ß-D-xylofuranosyl)adenine (5b) have been synthesized in very high yields using a new efficient reagent, tris(2,4,6-tribromophenoxy)dichlorophosphorane (BDCP). The reaction time was greatly reduced to 5-8 min. NOE data suggested an inversion of configuration at $C_{3'}$-position and thus an SN2 mechanism has been proposed for the chlorination reaction.

INTRODUCTION

Halodeoxynucleosides, because of their paramount importance as potential synthetic intermediates, have been extensively exploited for their use in the synthesis of cyclonucleosides[1], nucleotides[2], sulfur analogoues[3], phosphonic acid derivatives[3], deoxynucleosides[4], unsaturated and amino sugars[5,6], and recently in many cases, they proved to be of primary biological importance[7,8,9] and as anti-viral agents[10]. Traditionally the preparation of halodeoxynucleosides has been achieved by introduction of p-toluenesulfonyl or methanesulfonyl onto the respective hydroxyl position of suitably protected nucleosides and subsequent replacement of sulfonyl group by heating in a solvent like acetone with metallic halides[2,11,12], the latter being better since readily replaced. In

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another approach, halogenation has been effected by ring opening reaction of O^2,2'-cyclonucleosides with hydrogen halides[13] at higher temperatures.

A major breakthrough was introduced in this field by Rydon *et al* in 1953 using quasi-phosphonium halides[14-20], viz. methyltriphenoxyphosphonium iodide and iodotri-phenoxyphosphonium iodide in benzene. The Vilsmeier reagent, i.e., phosphoryl halide in DMF[21] also effected chlorination and bromination by heating the reaction mixture at 60 °C for 2 h. A slight modification, i.e., Vilsmeier-Haack reagent, viz. thionyl halide in DMF also did not work very well[22,23].

The halodeoxynucleosides chemistry entered a new phase with the advent of carbon tetrahalides and triphenylphosphine in DMF[24-26]. Several other modifications[27,28,29] have also been used from time to time for synthesizing halodeoxynucleosides. All the reagents used for halogenation so far either required heating or prolong time for the completion of the reactions or gave a plethora of side products.

These reports for the halogenation of nucleoside sugar moieties prompted us to develop a suitable mild halogenating agent, and in this pursuance, we have introduced here a new, efficient and versatile reagent, viz. tris(2,4,6-tribromophenoxy)dichlorophosphorane (BDCP), FIG.1, for rapid chlorination of deoxynucleosides with much better yields under milder conditions than reported earlier.. The preparation and operational handling of this reagent are very easy except that absolutely anhydrous conditions should be observed during the entire course of the reactions in order to get good results. A preliminary report of this work has already been published[30].

RESULTS AND DISCUSSION

The reaction of 5'-hydroxyl group of suitably protected purine and pyrimidine deoxyribonucleosides with BDCP proceeded frequently very rapidally and gave the corresponding 5'-chloro-5'-deoxyribonucleosides in high yield, FIG. 2. The reaction was complete within 5 min at room temperature using 1.8 equiv of BDCP, unlike other chlorinating reagents where a long time is required. Thus the compound **3a** was obtained in 92% yield as a crystalline solid. The confirmation of the product was done by NMR data where 5'-protons appeared as a double doublet (J= 12.2 and 21.2 Hz) unlike a multiplet in the case of starting material. This was, most probably, attributed to the electronegative character of the chlorine atom. A possibility of formation of O^2, 5'- cyclothymidine was completely ruled out by N^3- benzoylaiton. The compound **3b** showed a hypsochromic shift in the UV spectrum as shown in TABLE 1.

NMR spectrum of this compound showed a well separated double doublet for 5'-protons (J=12.05 Hz each) which appeared as a doublet in the starting material. Further separation of peaks of 2' and 2'' protons by 0.2 ppm seemed to be another support for formation of this compound. Similarly, the compound **3c** also showed a hypsochromic shift in its UV spectrum (table 1). In its NMR spectrum, the effect of 5' - chlorination was not so

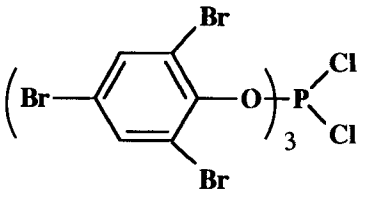
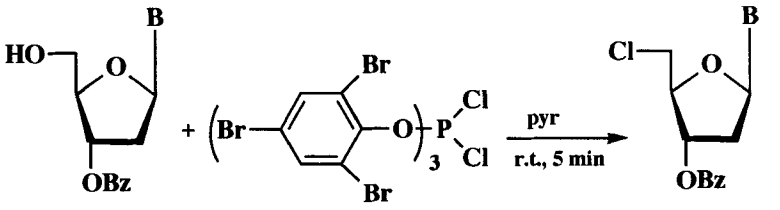


FIGURE 1. Structure of tris(2,4,6-tribromophenoxy)dichlorophosphorane (BDCP)

TABLE 1. UV spectral data of 5'- chlorinated nucleosides and their respective substrates

Compound	UV (MeOH) nm	
	λ max	λ min
2b	302	286
3b	301	285
2c	278	250
3c	277	248



2a. B= N^3- bz dT
2b. B= N^4- bz dC
2c. B= N^6- bz dA

3a. B= N^3- bz dT
3b. B= N^4- bz dC
3c. B= N^6 - bz dA

FIGURE 2. 5'-Chlorination of deoxynucleosides

pronounced as in the case of pyrimidine nucleosides but the 4'-protons appeared as a doublet (J=1.98 Hz), unlike a singlet in the case of starting material. The structures of all these compounds were further confirmed by elemental analyses.

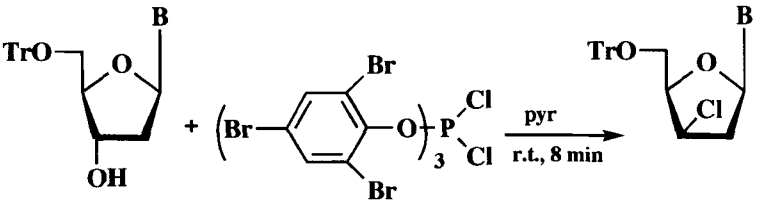
The 3'-halogenation of nucleosides has been rather complicated compared to 5'-halogenation because of 3'- hydroxyl being a secondary hydroxyl group, it is less reactive and less accessible to the halogenating reagents. The best method reported previously[20] required treatment of suitably protected nucleosides with the Rydon reagent, methyltriphenoxyphosphonium iodide in DMF for 10-15 h and resulted in many side products. Another problem with the use of this reagent was that during the course of the reaction, hydrogen iodide was released and it resulted in the partial loss of trityl group. In our case, the time limit was drastically reduced without any loss of trityl group. When pyridine was used as a solvent, hydrogen chloride formed during the course of the reaction was trapped as pyridinium hydrochloride and thus there was no trityl loss detected.The corresponding 3'-chloro-3'-deoxyribonucleosides were obtained in high yield using two equiv of BDCP at room temperature in just 8 min, FIG. 3. The proton NMR spectrum of the compound **5a** showed an appreciable change in the chemical shift of C_2, protons which are separated by 0.51 ppm. It indicated a threo configuration for this compound whereas in erythro configuration, the C_2, protons frequently occur as overlapping signals. Furthermore, the C_1, protons which occur as triplet in erythro configuration has been observed as a quartet in this case. All these observations are well in harmony with previous reports[20]. Similarly, the compound **5b** also showed a quartet unlike a triplet in erythro configuration and a down field shift in C_2, protons in its NMR spectrum. Also, the UV spectrum of this compound showed a hypsochromic shift of 2 nm in λ max and 1 nm in λ min. These compounds were again confirmed by their elemental analyses. Based on extensive NOE studies, FIG. 4, an SN2 mechanism has been proposed for the chlorination reactions.

CONCLUSIONS

From the foregoing results and discussion, it is crystal clear that the use of BDCP has many advantages over the other reagents used so far, e.g., greatly reduced reaction time with higher yields and the reactions can be carried out at ambient temperature, and thus the problem of racemization at higher temperature has been completely eliminated. Further, the problem of trityl loss during 3'-chlorination has also been solved by the use of a basic solvent like pyridine and a neat and clean reaction without formation of any side products or reactive species has an obvious advantage during purification of the products.

EXPERIMENTAL

General. All the nucleosides were purchased from Yoshitomi Pharmaceutical Co. Ltd while all other reagents were purchased from Tokyo Chemical Co. Ltd. Toluene was dried by refluxing over CaH_2 overnight, distilled and stored over molecular seive, 4 Å. Pyridine



1a. B= N^3-bz dT
1b. B= N^6-bz dA

2a. B= N^3-bz dT
2b. B= N^6-bz dA

FIGURE 3. 3'-chlorination of deoxynucleosides

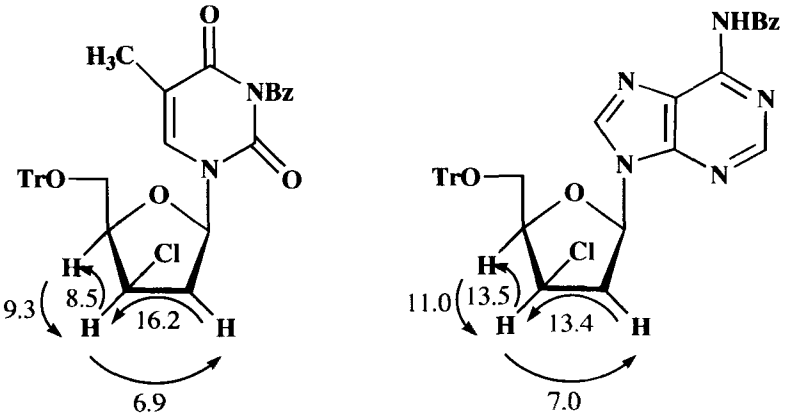


FIGURE 4. Intramolecular NOE correlations: The arrows start from the atom irradiated and the numbers below show the NOE(%) effects

was purified by refluxing over p-toluenesulfonyl chloride for 5h and dried by refluxing over CaH_2 overnight, distilled and stored. Silica gel 60 F-254 tlc plates (Merck Co. Ltd) were used and exposed to UV light and charred with 5% H_2SO_4 in methanol for differentiation and identification of spots. Hitachi 220A UV spectrophotometer was used for recording UV spectra. ^1H and ^{31}P NMR were recorded on JEOL JNM-270 EX spectrophotometer. Degassed solvents were used for NOE studies and chemical shifts are shown in ppm with respect to tetramethyl silane as standard. The following notations have been used in describing the chemical shifts: s - singlet, d - doublet, dd - double doublet, t - triplet, q - quartet, m - multiplet and br - broad. Elemental analyses were carried out at Research Institute of Resourse, Tokyo Institute of Technology.

The suitably protected deoxynucleosides used in this study were prepared according to the literature procedure[31]. These compounds were fully characterised by their UV and NMR analyses.

Tris(2,4,6-tribromophenoxy)dichlorophosphorane (BDCP). Phosphorus pentachloride (5.2 g, 0.025 mmol), freshly distilled , was allowed to stand at 50 °C under vacuum for 10 min. To this was added recrystallized 2,4,6-tribromophenol (24.81 g, 0.075 mmol) dissolved in dry toluene (150 mL; this solution was kept over molecular seive, 4 Å , for 24 h) with the help of a syringe. This solution was refluxed on an oil bath for 6h with the condenser top bearing an argon balloon. A yellow color solution resulted. The reaction mixture was allowed to cool at room temperature and then the toluene was removed using aspirator putting a dry ice trap in between, while the flask was connected to an argon balloon. At this stage, an yellowish solid resulted. This solid material was dissolved in just sufficient amount of dry toluene. The flask was connected to an argon balloon and the product was allowed to crystallize at ambient temperature.After two days, BDCP appeared in the form of white flakes, which were broken, filtered off and washed well with dry toluene under N_2 atmosphere. It was further dried over P_2O_5 *in vacuo* for 2 days.Yield 19.51 g (72%); m.p. 229 °C; purity 82%; ^{31}P NMR (pyr-*d*6) -64.26 ppm.

5'-Chloro-5'-deoxy-N^3,3'-*O*-dibenzoylthymidine (3a). To a solution of BDCP (165 mg; 0.36 mmol) in anhydrous pyridine (10 mL) was added a solution of N^3,3'-*O*-dibenzoylthymidine (90 mg; 0.2 mmol) in pyridine (5 mL) with the help of a syringe. Completely anhydrous conditions were observed during the preparation of the solutions and their addition. The resulting mixture was stirred at room temperature for 5 min under argon atmosphere when the reaction was found to be complete as suggested by tlc. Reaction was quenched by addition of water (0.5 mL) and the resulting reaction mixture was evaporated to dryness *in vacuo*. Pyridine was completely removed by co-evaporation with toluene. This dry mass was subjected to silica gel (15 g) chromatography and the product was eluted in 80% DCM in hexane to 0.3% methanol in DCM. The desired fractions were pooled and evaporated to dryness when the product was obtained as white crystalline solid. Yield 87 mg (92%); R_f 0.22 (DCH/Hex, 10:1, v/v); ^1H NMR ($CDCl_3$): δ =1.99 (s, 3H, 5-CH_3), 2.35-2.46, 2.58-2.65 (m, 2H, 2'-H), 4.00 (dd, J = 12.2 and 21.1 Hz, 2H, 5'-H), 4.43-4.44 (m, 1H, 4'-H), 5.51-5.49 (m, 1H, 3'-H), 6.41-6.47 (m, 1H, 1'-H), 7.26-8.01 (m, 11H, Ar-H and 6-H of Th). Anal: calcd for $C_{24}H_{21}ClN_2O_6$: C, 61.46; H, 4.54; Cl, 7.56; N, 5.97. Found; C, 61.29; H, 4.98; Cl, 7.01; N, 5.27.

5'-Chloro-5'-deoxy-N^4,3'-*O*-dibenzoyldeoxycytidine (3b). A solution of N^4,3'-*O*-dibenzoyldeoxycytidine (172 mg, 0.4 mmol) in dry pyridine (5 mL) was added to a solution of BDCP (326 mg, 0.7 mmol) with the help of a syringe and the reaction mixture was stirred at room temperature for 5 min under argon atmosphere. After usual work-up and silica gel column chromatography using 1% methanol in DCM, the product was obtained as white shining glass. Yield 154 mg (86%); R_f 0.5 (DCM/Hex, 19:1, v/v); UV (MeOH) λ max 301 nm, λ min 285 nm; ^1H NMR ($CDCl_3$): δ = 2.25-2.35, 2.97-3.04 (m, 2H, 2'-H), 3.95-4.01, 4.07-4.12 (dd, J = 12.00 Hz, 2H, 5'-H), 4.5 (d, J = 2.31 Hz, 1H, 4'-H), 5.5 (d, J = 6.9 Hz, 1H, 3'-H), 6.48 (q, 1H, 1'-H), 7.89 (d, J = 7.3 Hz, 1H, C5H), 8.28 (d, J = 7.9 Hz, 1H, C_6H), 7.45-7.65 (m,10H, Ar-H). Anal:. Calcd

for $C_{23}H_{20}ClN_3O_5$: C, 60.8; H, 4.4; Cl, 7.71; N, 9.24. Found; C, 60.86; H, 4.37; Cl, 7.81; N, 8.96.

5'-Chloro-5'-deoxy-N^6,3'-*O*-dibenzoyldeoxyadenosine (3c). To a solution of BDCP (165 mg; 0.36 mmol) in anhydrous pyridine (10 mL) was added a solution of N^6,3'-*O*-dibenzoyldeoxyadenosine (91.8 mg, 0.2 mmol) in dry pyridine (5 mL) and the mixture was stirred for 5 min at ambient temperature. After usual work-up and silica gel column chromatography using 0.6% methanol in DCM, the product was obtained as white shining foam. Yield 86 mg (90%); R_f 0.77 (DCM/Hex, 20:1, v/v); UV (MeOH) λ max 277 nm, λ min 248 nm; ^{1}H NMR ($CDCl_3$): δ =2.63-2.70, 3.27-3.38 (m, 2H, 2'-H), 4.02-4.05 (m, 2H, 5'-H), 4.47 (s, 1H, 4'-H), 5.83-5.98 (m, 1H, 3'-H), 6.45-6.51 (m, 1H, 1'-H), 7.47-8.16 (m, 10H, Ar-H), 8.81 (s, 1H, 2-H of Ad), 9.12 (s, 1H, 8-H of Ad). Anal : Calcd for $C_{24}H_{20}ClN_5O_4$: C, 60.31; H, 4.22; Cl, 7.42; N, 13.66. Found; C, 59.9; H, 4.47; Cl, 7.44; N, 13.71.

N^3-Benzoyl-1-(3-chloro-2,3-dideoxy-5-*O*-trityl-ß-D-xylofuranosyl)-thymine (5a). To a stirred solution of BDCP (550 mg, 1.2 mmol) in anhydrous pyridine (15 mL) was added a solution of 5'-*O*-trityl-N^3-benzoylthymidine (353 mg, 0.6 mmol) in dry pyridine (7 mL) and the stirring continued at room temperature for 8 min. After usual work-up and silica gel column chromatography using 70-80% DCM in hexane, the product was obtained as white solid. Yield 306 mg (85%); R_f 0.36 (DCM/Hex, 10:1, v/v); ^{1}H NMR ($CDCl_3$): δ = 1.91-1.92 (m, 3H, 5-CH_3), 2.39-2.45, 2.86-2.97 (m, 2H, 2'-H), 3.42-3.48, 3.68-3.75 (m, 2H, 5'-H), 4.29-4.34 (m, 1H, 4'-H), 4.46-4.49 (m, 1H, 3'-H), 5.96-6.42 (m, 1H, 1'-H), 7.26-8.00 (m, 21H, Ar-H and 6-H of Th). Anal : Calcd for $C_{36}H_{22}ClN_2O_5$: C, 71.15; H, 5.1; Cl, 5.83; N, 4.61. Found; C, 71.2; H, 5.33; Cl, 5.69; N, 5.01.

N^6-Benzoyl-9-(3-chloro-2,3-dideoxy-5-*O*-trityl-ß-D-xylofuranosyl)adenine (5b). To a stirred solution of BDCP (367 mg, 0.8 mmol) in anhydrous pyridine (15 mL) was added a solution of 5'-*O*-trityl-N^6-benzoyldeoxyadenosine (239 mg, 0.4 mmol) in dry pyridine (6 mL) and the stirring continued at room temperature for 8 min under argon atmosphere. After usual work-up and silica gel column chromatography using 0.2-0.3% methanol in DCM , the product was obtained as white solid. Yield 197 mg (80%); R_f 0.73 (DCM/MeOH, 20:1, v/v); UV (MeOH) λ max 276 nm, λ min 241 nm; ^{1}H NMR ($CDCl_3$): δ = 2.73-2.79, 2.93-3.18 (m, 2H, 2'-H), 3.37-3.47, 3.56-3.70 (m, 2H, 5'-H), 4.34-4.40 (m, 1H, 4'-H), 4.58-4.62 (m, 1H, 3'-H), 6.42-6.78 (m, 1H, 1'-H), 7.14-7.86 (m, 20H, Ar-H), 8.42 (s, 1H, 2-H of Ad), 8.63 (s, 1H, 8-H of Ad). Anal : Calcd for $C_{36}H_{30}ClN_5O_3$: C, 70.1; H, 4.86; Cl, 5.71; N, 11.36. Found; C, 70.2; H, 4.97; Cl, 5.69; N, 11.47.

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