Pergamon

Tetrahedron Letters 39 (1998) 7123–7126

TETRAHEDRON LETTERS

A Convenient Method for Phosphorylation Involving a Facile Oxidation of *H*-Phosphonate Monoesters *via* Bis(trimethylsilyl) Phosphites

Takeshi Wada, Akira Mochizuki, Yuichi Sato and Mitsuo Sekine*

Department of Life Science, Faculty of Bioscience and Biotechnology, Tokyo Institute of Technology, Nagatsuta, Midoriku, Yokohama 226-8501, Japan

Received 13 June 1998; revised 12 July 1998; accepted 13 July 1998

Abstract: A new convenient route to phosphate monoesters from alcohols has been developed. *H*-Phosphonate monoesters, which are readily accessible by phosphonylation of the parent alcohols, were oxidized with *t*-BuOOH or *N*-sulfonyloxaziridines under anhydrous conditions *via* the corresponding bis(trimethylsilyl) phosphites. *N,O*-Bis(trimethylsilyl)benzamide (BSB) and (camphorsulfonyl)oxaziridine (CSO) were found to be highly effective for silylation of *H*-phosphonates and oxidation of silyl phosphites, respectively.

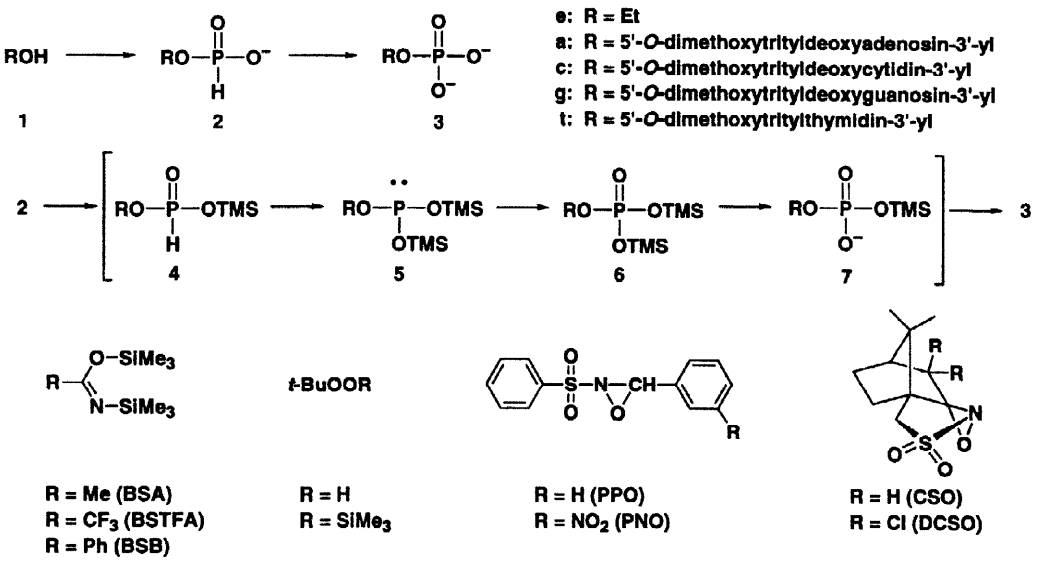
Keywords: phosphorylation; oxidation; oxaziridines; nucleic acids

In the chemical synthesis of natural products bearing phosphate groups, a number of methods for phosphorylation of the hydroxyl group have been explored [1]. In recent years, phosphate monoesters have frequently been synthesized *via H*-phosphonate diester [1,2] and phosphite triester intermediates [1,3] by use of highly reactive phosphonylating and phosphitylating reagents. After successive oxidation of these intermediates and removal of their phosphate protecting groups, the phosphorylated products can generally be obtained in good yields. On the other hand, although *H*-phosphonate monoesters **2** are readily accessible by the simple phosphonylation of the parent alcohols **1** with a wide variety of phosphonylating reagents (Scheme 1) [4], the direct oxidation of these compounds to the corresponding phosphates **3** has rarely been reported. One reason is the difficulty in the oxidation of *H*-phosphonate monoesters **2** compared with that of *H*-phosphonate diesters and phosphite triesters [5]. It is well known that the *H*-phosphonate monoesters **2** can be converted into trivalent silyl phosphites **5** [6] which are highly reactive to oxidizing reagents. Two practical methods have been reported to date for the oxidation of silyl phosphites **5**, which can be prepared from the corresponding *H*-phosphonates **2** with chlorotrimethylsilane in pyridine. Hata and Sekine have described the oxidation of thymidine 5'-bis(trimethylsilyl) phosphite with dipyridyl disulfide [7]. In this method, an additional hydrolytic step of the *S*-pyridyl phosphorothioate intermediate is required. Garegg *et al.* have described the use of aqueous iodine for the oxidation of silyl phosphite derivatives in the presence of chlorotrimethylsilane [8]. In this case, stringent reaction conditions are required. For instance, an excess amount of water results in the hydrolysis of the silyl group with incomplete reaction while excess silylating reagent results in the

0040-4039/98/$ - see front matter
PII: S0040-4039(98)01513-5

formation of pyrophosphate derivatives [9]. Both methods produce the water-soluble by-products from the silylating and oxidizing reagents. Unfortunately, most of the phosphate monoester derivatives **3** which can be obtained by the above methods are polar and water-soluble. Therefore, time-consuming purification of these products is generally required [7]. In this paper, we report an efficient method for the oxidation of *H*-phosphonate monoesters **2** *via* silyl phosphite **5** under mild anhydrous conditions, which can provide a new convenient route to phosphate monoesters from alcohols. (Scheme 1).

Scheme 1



First, triethylammonium ethyl phosphonate (**2e**) was treated with 5 equiv of *N,O*-bis(trimethylsilyl)acetamide (BSA) in CD_3CN and the reaction was monitored by ^{31}P NMR. After 2 min, the resonance signal of **2e** (3.28 ppm, $^1J_{PH}$ = 618.9 Hz, $^3J_{PH}$ = 8.3 Hz) completely disappeared and the signal of ethyl trimethylsilyl phosphonate **4e** (-1.92 ppm, $^1J_{PH}$ = 689.7 Hz, $^3J_{PH}$ = 8.5 Hz) was observed as a major product. The complete conversion of **4e** to ethyl bis(trimethylsilyl) phosphite **5e** (119.45 ppm, $^3J_{PH}$ = 7.3 Hz) required 70 min. Interestingly, use of a stronger silylating reagent *N,O*-bis(trimethylsilyl)trifluoroacetamide (BSTFA) required similar reaction time for the synthesis of **5e**. When **2e** was treated with *N,O*-bis(trimethylsilyl)benzamide (BSB) [10] which has not been used as a silylating reagent to date, quantitative formation of **5e** was observed after 5 min. On the other hand, when the DBU salt of **2e** was used as the starting material, the formation of **5e** was completed after 5 min by use of BSA. In this case, a free strong base DBU, which was released from **2e** upon the silylation, promoted the conversion of the inert species **4e** *via* tautomerism to a more reactive phosphite form that is quickly silylated to give **5e**.

The resulting silyl phosphite **5e** was treated *in situ* with 2 equiv of *t*-BuOOTMS. Monitoring of the reaction mixture by ^{31}P NMR indicated the formation of the corresponding bis(trimethylsilyl) phosphate **6e** (-16.11 ppm, $^3J_{PH}$ = 7.9 Hz). The reaction proceeded very slowly and the time required for the complete reaction was found to be

about 1 day. In contrast to this fact, addition of 50 equiv of anhydrous *t*-BuOOH to the reaction mixture resulted in the quantitative formation of trimethylsilyl ethyl phosphate **7e** (-6.32 ppm, $^3J_{PH}$ = 7.3 Hz) after 5 min. However, use of a small excess amount (2 equiv) of *t*-BuOOH with **5e** resulted in incomplete reaction after several hours with formation of **4e**. These results indicated that *t*-BuOOH was quickly silylated to give the less reactive *t*-BuOOTMS and the silyl phosphite **5e** was partially converted to **4e** which can not be oxidized with *t*-BuOOH or *t*-BuOOTMS. In the case of the reaction of **5e** with a large excess mount of *t*-BuOOH, the oxidation proceedrd faster than the desilylation of **5e** and the silylation of *t*-BuOOH.

In consideration of the above facts, an oxidizing reagent having no acidic hydrogens is apparently suitable for the oxidation of the silyl phosphites in the presence of silylating reagents. Quite recently, we have reported a new oxidation method for *H*-phosphonate diesters with 2-(phenylsulfonyl)-3-(3-nitrophenyl)oxaziridine (PNO) [12] *via* the corresponding trimethylsilyl phosphites under anhydrous conditions [11]. This method was next applied to the oxidation of the bis(trimethylsilyl) phosphite **5e**.

When **5e** was treated with 2 equiv of PNO in CD_3CN, the quantitative formation of **6e** was observed after 2 min. However, removal of the TMS group from **6e** by treatment with MeOH-Et_3N (9:1, v/v) resulted in the formation of an unidentified product (13.33 ppm). In this reaction, the phosphate anion might attack the imino carbon of *N*-(3-nitrobenzylidene)-phenylsulfonamide, which was generated from PNO, to form the by-product. In the case of using 2-(phenylsulfonyl)-3-phenyloxaziridine (PPO) [13] as an oxidizing reagent, a similar reaction was observed. In contrast to these facts, no side reactions were observed when (1*S*)-(+)-(10-camphorsulfonyl)oxaziridine (CSO) [14] and (1*S*)-(+)-(8,8-dichlorocamphor-sulfonyl)oxaziridine (DCSO) [15] were used as the oxidizing reagents. Treatment of the silyl phosphite **5e** with 2 equiv of CSO or DCSO resulted in the quantitative formation of **6e** after 2 min. Alternatively, treatment of the DBU salt of **2e** with CSO or DCSO in the presence of BSB in CD_3CN resulted in the direct formation of **6e** after 5 min. The product **6e** was treated with excess amounts of MeOH-Et_3N (9:1, v/v). After 5 min, almost quantitative formation of ethyl trimethylsilyl phosphate **7e** was observed. The complete removal of the TMS group from **7e** required several hours. When **6e** was treated with MeOH-DBU (97:3, v/v), the two TMS groups were quickly removed to give ethyl phosphate **3e** (5.37 ppm, $^3J_{PH}$ = 5.5 Hz) within 10 min.

In a similar manner, the DBU salts of nucleoside 3'-*H*-phosphonates (**2a**, **2c**, **2g**, and **2t**) bearing unmodified bases, which were synthesized in good yields by the phosphonylation of nucleosides (**1a**, **1c**, **1g**, and **1t**) with diphenyl phosphonate followed by the successive hydrolysis of the phenyl group [11], were converted to the corresponding 3'-phosphates (**3a**, **3c**, **3g**, and **3t**) by use of the optimized conditions with BSB and CSO as the silylating and oxidizing reagents, respectively [16]. All the reactions were monitored by ^{31}P NMR, which did not show any side reactions. The reaction mixtures containing **3** were diluted with H_2O-pyridine (1:1, v/v) and washed with $CHCl_3$. It is noteworthy that CSO, (camphorsulfonyl)imine, and benzamide, which exist in the reaction mixture, can be easily removed by simple extraction. In contrast to benzamide, acetamide derived from BSA is

difficult to remove by extraction or silica gel column chromatography. The aqueous layers were concentrated to dryness to give **3a**, **3c**, **3g**, and **3t** in 91, 92, 95, and 92% yields, respectively. These products were essentially pure (^{1}H and ^{31}P NMR) without further purification.

In conclusion, the present method is highly effective for the oxidation of *H*-phosphonate monoesters under mild conditions. A new silylating reagent BSB was found to be superior to the conventional BSA or BSTFA for the rapid silylation of *H*-phosphonate monoesters. It was found that commercially available CSO and less expensive DCSO were suitable for the present approach [17]. Moreover, these reagents do not react with purine, pyrimidine, and other modified bases of nucleic acids [11,18]. Therefore, the present method would be useful for the synthesis of various nucleotides and 5'-phosphorylated oligonucleotides as well as a wide variety of natural products bearing phosphate monoester functions *via H*-phosphonate intermediates.

Acknowledgments

This work was supported by Kanagawa Academy of Science and Technology (KAST) Research Grants, a Grant-in-Aid for Scientific Research on Priority Areas from the Ministry of Education, Science, Sports and Culture, Japan, and a grant from "Research for the Future" Program of the Japan Society for Promotion of Science (JSPS-RFTF 97I00301).

References and Notes

[1] Hayakawa Y. In: Trost BM, Fleming I. editors. Comprehensive organic synthesis. Oxford: Peragamon Press, 1991;6:601–630.
[2] Stawinski J. In: Engel R. editor. Handbook of organophosphorus chemistry. New York: Dekker, 1992:377–434.
[3] Beaucage SL, Iyer RP. Tetrahedron 1993;49:10441–10488.
[4] Schefield JA, Todd AR. J. Chem. Soc. 1961:2316–2310. Hata T, Sekine, M. J. Am. Chem. Soc. 1974;96:7363–7364. Sekine M, Hata T. Tetrahedron Lett. 1975;1711–1714. Sekine M, Mori H, Hata T. Tetrahedron Lett. 1979:1145–1148. Garegg, PJ, Regberg T, Stawinski J, Strönberg R. Chemica Scripta 1986;26:59–62. Garegg, PJ, Lindh I, Regberg T, Stawinski J, Strönberg R. Tetrahedron Lett. 1986;27:4051–4054. Froehler BC, Ng PG, Matteucci MD. Nucleic Acids Res. 1986;14:5399–5407. Marugg JE, Tromp M, Kuyl-Yeheskiely E, van der Marel GA, van Boom JH. Tetrahedron Lett. 1986;27:2661–2664. Sekine M, Narui S, Hata T. Tetrahedron Lett. 1988;29:1037–1040. Takaku H, Yamakage S, Sakatsme O, Ohtsuki M. Chem Lett. 1988:1675–1678. Sakatsume O, Ohtsuki M, Takaku H, Reese CB. Nucleic Acids Res. 1989;17:3689–3697. Sakatsume O, Yamane H, Takaku H, Yamamoto N. Tetrahedron Lett. 1989;30:6375–6378. Stawinski J, Thelin M. Nucleosides Nucleotides 1990;9:120–135. Jankowska J, Sobkowski M, Stawinski J, Kraszewski A. Tetrahedron Lett. 1994;35:3355–3358. Kers A, Kers I, Stawinski J, Sobkowski M, Kraszewski A. Synthesis 1995:427–430. Bhongle NN, Tang JY. Tetrahedron Lett. 1995;36:6803–6806. Yang Z, Xu Z, Shen N, Fang Z. Nucleosides Nucleotides 1995;14:167–173. Ozola V, Reese CB, Song Q. Tetrahedron Lett. 1996;37:8621–8624.
[5] Brown DM, Hammond PR. J. Chem Soc. 1960:4229–4232.
[6] Wozniak L, Chojnowski J. Tetrahedron 1989;45:2465–2524 and references cited therein.
[7] Hata T, Sekine M. Tetrahedron Lett. 1974;7363–7366.
[8] Garegg PJ, Regberg T, Stawinski J, Strömberg R. J. Chem. Soc. Perkin Trans. I. 1987;1269–1273.
[9] Wada T, Sekine M. unpublished results.
[10] Pump J, Rochow EG. Chem. Ber. 1964;97:627–635.
[11] Wada T, Sato Y, Honda F, Kawahara S, Sekine M. J. Am. Chem. Soc. 1997;119:12710–12721.
[12] For comprehensive reviews on the chemistry of oxaziridines, see Davis FA, Sheppard AC. Tetrahedron 1989;45:5703–5742.
[13] Davis FA, Lamendola Jr.J, Nadir U, Kluger EW, Sedergran TC, Panunto TW, Billmers R, Jenkins Jr.R, Truchi IJ, Watson WH, Chen JS, Kimura M. J. Am. Chem. Soc. 1980;102:2000–2005.
[14] Davis FA, Towson JC, Weismiller MC, Lal S, Carroll PJ. J. Am. Chem. Soc. 1988;110:8477–8482.
[15] Davis FA, Weismiller MC, Murphy CK, Reddy RT, Chen B. J. Org. Chem. 1992;57:7274–7285.
[16] A typical procedure: A DBU salt of **2a** (77 mg, 0.1 mmol) and CSO (60 mg, 0.2 mmol) were suspended in dry CH_3CN (1 ml) under argon atmosphere. BSB (266 mg, 1 mmol) was added to the above mixture and it was stirred at rt for 5 min to give **6a** (-12.64 ppm, $^3J_{PH}$ = 7.3 Hz) quantitatively (^{31}P NMR). MeOH-DBU (97:3, v/v, 1 ml, 0.1 mmol) was added to the mixture and it was stirred at rt for 10 min to give **3a** (4.79 ppm, $^3J_{PH}$ = 6.1 Hz) quantitatively (^{31}P NMR). The mixture was diluted with H_2O-pyridine (1:1, v/v, 10 ml) and washed with $CHCl_3$ (5 ml x 3). The aqueous layer was co-evaporated with EtOH and concentrated to dryness to give **3a** (86 mg, 91%) as a colorless foam.
[17] These reagents were found to be effective for the oxidation of oligodeoxyribonucleotides bearing *H*-phosphonate internucleotidic linkages on a solid-support in the presence of BSA or BSB [11].
[18] Hogrefe RI. (Genta Inc.) personal communicaton. In: Glen Report. Virginia: Glen Research, 1996;9:8–9.