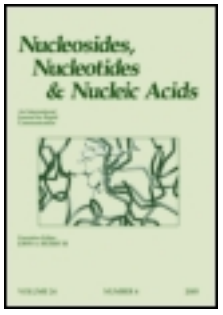


Nucleosides and Nucleotides
Publication details, including instructions for authors and subscription information:
http://www.tandfonline.com/loi/lncn19

Selective 5′-O-Acetylation of 2′-Deoxynucleosides and Nucleosides by a Modified Mitsunobu Procedure

Gong-Xin He [a] & Norbert Bischofberger [a]
[a] Gilead Sciences Inc. , 353 Lakeside Drive, Foster City, CA, 94404, USA
Published online: 22 Aug 2006.

To cite this article: Gong-Xin He & Norbert Bischofberger (1997) Selective 5′-O-Acetylation of 2′-Deoxynucleosides and Nucleosides by a Modified Mitsunobu Procedure, Nucleosides and Nucleotides, 16:3, 257-263, DOI: 10.1080/07328319708001346

To link to this article: http://dx.doi.org/10.1080/07328319708001346

NUCLEOSIDES & NUCLEOTIDES, 16(3), 257-263 (1997)

SELECTIVE 5'-O-ACETYLATION OF 2'-DEOXYNUCLEOSIDES AND NUCLEOSIDES BY A MODIFIED MITSUNOBU PROCEDURE

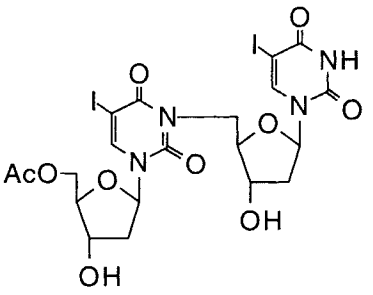
Gong-Xin He* and Norbert Bischofberger
Gilead Sciences Inc., 353 Lakeside Drive, Foster City, CA 94404, USA

Abstract: 5'-Hydroxyl groups of deoxynucleosides and nucleosides except for guanosine can be acetylated in high yields and selectivities through a modified Mitsunobu procedure by adding diethyl azodicarboxylate to a suspension of the deoxynucleoside or nucleoside in dioxane containing triphenylphosphine and excess acetic acid at 60°C.

Selective protection of the 5'-hydroxyl group versus the 3'- or/and 2'-hydroxyl groups of 2'-deoxynucleosides and nucleosides is a frequently utilized reaction. Several 5'-O-selective sterically hindered protecting groups, such as trityl, di(*t*-butyl)methylsilyl, etc., have been developed.[1] The presence of a bulky 5'-O-protecting group can cause steric hindrance to the subsequent reactions on either the base or the sugar moiety. Most of the current 5'-O-selective protecting groups are acid-labile and have to be removed under acidic conditions. One exception is the pivaloyl group, which has to be removed under relatively strong basic conditions.[2] Therefore, in some cases, it is desirable to have a small, base-labile protecting group, such as acetyl, which can be selectively introduced onto the 5'-hydroxyl group. However, a survey of the literature reveals that in most methods for selective acetylation of a primary hydroxyl group versus a secondary hydroxyl group, the diol reactant has to be completely dissolved in a relatively non-polar solvent

and/or the reaction must be conducted at a low temperature.[3-5] For example, Yamamoto recently reported a method using acetyl chloride/hindered amine in CH_2Cl_2 at -78°C.[3] The selective acetylation with *N*-acetylimidazole uses chloroform as the solvent.[4] These methods are not suitable for deoxynucleosides and nucleosides because of their low solubility in these solvents. In this paper, we would like to report that by using a modified Mitsunobu procedure, an acetyl group can be introduced to the 5'-O-position of deoxynucleosides and nucleosides, except for guanosine, through a simple procedure with high selectivity and high yield.

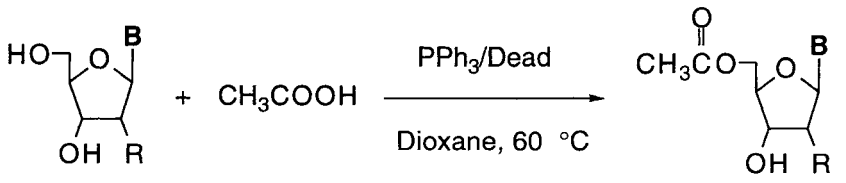
Mitsunobu reported that the selective acetylation of the 5'-hydroxyl group of thymidine can be achieved in 55% yield by adding a dioxane solution of acetic acid (1.0 eq) and diethyl azodicarboxylate (1.0 eq) to thymidine and triphenylphosphine (1 eq) suspended in dioxane.[6] Under similar conditions, selective aroylation of the 5'-hydroxyl group of thymidine was achieved with the optimal yield of 85% for *p*-nitrobenzoylation. The selective aroylation of the 5'-hydroxyl group of uridine and adenosine in 43 - 56% yield was also reported.[7] By using this procedure in the acetylation of 5-iodo-2'-deoxyuridine, we obtained 5'-O-acetyl-5-iodo-2'-deoxyuridine in about 50% yield along with a major impurity isolated in 30% yield and identified as the dimeric compound **1** by 2D NMR.[8] Apparently,



1

under the reaction conditions, the N^3 of 5-iodo-2'-deoxyuridine competes with acetic acid as a nucleophile. Based on this result, we modified the procedure to adding the activating agent to the diol containing excess acetic acid. Thus, when a dioxane solution of diethyl azodicarboxylate (1.5 eq) was added to a suspension

TABLE 1. Selective acetylation of 5'-hydroxyl group of nucleosides



R	B	Yield (%)
H	5-iodouracil	87
"	N^6-benzoyladenine	90
"	thymine	85
"	N^6-benzoylcytosine	90
"	N^2-isobutyrylguanine	88
"	N^2-diisobutylformamidineguanine	74
OH	uracil	85
"	N^6-benzoyladenine	93
"	N^6-benzoylcytosine	76
"	N^2-isobutyrylguanine	81[a]
"	N^2-diisobutylformamidineguanine	75[b]

[a] 1:1 mixture of 5'-O-acetyl-N^2-isobutyrylguanosine and 5'-N^3-cyclo-N^2-isobutyrylguanosine

[b] containing 20% of 3'-O-acetyl-N^2-diisobutylformamidineguanosine

containing 5-iodo-2'-deoxyuridine, triphenylphosphine (1.5 eq), and acetic acid (5.0 eq) in dioxane, the desired product 5'-O-acetyl-5-iodo-2'-deoxyuridine was obtained in 87% yield. Subsequently, this procedure was also applied to other deoxynucleosides, including N^6-benzoyl-2'-deoxyadenosine, thymidine, N^2-isobutyryl-2'-deoxyguanosine, and N^6-benzoyl-2'-deoxycytidine (Table 1). The reaction is very selective and no 3'-O-acetyl-isomer was detected in the reaction. Yields are generally high and no other by-products, such as 5'-N^3-cycloadenosine, 5'-O^4-cyclothymidine, etc., were found

for the deoxynucleosides. In addition, if the starting material is not completely anhydrous, more triphenylphosphine and diethyl azodicarboxylate can be added until TLC indicates the completion of the reaction. The reaction was successfully applied to nucleosides also, although more dioxane and reagents, including triphenylphosphine, diethyl azodicarboxylate, and acetic acid, must be used to compensate for the low solubility. As shown in Table 1, N^6-benzoyladenosine, uridine, and N^6-benzoylcytidine give good yields and selectivities. For N^2-isobutyrylguanosine, however, an 1:1 mixture of 5'-O-acetyl-N^2-isobutyrylguanosine and 5'-N^3-cyclo-N^2-isobutyrylguanosine[9] was obtained. By using N^2-diisobutylformamidineguanosine, the intramolecular cyclization was suppressed, but the product obtained was contaminated by 3'-O-acetyl-N^2-diisobutylformamidineguanosine (20%).

In summary, by using this procedure, acetyl groups can be introduced onto the 5'-hydroxyl groups of deoxynucleosides and nucleosides with high yield and high selectivity. Since acetyl groups can be removed under very mild basic conditions, such as methanolic ammonia at room temperature for 30 minutes, the 5'-O-acetyl group can be used as an alternative to the acid-labile protecting groups, such as trityl and silyl.

EXPERIMENTAL SECTION

General

Deoxynucleosides and nucleosides were purchased from Chem-Impex International, IL and Peninsula Laboratories, Inc., CA. Other chemicals were obtained from Aldrich. ^{1}H-NMR and 2D NMR experiments (HMBC and HMQC) were conducted on GE QE-300 300 MHz or Varian Unityplus 500 MHz spectrometer using tetramethylsilane as an internal standard.

General procedure

The deoxynucleoside (2 mmol) or nucleoside (1 mmol) was suspended in dioxane (50 mL). Triphenylphosphine (3 mmol) and glacial acetic acid (10 mmol) were added. The resulting mixture was heated to 60°C under stirring, and a solution of diethyl azodicarboxylate (3 mmol) in dioxane (10 mL) was added dropwise.

Gradually, the suspension became a clear solution. The solution was subsequently stirred at 60°C for 1 hours, followed by cooling to room temperature and evaporation of the solvent. The oily residue was purified by silica gel chromatography (CH_2Cl_2/MeOH = 100/5 to 100/10 (v/v)) yielding the desired product. Structure assignments and purities of the products were confirmed by ^{1}H-NMR[10] and 2D NMR experiments.

Acknowledgment: This work was supported in part by SBIR Grant No. 1R43 HL484311-01.

REFERENCES AND NOTES:

1. Greene, T. W.; Wuts, P. G. M. *Protective Group in Organic Synthesis* 2nd Ed., p. 53 - 84, John Wiley & Sons, Inc. 1991.
2. Robins, M. J.; Hawrelak, S. D.; Kanai, T.; Siefert, J. M.; Mengel, R. *J. Org. Chem.* **1979**, *44*, 1317.
3. Ishihara, K.; Kurihara, H.; Yamamoto, H. *J. Org. Chem.* **1993**, *58*, 3791.
4. Lemieux, R. U.; Driguez, H. *J. Am. Chem. Soc.* **1975**, *97*, 4063.
5. Stork, G.; Takahashi, T.; Kawamoto, I.; Suzuki, T. *J. Am. Chem. Soc.* **1978**, *100*, 8272.
6. Mitsunobu, O.; Kimura, J.; Fujisawa, Y. *Bull. Chem. Soc. Jpn.* **1972**, *45*, 245.
7. Shimokawa, S.; Kimura, J.; Mitsunobu, O. *Bull. Chem. Soc. Jpn.* **1976**, *49*, 3357.
8. Compound **1**: ^{1}H-NMR (500 MHz, DMSO-d_6) δ 11.68(s, NH, 1H), 8.12(s, 6-H, 1H), 8.06(s, 6-H, 1H), 6.17(t, *J* = 7 Hz, 1'-H, 1H), 6.03(dd, *J* = 5.5, 8 Hz, 1'-H, 1H), 5.41(d, *J* = 4.5 Hz, OH, 1H), 5.35(d, *J* = 4 Hz, OH, 1H), 4.1 - 4.3(m, 3' and 4'-H, 4H), 3.9 - 4.1(m, 5' and 5"-H, 4H), 2.2 - 2.3(m, 2' and 2"-H, 2H), 2.11(s, CH_3, 3H), 2.0 - 2.1(m, 2' and 2"-H, 2H).
9. 5'-N^3-cyclo-N^2-isobutyrylguanosine: ^{1}H-NMR (500 MHz, DMSO-d_6) δ 8.02(s, 8-H, 1H), 6.26(s, 1'-H, 1H), 5.55(d, *J* = 4.5 Hz, OH, 1H), 5.50(d, *J* = 6 Hz, OH, 1H), 5.16(d, *J* = 14.5, 5'-H, 1H), 4.6(m, 4'-H, 1H), 4.3(m, 3'-H, 1H), 3.9(m, 2'-H, 1H), 3.86(d, *J* = 14.5, 5"-H, 1H). Formation of 5'-N^3-cyclo-N^2-isobutyryl-2'-deoxyguanosine under Mitsunobu conditions was reported recently.

Perbost, M.; Hoshiko, T.; Morvan, F.; Swayze, E.; Griffey, R. H.; Sanghvi, Y. S. *J. Org. Chem.* **1995**, *60*, 5150.

10. 5'-O-Acetyl-5-iodo-2'-deoxyuridine: ^{1}H-NMR (300 MHz, DMSO-d_6) δ 11.71(s, NH, 1H), 7.97(s, 6-H, 1H), 6.08(t, J = 6.5 Hz, 1'-H, 1H), 5.39(d, J = 4.0 Hz, OH, 1H), 4.2(m, 3'-H, 5'-H, and 5"-H, 3H), 3.9(m, 4'-H, 1H), 2.3(m, 2'-H, 1H), 2.2(m, 2"-H, 1H), 2.10(s, $COCH_3$, 3H).

5'-O-Acetyl-N^6-benzoyl-2'-deoxyadenosine: ^{1}H-NMR (300 MHz, DMSO-d_6) δ 11.18(s, NH, 1H), 8.74(s, 8-H, 1H), 8.63(s, 2-H, 1H), 7.5 - 8.1(m, Ph-H, 5H), 6.49(t, J = 6.5 Hz, 1'-H, 1H), 5.52(d, J = 4 Hz, OH, 1H), 4.5(m, 3'-H, 1H), 4.3(m, 5'-H, 1H), 4.1(m, 5"-H, 1H), 4.0(m, 4'-H, 1H), 2.9(m, 2'-H, 1H), 2.4(m, 2"-H, 1H), 1.97(s, $COCH_3$, 3H).

5'-O-Acetylthymidine: ^{1}H-NMR (300 MHz, DMSO-d_6) δ 11.31(s, NH, 1H), 7.43(s, 6-H, 1H), 6.17(t, J = 6.5 Hz, 1'-H, 1H), 5.40(d, J = 4.0 Hz, OH, 1H), 4.2(m, 3'-H, 5'-H, and 5"-H, 3H), 3.9(m, 4'-H, 1H), 2.4(m, 2'-H, 1H), 2.2(m, 2"-H, 1H), 2.04(s, $COCH_3$, 3H), 1.78(s, CH_3, 3H).

5'-O-Acetyl-N^6-benzoyl-2'-deoxycytidine: ^{1}H-NMR (300 MHz, DMSO-d_6) δ 11.24(s, NH, 1H), 8.14(d, J = 7.5 Hz, 6-H, 1H), 7.4 - 8.0(m, Ph-H, 5H), 7.38(d, J = 7.5 Hz, 5-H, 1H), 6.15(t, J = 6 Hz, 1'-H, 1H), 5.45(d, J = 4.5 Hz, OH, 1H), 4.2(m, 3'-H, 5'-H, and 5"-H, 3H), 4.0(m, 4'-H, 1H), 2.3(m, 2'-H, 1H), 2.1(m, 2"-H, 1H), 2.05(s, $COCH_3$, 3H).

5'-O-Acetyl-N^2-isobutyryl-2'-deoxyguanosine: ^{1}H-NMR (300 MHz, DMSO-d_6) δ 12.06(s, NH, 1H), 11.64(s, NH, 1H), 8.18(s, 8-H, 1H), 6.21(t, J = 6.5 Hz, 1'-H, 1H), 5.46(d, J = 3 Hz, OH, 1H), 4.4(m, 3'-H, 1H), 4.2(m, 5'-H, 1H), 4.1(m, 5"-H, 1H), 4.0(m, 4'-H, 1H), 2.7(m, 2'-H, 1H), 2.6(m, CH, 1H), 2.3(m, 2"-H, 1H), 2.00(s, $COCH_3$, 3H), 1.15(d, J = 6.5 Hz, CH_3, 6H).

5'-O-Acetyl-N^2-diisobutylformamidine-2'-deoxyguanosine: ^{1}H-NMR (300 MHz, DMSO-d_6) δ 11.28(s, NH, 1H), 8.57(s, CH=N, 1H), 7.97(s, 8-H, 1H), 6.24(t, J = 6.5 Hz, 1'-H, 1H), 5.45(d, J = 4 Hz, OH, 1H), 4.4(m, 3'-H, 1H), 4.3(m, 5'-H, 1H), 4.1(m, 5"-H, 1H), 4.0(m, 4'-H, 1H), 3.2 - 3.4(m, CH_2, 4H), 2.7(m, 2'-H, 1H), 2.3(m, 2"-H, 1H), 1.99(s, $COCH_3$, 3H), 1.8 - 2.2(m, CH, 2H), 0.8 - 1.0(m, CH_3, 12H).

5'-O-Acetyluridine: ^{1}H-NMR (300 MHz, DMSO-d_6) δ 11.34(s, NH, 1H), 7.61(d, J = 8.0 Hz, 6-H, 1H), 5.74(d, J = 5.0 Hz, 1'-H, 1H), 5.65(d, J = 8.0 Hz, 5-H, 1H), 5.5(bs, OH, 1H), 5.3(bs, OH, 1H), 4.3(m, 5'-H, 1H), 4.2(m, 5"-H, 1H), 4.1(m, 2'-H, 1H), 3.8 - 4.0(m, 3'-H and 4'-H, 2H), 2.04(s, $COCH_3$, 3H).

5'-O-Acetyl-N^6-benzoyladenosine: ^{1}H-NMR (300 MHz, DMSO-d_6) δ 11.22(s, NH, 1H), 8.76(s, 8-H, 1H), 8.66(s, 2-H, 1H), 7.5 - 8.1(m, Ph-H, 5H), 6.04(d, J = 4.5 Hz, 1'-H, 1H), 5.7(bs, OH, 1H), 5.4(bs, OH, 1H), 4.7(m, 2'-H, 1H), 4.3(m, 5'-H, 1H), 4.2(m, 5"-H, 1H), 4.1(m, 3'-H and 4'-H, 2H), 2.01(s, $COCH_3$, 3H).

5'-O-Acetyl-N^6-benzoylcytidine: ^{1}H-NMR (300 MHz, DMSO-d_6) δ 11.27(s, NH, 1H), 8.14(d, J = 7.5 Hz, 6-H, 1H), 7.4 - 8.0(m, Ph-H, 5H), 7.38(d, J = 7.5 Hz, 5-H, 1H), 5.80(d, J = 3.0 Hz, 1'-H, 1H), 5.7(bs, OH, 1H), 5.3(bs, OH, 1H), 4.2 - 4.4 (m, 5'-H and 5"-H, 2H), 4.1(m, 2'-H and 3'-H, 2H), 3.9(m, 4'-H, 1H), 2.07(s, $COCH_3$, 3H).

5'-O-Acetyl-N^2-isobutyrylguanosine: ^{1}H-NMR (300 MHz, DMSO-d_6) δ 8.00(s, 8-H, 1H), 5.81(d, J = 5.5 Hz, 1'-H, 1H), 5.6(bs, OH, 1H), 5.4(bs, OH, 1H), 4.5(m, 2'-H, 1H), 4.5(m, 5'-H, 1H), 4.2(m, 3'-H and 5"-H, 2H), 4.1(m, 4'-H, 1H), 2.02(s, $COCH_3$, 3H), 2.5 - 2.6(m, CH, 1H), 1.1 - 1.2(m, CH_3, 6H).

5'-O-Acetyl-N^2-diisobutylformamidineguanosine: ^{1}H-NMR (300 MHz, DMSO-d_6) δ 11.31(s, NH, 1H), 8.56(s, CH=N, 1H), 7.98(s, 8-H, 1H), 5.80(d, J = 5 Hz, 1'-H, 1H), 5.6(bs, OH, 1H), 5.5(bs, OH, 1H), 4.6(m, 2'-H, 1H), 4.3(m, 5'-H, 1H), 4.2(m, 3'-H, 1H), 4.1(m, 5"- H, 1H), 4.0(m, 4'-H, 1H), 3.2 - 3.4(m, CH_2, 4H), 2.00(s, $COCH_3$, 3H), 1.8 - 2.2(m, CH, 2H), 0.7 - 1.0(m, CH_3, 12H).

Received November 27, 1996
Accepted December 26, 1996