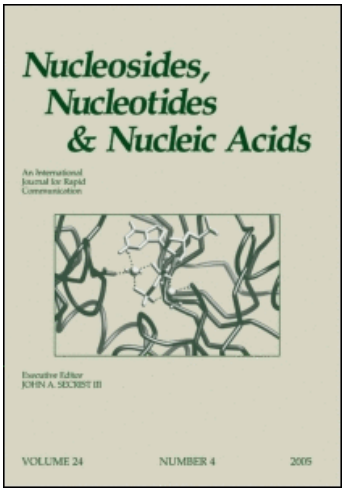


Nucleosides, Nucleotides and Nucleic Acids
Publication details, including instructions for authors and subscription information:
http://www.informaworld.com/smpp/title~content=t713597286

^{18}O-Labeled Adenosine and 9-(β-D-Arabinofuranosyl)Adenine (ARA-A): Synthesis, Mass Spectrometry, and Studies of ^{18}O-Induced ^{13}C NMR Chemical Shifts

Cong Jiang[a]; Robert J. Suhadolnik[b]; David C. Baker[a]
[a] Department of Chemistry, The University of Alabama, Tuscaloosa, AL [b] Department of Biochemistry, Temple University School of Medicine, Philadelphia, PA

To cite this Article Jiang, Cong , Suhadolnik, Robert J. and Baker, David C.(1988) '^{18}O-Labeled Adenosine and 9-(β-D-Arabinofuranosyl)Adenine (ARA-A): Synthesis, Mass Spectrometry, and Studies of ^{18}O-Induced ^{13}C NMR Chemical Shifts', Nucleosides, Nucleotides and Nucleic Acids, 7: 3, 271 — 294
To link to this Article: DOI: 10.1080/07328318808068710
URL: http://dx.doi.org/10.1080/07328318808068710

NUCLEOSIDES & NUCLEOTIDES, 7(3), 271-294 (1988)

^{18}O-LABELED ADENOSINE AND 9-(β-D-ARABINOFURANOSYL)ADENINE (ARA-A): SYNTHESIS, MASS SPECTROMETRY, AND STUDIES OF ^{18}O-INDUCED ^{13}C NMR CHEMICAL SHIFTS

Cong Jiang,[†] Robert J. Suhadolnik,[‡] and David C. Baker[†*]

[†]Department of Chemistry
The University of Alabama
Tuscaloosa, AL 35487-9671

and

[‡]Department of Biochemistry
Temple University School of Medicine
Philadelphia, PA 19140

ABSTRACT. [2'-^{18}O]- and [3'-^{18}O]-Adenosine and [2'-^{18}O]- and [3'-^{18}O]-9-(β-D-arabinofuranosyl)adenine were synthesized from appropriate nucleoside precursors. The sites of ^{18}O-incorporation were determined by mass spectrometry. ^{18}O-Induced ^{13}C NMR shifts were measured for 2'- and 3'-labeled adenosines as 1.2 and 1.6 Hz, respectively.

INTRODUCTION

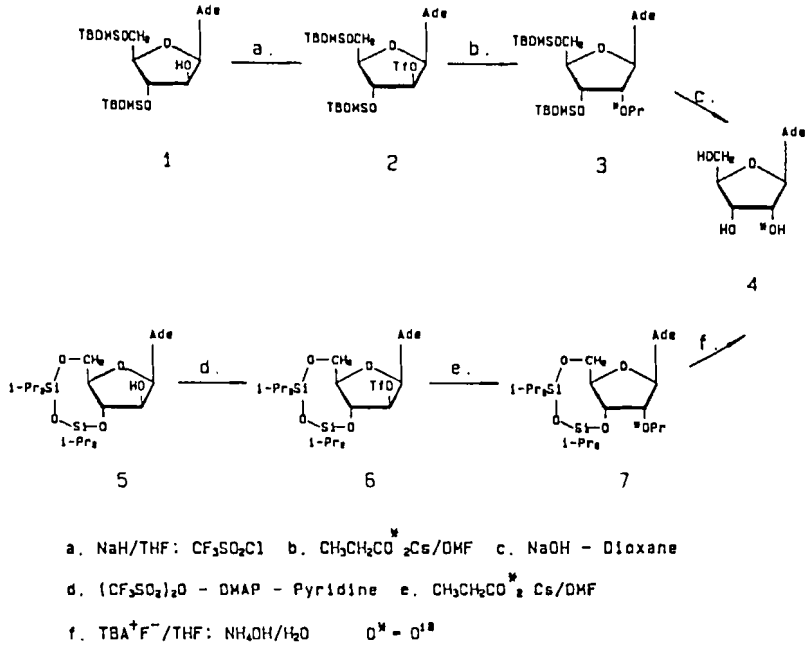
The use of isotopically labeled nucleosides, particularly those labeled with radioactive nuclides, has been well established in all facets of nucleic acid chemistry. Research on the biosynthesis of 2'-deoxycoformycin (2'-dCF)[1,2] and 9-(β-D-arabinofuranosyl)adenine (ara-A),[3,4] both products from the fermentation of Streptomyces antibioticus, utilized both ^{14}C- [1 - 4] and ^{13}C-labels[2] as biochemical markers. ^{13}C-labeled precursors were found especially useful in establishing the fact that 2'-dCF is derived from adenosine via a unique ring expansion, with D-ribose serving as C-donor.[2] In the case of the biosynthesis of ara-A, an "adenosine 2'-epimerase" has been identified[3,4] which converts adenosine to ara-A. However, details of

the mechanism of the epimerization remain obscure, and ^{18}O-labeled precursors were deemed necessary to establish the fate of the 2'-OH and 3'-OH in adenosine as well as the origin of the 2'-OH in the product, ara-A. It was anticipated that such ^{18}O-labeled products would be easily studied by mass spectrometry and possibly by observation of ^{18}O-induced ^{13}C NMR chemical shifts, a procedure[5] which has recently gained wide acceptance in the biosynthetic field. To this end, a workable synthesis of [2'-^{18}O]- (**4**) and [3'-^{18}O]-adenosine (**16**), as well as [2'-^{18}O]- (**20**) and [3'-^{18}O]-9-(β-D-arabinofuranosyl)adenine (**12**), preferably one which proceeded from intact nucleoside precursors, was required.[6]

DISCUSSION AND RESULTS

Synthesis. [2'-^{18}O]-Adenosine (4). The shortest and most direct approach to [2'-^{18}O]-adenosine (**4**) should be via 2'-inversion of a 3',5'-O-protected 9-β-D-arabinofuranosyladenine derivative with a suitable leaving group at C-2'. Such a process is shown in Scheme 1.

SCHEME 1



a. NaH/THF: CF_3SO_2Cl b. $CH_3CH_2CO^*_2Cs$/DMF c. NaOH - Dioxane

d. $(CF_3SO_2)_2O$ - DMAP - Pyridine e. $CH_3CH_2CO^*_2Cs$/DMF

f. TBA^+F^-/THF: NH_4OH/H_2O $O^* = O^{18}$

Thus 9-[3',5'-di-O-(tert-butyldimethylsilyl)-β-D-arabinofuranosyl]-adenine (**1**)[7,8] was converted to its 2'-O-(trifluoromethanesulfonyl) derivative (triflate) **2** using sodium hydride - trifluoromethanesulfonyl chloride in a yield of 43%. The ^{1}H NMR spectrum of **2** showed a pronounced downfield shift at δ 5.21 for H-2' (a pseudo triplet, with $J_{1',2'}$ = 2 Hz and $J_{2',3'}$ = 3.5 Hz) which served to pinpoint the site of triflation. Displacement of the triflyl group using cesium [^{18}O]-propionate in dry N,N-dimethylformamide (DMF) afforded, in 85% yield, [2'-^{18}O]-3',5'-di-O-(tert-butyldimethylsilyl-2'-O-(1-propionyl)-adenosine (**3**). As for **2**, the ^{1}H NMR spectrum of **3** reflected acylation of the 2'-O, with H-2' resonating at δ 5.40 (pseudo triplet, $J_{1',2'}$ = 5.3 Hz and $J_{2',3'}$ = 5.0 Hz). In each compound, H-1' appeared as a doublet, with $J_{1',2'}$ = 2.1 Hz for **2** and $J_{1',2'}$ = 5.3 Hz for **3**. Compound **3** was then desilylated with fluoride, and the 2'-ester was hydrolyzed in base (aqueous dioxane) to give [2'-^{18}O]-adenosine (**4**) in 89% yield. The product was identical by mp and ^{1}H NMR spectroscopy with authentic adenosine.

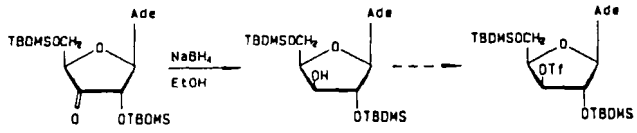
Although the above synthetic route furnished **4** in quantities sufficient for milligram-scale requirements, it became necessary, on account of demand for this compound for biosynthetic studies, to develop a higher yielding process. Drawing upon the work of Markiewicz and coworkers that utilizes the reagent, 1,3-dichloro-1,1,3,3-tetraisopropyldisiloxane, to simultaneously block the 3'- and 5'-positions of ribofuranosyl nucleosides,[9 - 11] ara-A was reacted with the reagent (using modified conditions specified by Robins and coworkers[12]) in pyridine to give **5** in a respectable yield (Scheme 1). There was no evidence of a possible 2',5'-isomer as determined by TLC and ^{1}H NMR spectroscopy. Triflation then proceeded using trifluoromethanesulfonic anhydride - 4-(N,N-dimethylamino)pyridine in dichloromethane to give glassy **6** in 75% yield after column chromatography. Displacement of triflate **6** with cesium [^{18}O]-propionate then gave **7** in 89% yield. As

for **2** and **3**, the ^{1}H NMR spectrum of **6** and **7** revealed the presence of 2'-O-triflyl and 2'-O-(1-propionyl) groups, as H-2' resonated at δ 5.48 and δ5.79, respectively, in **6** and **7**. A distinguishing feature of these spectra was the fact that inversion of **6** to **7** resulted in a dramatic alteration of $J_{1',2'}$ = 5.8 Hz (for **6**) to 0 Hz (for **7**). (See Experimental Section.) The small coupling between H-1' and H-2' for ribonucleosides derivatized with Markiewicz's protecting group has been noted by Robins and coworkers and has been advocated as a means of assigning anomeric configuration among such compounds.[12] These workers attribute the observed coupling to the greater population of the ^{3}E (C-3' endo) conformation for such rigid, fused-ring compounds. The inverted product **7** was then converted to [2-^{18}O]-adenosine (**4**) in 89% yield by a combination of (a) fluoride ion desilylation, followed by (b) base hydrolysis of the 2'-ester. The site of attachment for the ^{18}O label was firmly established by mass spectrometry and by observation of an appropriate ^{18}O-induced ^{13}C NMR shift for the C-2' resonance (vide infra).

The advantages offered by the sequence of **5** to **4** over that of **1** to **4** are a higher yield, greater reproducibility, and easier access to the 3',5'-protected derivative **5**, which requires no separation from isomeric bis-protected compounds as in the case of **1**. The synthetic advantages are somewhat offset, however, by the higher cost of Markiewicz's reagent. Nonetheless, the latter process is advocated as the route of choice for synthesis of **4**.

Attempted Synthesis of [3'-^{18}O]-Adenosine (16). Encouraged by the success of the synthesis of [2'-^{18}O]-adenosine (**4**), an analogous process was designed whereby the corresponding 9-[2',5'-di-O-(tert-butyldimethylsilyl)-β-D-xylofuranosyl]adenine (**9**,[7] Scheme 2) was to be (a) triflated and (b) displaced with cesium [^{18}O]-propionate to afford the 3'-labeled adenosine derivative. However, no more than a miniscule amount (i.e., an NMR sample) of the desired 3'-O-triflyl derivative in

SCHEME 2



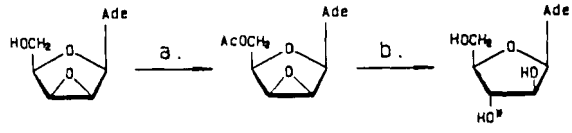
8 9

the xylo series could be obtained, and starting material was routinely recovered in high yield from attempted triflation reactions using either triflyl chloride or triflic anhydride - DMAP. The reason for this difficulty is not apparent; however, a survey of the literature on carbohydrate triflates[13] shows no examples of any 3'-O-triflyl-derivatized nucleosides.

Upon rethinking the strategy to [3'-^{18}O]-adenosine (**16**), it became apparent that, if one had access to [3'-^{18}O]-9-(β-D-arabinofuranosyl)-adenine (**12**), the process of **5** to **4** depicted in Scheme 1 would work beautifully to synthesize the desired 3'-labeled adenosine (**16**). Thus [3'-^{18}O]-ara-A (**12**) became the immediate target compound.

[3'-^{18}O]-9-(β-D-Arabinofuranosyl)adenine (12). Drawing upon the process used by Lee et al. for the original synthesis of ara-A,[14] whereby 9-(2',3'-anhydro-β-D-lyxofuranosyl)adenine (**10**) was ring-opened with sodium benzoate to give the arabinonucleoside, a synthesis of **12** was designed as depicted in Scheme 3. Compound **10** is readily available

SCHEME 3



10 11 12

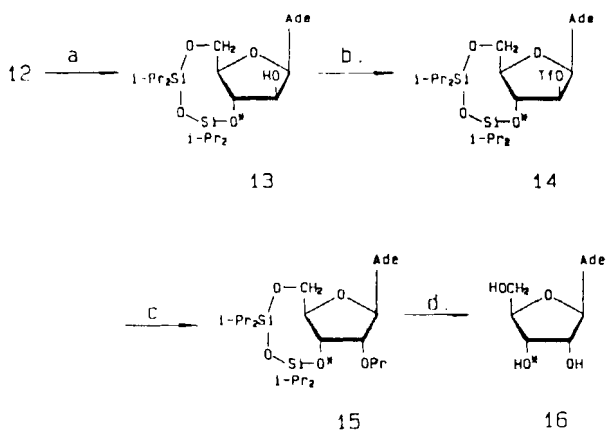
a. Ac_2O - Pyridine b. $CH_3CH_2CO^*{}_2Cs$/DMF; NH_4OH - MeOH $O^* = O^{18}$

from ara-A via a one-step, high-yielding process that uses the Mitsunobu reagent[15] to effect closure to the epoxide.[16] Acetylation of

10 furnished **11**, a compound of enhanced solubility, in 95% yield. Ring opening of epoxide **11** proceeded with cesium [$^{18}\underline{O}$]-propionate to give upon workup and hydrolysis in ammonium hydroxide a 54% yield of [3'-$^{18}\underline{O}$]-ara-A (**12**) that was purified by adsorption-desorption on Diaion HP-20 resin. The product was of high purity and was identical with authentic Ara-A by mp and and by ^{1}H NMR spectroscopy. The ^{18}O label was firmly established to be at the 3'-position by mass spectrometry (vide infra).

[3'-$^{18}\underline{O}$]-Adenosine (16). With sufficient quantities of [3'-$^{18}\underline{O}$]-ara-A (**12**) in hand, a sequence patterned after the synthesis of **4** (i.e., conversion of 5 to **4**) was devised as shown in Scheme 4. Thus by

SCHEME 4

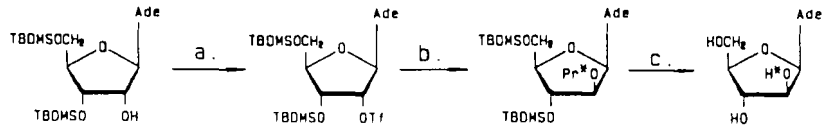


substituting [3'-$^{18}\underline{O}$]-ara-A (**12**) for ara-A, the reaction was carried out as for the synthesis of **4** to afford **16** in an overall yield of 40% from **12** (32% from ara-A). Compounds **14** and **15** gave ^{1}H NMR spectra which matched exactly those recorded for **6** and **7**. The product **16** had a mp and ^{1}H NMR spectrum identical with those for authentic adenosine. The site of the ^{18}O-labeling was established by mass spectrometry and by the

observed ^{18}O-induced ^{13}C NMR shift for the C-3' carbon resonance (vide infra).

[2'-^{18}O]-9-(β-D-Arabinofuranosyl)adenine (20). The synthesis of the remaining analogue, [2'-^{18}O]-9-(β-D-arabinofuranosyl)adenine (**20**), was patterned after the synthesis of ara-A described by Ranganathan and Larwood[17] who used 3',5'-di-O-(tetrahydropyranyl)adenosine prepared by a rather lengthy process to obtain a 2'-O-(triflyl) derivative that was inverted at the 2'-carbon by AcO^- in HMPA to give ara-A in a 40% overall yield. By the considerably shortened process shown in Scheme 5, 3',5'-di-O-(tert-butyldimethylsilyl)adenosine (**17**) is triflated with sodium

SCHEME 5



17 18 19 20

a. NaH/THF: CF_3SO_2Cl b. $CH_3CH_2CO^*{}_2Cs$/DMF c. NaOH - Dioxane $O^* = O^{18}$

hydride - trifluoromethanesulfonyl chloride to give **18** in 30% yield. The ^{1}H NMR spectrum for **18** gave a pseudo triplet for H-2' at δ 5.84, confirming triflation at O-2'. The observed $J_{1',2'}$ = 4.3 Hz (compare with $J_{1',2'}$ = 2.1 Hz for the D-arabino analogue **2**). Inversion using cesium [^{18}O]-propionate then gave the D-arabino nucleoside **19** in 89% yield. The ^{1}H NMR spectrum shows the H-2' at δ5.40, confirming acylation at O-2'. The observed $J_{1',2'}$ = 5.4 Hz. Deprotection of **19** was effected with fluoride and base as for **3** to give the [2'-^{18}O]-ara-A in 79% yield. The product was identical with authentic ara-A by mp and by ^{1}H NMR spectroscopy. The position of the ^{18}O label was firmly established by mass spectrometry (vide infra).

The above sequence from **17** to **20** is limited by the low yield of the triflation step to give **18**. It is anticipated that this situation can

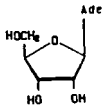
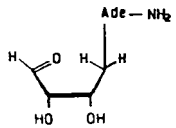
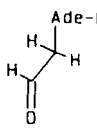
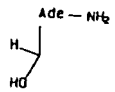
be improved through process development.[18] Possibly a sequence that utilizes 3',5'-O-(1,1,3,3-tetraisopropyldisilox-1,3-diyl)adenosine,[12] analogous to that employed for the synthesis of **4** (Scheme 1) which made possible a high-yield synthesis of triflate **6** using trifluoromethanesulfonic anhydride, would be advantageous.

Evidence of ^{18}O-Incorporation by Mass Spectrometry and by Gas Chromatography - Mass Spectrometry. Each of the ^{18}O-labeled nucleosides **4**, **12**, **16**, and **20** were examined by mass spectrometry (MS) and by gas chromatography - mass spectrometry of their per-O-trimethylsilyl (TMS) derivatives. As the fragmentation pathways for both free nucleosides[19] and their TMS derivatives[20] are well known, such can be used to establish the site of ^{18}O-labeling, as well as the level (i.e., per cent) of incorporation of the label into the molecule.

Examination of the MS of [2'-^{18}O]-adenosine (**4**, Table 1) shows a molecular ion at m/z 267 (0.7%, relative intensity), with an ^{18}O-isotopic peak at m/z 269 of equal intensity, indicating ca. 50% ^{18}O-incorporation. Other features of the spectrum were consonant with a [2'-^{18}O]-adenosine structure: an M - 30 peak at m/z 237 (4.1) and 239 (3.6) (loss of the 5'-CH_2OH group as $H_2C{=}O$),[21] and a strong M - 89 peak at m/z 178 (13) and 180 (14) (corresponds to AdeH-CH_2-CH=O and clearly shows ^{18}O-incorporation at C-2'), as well as the usual peaks characteristic of adenine nucleosides: B + 30 at m/z 164 (78) (Ade-CH_2OH, where the oxygen is from the pentose ether[19]) and the B + 1, B + 2 cluster at m/z 135 (100, base peak) and 136.

Similar results to those with the free nucleoside **4** were obtained when the compound was (trimethylsilyl)ated and the product was examined by GC/MS (Table 2). The fragments common for silylated nucleosides,[20] all showing ca. 50% ^{18}O-enrichment with P + 2 peaks, were observed as follows: a small molecular ion at m/z 555 (0.6) and 557 (0.5), a somewhat stronger M - 15 ion at 540 (3.3) and 542 (3.2) due to loss of Me from a TMS group, the whole glycosyl fragment (M - 221) at m/z 334

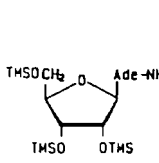
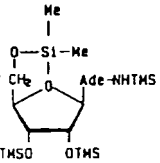
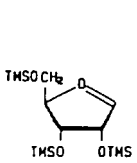
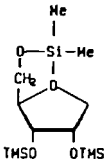
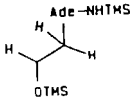
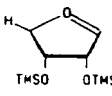
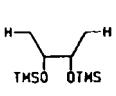
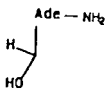
Table 1. Summary of Mass Spectral Data of [2'-^{18}O]- (4) and [3'-^{18}O]-Adenosine (16) and [2'-^{18}O]- (20) and [3'-^{18}O]-Ara-A (12)[a]

Compound[b]	M^+	M-30	M-89	B+30	B+1
					$[Ade-NH_3]^+$
4	269 (0.7) 267 (0.7)	239 (3.6) 237 (4.1)	180 (13.3) 178 (14.5)	164 (77.9)	135 (100)
12	269 (1.1) 267 (1.7)	239 (1.7) 237 (2.3)	180 (0.6) 178 (16.9)	164 (100)	135 (91.7)
16	269 (0.6) 267 (0.9)	239 (3.1) 237 (3.5)	180 (0.6) 178 (19.2)	164 (64.1)	135 (100)
20	269 (1.1) 267 (1.5)	239 (1.8) 237 (1.6)	180 (9.0) 178 (9.3)	164 (100)	135 (90)

a. m/z (relative intensity).

b. Only the D-ribo isomers are illustrated.

Table 2. Summary of Mass Spectral Data of Trimethylsilyl Derivatives of [2'-$^{18}\underline{O}$]- (**4**) and 3'-$^{18}\underline{O}$]-Adenosine (**16**) and [2'-$^{18}\underline{O}$]- (**20**) and [3'-$^{18}\underline{O}$]-Ara-A (**12**)[a]

Compound[b]	M^+	M-15	M-207	M-221	M-233	S-H-$CH_2O_8Me_3$	$C_4H_4O_2(Me_3Si)_2$	B+30	$Si(CH_3)_3$
									
4	557 (0.5)	542 (3.2)	350 (2.2)	336 (1.2)	324 (3.7)	247 (16.0)	232 (33.8)		
	555 (0.6)	540 (3.3)	348 (2.6)	334 (3.0)	322 (5.2)	245 (18.4)	230 (29.4)	236 (69.9)	73 (100)
12	557 (1.5)	542 (5.5)	350 (0.3)	336 (1.0)	324 (0.9)	247 (3.5)	232 (6.2)		
	555 (1.2)	540 (5.9)	348 (0.4)	334 (1.5)	322 (7.0)	245 (5.6)	230 (28.8)	236 (39.8)	73 (100)
16	557 (0.6)	542 (2.9)	350 (2.1)	336 (0.5)	324 (0.8)	247 (9.7)	232 (10.6)		
	555 (0.6)	540 (2.4)	348 (2.0)	334 (1.9)	322 (5.5)	245 (15.2)	230 (41.5)	236 (50.5)	73 (100)
20	557 (0.7)	542 (2.6)	350 (0.4)	336 (0.6)	324 (2.0)	247 (4.0)	232 (29.1)		
	555 (0.6)	540 (2.7)	348 (0.3)	334 (1.5)	322 (2.2)	245 (5.5)	230 (32.7)	236 (55.7)	73 (100)

a. m/z (relative intensity).

b. Only the D-ribo isomers are illustrated.

(3) and 336 (1.2), and, most importantly, a fragment at m/z 322 (5.2) and 324 (3.7) which showed the ^{18}O-incorporation at C-2' (i.e., fragment TMS-Ade-CH_2-CH-OTMS). Other fragments, which are common for TMS nucleosides, are listed in Table 2.

The mass spectra for [3'-^{18}O]-ara-A (**16**) and its per-O-TMS derivative were very similar (Tables 1 and 2) to those for **4**, with one notable exception: Whereas for **4** a strong peak at m/z 178 (13.3) and 180 (14.5) confirmed the incorporation of ^{18}O at C-2', m/z 178 for **16** was strong (19.2%) and m/z 180 was virtually nonexistent (0.6%). That the label in **16** was at C-3', not at C-2', was further substantiated by the MS of its TMS derivative (Table 2). Whereas the M - 233 fragment at m/z 322 (i.e., TMS-Ade-CH_2-CH-OTMS) was moderately strong (5.5%), m/z 324 was greatly diminished (0.8%). (These were of virtually equal intensity in **4**.) All other fragments (Table 2) where C-3' was believed to be included showed substantial to near-equivalent enrichment for the respective P + 2 peak. Deviations such as that observed for m/z 230 and 232 can possibly be attributed to competing MS fragmentation pathways[20] that give rise to ions of equivalent m/z that do not contain C-3'.

Mass spectra of the D-arabino nucleosides **12** and **20** (Table 1 and 2) closely resembled those of their adenosine counterparts **4** and **16**. Thus [2'-^{18}O]-ara-A (**20**) showed a pair of peaks as m/z 178 and 180 of near-equal intensity (Table 1), whereas [3'-^{18}O]-ara-A (**12**) gave rise to m/z 178 (16.9) and 180 (0.6). The mass spectra of the TMS-derivatives showed characteristics similar to their adenosine counterparts in that the C-2' ^{18}O label was easily recognized in **20** [m/z 322 (2.2) and 324 (2.0)], while the C-3' ^{18}O label was shown to be present in all major ions except m/z 324.

As TMS-ara-A and TMS-adenosine are conveniently separated by GC (T_R = 10.4 and 11.3 min, respectively), the ^{18}O-labeled compounds should prove invaluable as biochemical markers. It is anticipated that these [2'-^{18}O]-labeled compounds, in particular, with their peak pair at m/z

178 and 180 (or m/z 322 and 324 for their TMS derivatives) should serve for identification of products via MS or mass fragmentography in the biosynthesis of ara-A.

The ^{18}O-Isotope Effect in the ^{13}C NMR Spectroscopy of [2'-^{18}O]- (4) and [3'-^{18}O]-adenosine (16). Since the first observed ^{18}O-isotope effect on ^{13}C NMR chemical shifts was documented,[22] a flurry of activity in the area has resulted in structural and stereochemical studies,[23 - 25] in mechanistic studies,[26] as well as in the use of the effect to determine the biochemical origin of certain carbon and oxygen atoms in natural products.[27 - 29] The use of this technique has not been extended to nucleosides, the sole paper in the carbohydrate field being a mechanistic study of a monosaccharide rearrangement.[26] The field has also been recently reviewed.[5]

Our interest in this NMR phenomenon relates to elucidating the mechanism for the biosynthetic conversion of adenosine to ara-A. Our aim was to develop a technique as an adjunct to MS which could identify the position and stereochemistry of ^{18}O-incorporation in the biosynthesis of ara-A[3,4] from adenosine. It was envisioned that experiments using either ^{18}O-labeled adenosine, or ^{18}O-water and unlabeled adenosine, with Streptomyces antibioticus or the purified "adenosine epimerase"[4] could provide an ^{18}O-labeled ara-A which would be useful to elucidate the mode of epimerization.

To this end, both [2'-^{18}O]- (**4**) and [3'-^{18}O]-adenosine (**16**) were examined for ^{18}O-induced ^{13}C NMR shifts. The results are shown in Figure 1. The observed shift for C-2' of **4** was 1.19 Hz upfield, while that for the C-3' of (**16**) was 1.60 Hz upfield. To date the same experiments on the labeled ara-A's **12** and **20** have not been carried out due to difficulties encountered in solubilizing the products in deuterium oxide.

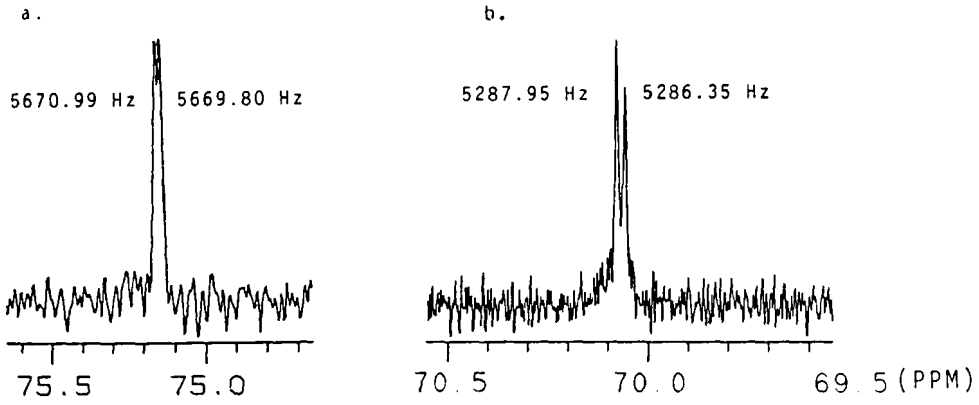


Fig. 1. ^{18}O-Induced ^{13}C NMR Chemical Shifts for [2'-^{18}O]-Adenosine (**4**) and [3'-^{18}O]-Adenosine (**16**). a. 2'-C for **4** and b. 3'-C for **16**.

EXPERIMENTAL

General Methods. Solvents were evaporated, except where otherwise noted, at aspirator vacuum at ~40 °C. Melting points were determined using a Thomas-Hoover "Unimelt" capillary melting point apparatus equipped with a Cole-Parmer model 8520-50 Digi-sense digital thermometer/8520-55 thermocouple combination that was calibrated with known standards. Ultraviolet (UV) spectra were recorded in 1-cm cells on a Varian DMS-100 UV/VIS spectrophotometer. Infrared (IR) spectra were recorded on a Perkin-Elmer 710B spectrophotometer. Mass spectrometry (MS) and gas chromatography - mass spectrometry (GC/MS) were carried out on a Hewlett-Packard 5840A GC unit equipped with a DB-5 (Durabond) fused silica capillary column (film thickness 0.25 μm, 30 m x 0.32 mm). Unless otherwise stated, temperature was isothermal at 225 °C with a helium carrier gas flowrate of 2 mL min^{-1}.

^{1}H NMR spectra were determined at 200 MHz using a Nicolet NT-200 instrument. Chemical shifts are reported in δ-units downfield from an internal standard of tetramethylsilane; multiplicities are first-order values (in Hz) and are indicated: d, doublet; s, singlet; t, triplet; m,

multiplet; ψt = a "pseudo" triplet, i.e., a dd with nearly equivalent J-values. Solutions were typically ~0.1% for ^{1}H NMR.

^{13}C NMR spectra were determined on a Nicolet NT-300 NMR spectrometer, operating in the Fourier transform mode, at 75.5 MHz. In a typical experiment for [2'-^{18}O]-adenosine (**4**), the proton-decoupled spectrum of a 20-mg sample, contained in 3 mL of deuterium oxide in a 10-mm tube, was measured with the following parameters: 2420 scans, 5 kHz sweep width, 4.0-sec post-acquisition delay time, 90° pulse angle, with a total of 32,768 k data points. The same procedure was used for [3'-^{18}O]-adenosine (**16**), except that 2816 total scans were used, with a post-acquisition delay time of 2.7 sec and 131,072 k data points.

Adsorption chromatography was carried out using E. Merck Silica Gel-60 products: (a) TLC on 0.2 mm aluminum-backed plates (catalog no. 5760); (b) open-column chromatography using 230 - 400 μm silica gel (catalog no. 7734): A, 80:20 ethyl acetate - petroleum ether (bp 30 - 60 °C); B, 90:10 chloroform - methanol; C, 95:5 chloroform - methanol; and D, 97:3 chloroform - methanol. Adsorption chromatography and desalting of free nucleosides was carried out on Diaion HP-20 resin (Mitsubishi Chemical Co., Ltd.), a porous polystyrene resin devoid of functional groups.

Preparation of [^{18}O]-propionic acid. The synthesis of [^{18}O]-propionic acid followed the procedure given by Gatenbeck et al.[30] To 1.85 g (1.74 mL, 20 mmol) of distilled propionyl chloride maintained under nitrogen with ice-bath cooling, 360 mg (0.36 mL, 20 mmol) of [^{18}O]-water was slowly added. The mixture was stirred at 0 °C for 30 min and at room temperature for 1 h. The light-yellow liquid which resulted was distilled to give 1.2 g (81.5%) of a colorless product: bp 67 °C (18 torr).

Preparation of cesium [^{18}O]-propionate. Using the published procedure for the preparation of cesium propionate,[31] 1.1 g (15 mmol) of

[^{18}O]-propionic acid was converted to 1.5 g of cesium [^{18}O] propionate, mp 158 - 160 °C.

9-[3',5'-Di-O-(tert-butyldimethylsilyl)-2'-O-(trifluoromethanesulfonyl)-β-D-arabinofuranosyl]adenine (2). To a stirred solution of 1.1 g (2.2 mmol) of 9-[3',5'-di-O-(tert-butyldimethylsilyl)-β-D-arabinofuranosyl]adenine (**1**)[7,8] in 25 mL of dry THF at 0 °C was added 106 mg (4.4 mmol) of sodium hydride. The mixture was stirred at 0 °C for 10 min and was warmed to room temperature to stir for 0.5 h until the solution became clear. The solution was then cooled to -70 °C, and 0.47 mL (4.22 mmol) of trifluoromethanesulfonyl chloride was added under nitrogen. The mixture was stirred at -70 °C under nitrogen for 1 h, at the end of which time an additional 0.2 mL of trifluoromethanesulfonyl chloride was added, and the reaction was allowed to proceed for an additional 1 h. At the end of this time, the mixture was poured into 20 mL of cold sodium bicarbonate solution, and the mixture was extracted with 2 x 50 mL of dichloromethane. The combined extracts were dried over magnesium sulfate. Evaporation of the solvent gave a white glass which was purified by column chromatography (75 g of silica gel and solvent E) to give 600 mg (43.4%) of pure product 2 as a glass: R_f = 0.42 (A); UV (CH_2Cl_2) 255 nm; ^{1}H NMR ($CDCl_3$) δ 0.11, 0.18 (12H, s, s, Me_2Si), 0.93, 0.94 (18H, s, s; Me_3C), 3.89 [2H, m (width = 35 Hz), H-5', 5'$_a$], 4.05 [1H, m (width = 17 Hz), H-4'], 4.79 (1H, ψt, $J_{3',4'}$ = 2 Hz, H-3'), 5.21 (1H, ψt, $J_{2',3'}$ = 3.5 Hz, H-2'), 5.76 [2H, bs, NH_2 (exchangeable with D_2O)], 6.60 (1H, d, $J_{1',2'}$ = 2.1 Hz, H-1'), 8.02, and 8.38 (s, s; 1H, 1H; H-2, H-8).

Anal. Calcd. for $C_{23}H_{40}F_3N_5O_6SSi_2$: C, 44.00; H, 6.37; N, 11.16; S, 5.19. Found: C, 43.98; H, 6.42; N, 11.10; S, 5.05.

[2'-^{18}O]-3',5'-Di-O-(tert-butyldimethylsilyl)-2'-O-(1-propionyl)-adenosine (3). To a stirred solution of 400 mg (0.64 mmol) of **2** in 5 mL of dry DMF at room temperature was added 197 mg (0.96 mmol) of cesium [^{18}O]-propionate. The mixture was stirred at room temperature for 2 h,

at the end of which time the DMF was evaporated at ca. 45 °C (ca. 1 torr), and the residue was chromatographed over silica gel with solvent A to give 300 mg (85.3%) of pure **3** as a white glass: R_f = 0.28 (A); UV (CH_2Cl_2) 256 nm; IR (KBr) 1600 (C=N) and 1700 cm^{-1} (C=O); ^1H NMR ($CDCl_3$ - D_2O) δ 0.08, 0.09, 0.11 (12H, 3 s's, Me_2Si), 0.91, 0.92 (18H, 2 s's, Me_3C), 1.73 (3H, t, $CH_3CH_2C=O$), 2.42 (2H, q, $CH_3CH_2C=O$), 3.87 (2H, ddd, $J_{5',5'a}$ = 11.5 Hz, H-5', H-5'a), 4.12 (1H, dd, $J_{4',5'a}$ = 2.7 Hz, $J_{4',5'}$ = 6.2 Hz, H-4'), 4.71 (1H, ψt, $J_{3',4'}$ = 4.2 Hz, H-3'), 5.62 (1H, ψt, $J_{2',3'}$ = 5.0 Hz, H-2'), 6.24 (1H, d, $J_{1',2'}$ = 5.3 Hz, H-1'), 8.14, and 8.35 (1H, 1H, s, s, H-2, H-8).

Anal. Calcd for $C_{25}H_{45}N_5O_5Si_2$: C, 54.26; H, 8.20; N, 12.67. Found: C, 54.26; H, 8.20; N, 12.62.

[2'-^{18}O]-Adenosine (4). To a solution of 200 mg (0.36 mmol) of **3** in 4 mL of dioxane was added 4 mL of 1 M aqueous sodium hydroxide. The mixture was stirred at room temperature overnight, at the end of which time 2 mL of 5% (v/v) aqueous hydrochloric acid was added to render the solution pH 8-9. The solvent was evaporated under vacuum to give a white solid that was dissolved in 3 mL of distilled water and loaded onto a column of Diaion HP-20 resin. The column was eluted with 100 mL of water, followed by 200 mL of 90:10 water - methanol to give 86 mg (89%) of pure **4** as a white solid: mp = 234 - 236 °C (lit.[32] 234 - 235 °C); ^1H NMR (D_2O) was identical with that of authentic adenosine; for MS data, see Table 1; for GC/MS data of TMS derivative see Table 2.

9-[3',5'-O-(1,1,3,3-Tetraisopropyldisilyl)-2'-O-(trifluoromethanesulfonyl)-β-D-arabinofuranosyl]adenine (6). By the same procedure used for the synthesis of **14** (vide infra), 800 mg (1.57 mmol) of **5**[12] was converted to 755 mg (75%) of **6**, isolated as a white glass: ^1H NMR of **6** was identical with that of **14**.

[2'-^{18}O]-3',5'-O-(1,1,3,3-Tetraisopropyldisilyl)-2'-O-(1-propionyl)adenosine (7). By the same procedure used for **15**, 600 mg (0.94 mmol) of **6** was converted, using 230 mg (1.13 mmol) of cesium [^{18}O]-propionate,

to 480 mg (89%) of **7**, isolated as a white solid: mp and ^{1}H NMR spectrum were identical with those of **15**.

[2'-^{18}O]-adenosine (4) from 7. By the same prodecure used for preparation of **16**, 400 mg (0.7 mmol) of **7** was deprotected to give 120 mg (64%) of **4**.

9-[2',5'-Di-O-(tert-butyldimethylsilyl)-β-D-xylofuranosyl]adenine (9). Using the procedure reported by Robins et al., 1.0 g (2.0 mmol) of 9-[2',5'-di-O-(tert-butyldimethylsilyl)-β-D-erythro-pentofuran-3-ulosyl]adenine (**8**)[33] was converted to 0.1 g (70%) of **9** as white glass: R_f = 0.23 (A); UV (MeOH) 258 nm; ^{1}H NMR (DMSO-d_6) δ -0.07, -0.05, -0.03 (12H, s, s, s, Me_2Si), 0.77, 0.82, 0.85 (18H, s, s, s, Me_3C), 3.88 [2H, m (width = 24 Hz), H-5',5a'], 4.00 [1H, m (width = 13 Hz), H-4'], 4.11 (1H, ψt, $J_{3,4}$ = 4.5 Hz), 4.39 (1H, s, H-2'), 5.82 (1H, s, H-1'), 8.11, and 8.21 (1H, 1H, s, s, H-2, H-8).

Anal. Calcd. for $C_{22}H_{29}N_5O_4Si_2 \cdot 0.3\ H_2O$: C, 52.72; H, 8.37; N, 13.98. Found: C, 52.65; H, 8.38; N, 13.92.

9-(5-O-Acetyl-2,3-anhydro-β-D-lyxofuranosyl)adenine (11). To 2.8 g (1.12 mmol) of 9-(2,3-anhydro-β-D-lyxofuranosyl)adenine (**10**)[16] in 40 mL of pyridine was added 1.0 mL of acetic anhydride at 0 °C. The mixture was stirred at 0 °C for 1 h, then at room temperature for an additional 5 h, at the end of which time all the solid material had dissolved. The solvent was evaporated, and two, 100-mL portions of toluene were sequentially added to the residue and evaporated to give a light yellow syrup. The syrup was stirred with 100 mL of ether for 30 min, resulting in the formation of 3.1 g (95%) of a white precipitate that was sufficiently pure for the next reaction: R_f = 0.32 (B); mp 187 - 189 °C; UV (CH_3OH) = 258 nm; ^{1}H NMR (DMSO-d_6) δ 2.05 (3H, s, Ac), 4.18 - 4.39 (5H, m, H-2', H-3', H-4', H-5', H-5'$_a$), 6.30 (1H, s, $J_{1',2'}$ = 0, H-1'), 7.39 (2H, s, NH_2), 8.17 and 8.18 (1H, 1H, s, s, H-2, H-8).

Anal. Calcd. for $C_{12}H_{13}N_5O_4$: C, 49.49; H, 4.50, N, 24.04. Found: C, 49.41; H, 4.52; N, 24.01.

[3'-^{18}O]-9-(β-D-Arabinofuranosyl)adenine (12). To 2.0 g (6.87 mmol) of **11** in 50 mL of dry DMF was added 1.84 g (8.81 mmol) of cesium [^{18}O]-propionate. The mixture was stirred at 100 °C under nitrogen for 4 h, at the end of which time the DMF was evaporated to give a yellow syrup to which was added 40 mL of methanol and 4 mL of concentrated ammonium hydroxide. The mixture was stirred at room temperature for 0.5 h, at the end of which time TLC showed the absence of starting material, with the major component having the same R_f as ara-A. The reagents were evaporated to give a yellow syrup to which were added 40 mL of ethyl acetate and 50 mL of water. The layers were separated, and the organic layer was washed with 40 mL of water. The combined aqueous layers were lyophilized to reduce the volume to ca. 30 mL, and a yellow precipitate was collected by filtration. The yellow solid was recrystallized from water to give 600 mg of pure **12** as a white solid. The combined mother liquors were concentrated to 3 mL and passed through a column of HP-20 resin (40 mL of HP-20 resin, 100 mL of water, followed by 200 mL of 90:10 water - methanol) to give an additional 380 mg of **12** (54%, total yield): mp = 255 - 257 °C (dec) (lit.[34] 257 - 257.5 °C); for MS data and GC/MS data on the per-O-trimethylsilyl ether, see Tables 1 and 2, respectively; ^{1}H NMR data (DMSO-d_6) were identical with those for authentic ara-A.

[3'-^{18}O]-9-[3',5'-O-(1,1,3,3-Tetraisopropyldisylyl)-β-D-arabinofuranosyl]adenine (13). To 900 mg (3.37 mmol) of **12** in 30 mL of dry pyridine was added 1.1 mL (1.1 g, 3.50 mmol) of 1,3-dichloro-1,1,3,3-tetraisopropyldisiloxane (Aldrich) under nitrogen at room temperature. The mixture was stirred at room temperature for 12 h, at the end of which time all solid material had dissolved. The solvent was removed, and two, 150-mL portions of toluene were sequentially added to the residue and evaporated to give a yellow syrup which was purified by column chromatography (80 g of silica gel, solvent D) to give 1.5 g (87%) of pure **13** which formed a white glass under high vacuum: R_f =

0.20 (C); mp = 115 - 117 °C; ^{1}H NMR spectrum (DMSO-d_6) was identical with that reported for the [^{16}O]-analogue of **13**.[12]

[3'-^{18}O]-9-[3',5'-O-(1,1,3,3-Tetraisopropyldisilyl)-2'-O-(trifluoromethanesulfonyl)-β-D-arabinofuranosyl]adenine (14). To 1.2 g (2.35 mmol) of 13 and 934 mg (8.32 mmol) of 4-(N,N-dimethylamino)pyridine in 60 mL of dry dichloromethane was added 1.6 mL (1.56 g, 19.8 mmol) of pyridine. The mixture was cooled to 0 °C, and 0.65 mL (1.09 g, 3.86 mmol) of trifluoromethanesulfonic anhydride was added over a period of 10 min. After stirring at 0 °C for 2 h, the mixture was poured into 50 mL of cold, saturated aqueous sodium bicarbonate, the two layers were separated, and the aqueous layer was extracted with 2 x 50-mL of dichloromethane. The combined organic layers were dried over magnesium sulfate and evaporated to give a syrup which was purified by column chromatography (70 g of silica gel, with chloroform as the eluant) to provide 1.08 g (71%) of **14** as a white glass: R_f = 0.39 (C); mp = 86 - 88 °C; ^{1}H NMR ($CDCl_3$) δ 0.99 - 1.19 (24H, m, Me_2CH), 3.96 (1H, m, H-4'), 4.15 (2H, m, $J_{4',5'}$ = 6.4 Hz, $J_{5',5'_a}$ = 12.3 Hz, H-5', H-5'$_a$), 5.40 (1H, t, $J_{3',4'}$ = 6.9 Hz, H-3'), 5.48 (1H, t, $J_{2',3'}$ = 6.5 Hz, H-2'), 5.62 [2H, s, NH_2 (exchangeable with D_2O)], 6.39 (1H, d, $J_{1',2'}$ = 5.8 Hz, H-1'), 7.93, and 8.33 (1H, 1H, s, s, H-2, H-8).

Anal. Calcd. for $C_{23}H_{38}F_3N_5O_7SSi_2$: C, 43.05; H, 5.97; N, 10.91; S, 5.00. Found: C, 43.00; H, 6.02; N, 10.86; S, 5.02.

[3'-^{18}O]-3',5'-O-(1,1,3,3-Tetraisopropyldisyl)-2'-O-(1-propionyl)adenosine (15). To a sirred solution of 800 mg (1.24 mmol) of **14** in 2.5 mL of dry DMF at room temperature was added 338 mg (1.64 mmol) of cesium propionate.[31] The mixture was stirred at room temperature for 4 h, at the end of which time the DMF was evaporated (ca. 45 °C and 1 torr) to give a white solid that was dissolved in 50 mL of dichloromethane, with clarification of the solution by filtration. The solvent was evaporated, and the residue was purified by column chromatography (50 g silica gel, solvent D) to provide 640 mg (91%) of

15 as a white solid: R_f = 0.34 (C); mp = 166 - 168 °C; UV (CH_2Cl_2) 256 nm; IR (KBr) 1600 (C=N), 1720 cm^{-1} (C=O); ^1H NMR ($CDCl_3$) δ 0.90 - 1.15 (28H, m, $\underline{Me}_2CH$), 1.19 (3H, t, $C\underline{H}_3CH_2CO_2$), 2.46 (2H, q, $CH_3C\underline{H}_2CO_2$), 3.98 - 4.21 (3H, m, H-4', H-5', H-5'$_a$), 5.07 (1H, dd, $J_{3',4'}$ = 8.5 H, H-3'), 5.51 [2H, s, NH_2 (exchangeable with D_2O)], 5.79 (1H, d, $J_{2',3'}$ = 5.3 Hz, H-2'), 6.03 (1H, s, $J_{1',2'}$ = 0 Hz, H-1'), 8.02, and 8.31 (1H, 1H, s, s, H-2, H-8).

Anal. Calcd. for $C_{25}H_{43}N_5O_6Si_2$: C, 53.06; H, 7.66; N, 12.38. Found: C, 52.96; H, 7.70; N, 12.32.

[3'-^{18}O]-Adenosine (16). To a solution of 550 mg (0.97 mmol) of **15** in 20 mL of THF was added 1.94 mL (2 equiv) of 1 M tetrabutylammonium fluoride in THF (Aldrich). The mixture was heated under reflux for 2 h at which time TLC showed absence of starting material. The mixture was cooled to room temperature, and 3 mL of concentrated ammonium hydroxide was added. The mixture was then stirred at room temperature for 2h, followed by heating under reflux for an additional 1 h. At the end of this time, TLC showed only one product, R_f = 0.16 (D). The solvent was evaporated to give a yellow syrup that was dissolved in 30 mL of water and equilibrated with 20 mL of dichloromethane. The two layers were separated, the organic layer was washed with 20 mL of water, and the combined aqueous layers were evaporated at ca. 1 torr to give a yellow syrup which was purified by passage over an HP-20 column (300 mL of water, followed by 200 mL of 90:10 water - methanol). The combined UV-active fractions were pooled and evaporated at ca. 1 torr to give a syrup which was crystallized from methanol to give 160 mg of pure **16**. A second crop of 25 mg was obtained after concentration of the mother liquors (total yield = 71%): mp = 232 - 234 °C (lit.[32] 234 - 235 °C for adenosine); for MS data, see Table 1; for GC/MS data of the per-O-trimethylsilyl derivative, see Table 2; ^1H NMR data (D_2O) were identical with those for authentic adenosine.

3',5'-Di-O-(tert-butyldimethylsilyl)-2'-O-(trifluoromethanesul-

fonyl)adenosine (18). By the same procedure used for the preparation of **2**, 0.92 g (1.86 mmol) of 3',5'-di-O-(tert-butyldimethylsilyl)adenosine (**17**)[35] was converted to 350 mg (30%) of glassy **18**: R_f = 0.44 (A); UV (CH_2Cl_2) 257 nm; ^{1}H NMR ($CDCl_3$) δ 0.07, 0.10, 0.17, 0.20 (12H, 4 s's, Me_2Si), 0.86, 0.96 (18H, s, s, Me_3C), 3.96 [2H, m (AB of ABX), H-5', $5'_a$], 4.16 [1H, m (width = 9.2 Hz), H-4'], 4.87 (1H, ψt, $J_{3',4'}$ = 4.7 Hz, H-3'), 5.78 [2H, s, NH_2 (exchangeable with D_2O)], 5.84 (1H, ψt, $J_{2',3'}$ = 4.5 Hz, H-2'), 6.29 (1H, d, $J_{1',2'}$ = 4.3 Hz, H-1'), 8.08, and 8.34 (1H, 1H, 2 s's, H-2, H-8).

Anal. Calcd. for $C_{23}H_{40}F_3O_6SSi_2$: C, 44.00; H, 6.37; N, 11.16; S, 5.19. Found: C, 44.10; H, 6.40; N, 11.15; S, 5.11.

[2'-^{18}O]-9-[3',5'-Di-O-(tert-butyldimethylsilyl)-2'-O-(1-propionyl)-β-D-arabinofuranosyl]adenine (19). By the procedure used for **3**, 200 mg (0.32 mmol) of **18** was converted to 150 mg (88.7%) of **19**, isolated as a white solid: R_f = 0.28 (A); UV (CH_2Cl_2) 256 nm; IR (KBr) 1600 (C=N), 1700 cm^{-1} (C=O); ^{1}H NMR ($CDCl_3$) δ 0.09, 0.12 (12H, s, s, Me_2Si), 0.83 (3H, t, CH_3CH_2C=O), 0.90, 0.95 (18H, s, s, Me_3C), 2.02 (2H, m, CH_3CH_2C=O), 3.93 [3H, m (width = 19.6 Hz), H-4', H-5', H-$5'_a$], 4.66 (1H, ψt, $J_{3',4'}$ = 4.6 Hz, H-3'), 5.40 (1H, ψt, $J_{2',3'}$ = 5.6 Hz, H-2'), 5.60 [2H, s, NH_2 (exchangeable with D_2O)], 6.56 (1H, d, $J_{1',2'}$ = 5.4 Hz, H-1'), 8.16 and 8.34 (1H, 1H, s, s, H-2, H-8).

Anal. Calcd. for $C_{25}H_{45}N_5O_5Si_2$: C, 54.26; H, 8.20; N, 12.67. Found: C, 54.17; H, 8.22; N, 12.62.

[2'-^{18}O]-9-β-D-Arabinofuranosyladenine (20). By the procedure used for preparation of **4**, 100 mg (0.18 mmol) of **19** was converted to 38 mg (78.5%) of **20**, isolated as a white solid: mp = 255 - 256 °C (dec) (lit.[34] 257 - 257.5 °C); ^{1}H NMR spectrum (DMSO-d_6) was identical with that of authentic 9-(β-D-arabinofuranosyl)adenine; for MS data, see Table 1; for GC/MS data on the per-O-trimethylsilyl ether, see Table 2.

***Preparation of trimethylsilyl derivatives of* [2'-^{18}O]- (4) and [3'-^{18}O]-adenosine (16), and [2'-^{18}O]- (20) and [3'-^{18}O]-9-(β-D-arabino-**

furanosyl)adenine (12). To 2 mg of the nucleoside in a 0.3-mL Reacti-Vial™ outfitted with a screw cap lined with Teflon™ was added 0.2 mL of dry acetonitrile and 0.1 mL of 1-(trimethylsilyl)imidazole (Aldrich). The mixture was allowed to stand overnight at room temperature, then it was heated to 60 °C for 1 h prior to GC/MS analysis.

In a typical GC experiment, per-O-trimethylsilyl-ara-A showed a retention time (T_R) of 10.4 min, while per-O-trimethylsilyladenosine was retained 11.3 min.

ACKNOWLEDGMENTS

This work was supported, in part, by grant no. AI-22296 from the National Institutes of Health. Anthony S. Serianni and Paul C. Kline of the Department of Chemistry, University of Notre Dame, are gratefully acknowledged for carrying out the ^{18}O-induced ^{13}C NMR shift studies.

REFERENCES

1. Hanvey, J. C.; Hardman, J. K.; Suhadolnik, R. J.; Baker, D. C. Biochemistry **1984**, 23, 904.

2. Hanvey, J. C.; Hawkins, E. S.; Tunac, J. B.; Dechter, J. J.; Baker, D. C.; Suhadolnik, R. J. Biochemistry **1987**, 26, in press.

3. Farmer, P. B.; Suhadolnik, R. J. Biochemistry **1972**, 11, 911.

4. Suhadolnik, R. J.; Wu, J. M.; Anderson, M. M.; von Minden, D.; McCloskey, J. A. Biochemistry **1987**, 26, submitted.

5. See, for example, Van Etten In Synthesis and Application of Isotopically Labeled Compounds, Proceedings of International Symposium; Muccino, R. R.; Ed.; Elsevier: Amsterdam, 1985; pp. 257 - 262, and references cited therein.

6. Both [2'-^{18}O]- and [3'-^{18}O]-adenosine have been synthesized by a lengthy process of $H_2{}^{18}O$ exchange with keto sugars, followed by nucleoside synthesis [Follman, H.; Hogenkamp, H. P. C. J. Am. Chem. Soc. **1970**, 92, 671. This process of ^{18}O exchange could be adapted to 2'- and 3'-keto nucleosides; however, the reduction of the keto nucleosides would not provide acceptable yields of D-ribo compounds.

7. Crews, R. P. Ph. D. Dissertation, University of Alabama, 1983.

8. Ogilvie, K. K.; McGee, D. P. C.; Boisvert, S. M.; Hakimelahi, G. H.; Proba, Z. A. Can. J. Chem. **1983**, 61, 1204.

9. Markiewicz, W. T.; Wiewiorowski, M. Nucleic Acids Symp. Ser. **1978**, No. 4, s185.

10. Markiewicz, W. T. J. Chem. Soc. Res. Symp. **1979** 24; J. Chem. Res. Miniprint. **1979**, 0181.

11. Markiewicz, W. T.; Padyukova, N. Sh.; Zdenek, S.; Smrt, J. Collect. Czech. Chem. Commun. **1980**, 45, 1860.

12. Robins, M. J.; Wilson, J. S.; Sawyer, L.; James, M. N. G. Can. J. Chem. **1983**, 61, 1911.

13. Binkley, R. W.; Ambrose, M. G. J. Carbohydr. Chem. **1984**, 3, 1.

14. Lee, W. W.; Benitez, A.; Goodman, L.; Baker, B. R. J. Am. Chem. Soc. **1980**, 82, 2648.

15. Mitsunobu, O. Synthesis **1981**, 1.

16. Mengel, R.; Bartke, M. Angew. Chem. Int. Ed. Engl. **1978**, 17, 679.

17. Ranganathan, R.; Larwood, D. Tetrahedron Lett. **1978**, 4341.

18. Ref. 17 cites a 70% yield of the 2'-O-(triflyl)derivative from 3',5'-di-O-(tetrahydropyranyl)adenosine. It is likely such success can also be achieved with **18**.

19. Shaw, S. J.; Desiderio, D. M.; Tsuboyama, K.; McCloskey, J. A. J. Am. Chem. Soc. **1970**, 92, 2510.

20. Pang. H.; Schram, K. H.; Smith, D. L.; Gupta, S. P.; Townsend, L. B.; McCloskey, J. A. J. Org. Chem. **1982**, 47, 3823.

21. The origin of the M - 30 peak has been generally attributed (ref. 19) to loss of 5'-CH_2OH as $H_2C{=}O$. This fact was firmly established through 5'-deuterium labeling by Baker, D. C.; Horton, D. Carbohydr. Res. **1972**, 21, 393.

22. Risley, J. M.; Van Etten, R. L. J. Am. Chem. Soc. **1979**, 101, 252.

23. Risley, J. M.; Van Etten, R. L. J. Am. Chem. Soc. **1980**, 102, 4609.

24. Risley, J. M.; Van Etten, R. L. J. Am. Chem. Soc. **1980**, 102, 6699.

25. Necleras, J. C. J. Am. Chem. Soc. **1980**, 102, 374.

26. Dessinges, A.; Castillon, S.; Olesker, A.; Thang, T. T.; Lukas, S. J. Am. Chem. Soc. **1984**, 106, 450.

27. Hutchinson, C. R.; Kurobane, I.; Cane, D. E.; Hasler, H.; McInnes, A. G. J. Am. Chem. Soc. **1981**, 103, 2477.

28. Cane, D. E.; Liang, T.-C.; Hasler, H. J. Am. Chem. Soc. **1982**, 104, 7274.

29. Cane, D. E.; Hasler, H.; Taylor, P. B.; Liang, T.-C. Tetrahedron **1983**, 39, 3449.

30. Gatenbeck, S.; Mosback, K. Acta. Chem. Scand. **1959**, 13, 1561. It is noteworthy that this process results in the incorporation of only one ^{18}O atom per molecule of propionic acid. Thus the nucleoside product is expected to show only 50% ^{18}O incorporation, due to the equivalent nature of the two oxygens of the cesium salt reagent (ref. 31) used in the triflate displacement.

31. Kruizinga, W. H.; Strijtveen, B.; Kellogg, R. M. J. Org. Chem. **1981**, 46, 4323.

32. The Merck Index, 10th ed.; Windholz, M. Ed.; Merck: Rahway, NJ, 1983; p. 23.

33. Hansske, F.; Madej, D.; Robins, M. J. Tetrahedron **1984**, 40, 125.

34. The Merck Index, 10th ed.; Windholz, M. Ed.; Merck: Rahway, NJ, 1983; p. 1426.

35. Ogilvie, K. K.; Beaucage, S. L.; Schifman, A. L.; Can. J. Chem. **1978**, 56, 2768.

Received June 8, 1987