Chemoenzymatic Synthesis of 4-Substituted Riboses. S-(4′-Methyladenosyl)-L-homocysteine†

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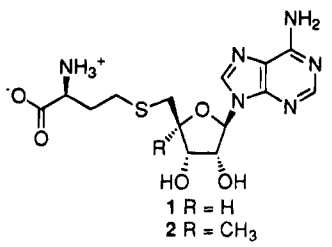
Summary: The synthesis of 4-*C*-methyl-D-ribose, 4-*C*-phenyl-D-ribose, D-ribose-*4-d*, and L-ribose derivatives as well as the title nucleoside by a chemoenzymatic strategy beginning from cyclopentadiene is described.

The level of *S*-adenosyl-L-homocysteine (AdoHcy) (**1**), a natural feedback inhibitor of *S*-adenosyl-L-methionine-dependent transmethylases, is controlled by AdoHcy hydrolase. The latter degrades AdoHcy by a NAD^+-dependent process involving (i) oxidation of the 3′-hydroxyl, (ii) β-elimination of homocysteine, (iii) addition of water to the resulting enone, and (iv) re-reduction of the 3′-keto group to produce adenosine.[1]



1 R = H
2 R = CH_3

Compounds which interfere with the transmethylation process have potential for therapeutic intervention in the viral replication process.[2] Among possible mechanism-based inhibitors of interest to us is the AdoHcy analog **2** in which the H at the 4′-ribose position is replaced by a methyl group. Such a compound could not participate in the elimination step of the Abeles–Palmer pathway described above and thus may function directly or indirectly as an inhibitor of transmethylations. The implementation of this strategy required a convenient route to 4-substituted riboses either by manipulation of natural carbohydrates or by direct chemoenzymatic synthesis. As the former process presents a formidable task in the stereocontrolled introduction of a substitutent at the configurational center,[3,4] we chose to develop a de novo chemoenzymatic strategy of 4-substituted riboses that would allow access to both the D and L series. We present here an efficient, stereoselective strategy to 4-substituted riboses involving the readily available enantiopure carbocyclic precursor **3** and its application to the synthesis of *S*-(4′-methyladenosyl)-L-homocysteine (**2**).

Syntheses of enantiopure enone **3** have been developed in this and other laboratories.[5] For the present purpose we prepared enone **3** from cyclopentadiene by a process involving the use of the very inexpensive crude porcine pancreatic lipase.[6] The transformations of enone (+)-**3**, $[\alpha]^{25}_D$ +66.3 (*c* 1.0, $CHCl_3$), into 4-methylribose is illustrated in Scheme 1. α-Bromination of the enone was nearly quantitative. Excellent diastereofacial selectivity (>20:1 by NMR) was achieved in the 1,2-addition of methylmagnesium bromide to **4** at −78 °C to produce the alcohol **5**. The ribose framework was unveiled from this carbocyclic precursor by ozonolysis of the vinyl bromide unit in methanol/pyridine followed by reductive workup; **6** was isolated in 91% yield as a single anomer. The intermediate acid bromide was converted in situ to the methyl ester during ozonolysis. Refluxing **6** with MeOH/acetone/H^+ gave the corresponding methyl glycosides (22:1, β:α),[7] and subsequent reduction of the methyl ester with lithium borohydride gave the protected β-methyl 4-*C*-methylribofuranoside (**7**)[8] in 74% overall yield from **3**.

The condensation of **7** and *N,N′*- bis(trifluoroacetyl)-L-homocystine, dimethyl ester in the presence of trialkylphosphines[9] proved unsuccessful, apparently due to the steric hindrance imparted by the 4-methyl group. The incorporation of the amino acid side chain was accomplished in good yield by formation of the 5-triflate and subsequent displacement with a suitably protected homocysteine in DMF. Initial studies utilized *N,N*-diallylhomocysteine, methyl ester and *N,N*-dibenzy-homocysteine, methyl ester; however, neither the allyl nor benzyl amino protecting groups could be removed efficiently at the end of the synthesis. The coupling of *N*-(trifluoroacetyl)-L-homocysteine, methyl ester[10] with the 5-triflate was quantitative and proceeded without epimerization to yield **8**. The acetonide was removed by

† Dedicated to Prof. J. Bryan Jones on the occasion of his 60th birthday.

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(2) A direct correlation between antiviral potency of adenine analogs and their inhibition of AdoHcy hydrolase has been demonstrated (De Clercq, E.; Cools, M. *Biochem. Biophys. Res. Commun.* **1985**, *129*, 306. Keller, B. T.; Borchardt, R. T. *Mol. Pharmacol.* **1987**, *31*, 485). For recent reviews see: Wolfe, M. S.; Borchardt, R. T. *J. Med. Chem.* **1991**, *34(5)*, 1521. Liu, S.; Wolfe, M. S.; Borchardt, R. T. *Antiviral Res.* **1992**, *19*, 247.

(3) Yoshimura, J. *Adv. Carbohydr. Chem. Biochem.* **1984**, *42*, 69–133. Gough, B. M.; Gunner, S. W.; Overend, W. G. *Carbohydr. Res.* **1970**, *14*, 173.

(4) De Voss, J. J.; Hangeland, J. J.; Townsend, C. A. *J. Org. Chem.* **1994**, *59*, 2715.

(5) (a) Johnson, C. R.; Penning, T. D. *J. Am. Chem. Soc.* **1988**, *110*, 4726. (b) Hudlicky, T.; Natchus, M. G.; Nugent, T. C. *Synth. Commun.* **1992**, *22*, 151. (c) Siddiqi, S. M.; Schneller, S. W.; Ikeda, S.; Snoeck, R.; Andrei, G.; Balzarini, J.; De Clercq, E. *Nucleosides Nucleotides* **1993**, *12*, 185. (d) Ohrui, H.; Konno, M.; Meguro, H. *Agric. Biol. Chem.* **1987**, *51*, 625. (e) Ali, S. M.; Ramesh, K.; Borchardt, R. T. *Tetrahedron Lett.* **1990**, *31*, 1509. (f) Belanger, P.; Prasit, P. *Tetrahedron Lett.* **1988**, *29*, 5521. (g) Deardorff, D. R.; Shambayate, S.; Myles, D. C.; Heerding, D. *J. Org. Chem.* **1988**, *53*, 3614.

(6) Laumen, K.; Schneider, M. P. *J. Chem. Soc., Chem. Commun.* **1986**, 1298. See also: Johnson, C. R.; Bis, S. J. *Tetrahedron Lett.* **1992**, *33*, 7287.

(7) The minor anomer (15–20%) was observed when the reaction was run at 25 °C but was reduced to 3–5% under refluxing conditions.

(8) The stereochemistry of the anomeric center in **7** was determined to be β by the observed NOE's between CH_3-4 and H-1 (0.9%) and the fact that no NOE was observed between H1 and H2 or H1 and H3. The anomeric center in **13** was determined to be α by the observed NOE's between H-5 and H-1 (0.4%) and between H-1 and H-2 (0.9%).

(9) Nakagawa, J.; Hata, T. *Tetrahedron Lett.* **1975**, *16*, 1409. Serafinowski, P. *Synthesis* **1985**, 926.

(10) We found the preparation of *N,N′*-bis(trifluoroacetyl)-L-homocystine dimethyl ester from L-homocystine by treatment with thionyl chloride in methanol followed by trifluoroacetic anhydride to be more efficient and convenient than that reported using successive treatments with dimethyl sulfite and trifluoroacetic anhydride (Cruickshank, P. A.; Sheehan, J. C. *Anal. Chem.* **1964**, *36*, 1191). Zinc/acetic acid cleavage of this disulfide gave the thiol, methyl *N*-(trifluoroacetyl)-L-homocysteine in 73% overall yield.

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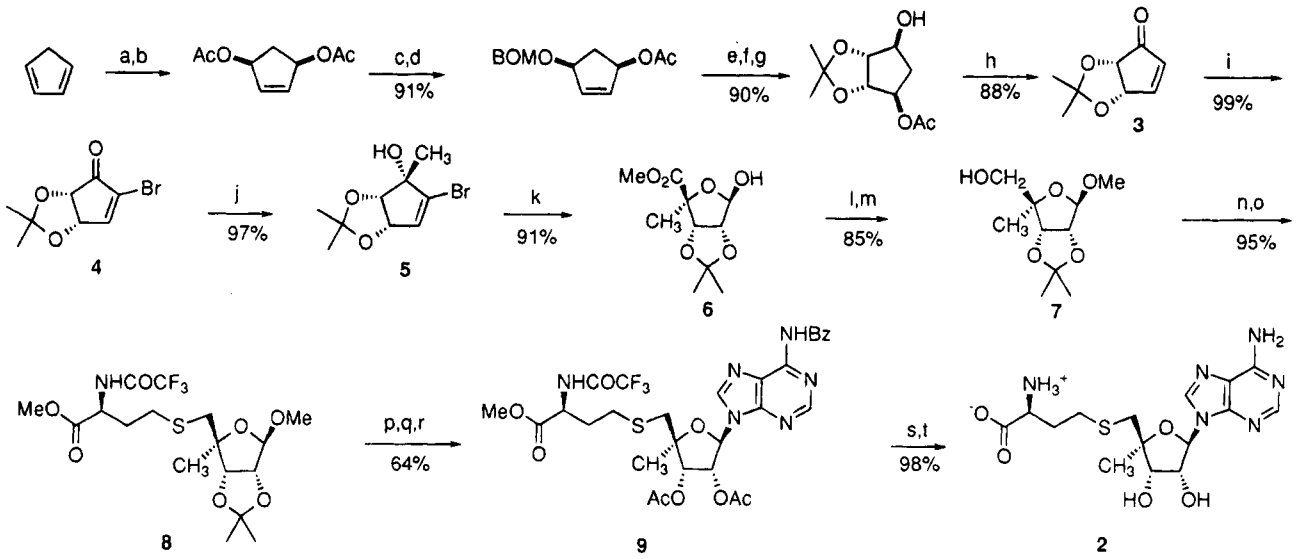
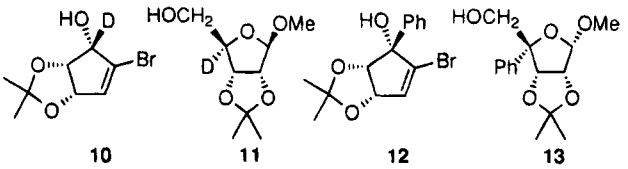
Scheme 1[a]



[a] (a) O_2, MeOH, rose bengal, thiourea $h\nu$; (b) Ac_2O; (c) crude porcine pancreatic lipase; (d) benzyloxymethyl chloride; (e) OsO_4, *N*-methylmorpholine *N*-oxide; (f) acetone, TsOH; (g) H2, Pd–C; (h) pyridinium dichromate, 4 Å molecular sieves, CH_2Cl_2; (i) Br_2, CH_2Cl_2 then Et_3N; (j) MeMgBr, Et_2O; (k) O_3, MeOH, pyridine, −78 → 0 °C then Me_2S; (l) MeOH, acetone, TsOH, Δ; (m) $LiBH_4$; (n) Tf_2O, Et_3N, −78 °C; (o) *N*-(trifluoroacetyl)-L-homocysteine, methyl ester, NaH, DMF; (p) MeOH, HCl; (q) Ac_2O, TEA; (r) N^6-benzoyladenine, TMS-OTf; (s) 0.2 M LiOH, 3 h; (t) NH_3, MeOH, 12 h.

two successive treatments of **8** with acidic methanol, and the methyl 2,3-*O*-diacetylglycoside was prepared with acetic anhydride. Presilylated N^6-benzoyladenine and the above methyl glycoside were treated at elevated temperature (65 °C, CH_3CN) with trimethylsilyl trifluoromethanesulfonate under Vorbrüggen[11] conditions to give the fully protected β-nucleoside **9** in 64% yield from **8** (three steps, no α-anomer was isolated). Treatment of **9** with aqueous LiOH for 3 h followed by NH_3/MeOH (to efficiently remove the final *N*-benzoyl protection group) and purification by ion exchange chromatography using Dowex 50 (H^+) followed by Dowex 50 (NH_4^+) gave clean *S*-(4′-methyladenosyl)-L-homocysteine (**2**) in 98% yield (44% overall yield from **3**).

Various 4-substituted riboses should be accessible by the strategy conveyed in Scheme 1. We found that a variety of nucleophiles including borohydrides, aryl Grignards, and alkyl Grignards provided excellent regio- and diastereoselective additions to α-bromo enone **4**. Syntheses of ribose-*d* derivatives are of particular interest, for example, for the investigation of calicheamicin cleavage of DNA by selective 5′- or 4′-hydrogen abstraction.[4] Treatment of **4** under Luche[12] conditions with $NaBD_4$ provided **10**. Subsequent ozonolysis, glycosidation (23:1, β:α), and reduction gave methyl 2,3-*O*-isopropylidene-β-D-ribofuranoside-4-*d* (**11**) (95% deuterium incorporation) in 67% overall yield from **3** (five-steps, not optimized). Similarly, methyl 2,3-*O*-isopropylidene-β-L-ribofuranoside was prepared from the enantiomer of **3**.[13] Chemoselective ozonolysis of the vinyl bromide **12**, prepared by addtion of phenylmagnesium bromide to **4**, followed by glycosidation (1:20, β:α)[7] and reduction gave methyl 4-phenyl-α-D-ribofuranoside (**13**)[8] in a modest 20% yield from **3**.



Compared to the manipulation of natural carbohydrates, this strategy offers the following advantages: (i) the stereogenic center at C-4 is set prior to unveiling the ribose framework; (ii) the availability of both enantiomers of enone **3** allows access to either D or L riboses; and (iii) the method by which the ribose is unveiled and protected as the methyl glycoside circumvents problems associated with pyranose–furanose equilibration, which for 4-substituted riboses would undoubtedly lay in favor of the pyranose form.[14] The biological activity of **2** will be reported in context with other mechanism based inhibitors based on the AdoHcy structure in a subsequent publication. Studies involving other 4′-alkylnucleosides and nucleotides are in progress.

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Supplementary Material Available: Experimental procedures for key reactions and compound characterization data along with copies of the ^1H and ^{13}C NMR spectra for compounds **2**, **7**, **8**, **9**, **11**, and **13** (16 pages). This material is contained in libraries on microfiche, immediately follows this article in the microfilm version of the journal, and can be ordered from the ACS; see any current masthead page for ordering information.

(11) Vorbrüggen, H.; Krolikiewicz, K.; Bennua, B. *Chem. Ber.* **1981**, *114*, 1234 and references cited therein.

(12) Gemal, A. L.; Luche, J.-L. *J. Am. Chem. Soc.* **1981**, *103*, 5454.

(13) The enantiomer of **3** was prepared by manipulation of the *cis*-hydroxy acetate described in Scheme 1 or by a modification of the procedure by Siddiqi *et al.*[3c] wherein an enzymatic resolution/acylation of (±)-*cis*-4-phenoxy-2-cyclopenten-1-ol was carried out in isopropenyl acetate using *Pseudomonas cepacia* lipase. Methyl 2,3-*O*-isopropylidene-β-L-ribofuranoside was prepared in 59% overall yield from the enantiomer of **3** when Luche reduction of **4** was carried out with $NaBH_4$. The optical rotation of the L-ribofuranoside was equal but of opposite sign to that of the well-known D-enantiomer.

(14) Angyal, S. J. *Adv. Carbohydr. Chem. Biochem.* **1984**, *42*, 15. The preparation of methyl 4-methyl-L-ribopyranoside from a L-*erythro*-pentopyranosid-4-ulose has been reported (Overend, W. G.; White, A. C.; Williams, N. R. *Carbohydr. Res.* **1970**, *15*, 185).