Bioorganic & Medicinal Chemistry Letters, Vol. 1, No 1, pp. 65-68, 1991
Printed in Great Britain

0960-894X/91 $3.00 + .00

PREPARATION OF THE GEOMETRIC ISOMERS OF DDC, DDA, D4C AND D4T AS POTENTIAL ANTI-HIV AGENTS

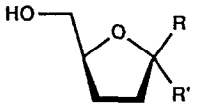
Muzammil M. Mansuri*, Vittorio Farina, John E. Starrett Jr.,
Daniel A. Benigni, Vera Brankovan# and John C. Martin

Bristol-Myers Squibb Company, 5 Research Parkway, Wallingford, CT 06492-7660
Oncogen, Seattle, WA 98121

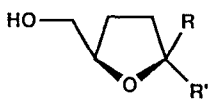
(*Received* 19 *October* 1990)

Abstract. ß-L-ddC (5), ß-L-ddA (6), ß-L-d4T (7) and ß-L-d4C (8) (enantiomers of natural dideoxynucleoside analogues with known potent HIV activity) were prepared as potential anti-HIV agents.

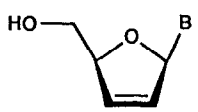
2',3'-Dideoxynucleosides analogues have been extensively studied as potential anti-HIV agents [1]. These analogues exert their antiviral effect by being converted to their corresponding triphosphates; these triphosphates then act as inhibitors of the unique viral enzyme reverse transcriptase (RT) [2-4]. While several structural analogues, with both sugar and base modifications, have been prepared as potential anti-HIV agents [5], little work has been done on the enantiomers of known anti-HIV agents [6]. We now report on the preparation and biological activities of compounds **5-12**, geometric isomers of four known (**1-4**) HIV agents.



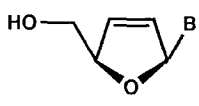
1 R = C, R' = H
2 R = A, R' = H
9 R = H, R' = C
10 R = H, R' = A



5 R = C, R' = H
6 R = A, R' = H
11 R = H, R' = C
12 R = H, R' = A



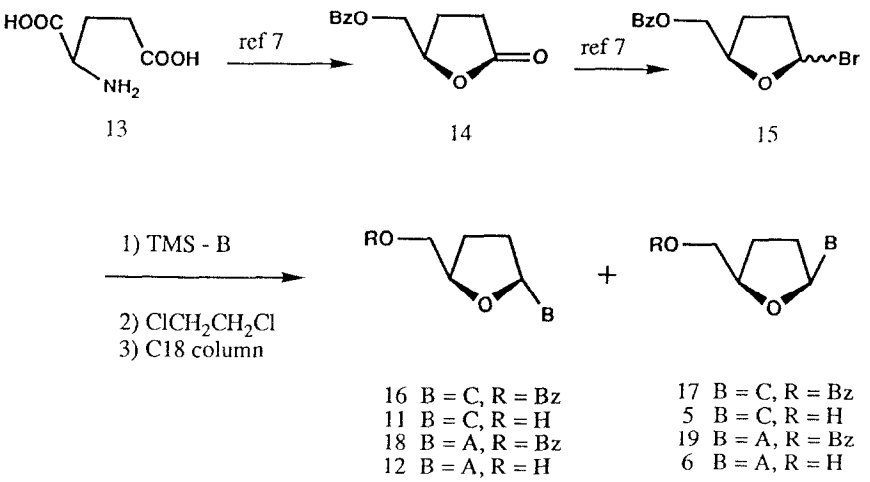
3 B = T
4 B = C



7 B = T
8 B = C

C = cytosine, T = thymine, A = adenine

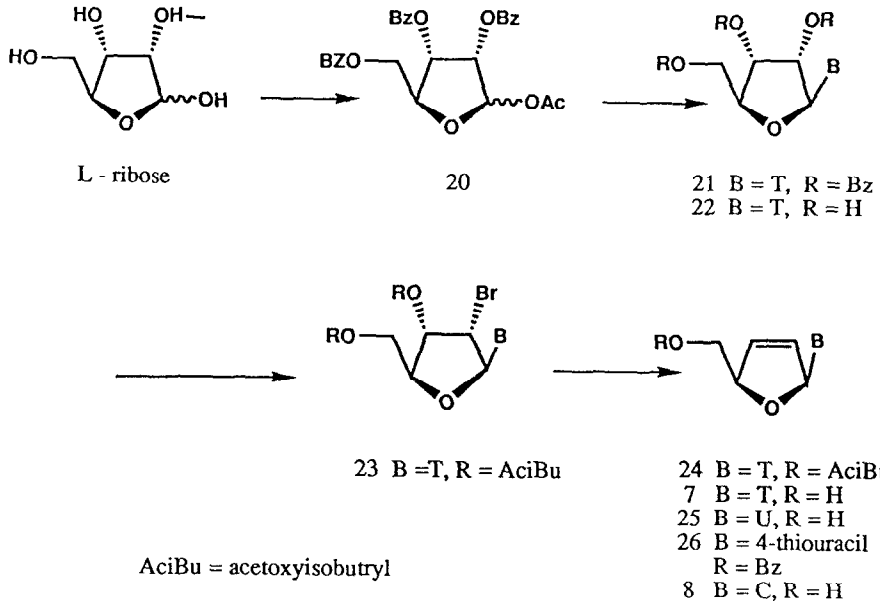
The preparation of the saturated ß-L-2',3'-dideoxynucleosides follows a sequence similar to one described earlier [7]. D-Glutamic acid (**13**) was converted, via lactone **14**, to a mixture of anomeric bromides **15** (Scheme 1) [8]. Coupling of **15** with bis-silylated cytosine gave a 2:3 mixture of **16:17** in nearly quantitative yield based on the sugar. The anomers were separated by reverse phase C_{18} column chromatography [9]. Deprotection of **16** and **17** under standard conditions (NH_3/MeOH) furnished **11** and **5** in 84% and 78% yields respectively. Alternatively, coupling with silylated adenine afforded a 1:1 mixture of **18:19** in 80% yield. Again, separation upon a reverse phase column gave the pure anomers **18** and **19**, which on deprotection furnished the pure nucleosides **12** and **6** [9,10].



The ß-L-enantiomer of d4T, compound 7, was prepared by coupling tribenzoyl acetate derivative (**20**) with silylated thymine under Vorbruggen conditions [12, 13], to give **21** in 88% yield, triflic acid was the catalyst. Compound **21** was converted to ß-L-5-methyluridine (**22**), which on treatment with acetoxyisobutyrylbromide furnished the bromoacetate **23**, in 86 % yield [14]. Reduction of **23** with a zinc/copper couple in DMF afforded **24**, which on deprotection gave the desired product, ß-L-d4T (**7**).

ß-L-D4U (**25**) was readily prepared by an analogous sequence to **7**, but using uracil, rather than thymine. Treatment of **25** with benzoyl chloride in pyridine, followed by reaction with Lawesson's reagent in chloroform heated to reflux, afforded the thioamide **26** [15]. One pot deprotection and amination at the 4 position of **26** with ammonia in methanol gave ß-L-d4C (**8**). Hydrogenation of **8** with palladium on carbon can also be used to prepare ß-L-ddC (**5**).

Compounds **5-12** were tested against HIV in CEM cells. ß-L-ddC (**5**) showed moderate activity (ID_{50} = 0.66μm, compared to ID_{50} = 0.04 μm for ß-D-ddC (**1**)), α-D-ddC (**9**) was also active but showed higher cellular toxicity (ID_{50} = < 0.1 μm, cell toxicity = 0.1 μm) [16]. None of the other analogues displayed activity at concentrations up to 100 μM. The anti-HIV activity of **5** is in contrast to the results of Okabe et al. [6], who report that **5** had no biological activity [17]. These results demonstrate that the enantiomers and diastereomers of known anti-HIV agents may also have anti-HIV activity [18].



Acknowledgement. We would like to thank Ed Pack, Bonnie Bleiberg and Susan Hill of the Analytical Department, Bristol-Myers Squibb, Wallingford for the help in establishing the enantiomeric purity of these compounds.

REFERENCES AND FOOTNOTES

[1] (a) Broder, S. Mitsuya; H. Proc. Natl. Acad. Sci. U.S.A. **1986**, 83, 1911.
(b) De Clercq,E. Antiviral Res. **1989**, 12, 1.

[2] Furman, P. Fyfe, J. A.; St. Clair, M. H; Weinhold, K.; Rideout, J. L.; Freeman, G. A. Lehrman, S. N.; Bolognesi, D. P.; Broder, S.; Mitsuya, H.; Barry, D. W. Proc. Natl. Acad. Sci., U.S.A. **1986**, 83, 8333.

[3] Johnson, M. A.; Ahluwalia, G.; Connelly, M. C.; Cooney, D. A.; Broder, S.; Johns, D. G.; Fridland, A. J. Biol. Chem. **1988**, 263, 15354.

[4] Mansuri, M. M.; Starrett, J. E.; Ghazzouli, I.; Hitchcock, M. J. M.; Sterzycki, R. Z.; Brankovan, V.; Lin, T-S.; August E. M. Prusoff, W. H.; Sommadossi, J-P.; Martin, J. C.; J. Med. Chem. **1989**, 32, 461.

[5] (a) Huryn, D. M.; Sluboski, B. C.; Tam, S. Y.; Todaro, L. J.; Weigele, M. Tetrahedron Letters **1989**, 30, 6259. (b) Norbeck, D. W.; Spanton, S.; Broder, S.; Mitsuya, H.; Tetrahedron Letters, **1989**, 30, 6263. (c) Kim, C-H.; Marquez, V.; Broder, S., Mitsuya, H.; Driscoll J. S. J. Med. Chem. **1987**, 30, 862.

[6] Okabe, M.; Sun R-C.; Tam, S. Y-K.; Todaro, L. J.; Coffen D. J. Org.Chem. **1988**, 53, 4780. The authors state that ß-L-ddC (**5**) was prepared and showed no activity.

[7] Farina, V.; Benigni, D. A. Tetrahedron Letters **1988**, 29, 1239.

[8] The D-glutamic acid was purchased from Sigma Fine Chemicals and was used without purification.

[9] The anomers were separated on a Waters LC-500 chromatography system using a reverse phase prep pak C-18 cartridge. After equilibration the column was eluted with 50% MeOH in either water or pH 6.5 buffer. The fractions were collected and analyzed by TLC (2 developments) in 5% MeOH/CH_2Cl_2.

[10] All the compounds prepared gave satisfactory elemental and spectroscopic results.

[11] A racemic mixture of D/L ß-ddA has previously been reported, see Robins, M.; Jones, R. A.; Mengel, R. J. Am. Chem. Soc. **1976**, 98, 8213.

[12] Recondo, R.; Rinderknecht, H. Helv. Chem. Acta. **1962**, 42, 1711.

[13] Vorbruggen, H.; Bennua, B. Chem. Ber. **1981**, 114, 1279.

[14] Jain, T. C.; Jenkins, I. D.; Russell, A. F.; Verheyden, J. P. H.; Moffatt, J. G. J. Org. Chem. **1974**, 39, 30.

[15] Kaneko, K.; Katayama, H.; Toshio, W.; Tadahiro, K. Synthesis **1988**, 152.

[16] The assay method used has previously been published. See reference [4].

[17] Chromatographic separation on a chiral column, to establish the optical purity of **5**, showed that there was less than 0.15% ß-D-ddC (**1**) present in the sample ß-L-ddC (**5**) [17]. The 0.15% represents the limit of detection of the column.

[18] A recent patent application from Medivir Pharmaceutical Co. (EP-A-0352248) reports an IC_{50} <1μm for **10** against HIV in H9 cells.

[19] An in situ prepared Cu-Proline column was used to estimate the enantiomeric purity. The conditioning mobile phase was 4 mM $CuSO_4$.0.5 water/50 mM KH_2PO_4 at pH 4.0. The operating mobile phase was 5/95 conditioning phase/water.