Bioorganic & Medicinal Chemistry Letters, Vol.3, No.4, pp. 543-546, 1993
Printed in Great Britain

0960-894X/93 $6.00 + .00

SYNTHESIS OF 1-(2-AMINOPROPYL)BENZIMIDAZOLES, STRUCTURALLY RELATED TO THE TIBO DERIVATIVE R82150, WITH ACTIVITY AGAINST HUMAN IMMUNODEFICIENCY VIRUS.[1]

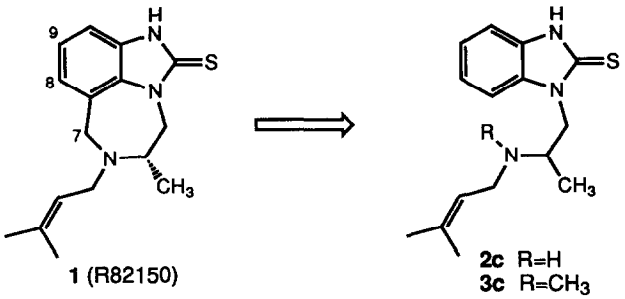
Eric E. Swayze,[a] Sithumini M. Peiris,[a] Louis S. Kucera,[d] E. Lucile White,[e] Dean S. Wise,[b] John C. Drach,[c] and Leroy B. Townsend.[a,b]*

[a]Department of Chemistry, College of Literature, Sciences, and Arts; [b]Department of Medicinal Chemistry, College of Pharmacy; [c]Department of Biologic and Material Sciences, School of Dentistry, The University of Michigan, Ann Arbor, Michigan 48109; [d]Department of Microbiology and Immunology, Bowman Gray School of Medicine, Winston-Salem, NC 27157; and [e]Microbiology Research Department, Southern Research Institute, Birmingham, AL 35205.

(*Received in USA 7 December* 1992)

Abstract: A number of 1-(2-aminopropyl)-2-mercaptobenzimidazoles related to the TIBO derivative R82150 have been prepared and tested for their activity against human immunodeficiency virus type 1 (HIV-1). These compounds were all modest inhibitors of the cytopathic effects of HIV-1*in vitro*, but only very weak inhibitors of HIV-1 reverse transcriptase (RT).

Treatment of infection by human immunodeficiency virus type 1 (HIV-1), the causative agent of AIDS,[2] is currently limited to the nucleoside analogs 3'-azido-3'-deoxythymidine (AZT, zidovudine), 2',3'-dideoxyinosine (DDI, didanosine), and most recently 2',3'-dideoxycytidine (DDC, zalcitabine). These drugs suffer from a number of limitations, including toxic side effects[3] and the emergence of drug-resistant strains of the virus.[4] Recently, non-nucleoside inhibitors of HIV-1 reverse transcriptase (RT) have been reported, including the TIBO analog R82150 (**1**)[5], nevirapine,[6] several 2-pyridinones,[7] a series of uracil derivatives,[8] and some bis heteroarylpiperazines.[9] Several of these non-nucleosides are active against HIV-1 (but not HIV-2) in the nanomolar range *in vitro*. This structurally diverse group of compounds have a similar antiviral profile, and inhibit HIV-1 RT via binding at an allosteric site, in contrast to active site inhibitors such as AZT.[10] The structure-activity relationships of the TIBO series of compounds have been well investigated,[11] but all modifications have conserved the tricyclic ring system. We were interested in studying whether the diazepine ring was necessary in order to maintain activity against HIV-1. Either removal of the C-7 carbon, or scission of the C-7 to C-7a bond in the diazepine ring of the TIBO derivative **1**, would lead to the 2-mercaptobenzimidazole derivatives **2c** and **3c**, respectively, which may be able to assume a TIBO-like conformation in the active site. In the present report, we describe the synthesis, cytotoxicity, and anti-HIV activity of several conformationally unrestrained TIBO analogs.

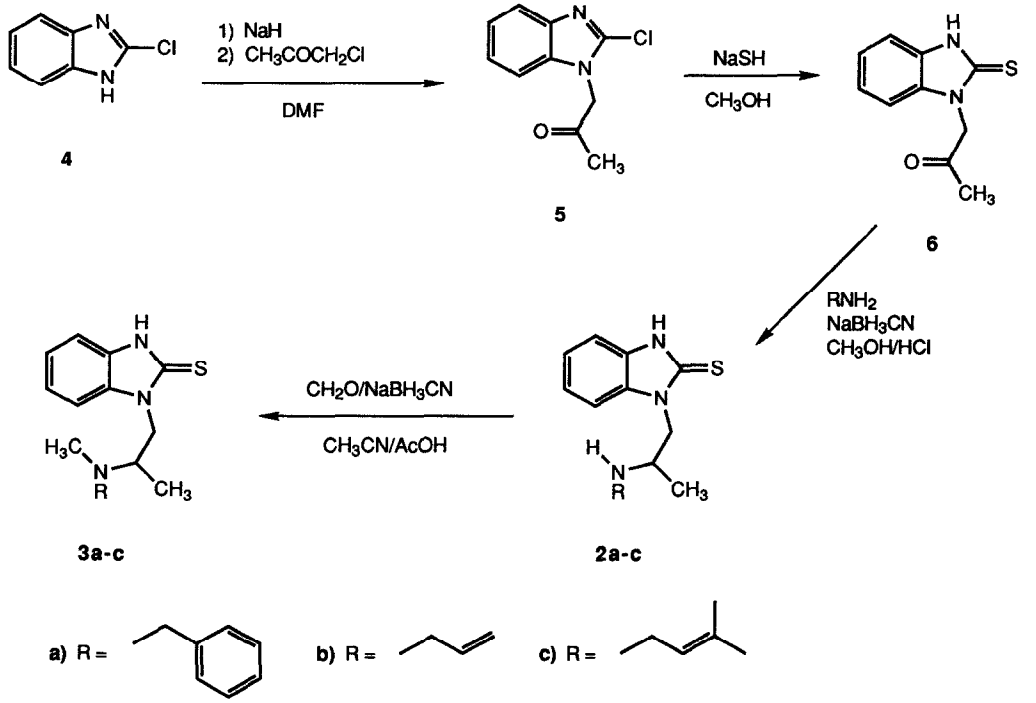


1 (R82150)

2c R=H
3c R=CH_3

Scheme I illustrates the synthetic route used to prepare the target compounds **2a-c** and **3a-c**. Treatment of the commercially available 2-chlorobenzimidazole (**4**) with sodium hydride (1.2 equivalents) in dimethylformamide at room temperature provided the sodium salt, which was alkylated with chloroacetone (1.2 equivalents) to afford the 1-substituted compound **5** in 93% yield. Displacement of the halogen proceeded smoothly at 80° C in methanolic sodium hydrogen sulfide (from 1.2 eqivalents sodium methoxide in methanol saturated with hydrogen sulfide) to give compound **6** in 81% yield. Reductive amination of the methyl ketone **6** with sodium cyanoborohydride[12] and an excess of the appropriate amine[13] in methanol at pH 7 gave a racemic mixture of the desired compounds **2a-c**. The free amines were purified by column chromatography on silica gel using either ethyl acetate/hexanes or methanol/chloroform as the mobile phase. Lyophilization of an acidified (with 1N HCl to pH=3) solution of the free amines in methanol/water provided the hydrochloride salts of **2a-c** in 57-59% yields. The methylated products (**3a-c**) were prepared by treatment of the free amines **2a-c** with excess formaldehyde and sodium cyanoborohydride in acetonitrile at pH 7, and then isolated as their hydrochloride salts after column chromatography.[14]

Scheme I. Synthesis of Some Substituted 2-Mercaptobenzimidazoles.

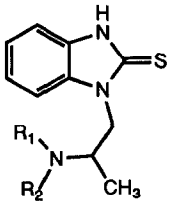


The 1-substituted benzimidazoles (**2a-c** and **3a-c**) were evaluated for cytotoxicity and activity against HIV-1 in CEM-SS cells.[15] All compounds tested were modest inhibitors of HIV-1 syncytial plaque formation, and fifty percent inhibitory concentrations (IC_{50}'s) ranged from 4 to 28 μM (Table I). Cytotoxicity was determined by measuring inhibition of [^{3}H]thymidine incorporation into DNA of uninfected cells. On this basis, all compounds exhibited a modest separation between antiviral activity and toxicity. Compounds **2a**, **2c**, and **3a** had similar toxicities, with 50% cytotoxic concentrations (CC_{50}) around 80 μM, while compounds **2b**,

3b, and **3c** were less toxic, not reaching a CC_{50} at drug concentrations of 100 μM. Compound **2a** was the most potent antiviral of the series, and had a selectivity index (CC_{50}/IC_{50}) of 19.

In order to ascertain whether these compounds were acting as HIV-1 RT inhibitors, compounds **2a-c**, **3a**, and **3c** were tested for their activity against HIV-1 RT using a ribosomal RNA template.[16] Shown in Table I is the inhibition of HIV-1 RT at a concentration of 10 μg/mL (approximately 30 μM), and at a concentration of 100 μg/mL (approximately 300 μM). All compounds tested were weak inhibitors of HIV-1 RT, even at concentrations higher than their *in vitro* IC_{50} values. In fact, at drug concentrations as high as 100 μg/mL (approximately 300 μM), the IC_{50} was not obtained. In contrast, the TIBO derivative R82913 (9-chloro derivative of **1**) was highly active against HIV-1 RT when tested as a positive control.[17] This dramatic difference in HIV-1 RT inhibition between the TIBO derivative R82913 and the new benzimidazoles demonstrates that an intact diazepine ring appears to be necessary for inhibition of HIV-1 RT by TIBO analogs. Furthermore, this minimal RT activity indicates that the modest *in vitro* anti-HIV-1 activity of the new 1-substituted mercaptobenzimidazoles is not solely due to the inhibition of HIV-1 RT, and that these compounds might function via a different mechanism from other non-nucleosides active against HIV-1. To the best of our knowledge, the compounds **2a-c** and **3a-c** are the first benzimidazoles with activity against HIV-1 at non-toxic concentrations. We are currently studying modifications of the N-1 substituent, as well as halogenation of the benzimidazole moiety, with the intent of increasing the potency and selectivity of this series of compounds.

Table I: Biological Activity of Some Substituted 2-Mercaptobenzimidazoles.



Compound	Substituent		% RT Inhibition[a]		50% Inhibitory Concentration (μM)[b]	
	R_1	R_2	10 μg/mL	100 μg/mL	HIV-1[c]	Cytotoxicity[d]
2a	H	$CH_2C_6H_5$	17	36	4.3	82
2b	H	$CH_2CH{=}CH_2$	4.5	29	28	>100
2c	H	$CH_2CH{=}C(CH_3)_2$	19	46	23	81
3a	CH_3	$CH_2C_6H_5$	13	31	20	79
3b	CH_3	$CH_2CH{=}CH_2$	ND[e]	ND	23	>100
3c	CH_3	$CH_2CH{=}C(CH_3)_2$	19	34	14	>100
R82150			ND	ND	0.026	79
R82913			IC_{50}=0.005 μg/mL		0.055	35

[a]% Inhibition of HIV-1 RT using a ribosomal RNA template versus control at the given concentration. [b]Average of two or three experiments. [c]Syncytial assay in CEM-SS cells. [d][^{3}H]thymidine uptake inhibition in CEM-SS cells. [e]Not determined.

Acknowledgement: The authors would like to thank Jack Hinkley for the preparation of starting materials. We also thank Nathan Iyer and Shelby Puckett (HIV-1 syncytial assay and cytotoxicity evaluations), and Larry J. Ross (HIV-1 RT assay) for technical support. This study was supported by the Department of Health and Human Services grant U01-AI-25739 from the National Institute of Allergy and Infectious Diseases.

References and Notes:

1) Portions of this study were presented at the 204th American Chemical Society National Meeting (Medi # 143), August 1992, Washington D. C.
2) Barre-Sinoussi, F.; Chermann, J. C.; Rey, F.; Nugeyre, M. T.; Chamaret, S.; Gruest, J.; Dauguet, C.; Axler-Blin, C.; Vezinet-Brun, F.; Rouzioux, C.; Rozenbaum, W.; Montagnier, L. *Science* **1983**, *220*, 868-871.
3) Yarchoan, R.; Mitsuya, H.; Myers, C. E.; Broder, S. *N. Engl. J. Med.* **1989**, *321*, 726-738.
4) Larder, B. A.; Darby, G.; Richman, D. D. *Science* **1989**, *243*, 1731-1734.
5) a) Pauwels, R.; Andries, K.; Debyser, Z.; Kukla, M.; Schols, D.; Desmyter, J.; De Clercq, E.; Janssen, P. A. J. *Biochemical Society Transactions* **1992**, *20*, 509-512. b) Pauwels, R.; Andries, K.; Desmyter, J.; Schols, D.; Kukla, M. J.; Bresline, H. J.; Raeymaekers, A.; Van Gelder, J.; Woestenborghs, R.; Heykants, J.; Schellekens, K.; Janssen, M. A. C.; De Clercq, E.; Janssen, P. A. J. *Nature* **1990**, *343*, 470-474.
6) Hargrave, K. D.; Proudfoot, J. R.; Grozinger, K. G.; Cullen, E.; Kapadia, S. R.; Patel, U. R.; Fuchs, V. U.; Mauldin, S. C.; Vitous, J.; Behnke, M. L.; Klunder, J. M.; Kollol, P.; Skiles, J. W.; McNeil, D. W.; Rose, J. M.; Chow, G. C.; Skoog, M. T.; Wu, J. C.; Schmidt, G.; Engel, W. W.; Eberlein, W. G.; Saboe, T. D.; Campbell, S. J.; Rosenthal, A. S.; Adams, J. *J. Med. Chem.* **1991**, *34*, 2231-2241.
7) Saari, W. S.; Hoffman, J. M.; Wai, J. S.; Fisher, T. E.; Rooney, C. S.; Smith, A. M.; Thomas, C. M.; Goldman, M. E.; O'Brien, J. A.; Nunberg, J. H.; Quintero, J. C.; Schleif, W. A.; Emini, E. A.; Stern, A. M.; Anderson, P. S. *J. Med. Chem.* **1991**, *34*, 2925-2928.
8) a) Baba, M.; De Clercq, E.; Tanaka, H.; Ubasawa, M.; Takashima, H.; Sekiya, K.; Nitta, I.; Umezu, K.; Walker, R. T.; Mori, S.; Ito, M.; Shigeta, S.; Miyasaka, T. *Mol. Pharmacol.* **1991**, *39*, 805-810. b) Baba, M.; De Clercq, E.; Tanaka, H.; Ubasawa, M.; Takashima, H.; Sekiya, K.; Nitta, I.; Umezu, K.; Nakashima, H.; Mori, S.; Shigeta, S.; Walker, R. T.; Miyasaka, T. *Proc. Natl. Acad. Sci. USA* **1991**, *88*, 2356-2360.
9) Romero, D. L.; Busso, M.; Tan, C.-K.; Reusser, F.; Palmer, J. R.; Poppe, S. M.; Aristoff, P. A.; Downey, K. M.; So, A. G.; Resnick, L.; Tarpley, W. G. *Proc. Natl. Acad. Sci. USA* **1991**, *88*, 8806-8810.
10) De Clercq, E. *Aids Research and Human Retroviruses* **1992**, *8*, 119-134.
11) a) Kukla, M. J.; Breslin, H. J.; Pauwels, R.; Fedde, C. L.; Miranda, M.; Scott, M. K.; Sherrill, R. G.; Raeymaekers, A.; Van Gelder, J.; Andries, K.; Janssen, M. A. C.; De Clercq, E.; Janssen, P. A. J. *J. Med. Chem.* **1991**, *34*, 746-751. b) Kukla, M. J.; Breslin, H. J.; Diamond, C. J.; Grous, P. P.; Ho, C. Y.; Miranda, M.; Rodgers, J. D.; Sherrill, R. G.; De Clercq, E.; Pauwels, R.; Andries, K.; Moens, L. J.; Janssen, M. A. C.; Janssen, P. A. J. *J. Med. Chem.* **1991**, *34*, 3187-3197.
12) Lane, C. F. *Synthesis* **1975**, 135-146.
13) Allylamine and benzylamine were purchased from Aldrich Chemical Company. 3-Methyl-2-butenylamine was prepared from 1-bromo-3-methyl-2-butene (Aldrich) via the literature method: Semenow, D.; Shih, C.-H.; Young, N. G. *J. Am. Chem. Soc.* **1958**, *80*, 5472-5475.
14) Melting point, ^{1}H NMR and elemental analysis data for compounds **2a-c** and **3a-c** are available from the authors upon request.
15) Kucera, L. S.; Iyer, N.; Leake, E.; Raben, A.; Modest, E. J.; Daniel, L. W.; Piantadosi, C. *Aids Research and Human Retroviruses* **1990**, *4*, 491-501.
16) White, E. L.; Buckheit, R. W.; Ross, L. J.; Germany, J. M.; Andries, K.; Pauwels, R.; Janssen, P. A. J.; Shannon, W. M.; Chirigos, M. A. *Antiviral Res.* **1991**, *16*, 257-266.
17) The TIBO derivatives R82150 and R82913 were found to be essentially equally potent inhibitors of HIV-1 RT when using a different template: Debyser, Z.; Pauwels, R.; Andries, K.; Desmyter, J.; Kukla, M.; Janssen, P. A. J.; De Clercq, E. *Proc. Natl. Acad. Sci. (USA)* **1991**, *88*, 1451-1455.