generation time of the cells compared to control cells without compound.

Mutagenicity Testing. The mutagenicity experiments were performed with *Salmonella typhimurium* strains TA 98 and TA 100 with and without S9 mix from rat liver by using the plate incorporation assay. Liver S9 fractions were prepared from male Wistar rats after induction with a combination of sodium phenobarbital (0.1% in the drinking water) and β-naphtoflavone (dissolved in corn oil, 12 mg/mL, and injected intraperitoneally on the fifth day of induction at a dosage of 80 mg/kg, or 0.666 mL corn oil solution/100 mg). The animals were fasted for 24 h immediately preceding death, and they were killed on the seventh day of induction by cervical dislocation. The efficacy of the S9 fraction was confirmed by a control experiment with strain TA 100 and benzo[a]pyrene. For mutagenicity testing a concentration of 20 μL of S9 per plate was used. Quercetin was included as a standard mutagen. Duplicate plates were poured for each dose of mutagen.[29-31]

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Synthesis and Anti-HIV-1 Activity of 4,5,6,7-Tetrahydro-5-methylimidazo[4,5,1-*jk*][1,4]benzodiazepin-2(1*H*)-one (TIBO) Derivatives

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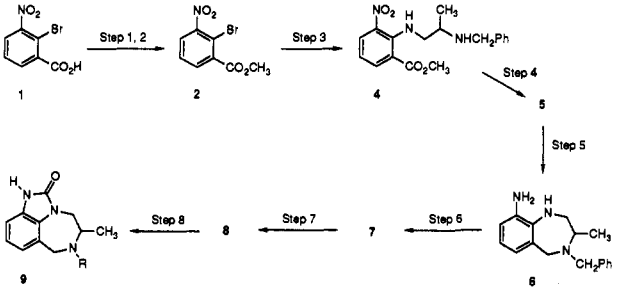
A series of 6-substituted 4,5,6,7-tetrahydro-5-methylimidazo[4,5,1-*jk*][1,4]benzodiazepin-2(1*H*)-ones (**9**) have been synthesized and tested for their ability to inhibit the replication of the HIV-1 virus in MT-4 cells. Two synthetic methods are described, one of which allows the synthesis of single enantiomers of the final products. A structure–activity study was done within the series of compounds to determine the optimum group for the 6-position substitution and to determine whether the activity was enantiospecific at the 5-position, which was substituted with a methyl group. The best analogue, **9jj**, inhibited HIV-1 with an IC_{50} of 4 μM, which is comparable to the activity level of DDI, a 2′,3′-dideoxynucleoside-type structure undergoing clinical trials as an anti-AIDS therapy.

Introduction

Therapeutic intervention for the treatment of HIV-1 infection which eventually results in AIDS (acquired immune deficiency syndrome) is limited to AZT (Zidovudine), the only approved drug for this disease. Unfortunately, it suffers from a number of limitations including limiting side effects and the revelation of the possible immergence of drug-resistant mutants of the virus.[1] Thus the need for new drugs to combat this disease is obvious, and several others are being evaluated for their effectiveness.[2] From the list of potential future therapeutic agents one is struck by the lack of good leads available for medicinal chemical optimization. The nucleoside analogues of which AZT is included are by far the most prevalent. However, the common mechanism of action of this group gives good reason to believe that all members of this structural class could suffer from some or all of the same limitations of AZT. Thus there is clearly a need for structures from other chemical families which inhibit the HIV-1 virus, possibly by alternative mechanisms.

Our approach to this problem was to choose a large group of representative structures from our collection of compounds for screening as inhibitors of HIV-1 replication. The limitations placed in choosing the compounds for initial screening were that they had little or no known pharmacological effect, including toxicity, from previous testing. Indeed, from 600 compounds that were evaluated in vitro for their ability to inhibit HIV-1 replication, **9a**

Scheme I



Step	Reagents and Conditions	Yield
1	$SOCl_2$, reflux, 2h	
2	CH_3OH, reflux, 1h	92% overall
3	$H_2NCH_2CH(CH_3)NHCH_2Ph$ (3), *n*-SuOH, Na_2CO_3, reflux, 1.5h	95%
4	2-hydroxypyridine, xylenes, reflux, 19h	93%
5	AlH_3, THF	87%
6	$Cl_3COCOCl$, N-methylmorpholine, CH_2Cl_2, 0°C	71%
7	H_2 (45 psi); 10% Pd/C, glacial HOAc	87%
8	Method : A; R-X, KI, Na_2CO_3, DMF	
	S; R-CHO, $NaCNBH_3$, CH_3CH	
	C; R-COOH, 1-hydroxybenzotriazole, DCC	

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§ Janssen Research Foundation, Belgium.

(1) Larder, B.; Kemp, S. *Nature* 1989, *246*, 1155. Larder, B.; Darby, G.; Richman, D. *Science* 1989, *243*, 1731.
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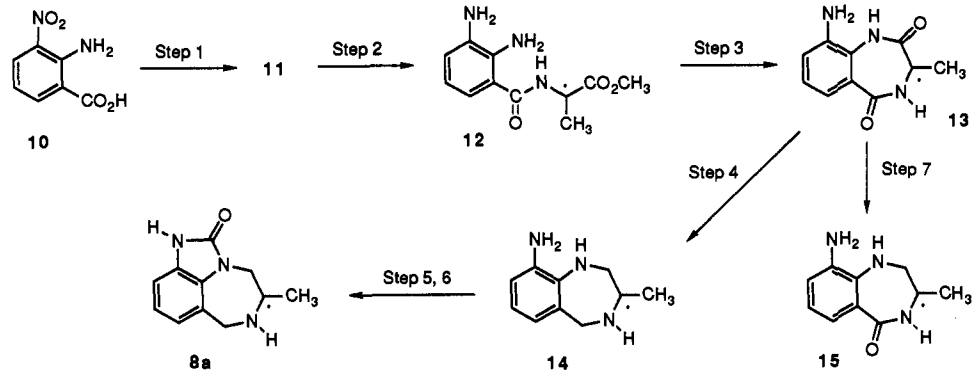
Scheme II



Step	Reagents and Conditions	Yield
1	1-Hydroxybenzotriazole, DCC, H_2N-CH(CH_3)-CO_2CH_3 · HCl	97%
2	H_2, EtOH,10% Pd/C	
3	200°C, neat under vacuum	58% overall
4	LAH, dioxane or glyme, reflux, 45h	
5	$Cl_3COCOCl$, N-methylmorpholine, CH_2Cl_2, 0°C	
6	H_2O, dioxane, 100°C	79% overall
7	LAH, THF, reflux	

· Denotes possible single enantiomer with the configuration determined by that of the alanine methyl ester used in step one.

blocked the virus at concentrations which were below the cytotoxic concentration. From that lead we initiated an ongoing program to optimize the potential anti-HIV-1 activity by systematic alteration of all portions of the lead structure. This first report deals with the variation of the group appended to the 6-position nitrogen of the diazepine ring.

Chemistry

Two different synthetic pathways have been used to prepare the common synthetic intermediate 8 which has been used to synthesize all the analogues (**9**) for testing. As indicated in Scheme I, 2-bromo-3-nitrobenzoic acid[3] was treated with thionyl chloride followed by methanol to give benzoate **2** in 92% yield. This was heated with 1,2-diaminopropane derivative **3**[4] to afford 4 by halogen displacement (95%). Cyclization to benzodiazepine **5** was accomplished in 93% yield by heating 4 in refluxing xylene in the presence of 2-hydroxypyridine catalyst. Reduction to triamine **6** was carried out smoothly with aluminum hydride at ambient temperature after initial attempts to carry out this reduction with lithium aluminum hydride were less successful. With the latter reagent, after 2.5 h in a mixture of THF and toluene at reflux, there was complete loss of starting material but only 35% of the desired **6** (GC analysis). The main product resulted from only partial reduction, the nitro to an amine, to yield the diamino lactam. Continued reflux gave increased amounts of **6**, but even after 5 days there was only 79% conversion. Several methods were explored to convert *o*-diamine **6** to cyclic urea **7** including fusion with urea at >200 °C, phosgene, and carbonyl diimidazole. Each gave desired product but were less effective or convenient than trichloromethyl chloroformate (diphosgene), which yielded **7** in 71%. Removal of the benzyl protecting group with catalytic reduction gave noramine 8 (87%). Three methods were used to append various groups to the secondary amine. Most often a standard alkylation procedure of an alkyl halide,[5] potassium iodide, and a base scavenger—sodium carbonate—in hot DMF gave the desired **9**. In a few cases, especially for the heteroaromatic derivatives **9h**, **9t**, and **9w**, the reductive alkylation method using the appropriate aldehyde and sodium cyanoborohydride was the method of choice. Because direct acetylation of 8 gave acetylation of the urea nitrogen as well, derivative **9g** was acquired from a coupling reaction with acetic acid (method C).

Scheme II describes an enantiospecific synthesis which allows obtention of either enantiomer of the 5-position substituent on the diazepine ring and is also versatile enough to allow future replacement of the methyl substituent with other groups. By using the readily available enantiomers of alanine methyl ester in a coupling reaction with 2-amino-3-nitrobenzoic acid,[6] the stereocenter at the carbon is fixed from the beginning. The reaction was accomplished under standard peptide coupling conditions with dicyclohexylcarbodiimide and 1-hydroxybenztriazole to give **11** in 97% yield. Catalytic reduction gave *o*-diamine **12**. A number of methods were explored to cyclize **12** to benzodiazepine derivative **13**. Heating **12** neat at ca. 200 °C under vacuum, to remove extruded methanol, turned out to be the best with an overall yield of 58% from **11**. The results of lithium aluminum hydride reduction of **13** were solvent dependent. One of the lactam carbonyls was reduced more readily than the other. Thus when the reduction of **13** was carried out in refluxing THF, **15** was the sole product. The higher reflux temperatures of dioxane

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Table I. Inhibition of HIV-1 Replication in MT-4 Cells

no.	R	IC_{50},[a] μM	CC_{50},[b] μM	SI[c]	n[d]
9a	$CH_2CH{=}CH_2$	70	670	10	44
9b	CH_2CN	I[e]	508		6
9c	$CH_2C(CH_3){=}CH_2$	47	665	15	19
9d	CH_2COCH_3	I	760		7
9e	$CH_2CO_2CH_3$	855	925		7
9f	$CH_2C{\equiv}CH$	580	990		7
9g	$COCH_3$	I	175		3
9h	2-furanylmethyl	106	529	5	2
9i	$CH_2C(CO_2C_2H_5){=}CH_2$	I	3		3
9j	$CH_2CH{=}CHCO_2CH_3$ (*E*)	I	252		6
9k	$CH_2CH{=}CH_2$[*R*-(−)]	I	592		3
9l	$CH_2CH{=}CH_2$[*S*-(+)]	66	670	10	11
9m	H[*R*-(−)]	I	901		4
9n	H[*S*-(+)]	I	901		4
9o	$CH_2CH_2CH{=}CH_2$	50	552	11	6
9p	$CH_2CH_2CH_3$	90	722	8	11
9q	CH_2CH_3	I	592		8
9r	$CH_2C(CH_3){=}CH_2$[S-(+)]	19	222	11	15
9s	CH_2-cyclopropyl	44	588	13	6
9t	1*H*-pyrrol-2-ylmethyl	I	92		4
9u	$CH(CH_3)_2$	I	65		4
9v	CH_2CH_2OH	I	776		4
9w	1*H*-imidazol-2-ylmethyl	I	685		4
9x	$CH_2CH{=}CHCH_3$ (*E*)	58	443	8	7
9y	$CH_2CH{=}CHCH_3$ (*Z*)	35	560	16	7
9z	$CH_2CH(CH_3)_2$	I	82		5
9aa	$CH_2CH_2CH_2CH_3$	100	501	5	7
9bb	$CH_2CH{=}C(CH_3)_2$	12.5	416	33	13
9cc	$CH_2CH{=}C(CH_3)CH_2CH_2$-$CH{=}C(CH_3)_2$ (*E*)	I	44		4
9dd	$CH_2CH{=}C(CH_3)C{\equiv}CH$	I	707		4
9ee	$CH_2CH{=}CHCH{=}CHCH_3$ (*E*,*E*)	I	378		3
9ff	$CH_2C(Br){=}CH_2$	62	416	7	4
9gg	$CH_2CH{=}C(CH_3)C{\equiv}CH$ (*E*)	I	18		3
9hh	$CH_2C(CH_3){=}CHCH_3$ (*E*)	29	106	4	4
9ii	$CH_2CH{=}C(CH_3)_2$[R-(−)]	22	450	20	8
9jj	$CH_2CH{=}C(CH_3)_2$[*S*-(+)]	4	427	105	8
9kk	$CH_2C(CH_2Ph){=}CH_2$	I	572		1
9ll	$CH_2C(CH_3){=}C(CH_3)_2$	I	>876		1
9mm	$CH_2CH{=}CHPh$ (*E*)	I	>783		1
9nn	$CH_2C(CH_2CH_3){=}CH_2$	37	66	2	2
9oo	$CH_2CH{=}CHC{\equiv}CH$ (*E*)	I	370		1
9pp	$CH_2C(i\text{-}Pr){=}CH_2$	I	18		2
9qq	$CH_2C(Ph){=}CH_2$	I	3		2
9rr	CH_2-1-cyclohexenyl	I	212		2
9ss	$CH_2CH{=}CHPh$ (*Z*)	122	388	3	2
9tt	$CH_2C(CH{=}CH_2){=}CH_2$	71	290	4	3
9uu	$CH_2CH_2OCH(CH_3)_2$	I	504		1
AZT		0.0015	9.3	6,200	
DDC		0.137	232	1,693	
DDI		5.5	1,053	191	

[a] Effective concentration of compound required to achieve 50% protection of MT-4 cells against the cytopathic effect of HIV-1. [b] Cytotoxic concentration of compound required to reduce the viability of mock-infected MT-4 cells by 50%. [c] Selectivity index (SI) is a ratio of CC_{50}/IC_{50}. [d] Number of experiments used to determine the average IC_{50} and CC_{50} values. [e] Inactive—does not inhibit virus replication by 50% at any concentration below the CC_{50}.

Table II. Product Purification and Characterization

no.	a	purification[b]	% yield	mp, °C	formula[c]
9a	A	+, Et_2O	73	138.5–140.5	$C_{14}H_{17}N_3O$
9b	A	+, MeOH/Et_2O (2:1)	57	188–190	$C_{13}H_{14}N_4O$
9c	A	EtOH	73	146–148	$C_{15}H_{19}N_3O$
9d	A	+, EtOH/Et_2O (1:2), EtOH	50	137.5–140.5	$C_{14}H_{17}N_3O_2$
9e	A	MeOH/Et_2O	57	142–144.5	$C_{14}H_{17}N_3O_3$
9f	A	EtOH/Et_2O, EtOH	60	142–143.5	$C_{14}H_{15}N_3O$
9g	C	+, MeOH	40	242–244	$C_{13}H_{15}N_3O_2$
9h	B	+, EtOH	35	142–144.5	$C_{16}H_{17}N_3O_2$
9i	A	EtOH	63	158.5–159.5	$C_{17}H_{21}N_3O_3$
9j	A	MeOH	70	187.5–188.5	$C_{16}H_{19}N_3O_3$
9k	A	Et_2O	23	110–112	$C_{14}H_{17}N_3O$
9l	A	Et_2O	34	110–112	$C_{14}H_{17}N_3O$
9m		*i*-PrOH	52	201.5–204	$C_{11}H_{13}N_3O$
9n		MeOH/*i*PrOH	73	202–204.5	$C_{11}H_{13}N_3O$
9o	A	+, Et_2O	56	102.5–104.5	$C_{15}H_{19}N_3O$[d]
9p	A	+, Et_2O	56	146–147.5	$C_{14}H_{19}N_3O$
9q	A	Et_2O	21	138–142	$C_{13}H_{17}N_3O$[e]
9r	A	EtOH	42	150–151	$C_{15}H_{19}N_3O$
9s	A	+, Et_2O	78	126–128	$C_{15}H_{19}N_3O{\cdot}H_2O$
9t	B	+, CH_3CN	29	80–90	$C_{16}H_{18}N_4O$[f]
9u	A	+, acetone	15	161–163.5	$C_{14}H_{19}N_3O$
9v	A	MeOH/Et_2O	32	152.5–155	$C_{13}H_{17}N_3O_2$
9w	B	MeOH	33	221–222	$C_{15}H_{17}N_5O$
9x	A	CH_3CN	64	125–126	$C_{15}H_{19}N_3O$
9y	A	+	60	105–106.5	$C_{15}H_{19}N_3O$[g]
9z	A	+, CH_3CN	39	123–124	$C_{15}H_{21}N_3O{\cdot}{}^1/_2H_2O$
9aa	A	+, Et_2O	20	135–136.5	$C_{15}H_{21}N_3O$
9bb	A	CH_3CN	60	155–157	$C_{16}H_{21}N_3O$
9cc	A	CH_3CN	64	99–100	$C_{21}H_{29}N_3O$
9dd	A	CH_3CN	49	129–130	$C_{17}H_{19}N_3O$
9ee	A	+, CH_3CN	48	127–128	$C_{17}H_{21}N_3O$
9ff	A	CH_3CN	48	146–148	$C_{14}H_{16}BrN_3O$
9gg	A	CH_3CN	87	166–168	$C_{17}H_{19}N_3O$
9hh	A	CH_3CN	80	148–150	$C_{16}H_{21}N_3O$
9ii	A	CH_3CN	50	131–135	$C_{16}H_{21}N_3O$
9jj	A	CH_3CN	45	134–136	$C_{16}H_{21}N_3O$
9kk	A	CH_3CN/CH_2Cl_2	85	166–169.5	$C_{21}H_{23}N_3O$
9ll	A	CH_3CN	72	178–180	$C_{17}H_{23}N_3O$
9mm	A	CH_3CN	73	159–161	$C_{20}H_{21}N_3O$[h]
9nn	A	+	45	108–110	$C_{16}H_{21}N_3O$
9oo	A	+ (twice)	41	138	$C_{16}H_{17}N_3O$
9pp	A	+, EtOH	28	138–141	$C_{17}H_{23}N_3O$
9qq	A	+, EtOH	37	144–147	$C_{20}H_{21}N_3O$
9rr	A	+, EtOH	32	151–155	$C_{18}H_{23}N_3O$
9ss	A	+, CH_3CN	45	134–135	$C_{20}H_{21}N_3O$
9tt	A	+, CH_3CN	44	115–117	$C_{16}H_{19}N_3O$
9uu	A	+	60	124–127	$C_{16}H_{23}N_3O_2$[i]

[a] Method/reagents used to synthesize the analogue: (A) RX, Na_2CO_3, KI, DMF: (B) RCHO, $NaCNBH_3$, MeOH; (C) RCO_2H, DCC, hydroxybenztriazole. [b] Purification method: If a silica flash chromatography was done on the crude product, it is indicated by a "+". The solvent used for recrystallization follows. [c] All products were analyzed for C, H. N. [d] C: calcd, 70.01; found, 68.54. N: calcd, 16.33; found, 15.90. [e] C: calcd, 67.51; found, 67.09. [f] C: calcd, 68.07; found, 66.66. N: calcd, 19.84; found, 19.41. [g] C: calcd, 70.01; found, 69.28. [h] C: calcd, 75.21; found, 74.52. [i] C: calcd, 65.41; found, 65.91.

or glyme are necessary to complete reduction to 14. Generally glyme was preferable because a significant amount of foaming with dioxane occurred during addition of the reagents. Carbonylation of 14 again was carried out with diphosgene in 79% yield. The unprotected secondary amine was converted to a carbamoyl chloride derivative during the course of this reaction. Consequently, a hydrolytic workup was used to regenerate amine 8a. The stereochemical purity of the 5-position enantiomers was confirmed by ^1H NMR studies using the chiral shift reagents (*R*)- and (*S*)-2,2,2-trifluoro-1-(9-anthryl)ethanol,[7] first on the racemic material 8 to show that the enantiomers could be differentiated and detected, and then on each of the pure enantiomers 8a to verify enantiomeric purity. In each case, the enantiomers 8a were free of the other enantiomer to the limits of detection of NMR. Thus the stereocenter fixed in the first reaction of the sequence has maintained its integrity throughout.

Results and Discussion

The anti-HIV-1 activity of this series was determined in MT-4 cells as described previously.[8] The cells were either infected with HIV-1 or mock infected and incubated in the presence of various concentrations of the test compounds (9). The number of viable cells was then determined 5 days after infection by staining with 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide as reported previously.[9]

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(8) Pauwels, R.; De Clerq, E.; Desmyter, J.; Balzarini, J.; Goubau, P.; Herdewijn, P.; Vandeputte, M. *J. Virol. Methods* **1987**, *16*, 171.

The anti-HIV-1 activity of this series is reported in Table I. The determinations for each compound include the concentration required to protect 50% of the cells against the HIV-1-induced cytopathic effects (IC_{50}) and the concentration of test compound which is cytotoxic (CC_{50}) to uninfected cells. The ratio of these two values is reported as a selectivity index (SI). Most of the determinations were run in duplicate as indicated and the reported values are an average. The nature of the assays make a determination of the standard deviations tenuous at best, although the values were usually quite consistent within the multiple determinations.

The original lead compound **9a** inhibited the cytopathic effect of HIV-1 at a concentration one-tenth its cytotoxic concentration. A large number of variations of the allyl appendage were made to explore the possible improvement in both the absolute activity as well as the selectivity index. For a discussion of the SAR (structure–activity relationship) the relative IC_{50} values will be used. An attached group at this 6-position is necessary for activity, since the unsubstituted compound (R = H, **9m** and **9n**) is inactive. Additionally, there seems to be the necessity for a certain amount of bulk at this position as the ethyl (**9q**) and linear propargyl (**9f**) substituents are ineffective. Although, in the latter case, there may be something disadvantageous with an acetylene group; four examples (**9f**, **9dd**, **9gg**, and **9oo**), contain that functionality and all are inactive. In general, the inclusion of a heteroatom in the "R" group caused a decrease in activity. Thus, when the methylene attached to the ring nitrogen is appended with a nitrile (**9b**), a ketone (**9d**), an ester (**9e**, **9i**, or **9j**), an alcohol (**9v**), an ether (**9uu**), or a heteroaromatic pyrrole (**9t**) or imidazole (**9w**), activity is lost. The exceptions seem to be the furan (**9h**) and bromide (**9ff**) derivatives, which have activity comparable to that of **9a**. The presence of an olefin enhances activity when a direct comparison can be made with the respective saturated group. Allyl (**9a**) is slightly better than propyl (**9p**), 2-butenyl (either E, **9x** or Z, **9y**) or 3-butenyl (**9o**) is more active than butyl (**9aa**), and 2-methylallyl (**9c**) is quite active, whereas the corresponding saturated analogue 2-methylpropane (**9z**) is surprisingly inactive. In contrast, to **9z**, the cyclized methylene cyclopropyl compound (**9s**) had good activity, presumably due to the near sp^2 character of the cyclopropyl bonds. Another series of compounds was synthesized to compare substitution of the allyl group. Addition of bulk at the 2-position of the allyl group was favorable or at least tolerated to an extent. Thus ethyl (**9nn**) > methyl (**9c**) = vinyl (**9tt**) = bromo (**9ff**) > H (**9a**), but 2-propyl (**9pp**), phenyl (**9qq**), benzyl (**9kk**), and ring fused cyclohexenyl (**9rr**) are all inactive. Substitution on the end of the allyl group was examined with a limited number of variations. As previously indicated, a single methyl group addition (**9x** and **9y**) gave improved activity. A combination of 2-methyl and 3-methyl addition (**9hh**) also gave improved activity. However, the best result came from dimethyl substitution (**9bb**) which turned out to be the best substitution we found in this series. When larger groups were attached to the end of the allyl (**9cc**, **9dd**, **9ee**, **9gg**, **9mm**, **9oo**, or **9ss**) activity was usually completely lost. In summary, we can draw the following conclusions about the SAR involved in the single change of an appended group at the 6-position of this novel structural type. A lipophilic group is mandatory for activity since the unsubstituted compound is inactive. Polarity due to heteroatoms diminishes activity and the presence of an olefin enhances activity. There is an optimum size requirement of the group, since activity is lost with increases or decreases in the length or in the breadth of the side chain. Among the compounds we have examined, the 3-methyl-2-butenyl group as exemplified in **9bb** was the best.

Another point we needed to explore was the enantiospecific nature of the activity against HIV-1. This would help to lend credence to a specific pharmacological intervention of these compounds at some undetermined point in the replication process of the virus[10] versus nonspecific toxic effects. We initially synthesized and tested each enatiomer of **9a**. The *S*-(+)-enatiomer **9l** was active and the isomer with the *R*-(−)-configuration (**9k**) was without effect against the virus. Interestingly, the cytotoxic effects (CC_{50}) of the two compounds were nearly equal. Another analogue with the correct configuration (*S*) of the methyl group at C-5 (**9r**) was more active than the racemic material (**9c**), but in this instance there was also a drop in the CC_{50} so that there was no improvement in the selectivity index. However, this could be construed as further evidence that the antiviral and cytotoxic effects were not linked since those two activities varied independently. Finally, the enantiomers (**9ii** and **9jj**) of the most active racemic analogue (**9bb**) were compared. Consistent with the previous data, the *S* configuration was the most active (3 times as active as the racemate), although in this case the *R* isomer also had activity (one-half as potent as the racemate).

From this work the optimal features were combined in **9jj**, which was improved over initially discovered lead compound **9a** in terms of IC_{50} (17.5-fold better, 4 μM vs 70 μM) and selectivity index (>10-fold better). This compound also compares favorably (see Table I) in those criteria with at least one nucleoside compound, DDI, which is currently being evaluated in clinical trials as another drug to add to anti-AIDS therapy.[11]

Research is ongoing to evaluate what effects other changes to the lead structure will have on the anti HIV-1 activity and also to elucidate the mechanism by which this structural type inhibits replication of the virus.[12]

Experimental Section

All final products included in the tables were characterized by 360-MHz ^1H NMR (Bruker AM 360WB), IR (Nicolet 60SX), and mass spectra (Finnegan 3300) as well as elemental analyses. The latter were performed by the internal Analytical Research Department of Janssen Research Foundation in Beerse, Belgium. Melting points were obtained on a Thomas-Hoover capillary immersion apparatus and are uncorrected. When NMR's are reported they are from primary spectra and the proton assignments are preliminary.

Methyl 2-Bromo-3-nitrobenzoate (2). Thionyl chloride (95 mL, 1.3 mol) was added to a stirred mixture of 1 (156.26 g, 0.635 mol) in 260 mL of toluene. The stirred mixture was heated to reflux and became homogeneous. After 2 h the mixture was allowed to cool to ambient temperature, which resulted in separation of a white solid. Concentration in vacuo yielded a residual solid which was stirred and diluted with 290 mL of methanol (exothermic). The resultant mixture was heated under reflux for 1 h and allowed to cool. The separated solid was further diluted with water and isolated on a filter. It was dissolved in CH_2Cl_2, washed with saturated $NaHCO_3$ (to remove unreacted 1), then brine, dried over K_2CO_3, and concentrated to yield a solid. It was

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recrystallized from 300 mL of methanol to yield 151.1 g (92%) of **2**. Mp: 76.5–78 °C.

Methyl 3-Nitro-2-[[2-[(phenylmethyl)amino]propyl]-amino]benzoate (4). In *n*-butanol (360 mL), **2** (93 g, 0.36 mol), **3** (58.74 g, 0.36 mol), and Na_2CO_3 (37.90 g, 0.36 mol) were combined and heated under reflux for 1.5 h. The reaction mixture was concentrated on a rotary evaporator, diluted with 180 mL of H_2O, and reconcentrated. The residue was diluted with 180 mL of H_2O and extracted twice with CH_2Cl_2 (600 mL total). The extracts were dried with $MgSO_4$ and concentrated to a red-orange oil that was suspended in 350 mL of *i*-PrOH and precipitated with the addition of 95 mL of HCl-saturated *i*-PrOH. The bright orange HCl salt was collected on a filter and washed with *i*-PrOH to yield 127.32 g (95%) of **4**. Mp: 200–201.5 °C. ^{1}H NMR (360 MHz, DMSO-d_6): δ 1.33 (d, J = 6 Hz, 3 H, CH_3), 3.1 (m, 1 H, C*H*-CH_3), 3.38 (m, 2 H, CH_2), 3.87 (s, 3 H, CH_3), 4.05 (m, 1 H, CH_2), 4.2 (m, 1 H, CH_2), 6.9 (m, 1 H, Ar), 7.4 (m, 3 H, Ar), 7.6 (m, 2 H, Ar), 8.1 (m, 3 H, Ar), 9.45 (bs, 1 H, NH), 9.9 (bs, 1 H, NH).

1,2,3,4-Tetrahydro-3-methyl-9-nitro-5*H*-1,4-benzodiazepin-5-one (5). Free base of **4** (108.86 g) was dissolved in 1.2 L of xylenes and 15.1 g of 2-hydroxypyridine was added. The mixture was heated under reflux for 19 h, allowed to cool, washed with H_2O, then brine, and gravity filtered. Crystals began forming from the filtrate which was further concentrated on a rotary evaporator. The product was isolated and dried at 78 °C to yield 91.6 g (93%) of **5**. Mp 121–123 °C. ^{1}H NMR (90 MHz, $CDCl_3$): δ 1.1 (d, J = 6 Hz, 3 H, CH_3), 3.25–3.95 (m, 3 H, CH_2NH, CH), 4.4 (d, J = 15 Hz, 1 H, PhCH_2), 5.05 (d, J = 15 Hz, 1 H, PhCH_2), 6.75 (t, 1 H, Ar), 7.3 (s, 5 H, Ar), 8.25–8.6 (m, 2 H, Ar), 9.05 (bs, 1 H, NH). MS: MH^+ (CI, CH_4) m/z 312.

2,3,4,5-Tetrahydro-3-methyl-4-(phenylmethyl)-1*H*-1,4-benzodiazepin-9-amine (6). A 1 M solution of lithium aluminum hydride in THF (800 mL, 0.8 mol; Aldrich) was cooled to 0 °C and stirred vigorously while concentrated H_2SO_4 (22.2 mL, 0.4 mol) was added dropwise over 30 min. The resultant mixture was stirred at ambient temperature for 2 h. A solution of **5** (43.1 g, 0.14 mol) in 500 mL of THF was added dropwise over 2.5 h at near room temperature. A cool water bath was used to moderate the exothermic reaction. After 24 h the reaction was cooled to 0 °C and quenched with the cautious addition of 58 mL of H_2O to the vigorously stirred mixture. The salts were removed by filtration and washed with THF (800 mL) and then stirred with CH_2Cl_2 (1 L) for 45 min and refiltered. Concentration of the filtrates yielded 32.64 g (87%) of **6** as a red-orange oil that was 87% pure by GLC (SE 30). ^{1}H NMR (360 MHz, $CDCl_3$): δ 1.2 (d, J = 6 Hz, 3 H, CH_3), 3.1–3.3 (m, 5 H, NH_2, NHCH_2, CH), 3.55–3.8 (m, 4 H, NH, ArCH_2, PhCH_2), 4.15 (d, J = 15 Hz, 1 H, PhCH_2), 6.45 (m, 1 H, Ar), 6.65 (m, 2 H, Ar), 7.3 (m, 5 H, Ph).

4,5,6,7-Tetrahydro-5-methyl-6-(phenylmethyl)imidazo[4,5,1-*jk*][1,4]benzodiazepin-2-(1*H*)-one (7). Trichloromethyl chloroformate (7.36 mL, 0.06 mol) was added dropwise via syringe over 20 min to a stirred, cold (0 °C) solution of **6** (32.64 g, 0.12 mol) in 400 mL of THF under argon. It was stirred another 10 min at 0 °C and then 1 h at ambient temperature. After dilution with 600 mL of CH_2Cl_2, saturated $NaHCO_3$ (400 mL) was added cautiously and the mixture stirred until homogeneous. The layers were separated, and the aqueous was washed with an additional portion of CH_2Cl_2. The combined organics were dried over K_2CO_3 and concentrated to a brown semisolid. Recrystallization from *i*-PrOH yielded 22 g (71%) of tan solid. Mp: 206.5–208 °C. ^{1}H NMR (360 MHz, DMSO-d_6): δ 1.25 (d, J = 7 Hz, 3 H, CH_3), 3.5 (d, J = 14 Hz, 2 H, CH_2C*H*), 3.7–3.8 (dd, J = 14 Hz, 1 H, CH_2NBz), 3.85 (d, J = 14 Hz, 1 H, CH_2CH), 4.0 (m, 3 H, CH_2Ph, CH_2NBz), 6.6 (m, 1 H, Ar), 6.85 (m, 2 H, Ar), 7.2–7.35 (m, 5 H, Ph), 10.9 (s, 1 H, NH). MS: MH^+ (CI CH_4) m/z 294.

4,5,6,7-Tetrahydro-5-methylimidazo[4,5,1-*jk*][1,4]benzodiazepin-2(1*H*)-one (8) from 7. A mixture of **7** (20.64 g), 10% Pd/C (1.01 g), and glacial acetic acid (130 mL) was placed in a Parr hydrogenator at ca. 60 psi of hydrogen pressure and 42 °C. After 6 h the mixture was filtered through a pad of Dicalite which was washed with HOAc/H_2O. The filtrate was concentrated to a clear light brown oil that was dissolved in 175 mL of H_2O, treated with Norit, and refiltered through Dicalite. The filtrate was basified to pH 8 with concentrated ammonium hydroxide and the separated solid collected on a filter and washed with H_2O. It was air-dried and then recrystallized from ca. 50 mL of CH_3OH/*i*-PrOH to yield 12.47 g (87%). ^{1}H NMR (360 MHz, DMSO-d_6): δ 1.15 (d, J = 6.5 Hz, 3 H, CH_3), 2.65 (bs, 1 H, NH), 3.0 (m, 1 H, C*H*CH_3), 3.1–3.2 (dd, 1 H, CH_2CH), 3.95 (d, J = 16.5 Hz, 1 H, CH_2NH), 4.0–4.15 (m, 2 H, CH_2NH, CH_2CH), 6.65–6.9 (m, 3 H, Ar), 10.85 (s, 1 H, CONH). MS: MH^+ (CI, CH_4) m/z 204.

(-)-Methyl (*S*)-2-[(2-Amino-3-nitrobenzoyl)amino]-propanoate (11). *N*-Methylmorpholine (5.49 mL, 0.05 mol) was added to a cold (−12 °C), stirred mixture of **10** (9.10 g, 0.05 mol), L-alanine methyl ester hydrochloride (6.95 g, 0.05 mol), and hydroxybenztriazole (13.50 g, 0.10 mol) in 200 mL of THF under argon. After 5 min, solid dicyclohexylcarbodiimide (10.30 g, 0.05 mol) was added. The mixture was allowed to warm to ambient temperature after 5.5 h and stirred an additional 16 h. It was recooled to 0 °C and filtered cold. The filtrate was concentrated and partitioned between ethyl acetate and saturated $NaHCO_3$ solution. The organic layer was washed with a second portion of saturated $NaHCO_3$, dried with $MgSO_4$, and concentrated to a solid that was triturated with hexane to yield 13.08 g (97%) of yellow solid. Mp: 129–130.5 °C. ^{1}H NMR (360 MHz, $CDCl_3$): δ 1.55 (d, 3 H, CH_3), 3.85 (s, 3 H, CH_3), 4.7–4.8 (m, 1 H, C*H*CH_3), 6.6–6.7 (m, 1 H, Ar), 6.75–6.8 (m, 1 H, NH), 7.7–7.75 (m, 1 H, Ar), 8.2 (bs, 2 H, NH_2), 8.25–8.3 (m, 1 H, Ar).

(+)-(*S*)-9-Amino-3-methyl-1*H*-1,4-benzodiazepine-2,5-dione (13). A mixture of **11** (12.58 g) and 10% Pd/C (3.5 g) in 200 mL of ethanol was placed in a Parr hydrogenator under 45 psi of hydrogen pressure. After 4 h the mixture was filtered through Dicalite and the filtrate concentrated to an oil. The stirred oil was placed under vacuum (25 mmHg) and the flask submersed in an oil bath at 150 °C. The temperature of the bath was raised to 202 °C over 10 min and maintained for 40 min. The oil solidified after 8 min at 202 °C. The solid was allowed to cool and was crushed, triturated with 15 mL of EtOH, and isolated on a filter. It was washed with cold EtOH, then Et_2O, and air-dried to yield 5.58 g (58%) of off-white solid. Mp: 281–286 °C (melting point obtained on the *R* isomer). ^{1}H NMR (360 MHz, DMSO-d_6): δ 1.2 (d, J = 6.5 Hz, 3 H, CH_3), 3.7–3.8 (m, 1 H, C*H*CH_3), 5.25 (s, 2 H, NH_2), 6.8–7.05 (m, 3 H, Ar), 8.3–8.4 (m, 1 H, NH), 9.35 (s, 1 H, NH). MS: MH^+ (FAB) m/z 206.

(+)-(*S*)-4,5,6,7-Tetrahydro-5-methylimidazo[4,5,1-*jk*]-[1,4]benzodiazepin-2(1*H*)-one (8a) from 14. Solid **13** (5.0 g, 0.024 mol) was added to a stirred suspension of lithium aluminum hydride (5.55 g, 0.146 mol) in 150 mL of dioxane at room temperature, under argon. The reaction mixture was heated to 100 °C and maintained for 45 h. It was then cooled to 10 °C and quenched with the sequential addition of 5.55 mL of H_2O, 5.55 mL of 15% NaOH solution, and 16.65 mL of H_2O. After 2 h the solid salts were removed by filtration and washed with hot THF and hot CH_2Cl_2. The combined filtrates and washes were dried with $MgSO_4$ and concentrated to an oil that was immediately combined with *N*-methylmorpholine (8.0 mL, 0.073 mol) in 100 mL of CH_2Cl_2. This solution was added over 15 min to a stirred, cold (0 °C) solution of trichloromethyl chloroformate (2.94 mL, 0.024 mol) in 120 mL of CH_2Cl_2 under argon. The resultant mix was stirred another 10 min at 0 °C and 20 min at ambient temperature before it was concentrated on a rotary evaporator to a solid. The solid was suspended in 70 mL of 85:15 H_2O/dioxane and heated on a steam bath for 45 min. When cool, the solution was washed twice with CH_2Cl_2, filtered, and basified with concentrated ammonium hydroxide. The solid that separated was collected on a filter, washed with cold H_2O and air-dried to yield 3.89 g (79%). Recrystallization from *i*-PrOH yielded an off-white solid. Mp: 201.5–204 °C.

6-Substituted 4,5,6,7-Tetrahydro-5-methylimidazo[4,5,1-*jk*][1,4]benzodiazepin-2(1*H*)-one (9). Typical Procedures. Method A. A solution of 1-bromo-3-methyl-2-butene (0.88 g, 5.9 mmol) in DMF (15 mL) was added to a stirred mixture of **8** (1.0 g, 4.92 mmol), potassium iodide (0.816 g, 4.92 mmol), and sodium carbonate (0.782 g, 7.38 mmol) in 60 mL of DMF under argon. After 22.5 h at ambient temperature the mixture was concentrated on a rotary evaporator. The residue was partioned between CH_2Cl_2 and $NaHCO_3$ solution (2×). The organic phase was washed with brine, dried with $MgSO_4$, and concentrated to yield 1.85 g. This was recrystallized twice from acetonitrile to yield 0.81 g (60%) of **9**. ^{1}H NMR (360 MHz, $CDCl_3$): δ 1.35 (d, J =

7 Hz, 3 H, CH_3), 1.45 (s, 3 H, CH_3), 1.75 (s, 3 H, CH_3), 3.1–3.25 (m, 2 H, $CH_2CH{=}C$), 3.45–3.55 (m, 1 H, $CHCH_3$), 3.85–3.95 (dd, 1 H, NCH_2CH), 4.05 (d, J = 17 Hz, 1 H, $ArCH_2N$), 4.1–4.15 (dd, 1 H, NCH_2CH), 4.25 (d, J = 17 Hz, 1 H, $ArCH_2N$), 5.2–5.25 (m, 1 H, $CH{=}C$), 6.75–6.8 (m, 1 H, Ar), 6.95–7.0 (m, 2 H, Ar), 9.75 (s, 1 H, NH). MS: MH^+ (CI, CH_4) m/z 272.

Method B. Furan-2-carboxaldehyde (1.41 mL, 17.2 mmol) was added to a stirred solution of **8** (1.39 g, 6.8 mmol) in 40 mL of methanol under argon. The mixture was stirred at ambient temperature for 3.5 h before addition of sodium cyanoborohydride (0.85 g, 14 mmol). The reaction was then heated to reflux and maintained for 17.5 h before another 0.56 mL (7 mmol) of furan-2-carboxaldehyde was added and reflux continued overnight. The mix was concentrated on a rotary evaporator and the residue partitioned between CH_2Cl_2 and saturated $NaHCO_3$. The organic layer was washed well with water, dried with K_2CO_3, and concentrated to yield crude product. This was recrystallized from ethanol, flash chromatographed (2.5% CH_3OH/CH_2Cl_2), and recrystallized again from ethanol to yield 0.67 g (35%) of **9**.

Method C. Glacial acetic acid (0.28 mL, 4.9 mmol) was added to a stirred solution of **8** (1.0 g, 4.9 mmol) and hydroxybenztriazole (1.33 g; 9.8 mmol) in 25 mL of DMF under argon at 0 °C. The solution was stirred at 0 °C for 1.5 h and at room temperature for 2 days before it was recooled to 0 °C and filtered. The filtrate was concentrated and the residual oil partitioned between CH_2Cl_2 and saturated $NaHCO_3$. The organics were washed with 2 N citric acid, then saturated $NaHCO_3$, dried over K_2CO_3, and concentrated. The crude product was recrystallized from methanol, flash chromatographed (2.5% CH_3OH/CH_2Cl_2), and recrystallized again from CH_3OH to yield 0.48 g (40%) of **9**.

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