Synthesis of 3-Nitrosoimidazo[1,2-*a*]pyridine Derivatives as Potential Antiretroviral Agents

Aziz Chaouni-Benabdallah[a,b], Christophe Galtier[a], Hassan Allouchi[c], Abdelhak Kherbeche[b], Jean-Claude Debouzy[d], Jean-Claude Teulade[e], Olivier Chavignon[e], Myriam Witvrouw[f], Christophe Pannecouque[f], Jan Balzarini[f], Erik de Clercq[f], Cécile Enguehard[a], and Alain Gueiffier[a]

[a] E.A. 3247 Groupe de Recherche en Chimie Hétérocyclique et Thérapeutique, Laboratoire de Chimie Thérapeutique, Faculté de Pharmacie, 31 Avenue Monge, 37200 Tours, France

[b] Laboratoire de Catalyse et Environnement, Université Sidi Mohamed Ben Abdellah, Ecole Supérieure de Technologie de Fès, B.P. 2427, Route d'Imouzzer, Fès, Maroc

[c] Laboratoire de Chimie Physique, Faculté de Pharmacie, 31 Avenue Monge, 37200 Tours, France

[d] CRSSA/BCM, 24 Av. des Maquis du Grésivaudan, B.P. 87, 38702 La Tronche, Cedex, France

[e] Laboratoire de Chimie Organique, Faculté de Pharmacie, 28 Place Henri Dunant, 63001 Clermont-Ferrand, France

[f] Rega Institute for Medical Research, Katholieke Universiteit Leuven, Minderbroedersstraat 10, B-3000 Leuven, Belgium

Key Words: *Imidazo[1,2-a]pyridine; nitrosation; antiretroviral; NCp7; crystal data*

Summary

Ten 2-aryl or heteroaryl-3-nitrosoimidazo[1,2-*a*]pyridine derivatives were synthesised as potential antiretroviral agents. The new compounds were characterized by elemental analysis, ^{1}H NMR, and by crystallography for (**14**). The compounds were devoid of any activity against HIV-1 or HIV-2.

Introduction

Since the demonstration of possible evasion to multidrug therapy with reverse transcriptase and protease inhibitors in HIV infection [1], the development of novel antiretroviral agents with other targets has been mandatory in the long-term therapy of AIDS.

From the different approaches investigated, the two retroviral Zn finger motifs of the HIV-1 nucleocapsid p7 (NCp7) protein appear to be of great interest [2]. Among all the known retroviruses, the spacing and metal chelating residues of the Cys-X_2-Cys-X_4-His-X_4-Cys (CCHC) Zn fingers are absolutely conserved [3], except for the spumaretroviruses [4]. Different families of compounds have been shown to be able to eject zinc from the two CCHC Zn fingers as a consequence of chemical modification at the sulfur atom of the Zn-coordinating Cys residues. From these compounds, C-nitrosoderivatives [3-nitrosobenzamide (NOBA)] [5], disulfide benzamides [2,2′-dithiobisbenzamides (DIBAS)] [6], dithiane and dithiolane derivatives [7], azodicarbonamide (ADA) [8] and pyridinoalkanoyl thiolesters (PATE) [9] have been documented. Recently, nucleomimetic strategy for the inhibition of HIV-1 nucleocapsid protein NCp7 was investigated [10]. Since it was then demonstrated that NOBA acts through its nitroso function, we were interested in continuation of our studies on imidazo[1,2-*a*]pyridine derivatives as antiviral agents [11], in the preparation of 3-nitroso derivatives as antiretroviral agents acting as potential zinc ejectors. It has been demonstrated that 2-phenylimidazo[1,2-*a*]pyridine derivatives could be nitrosated in the 3-position using sodium nitrite in acetic acid media at room temperature [12]. We now report the preparation and the activity of various 2-aryl or heteroaryl-3-nitrosoimidazo[1,2-*a*]pyridine derivatives as antiretroviral agents.

Correspondence: E.A. 3247 Groupe de Recherche en Chimie Hétérocyclique et Thérapeutique, Laboratoire de Chimie Thérapeutique, Faculté de Pharmacie, 31 Avenue Monge, 37200 Tours, France.
E-mail: enguehard@univ-tours.fr
Fax : +33 247367288

Results and Discussion

Chemistry

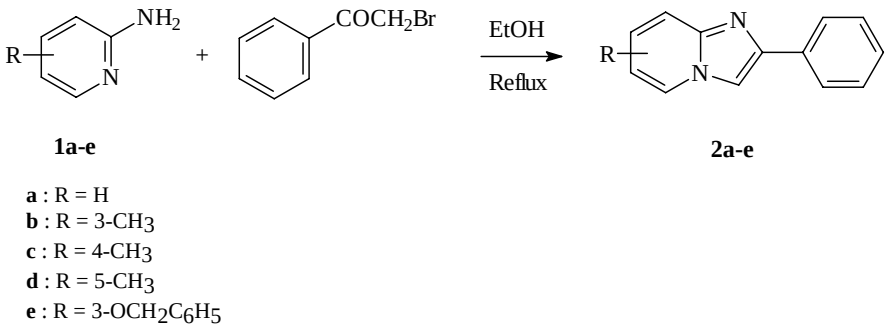
Substituted 2-aminopyridine **1a–e** were reacted with phenacyl bromide to give the desired imidazo[1,2-*a*]pyridines **2a–e** (Scheme 1). As a second alternative, influence of the

Table 1. Compounds synthesized and their mp and yields.

Compounds	R	Ar	Mp (°C)	Yield (%)
5	H	phenyl	165[a]	71
6	8-CH_3	phenyl	136	73
7	7-CH_3	phenyl	202[b]	70
8	6-CH_3	phenyl	199[c]	74
9	8-$OCH_2C_6H_5$	phenyl	203[d]	76
10	7-CH_3	naphth-2-yl	245	78
11	7-CH_3	biphen-4-yl	237	82
12	7-CH_3	thien-2-yl	208	90
13	7-CH_3	fur-2-yl	245	67
14	7-CH_3	pyridin-2-yl	197	87

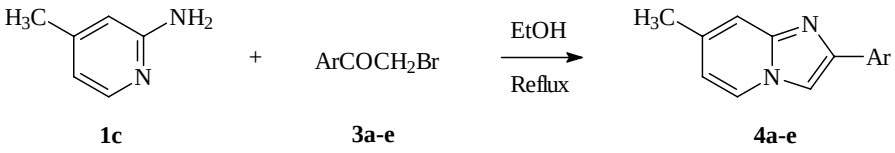
[a] mp 162–164°C[24], [b] mp 188°C[24], 194–196°C[12], [c] mp 180–182°C[24], [d] mp 193°C[25].

  0365-6233/01/0707/0224 $17.50 +.50/0



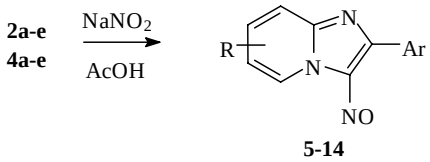
1a-e **2a-e**

a : R = H
b : R = 3-CH_3
c : R = 4-CH_3
d : R = 5-CH_3
e : R = 3-$OCH_2C_6H_5$



1c **3a-e** **4a-e**

a : Ar = pyridin-2-yl
b : Ar = napht-2-yl
c : Ar = biphen-2-yl
d : Ar = thien-2-yl
e : Ar = Fur-2-yl



5-14

Scheme 1. Synthesis of 2-aryl or heteroaryl-3-nitrosoimidazo[1,2-*a*]pyridines **5–14**.

2-substituent was investigated. Thus, 2-amino-4-picoline **1c** was reacted with aryl or heteroaryl-α-bromoethanone **3a–e** in refluxing ethanol according to Roe's procedure [13] to give the 2-substituted-7-methylimidazo[1,2-*a*]pyridines **4a–e**. Then nitrosation was achieved in a usual manner using sodium nitrite in acetic acid at room temperature to give the nitroso derivatives **5–14**. Slow addition rate of sodium nitrite in dilute solution prevented the formation of a red tar and thus exempted us from doing chromatography. Proof of the structure was easily given by ^{1}H NMR spectra with an upfield shift of H-5 in position peri from the NO group [14]. Finally, the crystal structure for compound **14** was investigated.

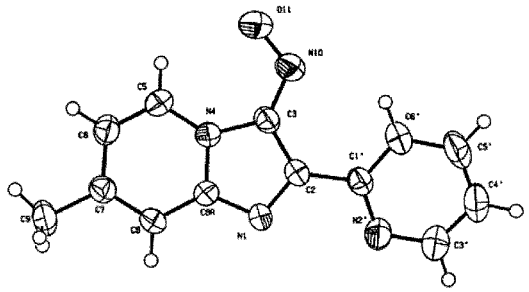


Figure 1. Ellipsoid representation of **14** and atomic labelling (X-Ray numbering).

Crystallographic Data

The thermal ellipsoid representation and the labelling of non-hydrogen atoms [15] of both conformations are presented in Figure 1. The bond lengths and bond angles agree quite well with literature data.

The imidazo[1,2-*a*]pyridine rings 1 (atoms N1 to C8A) and the pyridinic moiety 2 (atoms C1′ to C6′) are planar with the following dihedral angle : 2/1 = 26.4(1)°.

The packing of the molecule and the projection of the structure on the (x0z) and the (y0z) planes are shown in Figures 2 and 3a,b. There are numerous weak Van der Waals interactions so that the crystal packing is rather loose in ageement with the low observed density.

Antiviral Activities

All the synthesised compounds were inactive against HIV-1 (strain III_B) and HIV-2 (strain ROD) in MT-4 cells at subtoxic concentrations. Since the mechanistic pathway of NCp7 protein Zinc fingers of VIH is not completely established, interpretation of these results appears difficult. However, from the steric hinderance of the substituent in position 2, the nitroso

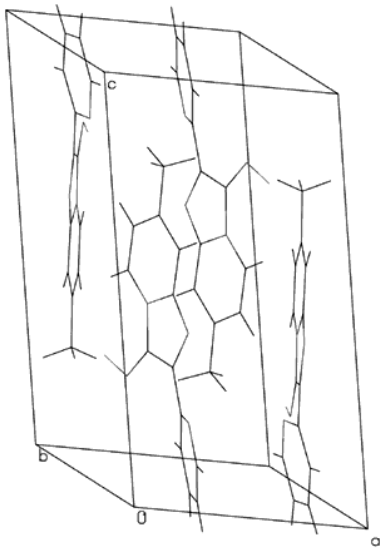


Figure 2. Packing representation of **14.**

group could not be located in the close vicinity of the thiolate group of the zinc finger cysteine. In order to verify this hypothesis, the synthesis of 2-unsubstituted derivatives would be of interest.

Acknowledgments

We thank Kristien Erven and Cindy Heens for excellent technical assistance. These investigations were supported by grants from the Fonds voor Wetenschappelijk Onderzoek (FWO) – Vlaanderen, the Geconcerteerde Onderzoeksacties (GOA) – Vlaamse Gemeenschap, and ISEP/FORTIS.

Experimental Part

Chemistry

Melting points were determined on a Totolli capillary apparantus and are uncorrected. Elemental analyses for C, H, N were performed by the Microanalytical Center, ENSCM, Montpellier, France and were in the range of 0.4% of the theoretical values. NMR spectra were recorded on a Bruker DPX 200 spectrometer. Chemical shifts are expressed from TMS using residual $CHCl_3$ at δ 7.30 ppm, and from the central resonance of $CDCl_3$ at δ 77.1 ppm. The following compounds were obtained according to described procedures: 2-phenylimidazo[1,2-*a*]pyridine **2a** [16], 8-methyl-2-phenylimidazo[1,2-*a*]pyridine **2b** [17], 7-methyl-2-phenylimidazo[1,2-*a*]pyridine **2c** [17], 6-methyl-2-phenylimidazo[1,2-*a*]pyridine **2d** [17], 8-benzyloxy-2-phenylimidazo[1,2-*a*]pyridine **2e** [18], 2-bromoacetylpyridine **3a** [19], 2-bromo-1-(naphth-2-yl)ethanone **3b** [20], 7-methyl-2-(biphen-4-yl)imidazo-[1,2-*a*]pyridine **4c** [21], 7-methyl-2-(thien-2-yl)imidazo[1,2-*a*]pyridine **4d** [22], 7-methyl-2-(fur-2-yl)imidazo[1,2-*a*]pyridine **4e** [23], 3-nitroso-2-phenylimidazo[1,2-*a*]pyridine **5** [24], 7-methyl-3-nitroso-2-phenylimidazo[1,2-*a*]pyridine **7** [12], and 8-benzyloxy-3-nitroso-2-phenylimidazo[1,2-*a*]pyridine **9** [25]. Original ^{1}H NMR for previously reported compounds are given in Table 2.

General Procedure for Cyclisation

Mixtures of 2-aminopyridine or substituted derivatives (10 mmol) and phenacylbromide (1.99 g, 10 mmol) or aryl/heteroaryl-α-bromoethanone (10 mmol) in ethanol (90 mL) were refluxed for 5 (**4a**) or 8 hours (**4b**). After cooling, the mixtures were evaporated to dryness, the residues taken up in water, made basic with sodium carbonate and extracted with dichloromethane. The organic layers were dried over calcium chloride, evaporated to dryness and chromatographed.

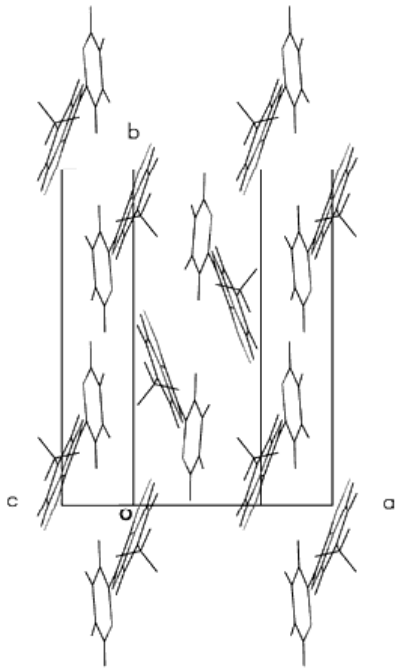


Figure 3a. Projections of **14** on the (x0y) plane.

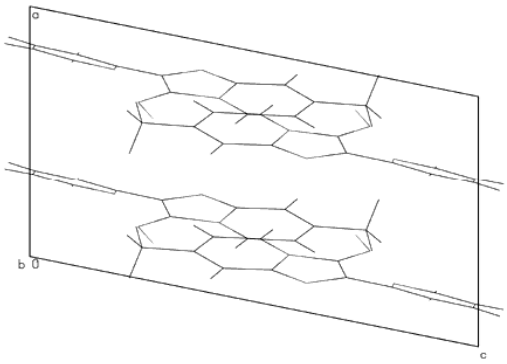


Figure 3b. Projections of **14** on the (x0z) plane.

*7-Methyl-2-(pyridin-2-yl)imidazo[1,2-a]pyridine **4a***

This compound was obtained from 2-amino-4-picoline **1c** and **3a** in 48% yield ; mp 183–185 °C; ^{1}H NMR ($CDCl_3$, 200 MHz); δ = 8.63 (d, $J_{5',6'}$ = 4.8 Hz, 1H, H-6′), 8.19 (d, $J_{3',4'}$ = 7.6 Hz, 1H, H-3′), 8.18 (s, 1H, H-3), 8.03 (d, $J_{5,6}$ = 6.9 Hz, 1H, H-5), 7.78 (td, $J_{3',4'}$ = $J_{4',5'}$ = 7.6 Hz, $J_{4',6'}$ = 1.8 Hz, 1H, H-4′), 7.40 (br.s, 1H, H-8), 7.22 (ddd, $J_{4',5'}$ = 7.6 Hz, $J_{5',6'}$ = 4.8 Hz, $J_{3',5'}$ = 1 Hz, 1H, H-5′), 6.63 (dd, $J_{5,6}$ = 6.9 Hz, $J_{6,8}$ = 1.4 Hz, 1H, H-6), 2.44 (s, 3H, CH_3) ; ^{13}C NMR ($CDCl_3$, 50 MHz); d = 153.1 (C-2′), 149.4 (C-6′), 146.1 (C-2*), 145.9 (C-8a*), 136.8 (C-4′), 133.8 (C-7), 125.1 (C-5), 122.5 (C-5′), 120.4 (C-3′), 116.1 (C-8), 115.4 (C-6), 110.3 (C-3), 21.4 (CH_3).

*7-Methyl-2-(naphth-2-yl)imidazo[1,2-a]pyridine **4b***

This compound was obtained from 2-amino-4-picoline **1c** and **3b** in 54% yield ; mp 200°C ; ^{1}H NMR ($CDCl_3$, 200 MHz); δ = 8.50 (br.s, 1H, H-1′), 7.96 (dd, $J_{3',4'}$ = 8.6 Hz, $J_{1',3'}$ = 1.7 Hz, 1H, H-3′), 7.87 (m, 4H, H-5, 4′, 5′, 8′), 7.80 (s, 1H, H-3), 7.49 (m, 2H, H-6′, 7′), 7.42 (br.s, 1H, H-8), 6.54 (dd, $J_{5,6}$ = 6.9 Hz, $J_{6,8}$ = 1.6 Hz, 1H, H-6), 2.38 (s, 3H, CH_3) ; ^{13}C NMR ($CDCl_3$, 50 MHz); = 146.2 (C-8a*), 145.2 (C-2*), 135.9 (C-8a′), 133.7 (C-4a′), 133.1 (C-7), 131.2 (C-2′), 128.3 (C-5′*), 128.2 (C-8′*), 127.7 (C-4′), 126.2 (C-7′*), 125.9 (C-6′*), 124.8 (C-5), 124.5 (C-1′), 124.1 (C-3′), 115.7 (C-8), 115.1 (C-6), 108.0 (C-3), 21.4 (CH_3).

Table 2. Original ^{1}H NMR for previously reported compounds.

Compounds	H-3	H-5	H-6	H-7	H-8	CH_3 or CH_2	Others		
2b	7.83 s	8.02 dd $J_{5,6}$ = 6.8 Hz $J_{5,7}$ = 1 Hz	6.68 t $J_{5,6}$ = 6.8 Hz $J_{6,7}$ = 6.8 Hz	6.96 dd $J_{6,7}$ = 6.8 Hz $J_{5,7}$ = 1 Hz	–	2.70 s	7.40 m H-3',4',5'	7.99 m H-2',6'	
2c	7.81 s	7.97 d $J_{5,6}$ = 6.8 Hz	6.64 dd $J_{5,6}$ = 6.8 Hz $J_{6,8}$ = 1.6 Hz	–	7.47 br.s	2.43 s	7.43 m H-3',4',5'	8.02 m H-2',6'	
2d	7.81 s	7.97 d $J_{5,7}$ = 1.7 Hz	–	7.05 dd $J_{7,8}$ = 9.2 Hz $J_{5,7}$ = 1.7 Hz	7.56 d $J_{7,8}$ = 9.2 Hz	2.36 s	7.40 m H-3',4',5'	7.93 m H-2',6'	
4c	7.85 s	8.04 m	6.65 dd $J_{5,6}$ = 7 Hz $J_{6,8}$ = 1.8 Hz	–	7.53 br.s	2.44 s	7.53 m H-3'',4'',5''	7.70 m H-3',5',2'',6''	8.04 m H-2',6'
4d	7.70 s	7.96 d $J_{5,6}$ = 7 Hz	6.62 dd $J_{5,6}$ = 7 Hz $J_{6,8}$ = 1.6 Hz	–	7.39 br.s	2.40 s	7.11 dd H-4' $J_{4',5'}$ = 5 Hz $J_{3',4'}$ = 3.6 Hz	7.31 dd H-5' $J_{4',5'}$ = 5 Hz $J_{3',5'}$ = 1.2 Hz	7.47 dd H-3' $J_{3',4'}$ = 3.6 Hz $J_{3',5'}$ = 1.2 Hz
4e*	7.58 s	7.80 d $J_{5,6}$ = 6.9 Hz	6.42 dd $J_{5,6}$ = 6.9 Hz $J_{6,8}$ = 1.5 Hz	–	7.29 br.s	2.28 s	6.51 dd H-4' $J_{3',4'}$ = 3.3 Hz $J_{4',5'}$ = 1.8 Hz	6.90 d H-3' $J_{3',4'}$ = 3.3 Hz	7.48 d H-5' $J_{4',5'}$ = 1.8 Hz
7	–	9.84 d $J_{5,6}$ = 6.8 Hz	7.10 dd $J_{5,6}$ = 6.8 Hz $J_{6,8}$ = 1.4 Hz	–	7.58 m	2.56 s	7.58 m H-3',4',5'	8.68 m H-2',6'	
9*	7.80 s	7.69 dd $J_{5,6}$ = 6.6 Hz $J_{5,7}$ = 1 Hz	6.55 dd $J_{6,7}$ = 7.6 Hz $J_{5,6}$ = 6.6 Hz	6.43 dd $J_{6,7}$ = 7.6 Hz $J_{5,7}$ = 1 Hz	–	5.42 s	7.40 m H-3',4',5' H-3'',4'',5''	7.55 m H-2'',6''	8.05 m H-2',6'

* reported in DMSO-D_6.

General Procedure for Nitrosation

To a solution of imidazo[1,2-*a*]pyridine derivative **2a–e** or **4a–e** (2.5 mmol) in acetic acid was slowly added a solution of sodium nitrite (25 mmol) in water (7 mL). The mixture was stirred for 30 minutes. The green precipitate was collected to give the pure nitroso derivative which was dried in an oven. In some cases chromatography on neutral alumina eluted with dichloromethane was necessary.

8-Methyl-3-nitroso-2-phenylimidazo[1,2-a]pyridine ***6***

^{1}H NMR ($CDCl_3$, 200 MHz); δ = 9.83 (d, $J_{5,6}$ = 7 Hz, 1H, H-5), 8.72 (m, 2H, H-2′, 6′), 7.60 (m, 4H, H-7, 3′, 4′, 5′), 7.17 (t, $J_{5,6}$ = $J_{6,7}$ = 7 Hz, 1H, H-6), 2.78 (s, 3H, CH_3).

6-Methyl-3-nitroso-2-phenylimidazo[1,2-a]pyridine ***8***

^{1}H NMR ($CDCl_3$, 200 MHz); δ = 9.89 (br.s, 1H, H-5), 8.66 (m, 2H, H-2′,6′), 7.77 (m, 2H, H-7, 8), 7.62 (m, 3H, H-3′,4′,5′), 2.49 (s, 3H, CH_3).

7-Methyl-2-(naphth-2-yl)-3-nitrosoimidazo[1,2-a]pyridine ***10***

^{1}H NMR ($CDCl_3$, 200 MHz); δ = 9.91 (d, 1H, $J_{5,6}$ = 6.8 Hz, H-5), 9.32 (br.s, 1H, H-1′), 8.76 (dd, $J_{3',4'}$ = 8.7 Hz, $J_{1',3'}$ = 1.7 Hz, 1H, H-3′), 7.91–8.09 (m, 3H, H-4′, 5′, 8′), 7.66 (br.s, 1H, H-8), 7.57–7.63 (m, 2H, H-6′, 7′), 7.10 (dd, $J_{5,6}$ = 6.8 Hz, $J_{6,8}$ = 1.6 Hz, 1H, H-6), 2.57 (s, 3H, CH_3).

2-(Biphen-4-yl)-7-methyl-3-nitrosoimidazo[1,2-a]pyridine ***11***

^{1}H NMR ($CDCl_3$, 200 MHz); δ = 9.88 (d, $J_{5,6}$ = 6.8 Hz, 1H, H-5), 8.79 (d, *J* = 8.6 Hz, 2H, H-2′, 6′), 7.71–7.84 (m, 4H, H-3′, 5′, 2″, 6″), 7.65 (br.s, 1H, H-8), 7.43–7.56 (m, 3H, H-3″, 4″, 5″), 7.11 (dd, $J_{5,6}$ = 6.8 Hz, $J_{6,8}$ = 1.4 Hz, 1H, H-6), 2.58 (s, 3H, CH_3).

7-Methyl-3-nitroso-2-(thien-2-yl)imidazo[1,2-a]pyridine ***12***

^{1}H NMR ($CDCl_3$, 200 MHz); δ = 9.74 (d, 1H, $J_{5,6}$ = 6.8 Hz, H-5), 8.46 (dd, $J_{3',4'}$ = 3.8 Hz, $J_{3',5'}$ = 1.2 Hz, 1H, H-3′), 7.73 (dd, $J_{4',5'}$ = 5 Hz, $J_{3',5'}$ = 1.2 Hz, 1H, H-5′), 7.52 (br.s, 1H, H-8), 7.27 (dd, $J_{4',5'}$ = 5 Hz, $J_{3',4'}$ = 3.8 Hz, 1H, H-4′), 7.02 (dd, $J_{5,6}$ = 6.8 Hz, $J_{6,8}$ = 1.5 Hz, 1H, H-6), 2.52 (s, 3H, CH_3).

2-(Fur-2-yl)-7-methyl-3-nitrosoimidazo[1,2-a]pyridine ***13***

^{1}H NMR ($CDCl_3$, 200 MHz); δ = 9.80 (d, $J_{5,6}$ = 6.8 Hz, 1H, H-5), 7.88 (dd, $J_{4',5'}$ = 1.7 Hz, $J_{3',5'}$ = 0.7 Hz, 1H, H-5′), 7.85 (d, $J_{3',4'}$ = 3.5 Hz, 1H, H-3′), 7.61 (br.s, 1H, H-8), 7.11 (dd, $J_{5,6}$ = 6.8 Hz, $J_{6,8}$ = 1.5 Hz, 1H, H-6), 6.72 (dd, $J_{3',4'}$ = 3.5 Hz, $J_{4',5'}$ = 1.7 Hz, 1H, H-4′), 2.58 (s, 3H, CH_3).

7-Methyl-3-nitroso-2-(pyridin-2-yl)imidazo[1,2-a]pyridine ***14***

^{1}H NMR ($CDCl_3$, 200 MHz); δ = 9.84 (d, $J_{5,6}$ = 6.9 Hz, 1H, H-5), 8.98 (d, $J_{5',6'}$ = 4.6 Hz, 1H, H-6′), 8.90 (d, $J_{3',4'}$ = 7.8 Hz, 1H, H-3′), 7.94 (td, $J_{4',5'}$ = $J_{3',4'}$ = 7.8 Hz, $J_{4',6'}$ = 1.9 Hz, 1H, H-4′), 7.76 (br.s, 1H, H-8), 7.54 (ddd, $J_{4',5'}$ = 7.8 Hz, $J_{5',6'}$ = 4.6 Hz, $J_{3',5'}$ = 1 Hz, 1H, H-5′), 7.19 (dd, $J_{5,6}$ = 6.9 Hz, $J_{6,8}$ = 1.5 Hz, 1H, H-6), 2.61 (s, 3H, CH_3).

X-Ray Diffraction

Green plate crystals were grown by slow evaporation of ethanol/chloroform solutions. The crystal used for X-ray measurement was lamellar, with dimensions: 0.025×0.1×0.2 mm. The studied compound, $C_{13}H_{10}N_4O$, Mx =

228.25 g.mole^{-1}, crystallized in the monoclinic system, space group $P2_1/a$ (Z = 4). The unit cell parameters were obtained by a least-squares fit of the setting angles of 25 reflections with θ between 15 and 35° and are as follows : a = 7.155(3), b = 12.106(3), c = 13.022(4), and β = 101.37(3) with a cell volume of 1105.8(6) Å^3.The calculated density is equal to 1.431 g.cm^{-3}. The linear absorption coefficient was μ = 0.7585 mm^{-1} for the CuKα radiation and an ω–2θ scan.

The diffracted intensities were collected with a CAD-4 Enraf-Nonius diffractometer equipped with a graphite monochromator for θ_{max} = 60: $0 \leq h \leq 7, 0 \leq k \leq 13, -14 \leq l \leq 14$. Three standard reflections were used to monitor the data and to detect any decrease of intensity (–4 3 1, –1 –4 5, 1 4 5). There were 1598 independent reflections of which 797 were considered as observed (I>2σ (I) and R_{int} = 0.0206).

The crystal structure was solved and refined using the SHELX97 program [26]. Scattering factors were taken from the International Tables for Crystallography [27]. The hydrogen atoms were introduced in their theoretical positions and allowed to ride with the atoms to which they are attached; the refinement was then resumed. The final reliability factors were: R = 0.059, wR = 0.129 and the goodness of fit was equal to 0.967. The weight was equal to: $w = 1/[\sigma^2(F_o^2) + (0.0787P)^2 + 0.1435P]$ where $P = (F_o^2 + 2F_c^2)/3$. The minimum and maximum residual density were equal –0.231 and 0.239 e.Å^{-3} respectively.

Inhibition of HIV-1-Induced Cytopathicity in MT-4 and CEM Cells

The methodology of the anti-HIV assays has been described previously. Briefly, human MT-4 (ca. 4×10^5 cell mL^{-1}) cells were infected with 100 $CCID_{50}$ of HIV-1 (III_B)/mL or HIV-2(ROD)/mL and seeded in 200-μl wells of a microtiter plate, containing appropriate concentrations of the test compounds. After 5 days of incubation at 37°C, the number of viable (MT-4) cells was determined by an automated MTT dye staining of living cells.

References

[1] J. Cohen, *Science* **1997**, *277*, 32–33; J. K. Wong, M. Hezareh, H. F. Gunthard, D. V. Havlir, C. C. Ignacio, C. A. Spina, D. D. Richman, *Science* **1997**, *278*, 1291–1295; D. Finzi, M. Hermankova, T. Pierson, L. Canuth, C. Buck, R.-E. Chaisson, T. C. Quinn, K. Chadwick, J. Margolick, R. Brookmayer, J. Gallant, M. Markowitch, D. D. Ho, D. D. Richman, R. F. Silicano, *Science* **1997**, *278*, 1295–1300.

[2] J. L. Darlix, M. Lapadat-Tapolsky, H. de Rocquigny, B. P. Roques, *J. Mol. Biol.* **1995**, *254*, 523–537.

[3] T. L. South, P. R. Blake, R. C. Sowder, L. O. Arthur, L. E. Henderson, M. F. Summer, *Biochemistry* **1990**, *29*, 7786–7789; T. L. South, M. F. Summers, *Adv. Inorg. Biochem. Ser*. **1990**, *8*, 199–248.

[4] A. Rein, D. E. Ott, J. Mirro, L. O. Arthur, W. G. Rice, L. E. Henderson, *J. Virol*. **1996**, *70*, 4966–4972.

[5] [a] W. G. Rice, C. A. Schaeffer, B. Harten, F. Villinger, T. L. South, M. F. Summers, L. E. Henderson, J. W. Bess Jr, L. O. Arthur, J. S. McDougal, S. L. Orloff, J. Mendeleyev, E. Kun, *Nature* **1993**, *361*, 473–475 ; [b] W. G. Rice, C. A. Schaeffer, L. Graham, M. Bu, J. S. McDougal, S.L. Orloff, F. Villinger, M. Young, S. Oroszlan, M. R. Fesen, Y. Pommier, J. Mendeleyev, E. Kun, *Proc. Natl. Acad. Sci. USA* **1993**, *90*, 9721–9724.

[6] [a] W. G. Rice, J. G. Supko, L. Malspeis, R. W. Buchheit Jr., D.J. Clanton, M. Bu, L. Graham, C. A. Schaeffer, J. A. Turpin, J. Domagala, R. D. Gogliotti, J. P. Bader, S. M. Halliday, L. Coren, R. C. Sowder II, L. O. Arthur, L. E. Henderson, *Science* **1995**, *270*, 1194–1197 ; [b] J. M. Domagala, J. P. Bader, R. D. Gogliotti, J.P. Sanchez, M. A. Stier, Y. Song, J. V. N. Vara Prasad, P. J. Tummino, J. Scholten, P. Harvey, T. Holler, S. Gracheck, D. Hupe, W. G. Rice, R. Schultz, *Bioorg. Med. Chem.* **1997**, *5*, 569–579 ; [c] J. M. Domagala, R. D. Gogliotti, J. Sanchez, M. A. Stier, K. Musa, Y. Song, J. Loo, M. Reily, P. Tummino, P. Harvey, D. Hupe, L. Sharmeen, D. Mack, J. Scholten, J. Saunders, T. McQuade, *Drug Design Discov*., **1997**, *15*, 49–61 ; [d] J. V. N. Vara Prasad, J. A. Loo, F. E. Boyer, M. A. Stier, R. D. Gogliotti, W. J. Turner, P. J. Harvey, M. R. Kramer, D. P. Mack, J. D. Scholten, S. J. Gracheck, J. M. Domagala, *Bioorg. Med. Chem*. **1998**, *6*, 1707–1730.

[7] W. G. Rice, D. Baker, C. A. Schaeffer, L. Graham, M. Bu, S. Terpening, D. Clanton, R. Schultz, J. P. Bader, R. W. Buckheit Jr., L. Field, P. K. Singh, J. A. Turpin, *Antimicrob. Agents Chemother*. **1997**, *41*, 419–426.

[8] [a] M. Vandevelde, M. Witvrouw, J. C. Schmit, S. Sprecher, E. de Clercq, J.P. Tassignon, *AIDS Res. Hum. Retro. Vir*. **1996**, *12*, 567–568 ; [b] W. G. Rice, J. A. Turpin, M. J. Huang, D. Clanton, R. W. Buckhert Jr., D. G. Covell, A. Wallqvist, N. Mc Donnel, R. N. de Guzman, M. F. Summers, L. Zalkow, J. P. Bader, R. D. Haugwitz, E. A. Sausville, *Nat. Med*. **1997**, *3*, 341–345.

[9] J. A. Turpin, Y. Song, J. K. Inman, M. Huang, A. Wallqvist, A. Maynard, D. G. Covell, W. G. Rice, E. Appella, *J. Med. Chem*. **1999**, *42*, 67–86.

[10] S. Druillennec, H. Meudal, B. P. Roques, M. C. Fournié-Zaluski, *Bioorg. Med. Chem. Lett*. **1999**, *9*, 627–632.

[11] A. Gueiffier, M. Lhassani, A. Elhakmaoui, R. Snoeck, G. Andrei, O. Chavignon, J. C. Teulade, A. Kerbal, E. M. Essassi, J. C. Debouzy, M. Witvrouw, Y. Blache, J. Balzarini, E. de Clercq, J. P. Chapat, *J. Med. Chem*. **1996**, *39*, 2856–2859; A. Gueiffier, S. Mavel, M. Lhassani, A. Elhakmaoui, R. Snoeck, G. Andrei, O. Chavignon, J. C. Teulade, M. Witvrouw, J. Balzarini, E. de Clercq, J. P. Chapat, *J. Med. Chem*. **1998**, *41*, 5108–5112.

[12] J. C. Teulade, R. Escale, H. Viols, J. P. Chapat, G. Grassy, A. Carpy, J. M. Leger, *J. Chem. Soc. Perkin Trans. I* **1983**, 2663–2667.

[13] A. M. Roe, *J. Chem. Soc*. **1963**, 2195–2200.

[14] J. C. Teulade, A. Gueiffier, H. Viols, J. P. Chapat, G. Grassy, B. Perly, G. Dauphin, *J. Chem. Soc. Perkin Trans. I* **1989**, 1895–1899.

[15] A. L. Spek, Platon99, Program for Drawing Crystal and Molecular Diagrams, (1999), Univ. Of Ultrecht, Netherlands.

[16] N. Campbell, E.B. MacCall, *J. Chem. Soc*. **1951**, 2411–2416.

[17] F. Mattu, E. Marangiu, *Ann. Chim. (Rome)* **1964**, *54*, 496–509.

[18] R. Rydzkowski, D. Blondeau, H. Sliwa, *Tetrahedron Lett*. **1985**, *26*, 2571–2574.

[19] R. Menasse, G. Klein, H. Erlenmeyer, *Helv. Chim. Acta* **1955**, *38*, 1289–1291.

[20] A. Willemart, *Ann. Chim. (Paris)* **1929**, *12*, 345–422.

[21] Ng. Ph. Buu-Hoi, Ng. Hoan, R. Royer, *Bull. Soc. Chim. Fr*. **1950**, 489–493.

[22] Ng. Ph. Buu-Hoi, Ng. Hoan, *Rec. Trav. Chim. Pays-Bas* **1949**, *68*, 441–472.

[23] P. R. Loiseau, M. Payard, G. Grassy, Z. Dornelles Pinto, C. Advenier, J. P. Gnassounou, Y. Adam, *Eur. J. Med. Chem*. **1987**, *22*, 457–462.

[24] C. Párkányi, A. O. Abdelhamid, J. C. S. Cheng, A. S. Shawali, *J. Heterocyclic Chem*. **1984**, *21*, 1029–1032.

[25] R. Rydzkowski, D. Blondeau, H. Sliwa, *J. Chem. Res. (M)* **1986**, *11*, 3368–3390.

[26] G. M. Sheldrick, Program for the determination and the refinement of crystal structures, Univ. Gottingen, Germany **1997**.

[27] International Tables for X-Ray Crystallography, Kynoch Press, Birmingham, 1974, Vol. IV.

Received: May 7, 2001 [FP572]