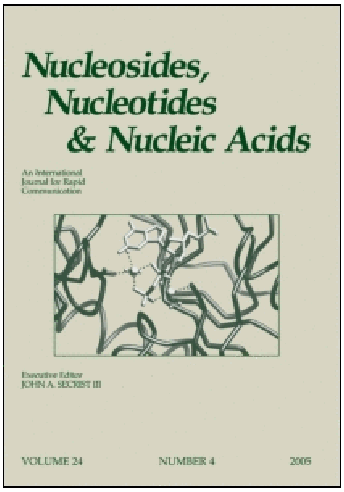


Nucleosides, Nucleotides and Nucleic Acids
Publication details, including instructions for authors and subscription information:
http://www.informaworld.com/smpp/title~content=t713597286

Design and Synthesis of Heterocyclic Carboxamides as Natural Nucleic Acid Base Mimics

Donald E. Bergstrom[ab]; Peiming Zhang[ab]; W. Travis Johnson[ab]
[a] Walther Cancer Institute, Indianapolis, Indiana [b] Department of Medicinal Chemistry and Pharmacognosy, Purdue University, West Lafayette, Indiana

To cite this Article Bergstrom, Donald E. , Zhang, Peiming and Johnson, W. Travis(1996) 'Design and Synthesis of Heterocyclic Carboxamides as Natural Nucleic Acid Base Mimics', Nucleosides, Nucleotides and Nucleic Acids, 15: 1, 59 — 68

To link to this Article: DOI: 10.1080/07328319608002370

URL: http://dx.doi.org/10.1080/07328319608002370

NUCLEOSIDES & NUCLEOTIDES, 15(1-3), 59-68 (1996)

DESIGN AND SYNTHESIS OF HETEROCYCLIC CARBOXAMIDES AS NATURAL NUCLEIC ACID BASE MIMICS

Donald E. Bergstrom*, Peiming Zhang, and W. Travis Johnson
Walther Cancer Institute, Indianapolis, Indiana 46208 and Department of Medicinal Chemistry and Pharmacognosy, Purdue University, West Lafayette, Indiana 47907.

Abstract: A series of heterocyclic carboxamides have been designed as mimics for the natural nucleic acid bases. The nucleosides 1-(2′-deoxy-β-D-ribofuranosyl)imidazole-4-carboxamide (**1**), 1-(2′-deoxy-β-D-ribofuranosyl)pyrazole-3-carboxamide (**2**), and 1-(2′-deoxy-β-D-ribofuranosyl)pyrrole-3-carboxamide (**3**) were synthesized and their structures confirmed by spectroscopic and analytical means.

Since an initial report by Ohtsuka et al. in 1985 [1], 2′-deoxyinosine (dI) has been widely employed as a universal nucleoside at ambiguous position in DNA primers and probes. However, dI does not base-pair identically with each of the four naturally occurring nucleosides[2,3] but shows a significant preference for pairing with dC[4]. This preference is to be expected because of the close structural resemblance to guanosine, from which it differs only by the absence of the NH_2 at C-2. The base-pairs G:I and T:I are significantly less stable than C:I and A:I base-pairs[2,3]. Crystal structures have been determined at high resolution for B-DNA duplexes containing C:I, A:I, and I:T base-pairs[4,5,6]. Although the C:I base-pair causes little perturbation in helix structure, A:I, I:T, and G:I base-pairs effect the integrity of the helix more adversely. A(*anti*):I(*anti*), AH^+(*anti*):I(*syn*), and A(*syn*):I(*anti*) have been observed by either X-ray crystallography or NMR[7,8]. In the base-pair with guanosine the preferred form is apparently I(*syn*):G(*anti*). I:T base-pairs assume a "wobble" conformation[6].

It is conceivable that inosine could be altered to decrease the amount of duplex structure reorganization that is required when one base-pair is forced to assume a wobble conformation, or when two purine bases are placed opposite one another. When two

---

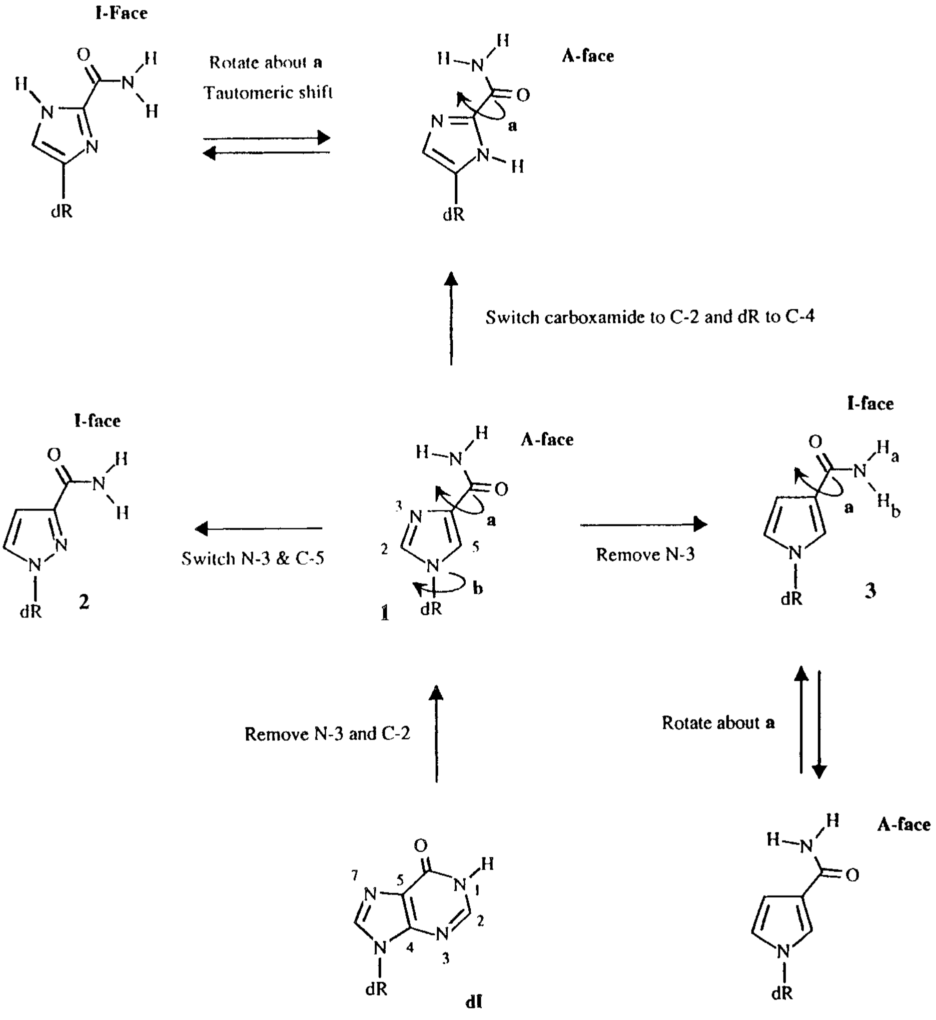
This paper is dedicated to Dr. Yoshihisa Mizuno.

purines base-pair in a normal B-DNA duplex, and both nucleosides are in the *anti* conformation, then the phosphodiester back bone must be expanded to accommodate the greater spacing between the C-1′ ′s of each nucleotide. Alternatively one of the two purine bases may assume a *syn* conformation which reduces extensive helix distortion. By removing the conformational restriction about the glycosidic bond (C-1′ to N-9) and between C-5 and C-6 one could conceivably generate a species with greatly enhanced flexibility for base-pairing. This could be achieved by opening the six-membered ring between N-3 and C-4. The most abbreviated form of dI, obtained by eliminating both C-2 and N-3, would be 1-(2′-deoxy-β-D-ribofuranosyl)imidazole-4-carboxamide (**1**). Although this structural modification should eliminate any constraints for conformational preference about the glycosidic bond, it is unlikely that the amide group would have similar conformational freedom. In particular, one would predict that the amide group should preferentially assume the A-face conformation shown in Figure 1. This conformational preference could be significantly altered by either removing N-3 (as in pyrrole-3-carboxamide, **3**) or changing the position of the nitrogen (as in pyrazole-3-carboxamide, **2**)[9]. Alternatively, the position of attachment of the imidazole heterocycle to the deoxyribose moiety and the amide position can be altered to achieve greater flexibility in H-bonding[10,11].

In this paper we describe the synthesis of a set of three nucleosides designed to allow us to explore the effect of heterocyclic structure on conformational preference of the attached carboxamide group and base-pairing in duplex DNA.

RESULTS AND DISCUSSION

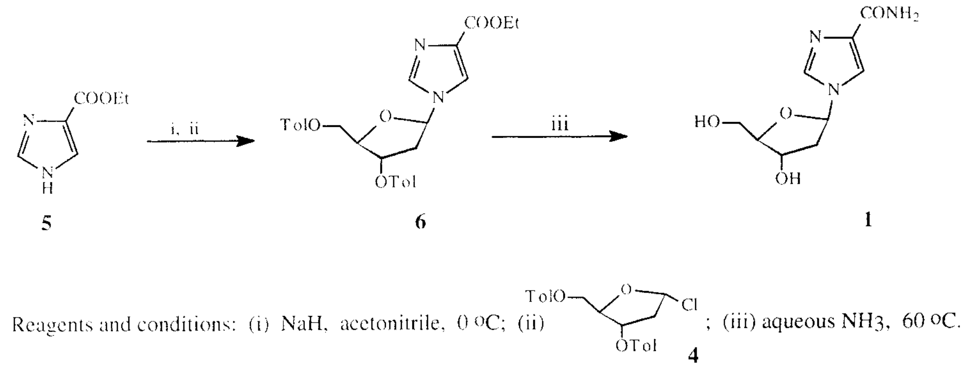
The synthesis of 1-(2′-deoxy-β-D-ribofuranosyl)imidazole-4-carboxamide (**1**) is summarized in Scheme 1. Compound **6** was synthesized by a slight modification of the sodium salt glycosylation procedure[12]. The sodium salt of ethyl imidazole-4-carboxylate (**5**)[13] was treated with 1-chloro-2-deoxy-3,5-di-*O*-*p*-toluoyl-α-D-*erythro*-pentofuranose (**4**)[14] at 0 °C to give the 1,4 disubstituted imidazole derivative **6** in 69% overall yield. The anomeric configuration of **6** was assigned as β based on the analysis of ^{1}H NMR signals for the anomeric and 2′ protons[15]. The presence of the α anomer was not detected when the reaction was performed at 0 °C. When this reaction was performed at ambient temperature the α isomer, which gave an ^{1}H NMR spectra in agreement with those previously published for other α nucleosides[15], was also obtained (data not shown). The regiochemical assignment of **6** was determined based on a comparison of cross-ring coupling constants to those published for other 1,4 and 1,5-disubstituted imidazoles[16]. The treatment of **6** with hot aqueous ammonia gave the desired free nucleoside **1** in 85% yield.



dR = 2'-deoxy-β-D-ribofuranosyl

A-face	Carboxamide mimics H-bonding pattern of A
I-Face	Carboxamide mimics H-bonding pattern of I (or G)

Figure 1. Carboxamide substituted heterocycles as A and I(G) mimics

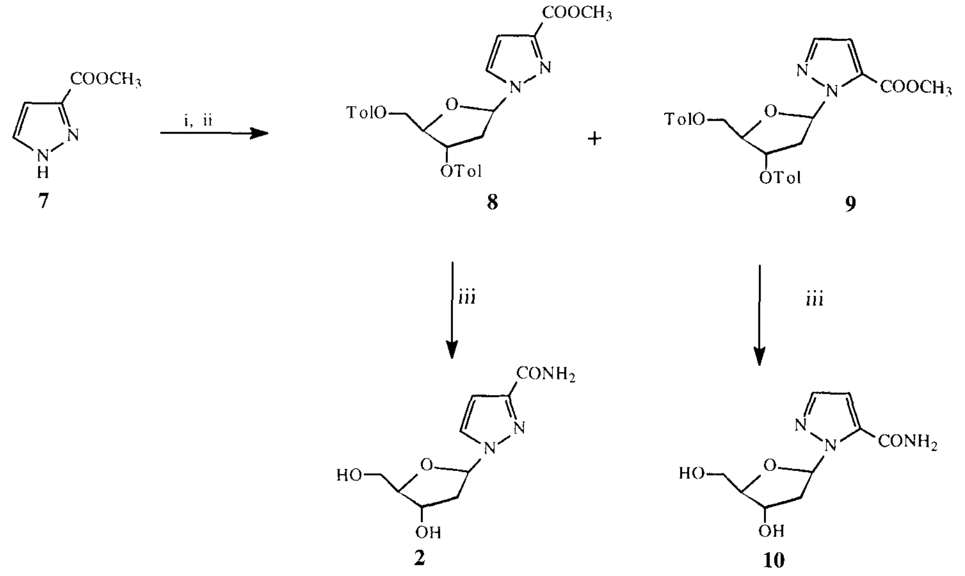


Reagents and conditions: (i) NaH, acetonitrile, 0 °C; (ii) 4; (iii) aqueous NH_3, 60 °C.

Scheme 1

The synthesis of 1-(2′-deoxy-β-D-ribofuranosyl)pyrazole-3-carboxamide (**2**) is outlined in Scheme 2. Methyl pyrazole-3-carboxylate (**7**) was prepared by the 1,3-dipolar cycloaddition of CH_2N_2 with methyl propiolate[17]. The sodium salt of **7** was generated *in situ* and reacted with **4** at ambient temperature to give two regioisomeric products. The 1,3-disubstituted product (**8**) was isolated as the major product in 51% overall yield, while the 1,5-disubstituted product (**9**) was obtained in 38% yield. Both products were assigned β configurations on the basis of their ^1H NMR spectra. The presence of α anomeric products were not detected. The regiochemical assignments of **8** and **9** were also accomplished by analysis of their ^1H NMR spectra. The signal for the anomeric proton of **9** (7.10 ppm) is shifted downfield compared to **8** (6.42 ppm), which is in agreement with known spectra of other 1,5- and 1,3-disubstituted pyrazole nucleosides[18], respectively. Compound **8** was heated at 110 °C in methanolic ammonia to give 1-(2′-deoxy-β-D-ribofuranosyl)pyrazole-3-carboxamide (**2**) in 80% yield. Similarly, the treatment of **9** with methanolic ammonia gave 1-(2′-deoxy-β-D-ribofuranosyl)pyrazole-5-carboxamide (**10**) in 76% yield.

1-(2′-Deoxy-β-D- ribofuranosyl)pyrrole-3-carboxamide (**3**) was synthesized according to the procedure reported by Robins and coworkers[19]. The product was identical to that prepared by Robins et al., as evidenced by ^1H NMR and mass spectrometry. As predicted by semi-empirical calculations [9] and shown in Figure 1, nucleoside **1** should prefer the A-face conformation in which one of the amide protons interacts with the lone pair electrons on N-3. In a similar fashion nucleoside **2** should prefer the I-face conformation in which one amide proton interacts with the lone pair electrons on N-2. In contrast, neither of the amide conformations for pyrrole 3-carboxamide deoxyribonucleoside (**3**) involve an



Reagents and conditions: (i) NaH, acetonitrile; (ii) **4**; (iii) ammonia / MeOH, 110oC.

Scheme 2

intramolecular H-bond interaction. One would predict a significant difference in the chemical shifts between the intramolecular H-bonded amide N-H of nucleosides **1** and **2**, and the corresponding N-H of nucleoside **3**. In fact, the amide protons of compound **3** appear dramatically different from those of **1** and **2** in the ^{1}H NMR (Figure 2). As shown in figure 2 the chemical shift of the two amide protons for nucleoside **3** fall at 6.70 ppm and 7.28 ppm. The downfield proton, H_a is separated from the upfield proton by 0.58 ppm. This is in marked contrast to the amide protons of nucleosides **1** and **2** which are separated by only 0.22 and 0.20 ppm respectively. In both cases the upfield proton has shifted significantly downfield compared to the corresponding proton on nucleoside **3**, while the downfield protons remain in nearly the same position. Although by no means sufficient to prove the predominance of one of the two rotamers, the NMR data is consistent with the semi-empirical calculations which predict that **1** exists primarily in the A-face rotamer form, while **2** exists primarily in the I-face rotamer form.

In summary, we have described the design and synthesis of three heterocyclic nucleoside analogs. **1**, **2**, and **3** which although superficially appear quite similar, may significantly differ in base-base interactions in a DNA double-helix because of

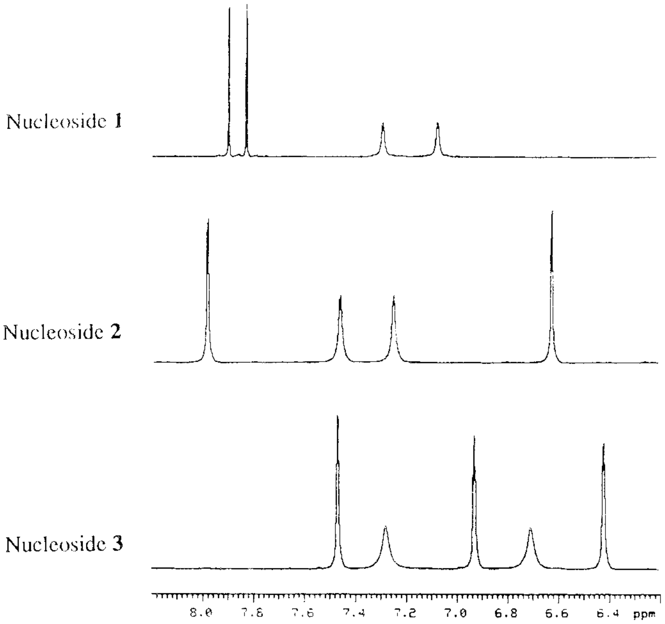


Figure 2. ^{1}H NMR Spectra (DMSO-d_6) of Nucleosides **1-3** Showing Aromatic Ring and Amide Protons

differences in conformational preferences, π-electron density, and molecular dipole. It is probable that the conformational flexibility about the amide group is greater for **3** than for **1** or **2** which would imply potential for less discriminate base-pairing. In fact, the pyrrole derivative (**3**) has shown little discrimination in base-pairing to the natural nucleosides as determined by Tm measurements of oligonucleotides containing this analog[20].

Further studies will be directed at the derivatization of these nucleosides for oligonucleotide synthesis, conformational preferences, physiochemical, and biochemical evaluations of these nucleosides in oligonucleotides.

EXPERIMENTAL

General Information. NMR spectra were obtained on a Varian VXR-500S spectrometer and signals were internally referenced to TMS. FAB mass spectra were recorded by the Mass Spectroscopy Laboratory, Department of Medicinal Chemistry and Pharmacognosy, Purdue University. Elemental analysis was performed by the Microanalysis Laboratory, Department of Chemistry, Purdue University. 2′-

Deoxyribose was purchased from Crystal Chem. Melting points were determined on a Buchi 510 apparatus and are uncorrected. Analytical thin layer chromatography (TLC) was carried out on pre-coated Whatman 60 F_{254} plates. Anhydrous acetonitrile was obtained from Aldrich, all other chemicals were of reagent grade or better quality and used as received.

Ethyl 1-(2′-deoxy-3,5-di-*O*-*p*-toluoyl-β-D-ribofuranosyl)imidazole-4-carboxylate (6). Ethyl imidazole-4-carboxylate (**5**) (0.655 g, 4.68 mmol) was dissolved in anhydrous acetonitrile (15 ml) and stirred under a nitrogen atmosphere until almost complete dissolution had occurred. 95% NaH (0.141 g, 5.62 mmol) was added and the mixture was stirred 10 min. until foaming had ceased. The grey mixture was cooled to 0 °C and 1-chloro-2-deoxy-3,5-di-*O*-*p*-toluoyl-α-D-*erythro*-pentosyl chloride (**4**) (2.18 g, 5.62 mol) was added portionwise over a 5 min. period. The reaction was monitored by TLC (Acetone/Hexane, 3:2) and the mixture stirred for 2 hours. Insoluble salts were removed by filtration through cellulose and the filtrate was evaporated under reduced pressure to a brown oil. The oily mixture was separated by chromatography on silica gel (Ethyl acetate/Hexane) and evaporated under reduced pressure. The residue was dried *in vacuo* over P_2O_5 to give **6** as a white solid (1.59 g, 69%): $R_f = 0.63$ (Hexane/Acetone 2:3); ^{1}HNMR ($CDCl_3$) δ 7.94 (d, *p*-Tol, J = 8.5 Hz, 2 H), 7.89 (d, *p*-Tol, J = 8.5 Hz, 2 H), 7.78 (d, H-2, $J_{2,5}$ = 1.0 Hz, 1 H), 7.74 (d, H-5, $J_{5,2}$ = 1.0 Hz, 1 H), 7.28 (d, *p*-Tol, J = 8.5 Hz, 2 H), 7.25 (d, *p*-Tol, J = 8.5 Hz, 2 H), 6.15 (pseudotriplet, H-1′, J = 13.7 Hz, 1 H), 5.67 (m, H-3′, 1 H), 4.66-4.58 (m, H-4′, H-5′, 3 H), 4.53 (q, OCH_2, J = 7.0 Hz, 2 H), 2.80-2.73 (m, H-2′a, 1 H), 2.70-2.62 (m, H-2′b, 1 H), 2.43 (s, CH_3 *p*-Tol, 3 H), 2.41 (s, CH_3 *p*-Tol, 3 H), 1.34 (t, CH_3, J = 7.0 Hz, 3 H); MS-FAB *m/z* (M+H) 493.2. Anal. Calcd. for $C_{27}H_{28}N_2O_7$: C, 65.84; H, 5.73; N, 5.69. Found: C, 65.60; H, 5.83; N, 5.59.

1-(2′-Deoxy-β-D-ribofuranosyl)imidazole-4-carboxamide (1). Compound **6** (1.5 g, 3.05 mmol) was suspended in 30% ammonium hydroxide (100 ml), sealed in a stainless steel Parr reaction vessel, and heated at 60 °C for 2 days. The resulting solution was evaporated to dryness under reduced pressure. The residue was separated by chromatography on silica gel ($CHCl_3$/MeOH), evaporated under reduced pressure, and dried *in vacuo* over P_2O_5 to give (**1**) as a hygroscopic white foam (0.591 g, 85%): R_f = 0.57 ($CHCl_3$/MeOH 1:1). ^{1}HNMR (DMSO- d_6): δ 7.90 (d, H-2, $J_{2,5}$ = 1.5 Hz, 1 H), 7.83 (d, H-5, $J_{5,2}$ = 1.5 Hz, 1 H), 7.30 (s, NH, 1 H); 7.08 (s, NH, 1 H), 6.05 (pseudotriplet, H-1′, J = 13.5 Hz, 1 H), 5.29 (s, OH-3′, 1 H), 4.96 (s, OH-5′, 1 H); 4.30-4.28 (m, H-3′, 1 H), 3.82-3.80 (m, H-4′, 1 H), 3.53-3.46 (m, H-5′, 2 H); 2.37-2.32 (m, H-2′a, 1 H), 2.25-2.20 (m, H-2′b, 1 H); MS-FAB High Res. Calcd. (M+H): 228.0984. Found: 228.0980. Anal. Calcd. for $C_9H_{13}N_3O_4 \cdot 1/4\ H_2O$: C, 46.65; H, 5.87; N, 18.13. Found: C, 46.40; H, 6.21; N, 17.95.

Methyl pyrazole-3-carboxylate (7). To a freshly prepared solution of diazomethane (14.00 mmol) in ether (50 ml) was added methyl propiolate (0.89 ml, 10.00 mmol). The solution was stirred overnight and evaporated to dryness. The residue was recrystallized in toluene to give **5** as a white solid (1.23 g, 89%): mp 140 °C (lit. 139-140 °C); ^{1}H NMR (DMSO-d_6) δ 7.85 (s, H-5, 1 H), 6.74 (s, H-4, 1 H), 3.78 (s, CH_3, 3 H).

Methyl 1-(2′-deoxy-3,5-di-*O*-*p*-toluoyl-β-D-ribofuranosyl)pyrazole-3-carboxylate (8) and methyl 1-(2′-deoxy-3,5-di-*O*-*p*-toluoyl-β-D-ribofuranosyl)pyrazole-5-carboxylate (9). To a stirred solution of **7** (0.50 g, 3.62 mmol) in anhydrous acetonitrile (25 ml) was added 60% NaH in mineral oil (0.17 g, 4.25 mmol) under a nitrogen atmosphere. Compound **4** (1.55 g, 3.99 mmol) was added after 30 min. The mixture was stirred for 30 minutes, filtered, and the filtrate evaporated to dryness. TLC indicated the residue was a mixture of two major products. They were separated by chromatography on silica gel (Hexane/Ethyl acetate). Compound **9** was obtained from the first fraction as a thick oil (0.422 g, 38%): $R_f = 0.73$ (Hexane/Ethyl acetate 1:1). ^{1}H NMR (DMSO-d_6) δ 7.90 (d, *p*-Tol, $J = 8.5$ Hz, 2 H), 7.82 (d, *p*-Tol, $J = 8.5$ Hz, 2 H), 7.74 (d, H-3, $J_{3,4} = 2.0$ Hz, 1 H), 7.35 (d, *p*-Tol, 2 H), 7.29 (d, *p*-Tol, 2 H), 7.10 (t, H-1′, $J_{1',2'} = 7.0$ Hz, 1 H), 6.96 (d, H-4, $J_{4,3} = 2.0$ Hz, 1 H), 5.84-5.81 (m, H-3′, 1 H), 4.53-4.50 (m, H-4′, 1 H), 4.42 (dd, H-5′, $J_{5'a,5'b} = 12.0$ Hz, $J_{5'a,4'} = 5.0$ Hz, 1 H), 4.29 (dd, H-5′b, $J_{5'a,5'b} = 12.0$ Hz, $J_{5'b,4'} = 5.5$ Hz, 1 H), 3.84 (s, OCH_3, 3 H), 3.24-3.19 (m, H-2′a, 1 H), 2.73-2.67 (m, H-2′b, 1 H), 2.39 (s, CH_3 *p*-Tol, 3 H), 2.36 (s, CH_3 *p*-Tol, 3 H); MS-FAB *m/z* (M+H) 479.0. Anal. Calcd. for $C_{26}H_{26}N_2O_7$: C, 65.28; H, 5.48; N, 5.85. Found: C, 65.48; H, 5.41; N, 5.48. Compound **8** was obtained as a thick oil (0.760 g, 51%): $R_f = 0.62$ (Hexane/Ethyl acetate 1:1). ^{1}H NMR (DMSO-d_6) δ 8.11 (d, H-5, $J_{5,4} = 2.5$ Hz, 1 H), 7.92 (d, *p*-Tol, $J =$ 8.0 Hz, 2 H), 7.85 (d, *p*-Tol, $J = 8.0$ Hz, H), 7.35 (d, *p*-Tol, $J = 8.0$ Hz, 2 H), 7.29 (d, Tol-H, $J = 8.0$ Hz, 2 H), 6.79 (d, H-4, $J_{4,5} = 2.5$ Hz, 1 H), 6.48 (t, H-1′, $J_{1',2'} = 6.5$ Hz, 1 H), 5.73-5.70 (m, H-3′, 1 H), 4.56-4.53 (m, H-4′, 1 H), 4.48-4.41 (m, H-5′, 2 H), 3.80 (s, OCH_3, 3 H), 3.13-3.08 (m, H-2′a,, 1 H), 2.75-2.70 (m, H-2′b, 1 H), 2.39 (s, CH_3 *p*-Tol, 3 H), 2.37 (s, CH_3 *p*-Tol, 3 H); MS-FAB *m/z* (M+H) 479.0. Anal. Calcd. for $C_{26}H_{26}N_2O_7$: C, 65.28; H, 5.48; N, 5.85. Found: C, 65.28; H, 5.23; N, 5.54.

1-(2′-Deoxy-β-D-ribofuranosyl)pyrazole-3-carboxamide (2). Compound **8** (0.760 g) was added to a solution of 50% ammonia in methanol (70 ml), sealed in stainless-steel Parr reaction vessel, and heated at 110 °C for 18 hours. The resulting solution was evaporated to dryness. The residue was separated by chromatography on silica gel with mixed eluents (Hexane/Ethyl acetate, Hexane/Acetone, and finally Ethyl acetate/Methanol). Compound **2** was obtained as a foam (0.285 g, 80%): ^{1}H NMR (CD_3OD) δ 7.89 (d, H-5, $J_{5,4}$= 2.5 Hz, 1 H), 6.74 (d, H-4, $J_{4,5} = 2.5$ Hz, 1 H), 6.17

(pseudotriplet, H-1′, $J_{1',2'}$ = 6.5 Hz, 1 H), 4.55 -4.52 (m, H-3′, 1 H), 3.98-3.96 (m, H-4′, 1 H), 3.71 (dd, H-5′a, $J_{5'a,5'b}$ = 12.0 Hz, $J_{5'a,4'}$ = 4.5 Hz, 1 H), 3.62 (dd, H-5′b, $J_{5'a,5'b}$ = 12.0 Hz, $J_{5'b,4'}$ = 5.0 Hz, 1 H), 2.75 (ddd, H-2′a, $J_{2'a,2'b}$ = 13.5 Hz, $J_{2'a,1'}$ = $J_{2'a,3'}$ = 6.0 Hz, 1 H), 2.38 (ddd, H-2′b, $J_{2'b,2'a}$ = 13.5 Hz, $J_{2'b,1'}$ = 6.5 Hz, $J_{2'b,3'}$ = 4.5 Hz, 1 H); ^{13}C NMR (CD_3OD) δ 166.72 (C=O), 147.64 (C-3), 132.07 (C-5), 107.61 (C-4), 91.46 (C-4′), 89.31 (C-1′), 72.50 (C-3′), 63.61 (C-5′), 41.29 (C-2′). MS-FAB High Res. Calcd (M+H): 228.0984. Found: 228.0991. Anal. Calcd. for $C_9H_{13}N_3O_4$: C, 47.57; H, 5.77; N, 18.49. Found: C, 47.39; H, 5.91; N, 18.21.

1-(2′-Deoxy-β-D-ribofuranosyl)pyrazole-5-carboxamide (10). Compound **10** was prepared from **9** (0.422 g) by the procedure for the synthesis of **2**. The residue was separated by chromatography on silica gel (Hexane/Ethyl acetate) to afford a white foam (0.152 g, 76%): ^{1}H NMR (CD_3OD) δ 7.55 (d, H-3, $J_{3,4}$ = 2.0 Hz, 1 H), 7.09 (dd, H-1′, $J_{1',2'b}$ = 7.0 Hz, $J_{1',2'a}$ = 6.0 Hz, 1 H), 6.79 (d, H-4, $J_{4,3}$ = 2.0 Hz, 1 H), 4.61-4.58 (m, H-3′, 1 H), 3.98-3.95 (m, H-4′, 1 H), 3.73 (dd, H-5′a, $J_{5'a,5'b}$ = 12.0 Hz, $J_{5'a,4'}$ = 3.5 Hz, 1 H), 3.59 (dd, H-5′b, $J_{5'b,4'}$ =12.0 Hz, $J_{5'b,4'}$ =5.5 Hz, 1 H), 2.80-2.75 (m, H-2′a , 1 H), 2.36 (ddd, H-2′b, $J_{2'a,2'b}$ = 13.5 Hz, $J_{2'b,1'}$ = 7.0 Hz, $J_{2'b,3'}$ = 5.0 Hz, 1 H). ^{13}C NMR (d_6 DMSO) δ 160.95 (C=O), 138.45 (C-3), 136.49 (C-5), 108.68 (C-4), 87.75 (C-4′), 86.00 (C-1′), 71.28 (C-3′), 62.60 (C-5′), 39.10 (C-2′). MS-FAB, *m/z* (M+H) 228.0. Anal. Calcd. for $C_9H_{13}N_3O_4$: C, 47.57; H, 5.77; N, 18.49. Found: C, 47.70; H, 6.00; N, 18.21.

Acknowledgments

The National Institutes of Health is gratefully acknowledged for support of this research.

REFERENCES

1. Ohtsuka, E.; Matsuki, S.; Ikehara, M.; Takahashi, Y.; Matsubara, K. *J. Biol. Chem.* **1985**, *260*, 2605.
2. Kawase, Y.; Iwani, S.; Inone, H.; Miura, K.; Ohtsuka, E. *Nucleic Acids Res.* **1986**, *14*, 7727.
3. Martin, F. H.; Castro, M. M.; Aboul-ela, F.; Tinoco, I., Jr. *Nucleic Acids Res.***1985**, *13*, 8927.
4. Xuan, J. C.; Weber, I. T. *Nucleic Acids Res.* **1992**, *20*, 5457.
5. Corfield, P. W. R.; Hunter, W. N.; Brown, T.; Robinson, P.; Kennard, O. *Nucleic Acids Res.* **1987**, *15*, 7935.
6. Cruse, W. B. T.; Aymani, J.; Kennard, O.; Brown, T.; Jack, A. G. C.; Leonard, G. A. *Nucleic Acids Res.* **1989**, *17*, 55.

7. Uesugi, S.; Oda, Y.; Ikehara, M.; Kawase, Y.; Ohtsuka, E. *J. Biol. Chem.* **1987**, *262*, 6965.

8. Leonard, G. A.; Booth, E. D.; Hunter, W. N.; T, B. *Nucleic Acids Res.* **1992**, *20*, 4753.

9. Using AM-1 we have calculated the enthalpies of formation for 1-methyl-4-imidazole carboxamide, 1-methyl-4-pyrazole carboxamide, and 1-methyl-4-pyrrole carboxamide. These calculations give evidence that C-4 carbamoyl-1-alkyl imidazoles and pyrazoles may prefer an *anti* conformation in the gas phase by 5.5 and 2.9 kcal/mole, respectively. For 1-methyl-4-pyrrole carboxamide the two conformations differ by only 1.3 kcal/mol. Semi-empirical calculations were performed using Hyperchem release 2 on a Gateway 2000 PC utilizing a Intel 486 DX2-66 microprocessor. The geometries of the model compounds were optimized and their enthalpies of formation were calculated in vacuo with a Fletcher-Reeves (conjugate gradient) algorithm to a final RMS gradient of 0.01 kcal/Å mol.

10. Bergstrom, D. E.; Zhang, P. *Tetrahedron Lett.* **1991**, *32*, 6485.

11. Bergstrom, D. E.; Zhang, P.; Zhou, J. *J. Chem. Soc. Perkins Trans. 1* **1994**, 3029.

12. Kazimierczuk, Z.; Cottam, H. B.; Revankar, G. R.; Robins, R. K. *J. Am. Chem. Soc.* **1984**, *106*, 6379.

13. Davis, D. P.; Kirk, K. L.; Cohen, L. A. *J. Heterocycl. Chem.* **1982**, *19*, 253.

14. Bhat, C. C. In *Synthetic Proceedures in Nucleic Acid Chemistry*; W. W. Zorbach and R. S. Tipson, Ed.; Interscience Publishers: NY, 1968; Vol. 1; pp 521.

15. Rousseau, R. J.; Robins, R. K.; Townsend, L. B. *J. Heterocycl. Chem.* **1970**, *7*, 367. Srivastava P. C., Robbins; R. K.; Takusagawa, F.; Berman, H. M. *J. Heterocycl. Chem.* **1981**, 18, 1659.

16. Matthews, H. R.; Rapport, H. *J. Am. Chem. Soc.* ***1973***, *95*, 2297.

17. Manecke, G.; Schemck, H. U. *Tetrahedron Lett.* **1969**, *8*, 617.

18. Earl, R. A.; Panzica, R. P.; Townsend, L. B. *J. Chem. Soc. Perkins Trans. 1* **1972**, 2672.

19. Ramasamy, K.; Robins, R. K.; Revankar, G. R. *Tetrahedron* **1986**, *42*, 5869.

20. Andrews, P.; Bergstrom, D. E.; Zhang, P. Unpublished Results.