Expanding the Genetic Alphabet: Pyrazine Nucleosides That Support a Donor–Donor–Acceptor Hydrogen-Bonding Pattern

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The 6-aminopyrazin-2(1*H*)-one, when incorporated as a pyrimidine-base analog into an oligonucleotide chain, presents a H-bond donor–donor–acceptor pattern to a complementary DNA or RNA strand. When paired with the corresponding acceptor–acceptor–donor purine in oligonucleotides, the heterocycle selectively contributes to the stability of the duplex, presumably by forming a base pair of *Watson–Crick* geometry joined by a nonstandard H-bonding pattern, expanding the genetic alphabet. Reported here is a short, high yielding, *β*-D-selective synthesis of a 6-aminopyrazin-2(1*H*)-one nucleoside *via* the glycine riboside derivative **28**. The key steps include a *Wittig–Horner* reaction of an appropriately protected ribose derivative (*Scheme 10*, **19** → **21**) followed by a *Michael*-like ring closure (*Scheme 12*, **30** → **1a** and **32** → **1b**). Thus, a variety of pyrazine nucleosides (*Scheme 13*) including the target 6-aminopyrazin-2(1*H*)-one riboside **1a**, and its 5-methyl derivative **1b**, 6-amino-5-methylpyrazin-2(1*H*)-one riboside, are obtained.

Introduction. – In its most general form, the *Watson–Crick* base pair joins a six-membered heterocyclic ring (in natural oligonucleotides, a pyrimidine) with a fused five/six-membered ring system (in natural oligonucleotides, a purine) *via* three H-bonds, one that joins the two central ring N-atoms of the paired heterocycles, and two that join flanking exocyclic functional groups (*Fig. 1*). To hold the pair together, H-bond donors in one heterocycle must be opposite H-bond acceptors in the other. With three H-bonds, eight ($=2^3$) H-bonding patterns and 16 independently replicable bases are conceivable within the *Watson–Crick* geometry. Six H-bonding patterns, or 12 independently replicable bases, are readily accessible by using amino and carbonyl functionality (*Fig. 2*) [1–3]. These form the components of an artificially expanded genetic-information system (AEGIS) [4].

In practice, pyrimidine analogs presenting acceptor–donor–donor and donor–donor–acceptor H-bonding patterns are difficult to obtain [2][5]. First, to be aromatic and, therefore, able to stack, the ring system must be joined to the sugar by a C–C bond ('*C*-nucleoside'). Several heterocycles might implement these H-bonding patterns on a *C*-nucleoside (*Fig. 3*). The 6-aminopyridin-2(1*H*)-one structure, which formally presents the correct H-bonding pattern, is readily oxidized, however, and did not appear to be suitable as a heterocyclic system to support these patterns [6]. Adding a ring N-atom to yield the 2-aminopyrimidin-4(3*H*)-one known as pseudocytidine decreases susceptibility to oxidation, but creates an unacceptable tautomeric ambiguity [7].

The donor–donor–acceptor H-bonding patterns can also be implemented on a pyrazine-ring system, for example as a 6-amino-pyrazin-2(1*H*)-one attached to ribose at the 3-position. We speculated that the additional N-atom in the pyrazine would

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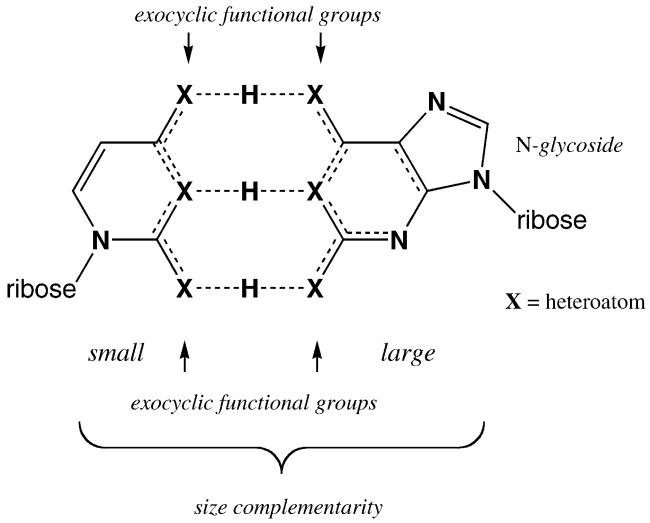


Fig. 1. *Generalized* Watson–Crick *nucleobase pair with three protons between a large heterocycle and a small heterocycle.* Two inter-base H-bonds are formed between exocyclic functional groups; one is formed between heteroatoms of the heterocycles. Dotted lines indicate the position of double bonds to complete the valences to the heteroatoms, and to make the heterocycles aromatic.

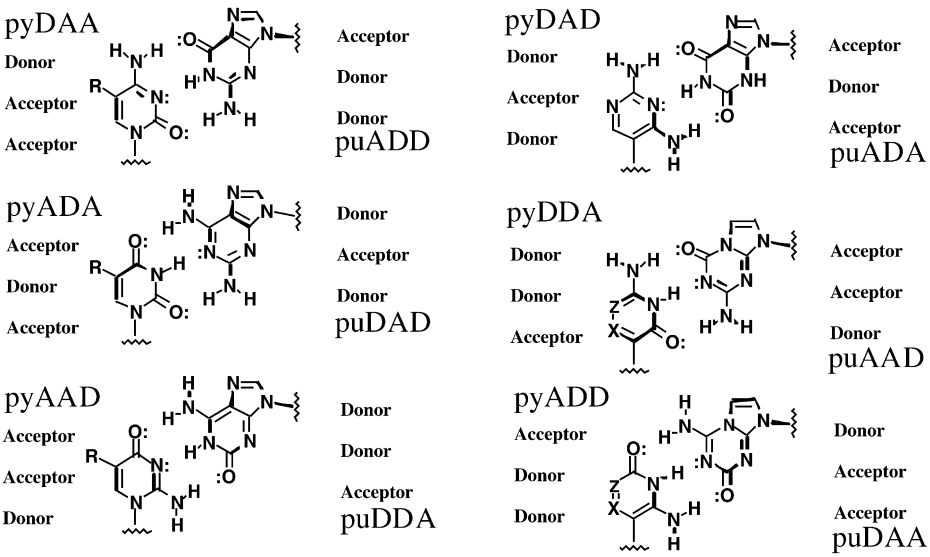


Fig. 2. Watson–Crick *geometry permits twelve nucleobases instead of the four found naturally.* Pyrimidines are designated by the prefix 'py', purines by the prefix 'pu'. Following the prefix is the order, from the major groove to the minor groove, of acceptor (A) and donor (D) groups. The standard A·T base pair is missing an amino group, of course. Amino A is shown to complete the H-bonding pattern. When Z and X are both CH, the heterocycle is a pyridine that is sensitive to oxidation. When X is N, then the heterocycle is a pyrazine, and Z can be either CH or substituted C.

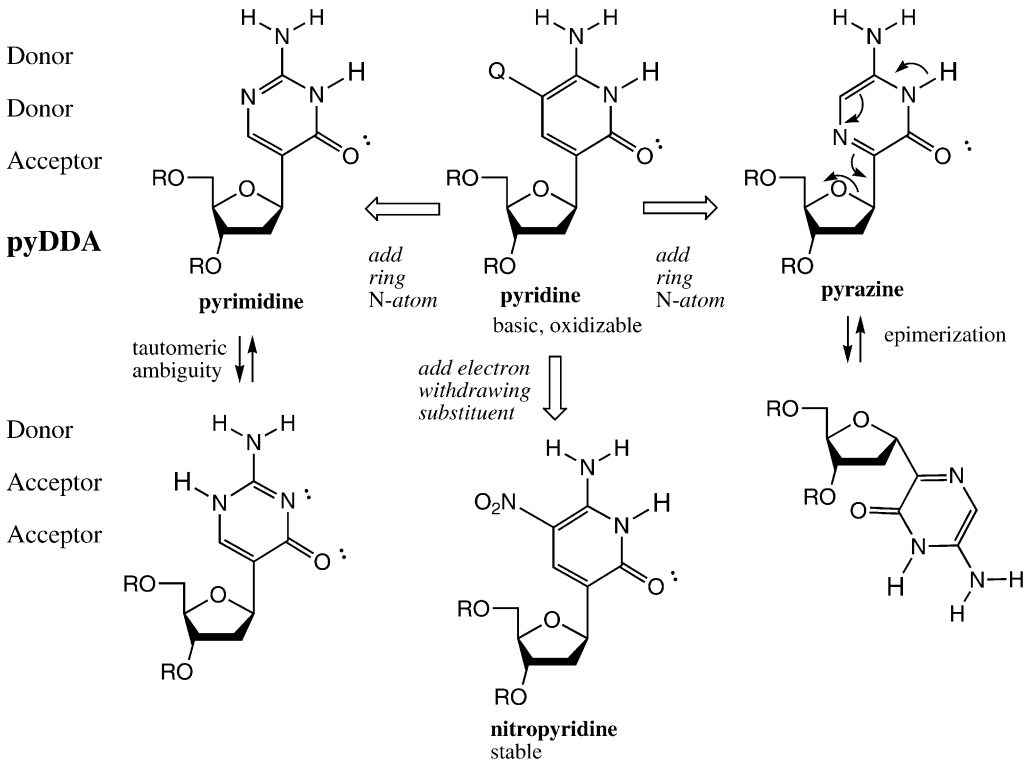


Fig. 3. *Different heterocycle* C*-glycosides implement the pyrimidine donor–donor–acceptor* (pyDDA) *H-bonding scheme.* Electron-withdrawing groups manage epimerization and oxidizability of these.

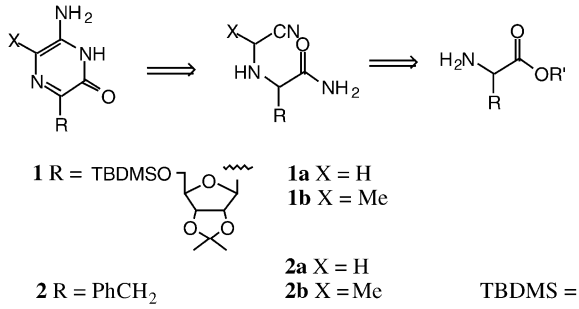
diminish both the basicity and the oxidizability of the system relative to the analogous pyridines. When this work began, however, neither the parent 6-aminopyrazin-2(1*H*)-one nor any pyrazine *C*-nucleosides were described in the literature. *Townsend* and co-workers have recently reported a metallation route to prepare halo-substituted pyrazine nucleoside analogs but without functionality suitable for supporting *Watson–Crick* base pairing [8].

Components of AEGIS have been used to increase the variety of amino acids that can be incorporated into proteins *via* ribosome-based translation [9], expand the potential of oligonucleotides binding in the major groove of double-stranded DNA [10], and explore the specificity and fidelity of polymerases [11]. AEGIS components are now exploited in FDA-approved diagnostic assays to monitor the viral load of patients infected with human immunodeficiency virus and hepatitis C [12]. For these and other reasons, nucleic acid analogs that implement nonstandard H-bonding patterns are attracting a new generation of researchers, eager to understand the rule-based molecular recognition properties [13], whose prominence in nucleic acids is emphasized by their absence in virtually every other class of organic molecule [14].

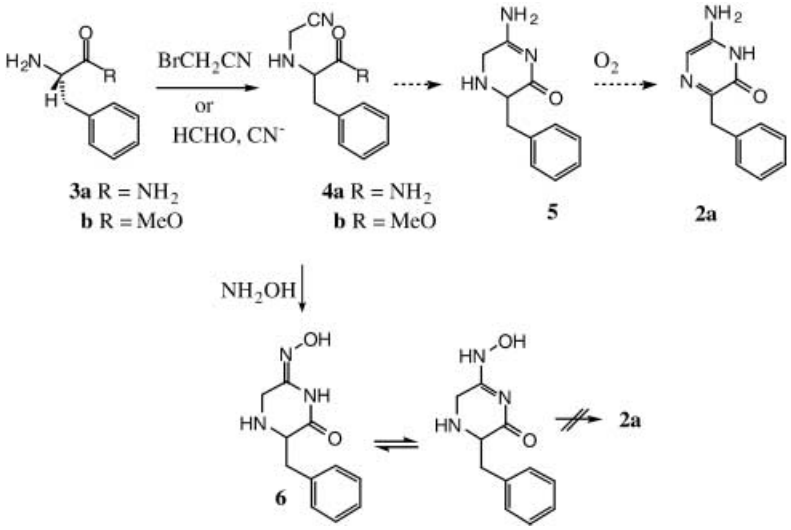
In view of these developments, we felt it timely to present here the results of a study of the synthesis of pyrazine nucleoside analogs that could serve as AEGIS components.

Synthesis. – Retrosynthetic analysis suggested that amino acids carrying a cyanomethyl substituent at the amino group might be readily cyclized to give pyrazine ribosides (*Scheme 1*). To explore this approach, a model compound, **2a**, was prepared from phenylalaninamide (**3a**) or phenylalanine methyl ester (**3b**) (*Scheme 2*). Compound **4a** was obtained in 40–50% yield by treating the amino group of a phenylalanine derivative with bromoacetonitrile in DMF. A *Strecker*-type condensation of **3a** or **3b**, formaldehyde, and KCN at pH 5–6 in dioxane/H_2O mixtures proved, however, to be more successful. The order of addition of the reagents was critical to the success of the *Strecker* route. Maintaining a slight excess of KCN relative to formaldehyde was needed to avoid the generation of substantial amounts of cyclic and dimeric products. Under these conditions, **4a** and **4b** were obtained in 89 and 95% yield, respectively.

Scheme 1



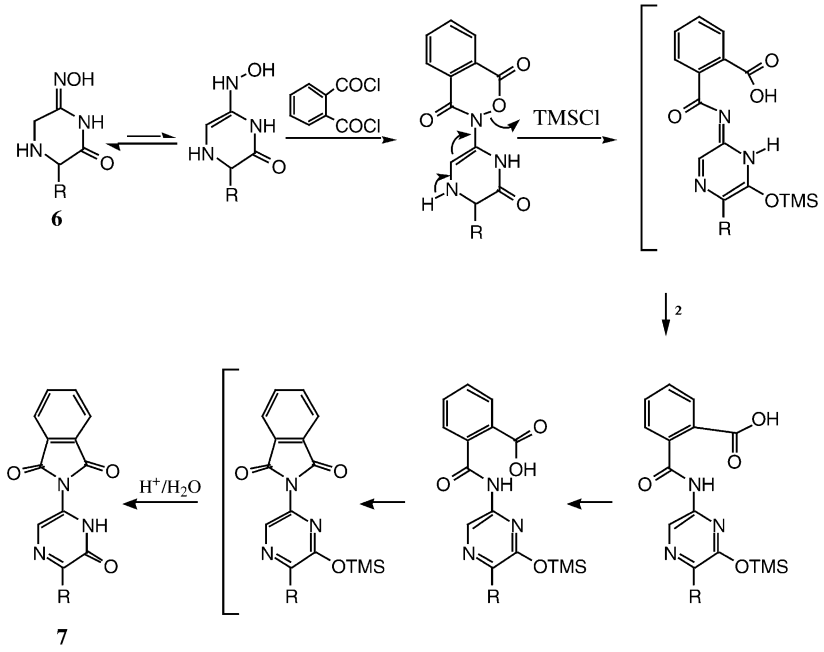
Scheme 2



Direct cyclization of **4a** should yield the dihydropyrazine **5** carrying keto and amino functionalities at the appropriate positions, but requiring oxidation to give the desired product **2a**. We considered the possibility that dioxygen in the air might serve as an oxidant. However, NaOMe in MeOH over Pd/C in the presence of air gave pyrazine **2a** in only 10% yield as an impure sample. Efforts were, therefore, undertaken to adjust the oxidation level of the precursor before cyclization.

To this end, hydroxylamine was used to initiate the cyclization of **4b** (*Scheme 2*). The desired product **6** dominated when the reaction was run with a large excess of reagent at 8°. The molecule overall was in the desired oxidation state, and various approaches were used to convert the dihydropyrazinedione monooxime to its aminopyrazine isomer. Recognizing that the oxime tautomer is strongly preferred over the *N*-imino-hydroxylamine tautomer, the compound was treated with phthaloyl dichloride in the hope of creating the cascade of reactions shown in *Scheme 3*. Unfortunately, only small amounts of a product that might possibly be assignable as the phthaloyl-protected pyrazine derivative **7** were obtained.

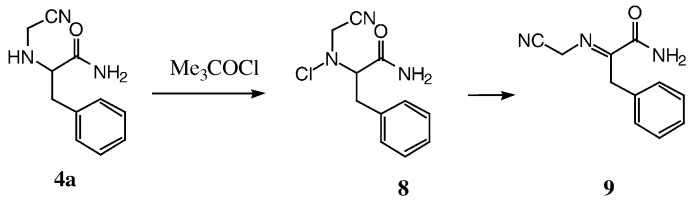
Scheme 3



R = $PhCH_2$, TMSCl = Me_3SiCl.

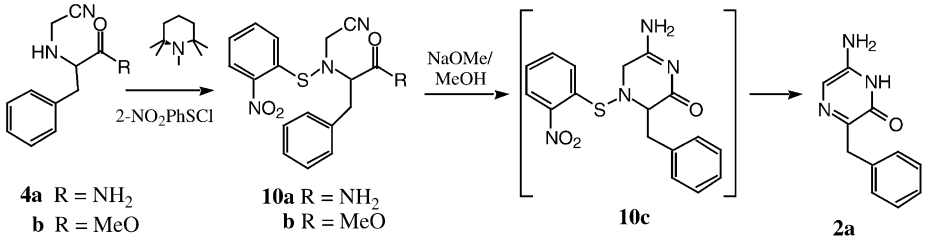
Efforts were then made to increase the oxidation state of the pyrazine precursor by attachment of a heteroatom to the secondary-amine N-atom. The *N*-chloro derivative **8** was obtained quantitatively by using *tert*-butoxy chloride (*Scheme 4*), and could be isolated and analyzed as a crude product. Elimination by treatment with Et_3N as a base yielded imine **9**. This compound could, however, not be cyclized, possibly because the (*E*)-configuration of the C=N bond of **9** is preferred.

Scheme 4



Alternative leaving groups were then examined. Treatment of both **4a** and **4b** with 2-nitrobenzenesulfenyl chloride and 1,2,2,6,6-pentamethylpiperidine (PMP) in THF under reflux yielded **10a** and **10b** in quantitative yield (*Scheme 5*). Even though the products appeared as a single compound by TLC, two sets of signals were observed in the NMR spectra at room temperature. In the ester **10b**, these signals coalesced at 100° in (D_6)DMSO, suggesting that the multiple signals arose from conformational isomers. Reaction of **10a** with NaOMe in MeOH at room temperature yielded pyrazine **2a** *via* cyclization and elimination of 2-nitrothiophenol in 92% yield. No by-products were detectable. An intermediate, seen only by TLC, was presumed to be the intermediate **10c**. Solutions of **2a** in the chromatography solvent mixture CH_2Cl_2/MeOH 95 : 5 or in $CDCl_3$ turned dark red upon standing at room temperature for 24 h. This decomposition was accompanied by broadening and, later, disappearance of NMR signals. In its solid form, **2a** decomposed more slowly. Nevertheless, the decomposition permitted only partial characterization of this 3-benzyl-pyrazin-2(1*H*)-one.

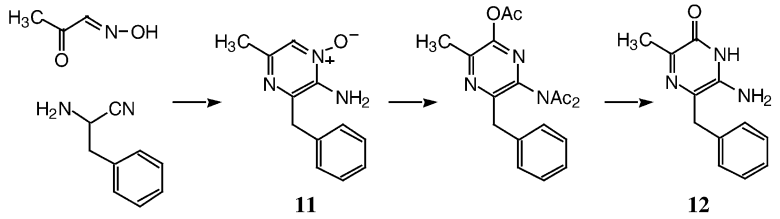
Scheme 5



An analogous decomposition was not observed with the 5-benzyl-4-methylpyrazin-2(1*H*)-one **12** prepared *via* rearrangement of the pyrazine *N*-oxide **11** (*Scheme 6*) [2]. Two possibilities to explain the differential stability were considered. First, decomposition might be catalyzed by contaminants arising from the reaction sequence used to prepare **2a**. Alternatively, the propensity to decompose might reflect the detailed nature of the substituents, with 5-benzyl-3-methylpyrazin-2(1*H*)-one **12** being more stable than 3-benzylpyrazin-2(1*H*)-one.

To test the possibility that the decomposition was catalyzed by contaminants arising from the reaction sequence used to prepare **2a**, **12** was prepared again *via* yet a different route (*Scheme 7*). Alaninamide **13** was converted *via* a *Strecker* reaction with benzeneacetaldehyde and KCN into **14**. Sulfenylation proceeded poorly with PMP in THF in this case, but proceeded in nearly quantitative yield in pyridine at room temperature. The resulting (phenylthio)amide **15** was cyclized with NaOMe in MeOH,

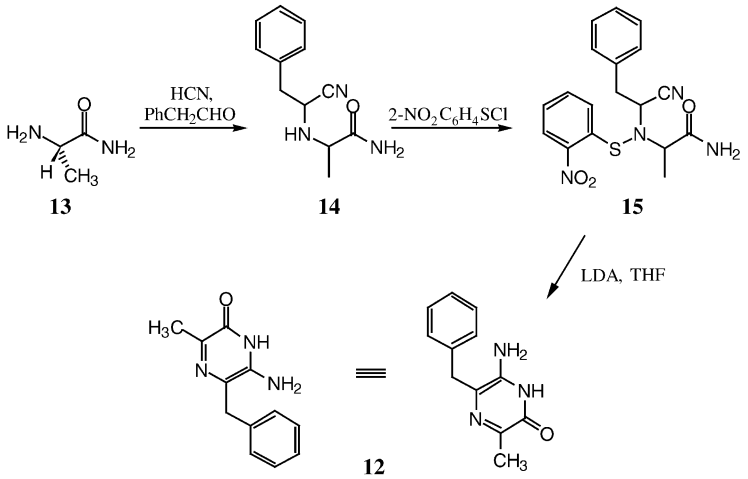
Scheme 6



11 **12**

or with lithium diisopropylamide (LDA) in THF, to give **12** as slightly yellow crystals after chromatography and recrystallization. This compound proved to be identical spectroscopically to the product from the rearrangement of the pyrazine *N*-oxide. As with the material **12** obtained *via* rearrangement of the *N*-oxide **11**, no decomposition was observed in $CDCl_3$ solution for three days at room temperature.

Scheme 7

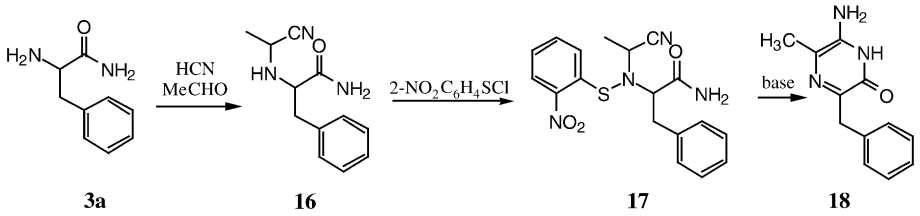


13 **14** **15** **12**

To prepare 6-amino-3-benzyl-5-methylpyrazin-2(1*H*)-one (**18**), phenylalaninamide (**3a**) was treated with acetaldehyde to give **16** (*Scheme 8*). Sulfenylation in pyridine and subsequent cyclization gave **18** as slightly beige crystals after recrystallization. Pyrazin-2(1*H*)-one **18** was also stable under the conditions where the methyl-unsubstituted **2a** was not. This suggested that alkyl substituents increase the stability of aminopyrazinones.

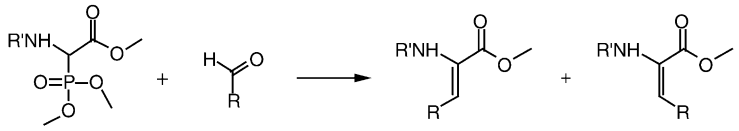
With an improved understanding of the reactivity of the aminohydroxypyrazine ring system, we turned to appending this system to a ribose skeleton. *Schmidt* and coworkers [15] developed a simple, general route for the synthesis of α-amino carboxylic acid derivatives, whose key step is the *Wittig–Horner* reaction of an α-phosphorylglycine ester **20** with various aldehydes to yield aminodihydro acid derivatives (*Scheme 9*). The scope of this method is very broad and includes aromatic, aliphatic, and heterocyclic aldehydes; ketone do not react [16]. In general, the (*Z*)-isomer is the

Scheme 8



3a **16** **17** **18**

Scheme 9



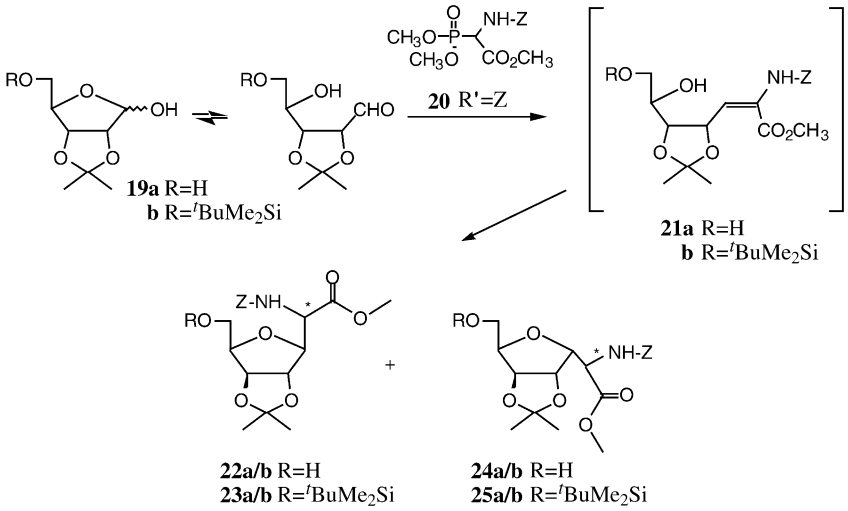
20 (*Z*)-isomer (*E*)-isomer

major product (*Scheme 9*). *Schmidt* and co-workers used phosphoranes obtained from the α-hydroxyglycine esters *via* α-phosphoryl esters **20** [17], and the *N*-[(benzyloxy)-carbonyl]-protected methyl ester of glycine is commercially available.

As starting materials for the *Wittig–Horner* reaction with **20** (R′=Z= $PhCH_2OCO$), both 2,3-*O*-isopropylideneribose **19a** and the 5-O-(*tert*-butyl)dimethylsilyl-protected derivative **19b** were considered (*Scheme 10*), based on the β/α-D-ratios reported by *Ohrui* and *Kane*. Both were readily prepared by known methods [18]. The *Wittig–Horner* reaction was found to proceed only poorly with the ribose derivative **19a** as the electrophile. The reaction was not complete and gave **22** and **24** (mixture of stereoisomers **a** and **b** with respect to the glycine moiety) in only 19% yield. In contrast, **19b** reacted with **20** (R′=Z) under the preferred conditions of *Schmidt* and co-workers to give a mixture of three products in *ca.* 60% yield. Better yields were achieved if **20** (R′=Z) was deprotonated first at $-78°$ in CH_2Cl_2 and then treated slowly with a solution of **19b** at 0°. The two major products **23a/b** were obtained in a combined yield of 90%, whose ratio varied between 1.5 : 1 and 3 : 1, and who could be partially resolved. The NMR spectra corresponded to those expected for **23** and/or **25** as a mixture of stereoisomers **a** and **b** with respect to the glycine moiety. Analysis of the configuration of the anomeric centers showed, however, that both major products **23a** and **23b** had the β-D-configuration. This suggested that their structure differed only by their configuration at C(2). Determination of this configuration was not undertaken, as the stereogenic center was subsequently lost in the synthetic sequence. The first substance to elute on chromatography (silica gel) was also the major stereoisomer and arbitrarily designated as **23a**; the other major product was designated as **23b**. The minor products were assigned by ^{1}H-NMR to be the α-D-anomers **25a/b**. A sufficient amount of these was not obtained pure for analysis. Nevertheless, we can conclude that the *Wittig-Horner* reaction had a β-D-selectivity of $\geq 30:1$ at a yield of *ca.* 90% in this case.

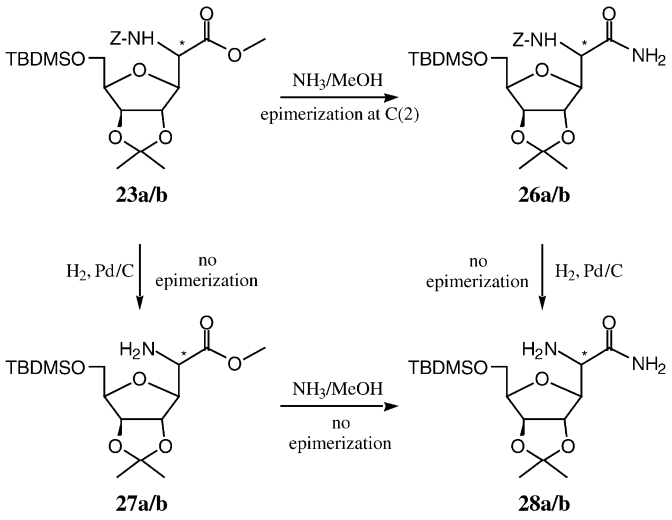
Ammoniolysis of esters **23a/b** in MeOH led in almost quantitative yield to the Z-protected amides **26a/b**, which could be partly separated (*Scheme 11*). The Z protecting

Scheme 10



group in **26a/b** could be removed without epimerization *via* catalytic hydrogenation with Pd and activated charcoal to yielded the free aminoamides **28a/b** in *ca*. 90% yield. The transformation of **23a/b** to **28a/b** proceeded better if the order of steps was reversed (*Scheme 11*). Thus, catalytic hydrogenation of esters **23a** and **23b** with Pd on activated charcoal led to the α-amino acid esters **27a** and **27b**, respectively, in 93% yield without epimerization. Reaction of the α-amino esters **27a** and **27b** with ammonia in

Scheme 11

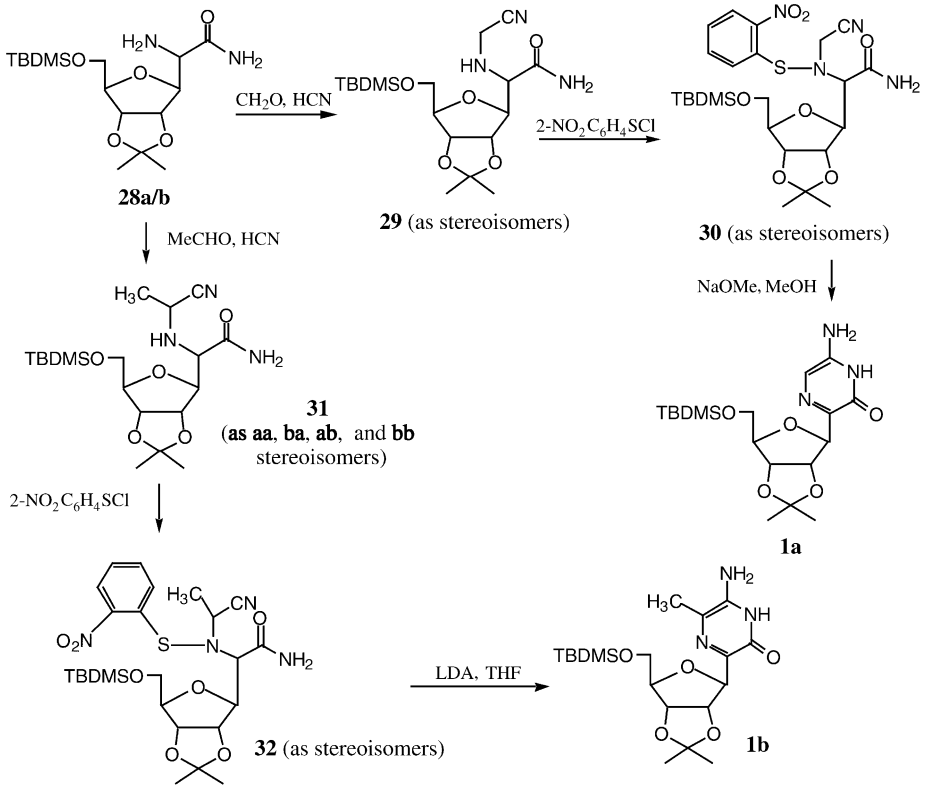


TBDMS = ^{t}BuMe$_2$Si

MeOH gave exclusively amides **28a** and **28b** in almost quantitative yield, again without epimerization. By this route, both aminoamides **28a/b** were obtained from ribose in an overall yield of 58% in five steps. This new route to glycine ribosides is short, should be applicable to the synthesis of both of the 6-aminopyrazin-2(1*H*)-one nucleosides needed, and should, with minor adaptations, be applicable to the synthesis of other *C*-glycosides.

The glycine riboside derivative **28** (stereoisomer mixture) was then treated under *Strecker* conditions with formaldehyde and cyanide to yield **29** as a mixture of stereoisomers (*Scheme 12*). Sulfenylation yielded **30**, which was transformed crude by treatment with MeONa/MeOH to give the target pyrazine riboside derivative **1a**. Through flash chromatography, **1a** was isolated in *ca.* 35% yield as an orange-brown solid, the color presumably arising through partial oxidation. As a component of a DNA molecule cannot have this property, no heroic efforts were made to exclude oxygen. Further, solutions of **1a** in MeCN colored quickly. *Via* reversed-phase chromatography, **1a** could be obtained pure as an almost colorless solid and characterized, even though the solid, when exposed to air, slowly turned yellow. A

Scheme 12



TBDMS = ^{t}BuMe$_2$Si

solution in $CDCl_3$ turned dark yellow in less than 1 h, and deep black after 2 days. This coloring led to broader signals and a shift in the aromatic H–C(5) signals in the NMR spectrum.

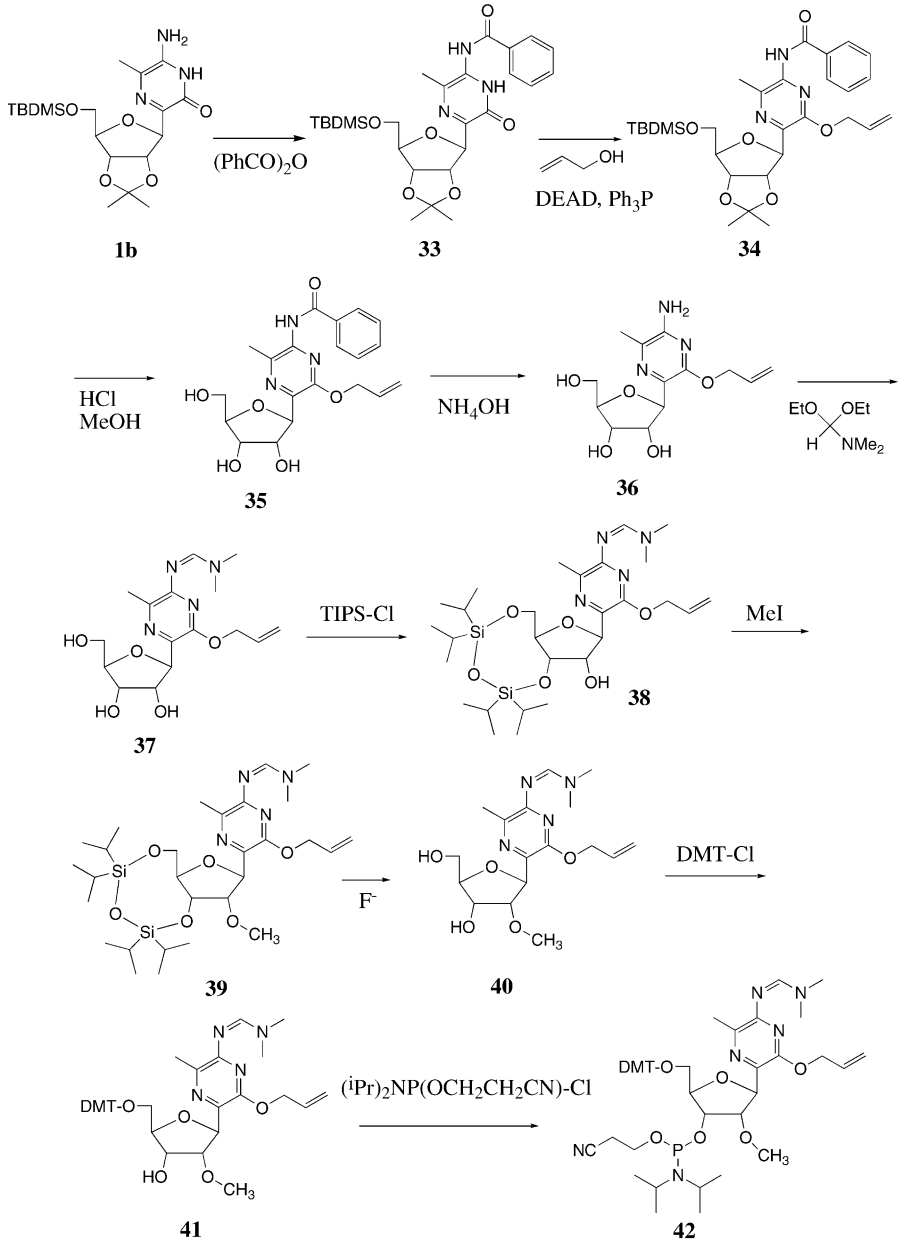
Work with the model system had suggested that an additional Me substituent at the pyrazine ring would suppress the decomposition reaction. To prepare the methylated pyrazine, **28a** was treated with acetaldehyde and HCN under the *Strecker* conditions developed in the model system to give a the secondary amines **31** in over 90% yield (from **28a/b**) as a mixture of stereoisomers arbitrarily designated as **aa**, **ab**, **ba**, and **bb** (*Scheme 12*). These could be transformed with 2-nitrobenzenesulfenyl chloride in pyridine in 99% yield to **32** (from ribose in an overall yield of 90%), again as a mixture of stereoisomers whose configurations were not assigned. Cyclization with LDA in THF led exclusively to the target pyrazine riboside **1b**, which was isolated by chromatography in 91% yield. Compound **1b** was then precipitated as nearly colorless flakes in 81% yield. Consistent with expectations based on the model studies, **1b** could be chromatographed without decomposition and without special care. Solutions of **1b** in $CDCl_3$ showed no change in either color or NMR spectrum over at least three days at room temperature. The same reaction sequence proceeded analogously with the C(2) epimer **28b** to yield **1b** in approximately the same yield. Significant differences in the reactivity of the two epimers could not be detected at any step.

Compound **1b** was then used as a starting material for the synthesis of protected pyrazine nucleoside derivatives suitable for the solid-phase synthesis of DNA (*Scheme 13*). Because the 2-deoxypyrazine nucleoside was prone to epimerization under conditions used to synthesize DNA, the 2′-OH group was retained. To prevent it from participating in base-catalyzed cleavage reactions, and to favor thermodynamically the β-D-epimer, the 2′-OH group was blocked as the methyl ether. The protecting group manipulations depicted in *Scheme 13* (*via* **33**–**41**) and described in the *Exper. Part* followed a strategy standard in the field, and yielded a 5′-dimethyoxytritylated 2-cyanoethyl diisopropylphosphoramidite **42** with the heterocyclic exocyclic amino group protected as its dimethylformamidine derivative and the exocyclic O-function protected as an allyl ether.

Discussion. – The experiments described provide a convenient procedure for preparing nucleoside analogs presenting a H-bond donor–donor–acceptor array. The pyrazine nucleoside **1b** is prepared in an overall yield of *ca.* 42% in just eight steps from ribose. The synthesis is selective for the β-D-epimer, which has a configuration analogous to that found in natural nucleosides. The compounds reported here also serve as starting points for oligonucleotide synthesis that contain the nonstandard base pair.

With respect to the stability of the pyrazine, it is remarkable that the addition of a single Me substituent (compare **1a** with **1b**) has such a significant effect on the stability of the nucleoside. This example illustrates the need, when designing extra letters in a genetic alphabet, to optimize the structure with respect to such substitution. Earlier work in these laboratories had shown that a Me substitution at the 5-position of isocytidine derivatives also stabilized the heterocycle, slowing deamination under alkaline conditions.

Scheme 13



TBDMS = ^{t}BuMe$_2$Si, DEAD = diethyl diazenedicarboxylate, TIPS-Cl = 1,3-dichloro-1,1,3,3-tetraisopropyldisiloxane, DMT = $(MeO)_2$Tr (Tr = trityl).

Outlook. – The particular pyrazine, **1b**, may be suited to develop a new class of molecules based on polymerases that incorporate an epimerizing nucleotide. Many polymerases are believed to identify, as a recognition element, an unshared pair of electrons (or, perhaps better termed, 'electron density') protruding from the nucleobase in the minor groove. The four standard nucleobases found in natural DNA (adenine, guanine, cytosine, and thymine) present this to the minor groove of the DNA double helix from N(3) of the purines and the exocyclic O-atom of the pyrimidines [19]. This pair of electrons is a H-bond acceptor and can, therefore, interact with a H-bond donating group presented by a polymerase to the minor groove. Because it is present in all standard nucleobases, this electron pair can be the basis of a 'common site' interaction between the polymerase and whatever nucleobase is present in the active site at any point in the polymerase catalytic cycle. Indeed, the electron pair appears to be the only such contact that all standard nucleobases can make in the same way. Therefore, the interaction between the unshared electron pair and the polymerase is expected to be used by polymerases generally to enforce the geometry of the base pair without discriminating between different substrates. This might be a key to polymerase fidelity.

Crystallographers have found evidence for such H-bonding interactions in the minor groove for various polymerases, including Taq [20][21] and Bst [22] from the A evolutionary family of polymerases, and RB69 from the family-B polymerases [23]. The residues from Taq and Bst that form H-bonds with the minor groove are conserved within most known family-A DNA polymerases [24]. These consist of *a*) an arginine (at position 573 in Taq) that forms a H-bond with a nucleotide immediately after incorporation and its template complement (N + 1, T + 1; *Fig. 1*), *b*) a glutamine (at position 754 in Taq) that can also form a H-bond with the template at position T + 1, *c*) an asparagine (at position 583 in Taq) that forms a H-bond with the elongating DNA strand three sites from the site of triphosphate addition (N + 3), and *d*) a lysine (at position 540 in Taq) that can form H-bonds with the nucleotides four and five positions away from the site of triphosphate addition (N + 4, N + 5).

Minor-groove contacts are also suggested for family-B polymerases by crystallography. For example, a lysine residue at position 706 in RB69 (a family-B polymerase) forms a H-bond with a nucleotide on the elongating strand two positions from the site of triphosphate addition (N + 2). A second lysine (at position 734 in RB69) forms a bond in the minor groove to a nucleotide four positions away from the addition site *via* a H_2O molecule (N + 4). A tyrosine (at position 567 in RB69) forms a bond to the minor groove of the template strand *via* a H_2O molecule one position away from the site of addition (T + 1). The lysine that interacts with the unshared electron pair carried by nucleobase N + 1 and the tyrosine that interacts with nucleobase T + 1 are conserved among family-B polymerases. The lysine that interacts in the minor groove with nucleobase N + 4 is an arginine in most archaebacterial and mammalian family-B DNA polymerases [25].

The pyrazine heterocycle reported here presents the unshared electron pair in the minor groove. Thus, it should be accepted as a triphosphate by polymerases where this electron pair is indeed a specificity determinant. Here, epimerization is not a key issue. The epimerization will interconvert the α- and β-D-isomers of the triphosphate substrates, and the polymerase will select the β-D-triphosphate for incorporation,

ignoring the α-D-triphosphate. As the β-D-triphosphate is consumed, however, the α-D-triphosphate will epimerize to create more.

Once incorporated, however, the epimerization reaction will lead to the formation of the α-D-anomer within the strand. As α-D-anomeric nucleotides accumulate within a strand, the affinity of the strand for its complement should be diminished. This, in turn, will cause the copy to disassociate from the template, permitting the template to serve as such for the synthesis of another product oligonucleotide.

Separating a product strand from a template strand is, of course, the purpose of the heating cycle in a polymerase chain reaction. Here, the product (having multiple α-D-anomeric nucleotides) would presumably not serve as a good template. Thus, the result would be a linear amplification without thermal cycling of an oligonucleotide containing nonstandard nucleotides. This may have applications in diagnostics and taggants.

Experimental Part

1. *General*. Unless otherwise mentioned, reagents were purchased from *Fluka* or *Aldrich* at highest quality (*puriss.* or *purum*). THF and toluene were freshly distilled from Na, MeCN and CH_2Cl_2 from CaH_2. All other solvents were purchased from *Fluka* or *Aldrich* in the highest quality. TLC: *Merck* TLC silica gel *60* F_{254} (d = 0.25 mm) and *Waters K6F* silica gel *60* (d = 0.25 mm); visualization either with UV light (λ 254 nm) or staining with either a soln. of phosphomolybdic acid/ceric(IV) sulfate tetrahydrate/conc. H_2SO_4 soln. or vanillin/EtOH/conc. H_2SO_4 soln. and subsequent heating. Flash chromatography (FC): 50–100-fold silica gel *60* (*Merck*, 0.040–0.063 mm, or *Fisher Davisil*, 0.035–0.070 mm) with 0.2–0.3 bar pressure. HPLC: semiprep. *Merck-Septech-Novaprep-5000* instrument, with silica-gel-*Merck-Lichrospher-Si* (60-7 μm) column; semiprep. *Waters-PrepLC-4000* instrument with *Waters-486* tunable absorbance detector on and *Waters-Prep-Nova-Pak-HR-C_{18}* column (60 Å, 25 × 100 mm); *Waters-616* pump with *Waters-600-S* controller, *Waters-996* photodiode-array detector, and *Shodex-RSpak-D18-613* column (6 × 150 mm) or *Waters-Nova-Pak-C_{18}* column (3.9 × 150 mm). UV/VIS Spectra: *Varian Cary-1-Bio* UV/VIS spectrophotometer with a *Cary* temperature controller and *Shimadzu-UV/VIS-160* spectrophotometer; λ_{max} (ε) in nm. NMR Spectra: *Bruker-AMX-500*, *Varian-Unity-500*, *Varian-EM-390*, *Varian-XL-300*, *Varian-Gemini-300*, and *Varian-VXR-300* instruments; δ in ppm rel. to $SiMe_4$ as internal standard, J in Hz; starred (* or **) attributions may be interchanged multiplicity of the ^{13}C-NMR signals by DEPT. Anal. GC/MS; *Hewlett-Packard* gas Chromatograph *5710A* combined with a mass spectrometer *5710B* as detector; t_R in min. MS: *VG-Tribrid* (EI, 70 eV), *VG-ZAB2-SEQ* and *Finnigan-MAT-95* (FAB, 3-nitrobenzyl alcohol (NOBA) matrix), *Finnigan-MAT-LCQ* (ESI), and *Bruker Reflex* (MALDI TOF; matrices mentioned below) instruments; in m/z (rel. %).

2. *Model Compounds.* N-*(Cyanomethyl)phenylalaninamide* (**4a**). KCN (716 mg, 11 mmol) was added to a soln. of phenylalaninamide (**3a**; 1.64 g, 10 mmol) in dioxane/H_2O 3:1 (40 ml). The pH was carefully adjusted to 6 with AcOH. Formalin (0.9 ml, *ca.* 11.7 mmol) was then slowly added within 1 h, and the mixture was stirred overnight at r.t. Sat. aq. $NaHCO_3$ soln. (*ca.* 50 ml) was then added, and the aq. phase was extracted with CH_2Cl_2. The combined org. phase was dried (Na_2SO_4) and evaporated, the residue suspended in CH_2Cl_2 (*ca.* 50 ml), the soln. filtered, and the filtrate evaporated. The residue was dried under high vacuum: **4a** (1.808 g, 89%), pure by TLC and ^{1}H-NMR. A smaller portion was recrystallized from acetone to give an anal. sample. Colorless solid. M.p. 150°. TLC (CH_2Cl_2/MeOH 9:1): R_f 0.4. IR (KBr): 3370, 3350, 3320, 3190, 3030, 2930, 2860, 2230, 1640, 1495, 1475, 1455, 1425, 1345, 1330, 1255, 1200, 1135, 1090, 1075, 1030, 1015, 960, 935, 920, 885, 875, 855, 845, 820, 780, 740, 700, 620, 575, 520, 485. ^{1}H-NMR ((D$_6$)DMSO): 2.65–2.75 (*m*, with *dd* at 2.72, J = 7.8, J_{gem} = 13.7, NH, 1 H of PhCH_2); 2.88 (*dd*, J = 5.7, J_{gem} = 13.7, 1 H of PhCH_2); 3.31–3.37 (*m*, > 1 H, H–C(2), H_2O); 3.43 (*dd*, J = 6.9, J_{gem} = 17.4, 1 H of CH_2CN); 3.59 (*dd*, J = 6.3, J_{gem} = 17.4, 1 H of CH_2CN); 7.10 (br. *s*, 1 H of $CONH_2$); 7.16–7.30 (*m*, 5 arom. H); 7.41 (br. *s*, 1 H of $CONH_2$). ^{13}C-NMR ((D$_6$)DMSO): 35.21 (*t*, CH_2CN); 38.67–40.34 (*m*, DMSO, and *t* of CH_2Ph); 62.06 (*d*, NCHCONH$_2$); 118.84 (*s*, CN); 126.13 (*d*, arom. CH); 128.00 (*d*, arom. CH); 129.17 (*d*, arom. CH); 138.08 (*s*, arom. C); 173.97 (*s*, $CONH_2$). MS: 204 (0.13, $[M+1]^+$), 203 (0.18, M^+), 176 (1, $[M-HCN]^+$), 160 (12), 159 (100, $[M-CONH_2]^+$), 147 (11), 132 (35), 119 (12), 112 (34), 105 (11), 92 (13), 91 (38), 85 (35), 65 (12). Anal. calc. for $C_{11}H_{13}N_3O$ (203.24): C 65.01, H 6.45, N 20.67; found: C 65.02, H 6.51, N 20.67.

N-*(Cyanomethyl)phenylalanine Methyl Ester* (**4b**). As described for **4a**, with KCN (0.72 g, 11 mmol), phenylalanine methyl ester hydrochloride (**3b**; 2.16 g, 10 mmol), dioxane/H_2O 1:1 (40 ml), and formalin (0.75 ml, 9.75 mmol; addition within 2.5 h). The residue obtained after the 1st evaporation was purified by FC (silica gel, AcOEt/hexane 3:7): **4b** (2.02 g, 95% rel. to formalin). Colorless oil. TLC (hexane/AcOEt 1:1): R_f 0.42. IR ($CHCl_3$): 3350, 3010, 2950, 1740, 1605, 1495, 1475, 1455, 1440, 1360, 1335, 1280, 1265, 1215, 1180, 1140, 1090, 1030, 1020, 995, 880. ^{1}H-NMR ($CDCl_3$): 1.86 (*s*, NH); 2.92 (*dd*, $J = 7.8$, $J_{gem} = 13.7$, 1 H of PhCH_2); 3.10 (*dd*, $J = 5.4$, $J_{gem} = 13.8$, 1 H of PhCH_2); 3.54 (*s*, CH_2CN); 3.72–3.76 (*m*, and *s* at 3.74, H–C(2), MeO); 7.17–7.25 (*m*, 5 arom. H). ^{13}C-NMR ($CDCl_3$): 35.96 (*t*, CH_2CN); 39.17 (*t*, PhCH_2); 52.16 (*q*, MeO); 61.12 (*d*, NCHCOOMe); 117.25 (*s*, CN); 127.16 (*d*, arom. CH); 128.71 (*d*, arom. CH); 129.16 (*d*, arom. CH); 136.25 (*s*, arom. C); 173.31 (*s*, COOMe). MS: 218 (4, M^+), 160 (12), 159 (100, $[M - COOMe]^+$), 132 (26), 127 (77, $[M - PhCH_2]^+$), 100 (23), 92 (10), 91 (34, $PhCH_2^+$), 66 (18), 65 (10). Anal. calc. for $C_{12}H_{14}N_2O_2$ (218.25): C 66.04, H 6.47, N 12.84; found: C 66.00, H 6.07, N 12.61.

3-Benzylpiperidine-26-dione 6-Oxime (**6**). A soln. of hydroxylamine hydrochloride (765 mg, 11 mmol) in MeOH (25 ml) was placed in a glass tube at 0° and treated with 5.4M MeONa in MeOH (1.8 ml, 9.72 mmol). After *ca.* 5 min stirring, **4b** (1.09 g, 5 mmol) was added, the tube sealed, and the mixture heated overnight at 60° in an oil bath with stirring. The products were then adsorbed on silica gel (2.3 g) and separated by FC (silica gel, CH_2Cl_2/MeOH 95:5): **6** (892.4 mg, 81%). Colorless crystals. M.p. 177°. TLC (CH_2Cl_2/MeOH 9:1): R_f 0.47. IR (KBr): 3300, 3240, 3025, 1660, 1640, 1495, 1465, 1455, 1430, 1400, 1380, 1320, 1245, 1145, 985, 960, 935, 900, 825, 790, 750, 740, 700, 590, 570, 500, 485. ^{1}H-NMR ((D$_6$)DMSO): 2.69 (*m*, NH); 2.78 (*dd*, $J = 9.7$, $J_{gem} = 14.1$, 1 H of PhCH_2); 3.18 (*dd*, $J = 3.7$, $J_{gem} = 14.1$, 1 H of PhCH_2); 3.34 (*dd*, J(5,NH) = 8.4, $J_{gem} = 15.5$, 1 H of NCH_2C(NOH)); 3.45 (*dd*, J(5,NH) = 5.5, $J_{gem} = 15.7$, 1 H of NCH_2C(NOH)); 3.58 (*dt*-like *m*, H–C(2)); 7.17–7.30 (*m*, 5 arom. H); 9.5, 10.1 (2 br. *s*, NH, NOH). ^{13}C-NMR ($CDCl_3$): 35.74 (*t*, PhCH_2); 41.92 (*t*, NCH_2C(NOH)); 59.79 (*d*, NCHCONH); 125.92 (*d*, arom. CH); 127.93 (*d*, arom. CH); 129.22 (*d*, arom. CH); 138.77 (*s*, arom. C); 144.21 (*s*, C(NOH)NH); 170.40 (*s*, CONH). MS: 219 (28, M^+), 129 (11), 128 (100, $[M - CH_2Ph]^+$), 100 (21), 91 (23, $PhCH_2^+$). Anal. calc. for $C_{11}H_{13}N_3O_2$ (219.24): C 60.26, H 5.98, N 19.17, O 14.60; found: C 59.87, H 6.02, N 19.03.

N-*Chloro*-N-*(cyanomethyl)phenylalaninamide* (**8**). A suspension of **4a** (20 mg, 103.4 μmol) in THF (1 ml) was cooled to −78° and treated with *tert*-butoxy chloride (12 mg, 110.53 μmol). The mixture was slowly (within 1 h) brought to r.t., yielding a clear colorless soln. The mixture was evaporated and dried under high vacuum to yield **8** (*ca.* 24 mg), which was analyzed only as crude product. Colorless oil. TLC (CH_2Cl_2/MeOH 9:1): R_f 0.53. ^{1}H-NMR (200 MHz, $CDCl_3$): 3.23 (*d*, $J = 6.8$, PhCH_2); 3.83 (*t*, $J = 6.8$, H–C(2)); 4.01 (*s*, CH_2CN); 5.69, 5.89 (2 br. *s*, each 1 H, $CONH_2$); 7.21–7.36 (*m*, > 5 H, arom. H). ^{13}C-NMR (50 MHz, $CDCl_3$): 39.79 (PhCH_2); 48.36 (CH_2CN); 74.07 (NCHCONH$_2$); 113.88 (CN); 127.11 (arom. CH); 128.64 (arom. CH); 128.98 (arom. CH); 139.71 (arom. C); 169.84 ($CONH_2$).

α-[(Cyanomethyl)imino]benzenepropanamide (**9**). A suspension of **4a** (51 mg, 251 μmol) in THF (2.5 ml) was cooled to −78° and treated with *tert*-butoxy chloride (30 mg, 276 μmol). The mixture was slowly brought to r.t. within 1 h. The resulting clear, colorless soln. was cooled again to −78° and treated with Et_3N (70 μl, 504 μmol). The mixture slowly became cloudy. After stirring at −78° for 1 h, the mixture was slowly (within *ca.* 2 h) brought to r.t., the yellow suspension filtered with a pipette and glass wool, and the residue washed several times with dry THF. Evaporation of the filtrate gave 56 mg of crude product which was purified by FC (silica gel, CH_2Cl_2/MeOH 95:5): **9** (35 mg, 69%). Dark yellow oil. TLC (CH_2Cl_2/MeOH 9:1): R_f 0.51. ^{1}H-NMR ($CDCl_3$): 4.00 (*s*, PhCH_2); 4.30 (*s*, CH_2CN); 5.88 (br. *s*, 1 H, $CONH_2$); 7.12 (*m*, 1 H of $CONH_2$, Ph); NH could not be determined with D_2O exchange because **9** decomposed in the presence of H_2O. ^{13}C-NMR ($CDCl_3$): 32.30, 39.69 (2*t*, CH_2CN, PhCH_2); 116.47 (*s*, CN); 127.25 (*d*, arom. CH); 128.51 (*d*, arom. CH); 129.23 (*d*, arom. CH); 133.63 (*s*, arom. C); 165.28, 168.53 (2*s*, $CONH_2$, C=NCH$_2$CN). MS ($C_{11}H_{11}N_3O$, 201.23): 201 (16, M^+), 158 (32), 157 (29), 117 (12), 116 (19), 92 (13), 91 (100), 65 (13).

N-*(Cyanomethyl)*-N-*[(2-nitrophenylthio]phenylalaninamide* (**10a**). A soln. of 2-nitrobenzenesulfenyl chloride (73 mg, 385 μmol), **4a** (50 mg, 246 μmol), and 1,2,2,6,6-pentamethylpiperidine (PMP; 70 μl, 387 μmol) in THF (3 ml) was heated under reflux (90° oil bath), until consumption of **4a** was complete (3 h, TLC monitoring). The mixture was diluted with a little MeOH, and the entire mixture was adsorbed on silica gel (0.12 g) and subjected to FC (silica gel, CH_2Cl_2/MeOH 95:5): **10a** (86 mg, 98%). Yellow crystals. TLC (CH_2Cl_2/MeOH 95:5): R_f 0.38. ^{1}H-NMR ((D$_6$)DMSO): not evaluated due to complexity (inversion and rotation isomers). ^{13}C-NMR ((D$_6$)DMSO; all signals doubled at r.t.): 36.07, 36.28 (2*t*, CH_2CN); 40.69, 44.60 (2*t*, PhCH_2); 68.10, 71.74 (2*d*, NCHCONH$_2$); 117.08, 117.24 (*s*, CN); 124.01, 124.84, 125.25, 125.40, 125.62, 125.80, 126.20, 126.69, 128.02, 128.50, 128.83, 129.22, 133.89, 134.34 (14*d*, arom. CH); 137.26, 138.05, 141.19, 141.62, 141.82, 143.37 (6*s*, arom. C); 172.42, 172.81 (*s*, $CONH_2$). MS ($C_{17}H_{16}N_4SO_3$, 356.40): 356 (< 1, M^+), 312 (10,

$[M - CONH_2]^+$), 202 (24), 159 (20), 157 (10), 155 (15), 154 (100), 148 (11), 138 (14), 106 (23), 98 (29), 96 (16), 92 (13), 91 (67), 65 (13).

N-*(Cyanomethyl)*-N-*[(2-nitrophenylthio]phenylalanine methyl ester* (**10b**). As described for **10a**, with 2-nitrobenzenesulfenyl chloride (280 mg, 1.48 mmol), **4b** (218.3 mg, 1 mmol), PMP (0.27 ml, 1.49 mmol), and THF (5 ml) for 2 h (by TLC). FC (residue adsorbed on silica gel (1.5 g); silica gel, CH_2Cl_2) gave **10b** (352 mg, 95%). Yellow oil which slowly solidified upon standing. TLC (CH_2Cl_2): R_f 0.35. ^{1}H-NMR ((D$_6$)DMSO): 2.97–3.24 (*m*, PhCH_2); 3.70, 3.76 (2*s*, MeO); 4.21–4.60 (*m*, H–C(2), CH_2CN); 6.66, 7.14–7.24, 7.28–7.51 (*d*, *m*, *m*, 8 arom. H); 8.23 (*dd*, 1 arom. H). ^{1}H-NMR (100°, (D$_6$)DMSO): 3.10 (*dd*, $J = 9.1$, $J_{gem} = 14.5$, 1 H of PhCH_2); 3.22 (*dd*, $J = 6.4$, $J_{gem} = 14.5$, 1 H of PhCH_2); 3.70 (*s*, MeO); 4.33 (*s*-like *m*, CH_2CN, H–C(2)); 7.26–7.43 (*m*, 8 arom. H); 8.21 (*dd*, $J_1 = 1.2$, 8.3, 1 arom. H). ^{13}C-NMR ($CDCl_3$; all signals doubled): 36.31, 37.18, 41.60, 45.76 (4*t*, PhCH_2, CH_2CN); 52.61 (*q*, Me); 68.22, 72.01 (2*d*, NCHCOOMe); 115.73, 115.88 (2*s*, CN); 124.02, 124.66, 125.32, 125.76, 126.96, 127.49, 128.64, 128.83, 129.21, 134.05, 134.30 (11*d*, arom. CH); 136.35, 136.48, 141.49, 141.92, 142.35 (5*s*, arom. C); 171.65, 171.79 (2*s*, COOMe). GC/MS (P10020): t_R 16.73; 312 (3, $[M - COOCMe]^+$), 248 (15), 235 (12), 154 (75), 145 (15), 130 (14), 106 (27), 98 (62), 96 (24), 92 (15), 91 (100), 78 (20). MS ($C_{18}H_{17}N_3SO_4$, 371.41): 371 (0.7), 313 (3), 248 (13), 154 (92), 145 (16), 106 (26), 98 (35), 96 (18), 92 (14), 91 (100), 65 (11).

*6-Amino-3-benzylpyrazin-2(1*H*)-one* (**2a**). At 0°, 5.4M MeONa (0.5 ml, 2.7 mmol) was added dropwise to a soln. of **10a** (387.6 mg, 1.088 mmol) in MeOH (15 ml). The mixture was slowly warmed under stirring to r.t. (→ orange and precipitation). After total consumption of **10a** (2 h, TLC monitored), the mixture was acidified with 2N HCl (pH *ca.* 6), concentrated *in vacuo*, and then evaporated under high vacuum. The residue was taken up in CH_2Cl_2/MeOH 9:1 and subjected to FC (silica gel, CH_2Cl_2/MeOH 9:1): **2a** (184.4 mg, 92%). Brownish foam. TLC (CH_2Cl_2/MeOH 9:1): R_f 0.29. ^{1}H-NMR (200 MHz, $CDCl_3$): 3.97 (*s*, PhCH_2); 4.7 (br. *s*, NH_2); 6.90 (*s*, H–C(5)); 7.17–7.27 (*m*, Ph). ^{13}C-NMR (50 MHz, $CDCl_3$): 38.81 (*t*, CH_2); 110.71 (*d*, C(5)); 126.56 (*d*, arom. CH); 128.79 (*d*, arom. CH); 128.84 (*d*, arom. CH); 139.63, 139.69 (2*s*, arom. C, arom. C(py)); 144.84 (*s*, arom. C(py)); 156.42 (*s*, arom. C(py)). GC/MS (P15020): t_R 5.7; 201 (100, M^+), 173, 156, 130, 91.

N-*(1-Cyano-2-phenylethyl)alaninamide* (**14**). KCN (360 mg, 5.53 mmol) was added to a soln. of L-alaninamide (**13**; 641 mg, 5 mmol) in dioxane/H_2O 5:3 (40 ml). The pH was carefully adjusted to 6 with AcOH. A soln. of benzeneacetaldehyde (50% in diethyl phthalate; 1.3 ml, 5.8 mmol) was added dropwise within 1 h, with an additional amount (0.5 ml, 2.2 mmol) added after 1 h. The mixture was poured into an aq. Na_2CO_3 soln., the aq. phase extracted with CH_2Cl_2, and the combined org. phase washed with sat. aq. NaCl soln., dried, and evaporated: **12** (811.7 mg, 75%). Colorless crystals. M.p. 107–108°. TLC (CH_2Cl_2/MeOH 95:5): R_f 0.29. IR (KBr): 3380, 3350, 3200, 3030, 2990, 2970, 2930, 2870, 2230, 1635, 1495, 1455, 1400, 1370, 1325, 1270, 1150, 1100, 1075, 1030, 1010, 985, 915, 840, 810, 790, 740, 700, 600, 575, 560, 510, 475. ^{1}H-NMR ($CDCl_3$): 1.32, 1.35 (2*d*, 3 Me); 2.96–3.15 (*m*, 2 H, PhCH_2); 3.37–3.47 (*m*, 1 H, CHCONH$_2$); 3.61–3.70, 3.82–3.90 (2*m*, 1 H, CHCN); 5.53, 5.77, 6.22, 6.52 (4 br. *s*, 2 H, NH); 7.26–7.39 (*m*, 5 H, *Ph*CH_2). ^{13}C-NMR ($CDCl_3$): 18.80, 19.84 (2*q*, Me); 39.33, 40.04 (2*t*, PhCH_2); 50.08, 50.79 (2*d*, CHCN); 55.88, 56.74 (2*d*, CHCONH$_2$); 119.19, 119.58 (2*s*, CN); 127.86, 128.92, 129.03, 129.37, 129.52 (5*d*, arom. CH); 134.60, 134.95 (2*s*, arom. C); 176.10 (*s*, $CONH_2$). MS: 217 (<1), 173 (16), 146 (53), 136 (11), 130 (13), 99 (100), 91 (26), 73 (22), 71 (20). Anal. calc. for $C_{12}H_{15}N_3O$ (217.27): C 66.34, H 6.96, N 19.34; found: C 66.60, H 6.78, N 19.24.

N-*(1-Cyano-2-phenylethyl)*-N-*[(2-nitrophenyl)thio]alaninamide* (**15**). Amide **14** (1.00 g, 4.6 mmol) was taken up in pyridine (4 ml) and then evaporated under high vacuum. The residue was redissolved in pyridine, and the soln. was treated with 2-nitrobenzenesulfenyl chloride (1.3 g, 6.86 mmol) and a few crystals of *N,N*-dimethylpyridin-4-amine (DMAP). After 1 h, the mixture was diluted with MeOH, dried under high vacuum, and subjected to FC (silica gel, CH_2Cl_2/MeOH 95:5): **15** (1.64 g, 96%). Yellow foam. TLC (CH_2Cl_2/MeOH 95:5): R_f 0.34. ^{1}H- and ^{13}C-NMR ($CDCl_3$): not assigned due to complexity (rotation and inversion isomers). MS ($C_{18}H_{18}N_4O_3S$, 370.43): 370 (1, M^+), 326 (13), 216 (14), 171 (12), 155 (10), 153 (100), 106 (19), 98 (26), 96 (10), 91 (53), 84 (10), 49 (10).

*6-Amino-5-benzyl-3-methylpyrazin-2(1*H*)-one* (**12**). A soln. of **15** (270 mg, 729 μmol) in THF (2 ml) was treated with 1.5M LDA in cyclohexane (1 ml, 1.5 mmol) at –78°. The mixture, which immediately turned deep red, was stirred at r.t. overnight. The entire soln. was then added dropwise to 2M phosphate buffer (pH 7.1; 5 ml). The mixture was extracted several times with AcOEt, the combined org. extract dried (Na_2SO_4) and evaporated, and the residue recrystallized from CH_2Cl_2 to yield the first batch of **12** (77.9 mg, 50%). The material in the mother liquor was subjected to FC (silica gel, CH_2Cl_2/MeOH 95:5): **12** as an orange foam which was recrystallized from MeCN (28.7 mg, 18%). The material was identical to an authentic sample [2]. Total yield of **12**: 106.6 mg (68%). M.p. 207–208°. TLC (CH_2Cl_2/MeOH 9:1): R_f 0.30. ^{1}H-NMR (200 MHz, (D$_6$)DMSO): 2.10 (*s*, Me); 3.82 (*s*, PhCH_2); 5.57 (br. *s*, NH_2); 7.14–7.28 (*m*, Ph); 10.7 (br. *s*, NH). ^{13}C-NMR (50 MHz,

(D_6)DMSO): 18.17 (*q*, Me); 36.79 (*t*, CH_2); 122.74 (*s*, C(5)); 125.95 (*d*, C(4′)); 128.35, 128.64 (2*d*, C(2′), C(3′)); 130.78 (*s*, C(3)); 140.52 (*s*, C(1′)); 146.23 (*s*, C(6)); 155.35 (*s*, C(2)). GC/MS (P10020); t_R 7.82; 2.15 (100, M^+), 186, 169, 145, 129, 110, 91.

N-*(1-Cyanoethyl)phenylalaninamide* (**16**). A soln. of **3a** (1.64 g, 10 mmol) in dioxane/H_2O 3:1 (60 ml) was treated with KCN (720 mg, 11.06 mmol) and its pH carefully adjusted with AcOH (*ca.* 4 ml) to pH 6. Within 1 h was slowly added acetaldehyde (1 ml, 17.7 mmol) by means of a cooled syringe, and the mixture was stirred at r.t. for *ca.* 1 h, until complete consumption of **3a** (TLC monitoring). Sat. aq. Na_2CO_3 soln. (*ca.* 50 ml) was added, the mixture extracted with CH_2Cl_2, the combined org. phase dried (Na_2SO_4) and evaporated, the residue taken up in CH_2Cl_2, the soln. treated with hexane until it became cloudy, and stored in the refrigerator for crystallization. More product was isolated by repeated recrystallization of the mother liquor. From the rest of the mother liquor, a second batch of **16** (295 mg, 14%) was isolated by FC (silica gel, CH_2Cl_2/MeOH 95:5). The amounts of the two diastereoisomers varied significantly from batch to batch. Total yield of **16**: 1.787 g (82%). Small, colorless needles. M.p. 109°. TLC (CH_2Cl_2/MeOH 9:1): R_f 0.41. IR (KBr): 3380, 3315, 3200, 3085, 3030, 2980, 2930, 2870, 2230, 1635, 1495, 1470, 1460, 1440, 1390, 1380, 1325, 1295, 1255, 1200, 1160, 1150, 1125, 1090, 1070, 1030, 935, 860, 820, 790, 720, 695, 605, 580, 550, 515, 485. ^{1}H-NMR ($CDCl_3$): 1.31, 1.42 (2*d*, $J = 7.0$, 7.1, 3 H, Me); 1.59, 1.78 (*d s*, 1 H, NH); 2.82, 2.87 (2*dd*, 1 H of PhCH_2); 3.22, 3.27 (2*dd*, 1 H of PhCH_2); 3.46–3.69 (*m*, 2 H, Me*CH*CN, H–C(2)); 5.9, 6.0 (br. *s*, 1 H of $CONH_2$); 6.6, 6.7 (br. *s*, 1 H of $CONH_2$); 7.22–7.39 (*m*, *Ph*CH_2). ^{13}C-NMR ($CDCl_3$): 19.20, 19.94 (2*q*, Me); 39.10, 39.26 (2*t*, Ph*C*H_2); 44.41, 44.51 (2*d*, Me*C*HCN); 61.57, 61.99 (2*d*, N*C*HCONH$_2$); 119.87, 120.10 (2*s*, CN); 127.38, 127.48 (2*d*, arom. CH); 128.10, 129.09, 129.22 (4*d*(= 3*d*), arom. CH); 135.89, 136.44 (2*s*, arom. C); 175.04, 175.32 (2*s*, $CONH_2$). MS: 218 (0.22, $[M+1]^+$), 190 (5.4, $[M-HCN]^+$), 173 (34, $[M-CONH_2]^+$), 149 (20), 147 (20), 146 (94), 131 (12), 130 (14), 126 (10), 105 (18), 104 (13), 103 (13), 99 (100), 91 (42), 77 (14), 65 (14), 54 (13), 51 (10), 44 (39). Anal. calc. for $C_{12}H_{15}N_3O$ (217.27): C 66.34, H 6.96, N 19.34; found: C 66.58, H 6.78, N 19.35.

N-*(1-Cyanoethyl)*-N-*[(2-nitrophenyl)thio]phenylalaninamide* (**17**). Compound **16** (434.5 mg, 2 mmol) was dissolved in dry pyridine (2 ml) and evaporated under high vacuum. The residue was again taken up in dry pyridine (2 ml), treated with a few crystals of DMAP and 2-nitrobenzenesulfenyl chloride (417 mg, 2.2 mmol), and stirred at r.t. Following complete reaction (*ca.* 1 h, TLC monitoring), a few drops of MeOH were added, and the mixture was evaporated under high vacuum. FC of the residue (silica gel, CH_2Cl_2/MeOH 95:5) gave **17** (735.7 mg, 99%). Light yellow foam. TLC (CH_2Cl_2/MeOH 95:5): R_f 0.22–0.45. ^{1}H- and ^{13}C-NMR ($CDCl_3$): not assigned due to complexity (rotation and inversion isomers). MS: 370 (< 1, M^+), 326 (2.7, $[M-CONH_2]^+$), 216 (19), 215 (100, $[M-NO_2C_6H_4SH]^+$), 214 (13), 186 (11), 154 (11), 153 (23), 137 (17), 91 (29). Anal. calc. for $C_{18}H_{18}N_4O_3S$ (370.43): C 58.36, H 4.90, N 15.12; found: C 59.06, H 5.15, N 15.23.

*6-Amino-3-benzyl-5-methylpyrazin-2(1*H*)-one* (**18**). *a*) To a soln. of **17** (180 mg, 486 μmol) in THF (1 ml), 1.5M LDA in cyclohexane (0.33 ml, 0.5 mmol) was added dropwise at −78° (→ immediately deep red soln. and precipitation (starting material)). Following complete addition, the mixture was slowly warmed, the starting material again went into soln., and the mixture was stirred for 2 h at r.t. Then 1-(trimethylsilyl)-1*H*-imidazole (0.37 ml, 2.52 mmol) was added dropwise, and the mixture was further stirred overnight at r.t. The entire mixture was added to a sat. aq. NH_4Cl soln. (10 ml), and the mixture was extracted with AcOEt. The combined org. phase was dried (Na_2SO_4) and evaporated, the residue (0.19 g) taken up in MeCN, and the product precipitated by scratching: **18** (61.3 mg, 59%) as a light yellow powder. The remaining mother liquor was subjected to FC (silica gel, CH_2Cl_2/MeOH 9:1): **18** (28 mg, 27%) as a light brown foam. Total yield of **18**: 85%.

b) To a soln. of **17** (196 mg, 529 μmol) in THF (2 ml), 1.5M LDA in cyclohexane (0.36 ml, 0.54 mmol) was carefully added at −78° (→ immediately deep red soln. and precipitation (starting material)). Following complete addition, the mixture was slowly warmed and stirred for 2 h at r.t. The mixture was again cooled to −78°, a further equivalent of LDA (0.36 ml,0.54 mmol) added dropwise, and the mixture again stirred at r.t. Following a further 2 h, the entire mixture was added to 2M phosphate buffer (pH 7.1; 10 ml), adjusted with 2N HCl to pH 7, and extracted with AcOEt. The combined org. phase was dried (Na_2SO_4) and evaporated, and the residue subjected to FC: **18** (113 mg, 99%) as a yellowish foam. Precipitation from MeCN yielded a light brown solid (92 mg, 80%).

Data of **18**: Yellowish to brown foam. M.p. 187–188°. TLC (CH_2Cl_2/MeOH 9:1): R_f 0.26. UV (EtOH): 373 (9366), 238.5 (10781), 335 (sh). IR (KBr): 3320, 3200, 3080, 3060, 3030, 2920, 1630, 1530, 1495, 1455, 1360, 1340, 1255, 1230, 1165, 1070, 1030, 1000, 950, 860, 750, 700, 590, 550, 505. ^{1}H-NMR ($CDCl_3$): 2.12 (*s*, Me); 3.79 (*s*, PhCH_2), 5.64 (br. *s*, NH_2); 7.10–7.25 (*m*, 5 arom. H). ^{13}C-NMR ((D_6)DMSO): 18.39 (*q*, Me); 37.06 (*t*, CH_2); 122.33 (*s*, arom. C(py)); 125.44, 127.93, 128.35 (3*d*, arom. CH); 130.46 (*s*, arom. C(py)); 140.68 (*s*, arom. C); 147.65 (*s*, arom. C(py)); 154.66 (*s*, arom. C(py)). MS: 216 (13), 215 (100, M^+), 214 (10), 187 (10), 186 (12), 99

(17), 91 (37), 77 (9), 44 (13), 43 (17), 42 (19). Anal. calc. for $C_{12}H_{13}N_3O$ (215.25): C 66.96, H 6.09, N 19.52; found: C 66.79, H 6.09, N 19.78.

3. *Pyrazine Nucleoside Analogs. 2,3-*O*-Isopropylidene-*D*-ribofuranose* (**19a**) [18]. A suspension of ribose (3.0 g, 20 mmol) in 0.2% H_2SO_4/acetone (60 ml) was stirred at r.t. until a clear soln. had formed and no more ribose was present (by TLC; *ca.* 1 h). For workup, solid anh. Na_2CO_3 (2 g, 18.9 mmol) was added, and the mixture was stirred until the soln. was neutral. Salts were filtered off, and the clear soln. was evaporated to a colorless oil (3.79 g), which could be used in the following reaction without further purification. For a higher-quality product, the crude material was purified by FC (CH_2Cl_2/acetone 8:2): **19a** (3.22 g, 85%) as an α/β-D-anomer mixture. TLC (CH_2Cl_2/MeOH 9:1): R_f 0.42. ^{1}H-NMR (200 MHz, $CDCl_3$; only signals of the main component; δ of OH varied): 1.32 (*s*, 3 H, Me_2CO_2); 1.49 (*s*, 3 H, Me_2CO_2); 3.55 (*dd*, J = 4.1, 6.1, OH–C(5)); 3.65–3.81 (*m*, CH_2(5)); 4.42 (*m*, H–C(4)); 4.59 (*d*, J = 5.9, H–C(2)*); 4.69 (*d*, J = 6.4, OH–C(1)); 4.84 (*d*, J = 5.9, H–C(3)*); 5.42 (*d*, J = 6.4, H–C(1)).

*5-*O*-[(*tert*-Butyl)dimethylsilyl]-2,3-*O*-isopropylidene-*D*-ribofuranose* (**19b**). *Method A:* To a soln. of **19a** (2.95 g, 15.51 mmol) and 1*H*-imidazole (2.112 g, 31 mmol) in CH_2Cl_2 (10 ml) at −50°, a soln. of ^{t}BuMe$_2$SiCl (2.57 g, 17.05 mmol) in CH_2Cl_2 (3 ml) was slowly added within 1 h. The mixture was stirred at −30 to −50° until the reaction was complete (*ca.* 6 h). Adsorption on silica gel (*ca.* 7 g) and FC (Et_2O/hexane 3:7) yielded **19b** as a colorless oil (3.53 g, 75%) which crystallized on standing, besides the 1,5-*O*-disilylated compound (α- and β-D together: 611.5 mg, 9%). Product **19b** crystallized after FC from a conc. soln. as fine white needles, which then were anomerically pure.

Method B: To a soln. of **19a** (2.91 g, 15.3 mmol) in CH_2Cl_2 (10 ml) at −70°, Et_3N (2.55 ml, 18.37 mmol) and DMAP (75 mg, 0.61 mmol) were slowly added. The mixture was warmed to r.t., and a soln. of ^{t}BuMe$_2$SiCl (2.538 g, 16.84 mmol) in CH_2Cl_2 (5 ml) was added within 1 h. After *ca.* 6 h of stirring at −30 to −50°, TLC (Et_2O/hexane 6:4) showed completion. Adsorption on silica gel (*ca.* 8 g) and FC (Et_2O/hexane 2:8) yielded **19b** (3.90 g, 84%) as a colorless oil besides a small amount of the 1-*O*-silylated product. Even though *Method B* was usually higher yielding, the product from *Method A* was easier to purify.

Data of **19b**: Crystals. M.p. 49–50°. TLC (Et_2O/hexane 3:7): R_f 0.22. ^{1}H-NMR ($CDCl_3$; the anomers are arbitrarily named a and b; ratio a/b typically 5:1): 0.06 (*s*, 6 H, b-Me_2Si); 0.15 (2*s*, 6 H, a-Me_2Si); 0.89 (*s*, 9 H, b-^{t}BuSi); 0.93 (*s*, 9 H, a-^{t}BuSi); 1.33, 1.49 (2*s*, each 3 H, a-Me_2CO_2); 1.40, 1.56 (2*s*, each 3 H, b-Me_2CO_2); 3.65 (*dd*, 1 H, b-CH_2(5)); 3.72–3.81 (*m*, 3 H, a-CH_2(5), b-CH_2(5)); 3.91 (*d*, J = 11.5, 1 H, b-OH); 4.15 (*m*, 1 H, b-H–C(4)); 4.36 (*m*, 1 H a-H–C(4)); 4.51 (*d*, J = 5.9, 1 H, a-H–C(2)*); 4.55 (*dd*, J = 4.0, 6.1, 1 H, b-H–C(2)); 4.70 (*d*, J = 5.9, 1 H, a-H–C(3)*); 4.73 (*dd*, J = 0.8, 6.1, 1 H, b-H–C(3)); 4.76 (*d*, J = 11.9, 1 H, a-OH); 5.29 (*d*, J = 11.9, 1 H, a-H–C(1)); 5.46 (*dd*, J = 4.0, 11.5, 1 H, b-H–C(1)). ^{13}C-NMR ($CDCl_3$): −5.65 (*q*, Me_2Si); 18.29 (*s*, Me_3CSi); 24.96, 26.51 (2*q*, Me_2CO_2); 25.80 (*q*, Me_3CSi); 64.87 (*t*, C(5)); 81.80, 87.04, 87.69 (3*d*, C(2), C(3), C(4)); 103.52 (*d*, C(1)); 112.10 (*s*, Me_2CO_2). MS: 303 (<1, M^+), 287 (11), 247 (19), 189 (16), 171 (23), 159 (12), 143 (50), 131 (15), 129 (72), 117 (57), 105 (10), 102 (13), 101 (31), 97 (13), 89 (25), 85 (10), 75 (100), 73 (51), 69 (26), 59 (43), 57 (10), 55 (21), 43 (33), 41 (15).

*Data of 1,5-*O*-Disilylated By-product:* TLC (Et_2O/hexane 3:7): R_f 0.64, 0.74.

*Data of 1-*O*-Silylated By-product:* TLC (hexane/Et_2O 4:6): R_f 0.54; R_f of **19b** 0.60.

N-*[(Benzyloxy)carbonyl]-2-(2,3-*O*-isopropylidene-β-*D*-ribofuranosyl)glycine Methyl Ester* (**22a/b**). KO^{t}Bu (134.3 mg, 1.21 mmol) was suspended at −78° in CH_2Cl_2 (1.5 ml). Phosphorylglycinate **20** (401.3 mg, 1.2 mmol) in CH_2Cl_2 (1.5 ml) was added dropwise. After *ca.* 15 min stirring, the soln. was taken up in a syringe and added dropwise under cooling *via* syringe (dry ice) at 0° within 25 min to a soln. of **19a** (190.4 mg, 1 mmol) in CH_2Cl_2 (1 ml). The mixture was stirred for 0.5 h at 0° and then for 6 days at r.t. Although the starting materials were not completely consumed, the mixture was worked up by dilution with CH_2Cl_2 (7 ml). The org. phase was washed with sat. aq. NaCl soln. (2 × 10 ml), dried ($MgSO_4$), and evaporated, and the residue subjected to FC (silica gel, CH_2Cl_2/acetone 8:2): stereoisomer mixture **22a/b** (75 mg, 19%). Colorless oil ^{1}H-NMR ($CDCl_3$): 1.32 (*s*, 3 H, Me_2CO_2); 1.53 (*s*, 3 H, Me_2CO_2); 2.90 (br. *s*, OH); 3.65–3.83 (*m*, and *s* at 3.77, 5 H, CH_2(5′), MeO); 4.02 (*m*, 1 H, H–C(2)*); 4.43 (*t*, J = 3.4, H–C(1′)*); 4.60–4.68 (*m*, 3 H, H–C(2′), H–C(3′), H–C(4′)*); 5.12 (2*s*, 2 H, PhCH_2); 6.23 (*d*, J = 2.4, 1 H, NH); 7.28–7.35 (*m*, 5 H, Ph). ^{13}C-NMR ($CDCl_3$): 25.46 (*q*, Me_2CO_2); 27.43 (*q*, Me_2CO_2); 52.79 (*q*, MeO); 55.96 (*d*, N$CHCO_2$Me); 62.41 (*t*, HOCH_2*); 67.23 (*t*, PhCH_2*); 81.01, 82.18, 84.32, 85.37 (4*d*, C(1′), C(2′), C(3′), C(4′)); 114.15 (*s*, Me_2CO_2); 128.03, 128.19, 128.52 (3*d*, arom. CH); 136.22 (*s*, arom. C); 156.93 (*s*, NHCO_2CH_2Ph); 170.59 (*s*, CO_2Me).

N-*[(Benzyloxy)carbonyl]-2-{5-*O*-[(*tert*-butyl)dimethylsilyl]-2,3-*O*-isopropylidene-β-*D*-ribofuranosyl}glycine Methyl Ester* (**23a/b**). KO^{t}Bu (1.557 g, 13.87 mmol) was suspended at −78° in CH_2Cl_2 (20 ml). Phosphorylglycinate **20** (4.596 g, 13.87 mmol) in CH_2Cl_2 (12 ml) was then injected. After *ca.* 1 h stirring, the cooling bath was replaced with an ice bath, and **19b** (3.52 g, 11.56 mmol) in CH_2Cl_2 (23 ml) was slowly added

dropwise within 1 h. The mixture was stirred for 6 h at 0° and then overnight at r.t. The mixture was diluted with CH_2Cl_2 (*ca.* 150 ml), the org. phase washed with sat. aq. NaCl soln. (2 × 80 ml), dried ($MgSO_4$), filtered, and the crude product adsorbed on silica gel (13 g). FC (silica gel, Et_2O/hexane 2:8, then 4:6) gave a small amount of starting material (*ca.* 5%) and stereoisomer mixture **23a/b** (5.313 g, 90%; ratio *ca.* 3:2) as a colorless oil. A third product, probably the α-D-anomer, was isolated in small amounts (< 3%). The mixture **23a/b** could be partly separated by FC.

Data of **23a**: Colorless oil. TLC (hexane/Et_2O 7:3): R_f 0.36. IR ($CHCl_3$): 3420, 2990, 2950, 2930, 2900, 2860, 1755, 1725, 1510, 1470, 1460, 1455, 1435, 1385, 1375, 1335, 1290, 1260, 1230, 1170, 1155, 1135, 1115, 1080, 1030, 1005, 975, 910, 885, 855, 840, 815, 700. ^{1}H-NMR (400 MHz, $CDCl_3$): 0.03 (*s*, 1 MeSi); 0.05 (*s*, 1 MeSi); 0.84 (*s*, ^{t}BuSi); 1.33 (*s*, 3 H, Me_2CO_2); 1.53 (*s*, 3 H, Me_2CO_2); 3.72 (*dd*, *J* = 2.4, J_{gem} = 11.4, 1 H, CH_2(5′)); 3.76 (*s*, MeO); 3.83 (*dd*, *J* = 2.6, J_{gem} = 11.4, 1 H, CH_2(5′)); 3.97 (*m*, H–C(4′)); 4.52–4.66 (*m*, H–C(2′), H–C(3′), H–C(1′), H–C(2)); 5.12 (*s*, 1 H of PhCH_2); 5.13 (*s*, 1 H of PhCH_2); 5.82 (*d*, *J* = 9.5, NH); 7.30–7.37 (*m*, Ph). ^{13}C-NMR (100 MHz, $CDCl_3$): −5.52 (*q*, MeSi); −5.42 (*q*, MeSi); 18.52 (*s*, Me_3*C*Si); 25.55 (*q*, *Me*$_2$CO$_2$); 25.94 (*q*, *Me*$_3$CSi); 27.60 (*Me*$_2$CO$_2$); 52.55 (*q*, MeO); 55.87 (*d*, N*C*HCO$_2$Me); 62.98 (*t*, C(5′)); 67.20 (*t*, PhCH_2); 80.32, 82.01, 83.30, 84.97 (4*d*, C(1′), C(2′), C(3′), C(4′)); 113.93 (*s*, Me_2*C*O$_2$); 128.12, 128.36, 128.39 (3*d*, arom. CH); 136.12 (*s*, arom. C); 156.75 (*s*, NH*C*O$_2$CH$_2$Ph); 170.32 (*s*, *C*O$_2$Me). GC/MS (P1001020): t_R 20.2; 452, 394, 344, 287, 213, 171, 117, 91 (100). MS: 509.2 (< 1, M^+), 452 (17, $[M - {}^t\text{Bu}]^+$), 287 (14), 129 (10), 91 (100), 73 (10).

Data of **23b**: Colorless oil. TLC (hexane/Et_2O 7:3): R_f 0.33. ^{1}H-NMR ($CDCl_3$): 0.07 (2*s*, Me_2Si); 0.89 (*s*, ^{t}BuSi); 1.33 (*s*, 3 H, Me_2CO_2); 1.51 (*s*, 3 H, Me_2CO_2); 3.68–3.76 (*m*, 5 H, MeO, CH_2(5′)); 4.06–4.11 (*m*, H–C(2), H–C(4′)*); 4.51–4.61 (*m*, H–C(1′)*, H–C(3′)**); 4.70–4.74 (*m*, H–C(2′)**); 5.11 (*s*, PhCH_2); 5.51 (br. *d*, *J* = 5.9, NH); 7.30–7.36 (*m*, Ph). GC/MS (P1001020): t_R 21.5; 452, 408, 394, 344, 287, 213, 171, 117, 91 (100).

N-*[(Benzyloxy)carbonyl]-2-{5*-O-*[(*tert*-butyl)dimethylsilyl]-2,3*-O-*isopropylidene-β*-D-*ribofuranosyl]glycinamide* (**26a/b**). A soln. of **23a/b** (8.715 g, 17.1 mmol) in MeOH (35 ml) was placed in a glass tube and sat. at 0° with NH_3 gas. The tube was sealed, and the mixture was stirred at r.t. for *ca.* 74 h. The soln. was evaporated and the residue adsorbed on silica gel. FC (silica gel, Et_2O/hexane 8:2, then AcOEt/hexane 1:1) yielded diastereoisomer mixture **26a/b** (7.788 g, 92%) as a colorless oil. The diastereoisomers could be separated only partially. Isomerically pure samples of **23** gave an epimer mixture **26a/b**, indicating that epimerization occurred upon ammoniolysis of the Z-protected derivative.

Data of **26a**: TLC (Et_2O/hexane 8:2): R_f 0.23. IR ($CHCl_3$): 3520, 3480, 3400, 3340, 3000, 2990, 2950, 2930, 2890, 2860, 1725, 1695, 1585, 1575, 1500, 1470, 1465, 1455, 1415, 1385, 1375, 1325, 1310, 1255, 1160, 1125, 1080, 1030, 1015, 980, 855, 835, 815, 700. ^{1}H-NMR ($CDCl_3$): 0.06 (*s*, Me_2Si); 0.87 (*s*, ^{t}BuSi); 1.32 (*s*, 3 H, Me_2CO_2); 1.52 (*s*, 3 H, Me_2CO_2); 3.73 (*dd*, *J* = 2.0, J_{gem} = 11.3, 1 H, CH_2(5′)); 3.91 (*dd*, *J* = 2.4, J_{gem} = 11.3, 1 H, CH_2(5′)); 3.95 (*m*, H–C(2)*); 4.46 (*m*, H–C(1′)*, H–C(4′)*); 4.53 (*m*, H–C(3′)**); 4.64 (*dd*, *J* = 4.9, 6.4, H–C(2′)**); 5.12, 5.14 (2*s*, 1 H each, PhCH_2); 5.82 (br. *s*, 1 H of CONH$_2$); 6.00 (br. *d*, *J* = 6.6, PhCH$_2$OCON*H*); 6.57 (br. *s*, 1 H of CONH$_2$); 7.30–7.37 (*m*, Ph). ^{13}C-NMR ($CDCl_3$): −5.38 (*q*, Me_2Si); 18.58 (*s*, Me_3*C*Si); 25.57 (*q*, *Me*$_2$CO$_2$); 26.01 (*q*, *Me*$_3$CSi); 27.55 (*Me*$_2$CO$_2$); 55.64 (*d*, N*C*HCONH$_2$); 62.49 (*t*, SiOCH$_2$); 67.33 (*t*, PhCH_2); 79.87, 81.74, 82.77, 84.48 (4*d*, C(1′), C(2′), C(3′), C(4′)); 114.53 (*s*, Me_2*C*O$_2$); 128.28, 128.51 (2*d*, arom. CH); 136.02 (*s*, arom. C); 156.38 (*s*, NH*C*O$_2$CH$_2$Ph); 171.46 (*s*, CONH$_2$). MS: 494.5 (2, M^+), 479.5 (1, $[M - \text{Me}]^+$), 450.5 (5, $[M - \text{CONH}_2]^+$), 437 (7, $[M - {}^t\text{Bu}]^+$), 406 (5), 379 (3), 348 (5), 287 (7, [*M* − substituent at C(1′)]$^+$), 171 (6), 129 (9), 117 (8), 101 (5), 97 (6), 92 (9), 91 (100), 75 (13), 73 (17), 59 (12), 57 (12), 43 (19).

Data of **26b**: TLC (Et_2O/hexane 8:2): R_f 0.29. ^{1}H-NMR ($CDCl_3$): 0.12 (*s*, MeSi); 0.13 (*s*, MeSi); 0.91 (*s*, ^{t}BuSi); 1.30 (*s*, 3 H, Me_2CO_2); 1.51 (*s*, 3 H, Me_2CO_2); 3.73 (*dd*, *J* = 4.9, J_{gem} = 11.5, 1 H, CH_2(5′)); 3.83 (*dd*, *J* = 3.3, J_{gem} = 11.5, 1 H, CH_2(5′)); 4.01 (*dd*, *J* = 2.7, 9.9, H–C(2)*); 4.18–4.26 (*m*, H–C(4′), H–C(1′)*); 4.64–4.66 (*m*, H–C(3′)*); 4.81–4.84 (*m*, H–C(2′)*); 5.12 (*s*, PhCH_2); 5.50 (br. *s*, 1 H of CONH$_2$); 5.72 (br. *s*, PhCH$_2$OCON*H*); 6.73 (br. *s*, 1 H of CONH$_2$); 7.30–7.37 (*m*, Ph). ^{13}C-NMR ($CDCl_3$): −5.40 (*q*, MeSi); −5.31 (*q*, MeSi); 18.48 (*s*, Me_3*C*Si); 25.48 (*q*, *Me*$_2$CO$_2$); 25.93 (*q*, *Me*$_3$CSi); 27.29 (*Me*$_3$CO$_2$); 55.65 (*d*, N*C*HCONH$_2$); 63.81 (*t*, SiOCH$_2$); 67.20 (*t*, PhCH_2); 81.22, 83.35, 85.49, 86.37 (4*d*, C(1′), C(2′), C(3′), C(4′)); 113.56 (*s*, Me_2*C*O$_2$); 127.99, 128.16, 128.51 (3*d*, arom. CH); 136.12 (*s*, arom. C); 156.67 (*s*, NH*C*O$_2$Ph); 171.44 (*s*, CONH$_2$).

2-{5-O-*[(*tert*-Butyl)dimethylsilyl]-2,3*-O-*isopropylidene-β*-D-*ribofuranosyl]glycine Methyl Ester* (**27a/b**). A soln. of **23a/b** (1.80 g, 3.53 mmol) in MeOH (14 ml) was treated with 10% Pd/C (120 mg) with stirring under H_2 (slight overpressure with balloon) at r.t. After complete consumption of **23a/b** (8 h, TLC monitoring), the soln. was filtered through a bed of *Celite* and evaporated. The crude **27a/b** (1.324 g, quant.) was sufficiently pure for the next reaction. Chromatography (Et_2O/hexane 8:2) partly separated the two isomers **27a/b** (yield after chromatography 93%). By using isomerically pure samples of **23**, epimerically pure products were obtained.

Data of **27a**: TLC (Et_2O/hexane 8:2): R_f 0.32. IR ($CHCl_3$): 2990, 2960, 2930, 2900, 2860, 1740, 1600, 1470, 1465, 1440, 1385, 1375, 1260, 1175, 1160, 1140, 1075, 1000, 975, 940, 900, 840, 815. ^{1}H-NMR ($CDCl_3$): 0.07 (*s*, Me_2Si); 0.90 (*s*, ^{t}BuSi); 1.35 (*s*, 3 H, Me_2CO_2); 1.54 (*s*, 3 H, Me_2CO_2); 1.67 (br. *s*, NH_2); 3.57 (*d*, *J* = 3.2, H–C(2)); 3.72 (*dd*, *J* = 3.5, J_{gem} = 11.2, 1 H, CH_2(5′)); 3.75 (*s*, MeO); 3.80 (*dd*, *J* = 3.3, J_{gem} = 11.2, 1 H, CH_2(5′)); 3.98 (*ddd*(= *dt*), *J* = 3.4, 4.6, H–C(4′)); 4.41 (*dd*(= *t*), *J* = 3.4, H–C(1′)); 4.67 (*dd*, *J* = 4.6, 6.4, H–C(3′)); 4.82 (*dd*, *J* = 3.6, 6.4, H–C(2′)); NOE data were used to assign configuration. ^{13}C-NMR ($CDCl_3$): −5.50 (*q*, MeSi); −5.41 (*q*, MeSi); 18.39 (*s*, Me_3*C*Si); 25.64 (*q*, *Me*$_2$CO$_2$); 25.90 (*q*, *Me*$_3$CSi); 27.61 (*Me*$_2$CO$_2$); 52.25 (*q*, MeO); 56.46 (*d*, N*C*HCO$_2$Me); 63.06 (*t*, SiOCH$_2$); 80.90, 82.42, 84.68, 85.17 (4*d*, C(1′), C(2′), C(3′), C(4′)); 113.72 (*s*, Me_2*C*O$_2$); 174.12 (*s*, *C*O$_2$Me). GC/MS (P15020): t_R 5.6; 360, 318, 287, 242, 200, 171, 129, 73 (100). MS: 377 (12), 376 (49, $[M+1]^+$); 360 (13, $[M-Me]^+$), 319 (20), 318 (98, $[M-{}^t\text{Bu}]$, 317 (17), 316 (19), 288 (10), 287 (48, $[M-NH_2CHCO_2Me]^+$), 286 (12), 260 (16, $[M-{}^t\text{BuMe}_2\text{Si}]^+$), 229 (10), 200 (10), 187 (13), 172 (10), 171 (17), 159 (10), 143 (10), 131 (12), 130 (16), 129 (81), 128 (14), 126 (10), 117 (55), 116 (18), 115 (23), 114 (11), 101 (27), 97 (24), 89 (39), 88 (51), 85 (16), 84 (10), 81 (10), 75 (72), 74 (12), 73 (100), 70 (11), 69 (16), 59 (42), 58 (14), 57 (25), 56 (13), 55 (23), 43 (35), 41 (21), 33 (14).

Data of **27b**: TLC (Et_2O/hexane 8:2): R_f 0.24. IR ($CHCl_3$): 2990, 2960, 2930, 2900, 2860, 1740, 1600, 1470, 1465, 1440, 1385, 1375, 1260, 1175, 1160, 1140, 1080, 1010, 975, 860, 840, 815. ^{1}H-NMR ($CDCl_3$): 0.07 (*s*, Me_2Si); 0.90 (*s*, ^{t}BuSi); 1.34 (*s*, 3 H, Me_2CO_2); 1.53 (*s*, 3 H, Me_2CO_2); 1.64 (br. *s*, NH_2); 3.72 (*dd*, *J* = 3.5, J_{gem} = 11.2, 1 H, CH_2(5′)); 3.73 (*d*, *J* = 5.9, H–C(2)); 3.75 (*s*, MeO); 3.78 (*dd*, *J* = 3.2, J_{gem} = 11.2, 1 H, CH_2(5′)); 4.03 (*q*-like *m*, *J* = 3.3, H–C(4′)); 4.18 (*dd*-like *m*, *J* = 3.1, 5.8, H–C(1′)); 4.64, 4.65 (2*m*, H–C(2′), H–C(3′)); NOE data were used to assign configuration. ^{13}C-NMR ($CDCl_3$): −5.50 (*q*, MeSi); −5.41 (*q*, MeSi); 18.42 (*s*, Me_3*C*Si); 25.61 (*q*, *Me*$_2$CO$_2$); 25.94 (*q*, *Me*$_3$CSi); 27.54 (*Me*$_2$CO$_2$); 52.12 (*q*, MeO); 56.46 (*d*, N*C*HCO$_2$Me); 63.42 (*t*, SiOCH$_2$); 81.28, 82.06, 85.10, 85.75 (4*d*, C(1′), C(2′), C(3′), C(4′)); 113.72 (*s*, Me_2*C*O$_2$); 173.35 (*s*, *C*O$_2$Me). MS: 3.77 (4), 376 (16, $[M+1]^+$), 360 (13, $[M-Me]^+$), 319 (12), 318 (62, $[M-{}^t\text{Bu}]^+$), 316 (12), 288 (12), 287 (56, $[M-NH_2CHCO_2Me]$), 260 (30, $[M-{}^t\text{BuMe}_2\text{Si}]^+$), 229 (11), 187 (15), 172 (18), 172 (12), 170 (20), 143 (12), 136 (10), 133 (11), 131 (10), 130 (15), 129 (92), 117 (52), 116 (12), 115 (21), 114 (12), 101 (28), 97 (22), 89 (37), 88 (62), 85 (17), 84 (10), 81 (11), 75 (72), 74 (11), 73 (100), 70 (11), 69 (15), 59 (39), 58 (13), 57 (24), 56 (12), 55 (22), 43 (33), 41 (19), 33 (14).

*2-{5-*O*-[(*tert*-Butyl)dimethylsilyl]-2,3-*O*-isopropylidene-β-*D*-ribofuranosyl}glycinamide* (**28a/b**). *Method A, by Removal of the Z-Group from* **26a/b**: To a soln. of **26a/b** (6.84 g, 13.8 mmol) in MeOH (80 ml) was added Pd/C (6.9 g). The mixture was stirred for 48 h under H_2 at r.t. The mixture was filtered through a bed of *Celite*, which was washed with MeOH. The filtrate and washings were evaporated. FC (silica gel, CH_2Cl_2/MeOH 95:5) of the residue yielded **28a/b** (4.45 g, 89%) as a colorless oil. The reaction proceeded without epimerization.

Method B, by Aminolysis of **27a/b**: A soln. of **27a/b** (1.132 g, 3.015 mmol) in MeOH (30 ml) in a glass tube was saturated at 0° with NH_3 gas. The tube was sealed and the mixture stirred for 2 d at r.t. The mixture was evaporated and the residue purified by FC (silica gel, CH_2Cl_2/MeOH 95:5): **28a/b** (1.082 g, quant.) as a colorless oil. The reaction proceeded without epimerization.

Data of **28a**: TLC (CH_2Cl_2/MeOH 9:1): R_f 0.44. IR ($CHCl_3$): 3500, 3380, 3000, 2960, 2935, 2860, 1685, 1590, 1550, 1470, 1465, 1385, 1375, 1260, 1160, 1135, 1080, 1005, 980, 970, 905, 860, 840, 815. ^{1}H-NMR ($CDCl_3$): 0.07 (*s*, MeSi); 0.08 (*s*, MeSi); 0.90 (*s*, ^{t}BuSi); 1.35 (*s*, 3 H, Me_2CO_2); 1.54 (*s*, 3 H, Me_2CO_2); 1.77 (br. *s*, NH_2); 3.52 (*d*, *J* = 3.9, H–C(2)); 3.75 (*dd*, *J* = 2.6, J_{gem} = 11.3, 1 H, CH_2(5′)); 3.86 (*dd*, *J* = 2.7, J_{gem} = 11.3, 1 H, CH_2(5′)); 4.04 (*ddd*(= *dt*), *J* = 2.6, 3.9, H–C(4′)); 4.36 (*dd*(= *t*), *J* = 4.1, H–C(1′)); 4.63 (*dd*, *J* = 4.4, 6.6, H–C(2′)); 4.69 (*dd*, *J* = 4.0, 6.7, H–C(3′)); 5.49 (br. *s*, 1 H of $CONH_2$); 7.31 (br. *s*, 1 H of $CONH_2$). ^{13}C-NMR ($CDCl_3$): −5.50 (*q*, MeSi); −5.41 (*q*, MeSi); 18.42 (*s*, Me_3*C*Si); 25.61 (*q*, *Me*$_2$CO$_2$); 25.93 (*q*, *Me*$_3$CSi); 27.54 (*Me*$_2$CO$_2$); 55.97 (*d*, N*C*HCONH$_2$); 63.02 (*t*, SiOCH$_2$); 80.83, 82.09, 84.59, 84.78 (4*d*, C(1′), C(2′), C(3′), C(4′)); 114.21 (*s*, Me_2*C*O$_2$); 175.19 (*s*, CONH$_2$). GC/MS (P1001020): t_R 9.4. GC/MS (P10010): t_R 15.1; 360, 345, 318, 303, 245, 171, 97, 73 (100). MS: 362 (4), 361 (5), 318 (11), 317 (36), 316 (57), 305 (24), 304 (51), 303 (53), 287 (22), 259 (11), 258 (21), 245 (10), 171 (39), 131 (10), 130 (12), 129 (55), 126 (10), 117 (39), 116 (11), 115 (18), 101 (18), 98 (11), 97 (25), 89 (35), 85 (16), 76 (10), 75 (70), 74 (23), 73 (100), 70 (10), 69 (12), 59 (33), 58 (13), 57 (20), 56 (11), 55 (16), 43 (23), 41 (14).

Data of **28b**: TLC (CH_2Cl_2/MeOH 9:1): R_f 0.43. IR ($CHCl_3$): 3500, 3380, 3010, 2950, 2930, 2900, 2880, 2860, 1685, 1585, 1555, 1470, 1460, 1385, 1375, 1255, 1160, 1135, 1080, 1005, 970, 935, 860, 835, 810. ^{1}H-NMR ($CDCl_3$): 0.08 (*s*, MeSi); 0.09 (*s*, MeSi); 0.91 (*s*, ^{t}BuSi); 1.35 (*s*, 3 H, Me_2CO_2); 1.53 (*s*, 3 H, Me_2CO_2); 1.77 (br. *s*, NH_2); 3.52 (*d*, *J* = 6.7, H–C(2)); 3.73 (*dd*, *J* = 3.3, J_{gem} = 11.3, 1 H, CH_2(5′)); 3.83 (*dd*, *J* = 2.8, J_{gem} = 11.3, 1 H, CH_2(5′)); 4.11 (*m*, H–C(4′), H–C(1′)); 4.64 (*dd*, *J* = 3.6, 6.5, H–C(2′)*); 4.75 (*dd*, *J* = 4.0, 6.5, H–C(3′)*); 5.7 (br. *s*, 1 H of $CONH_2$); 6.9 (br. *s*, 1 H of $CONH_2$). ^{13}C-NMR ($CDCl_3$): −5.48 (*q*, MeSi); −5.37 (*q*, MeSi); 18.39 (*s*, Me_3*C*Si);

25.57 (*q*, *Me*$_2$CO$_2$); 25.93 (*q*, *Me*$_3$CSi); 27.53 (*Me*$_2$CO$_2$); 56.58 (*d*, N*C*HCONH$_2$); 63.54 (*t*, SiOCH$_2$); 81.12, 82.41, 85.16, 85.81 (4*d*, C(1′), C(2′), C(3′), C(4′)); 113.88 (*s*, Me$_2$*C*O$_2$); 174.91 (*s*, CONH$_2$). GC/MS (P1001020): t_R 9.3. GC/MS (P10010): t_R 15.0; 360, 345, 318, 303, 258, 171, 97, 73 (100). MS: 361 (13), 345 (12), 317 (12), 316 (52), 304 (17), 303 (82), 287 (22), 258 (15), 245 (30), 173 (10), 171 (38), 129 (52), 117 (27), 116 (10), 115 (14), 101 (14), 97 (21), 89 (28), 86 (10), 85 (13), 84 (13), 75 (59), 74 (20), 73 (100), 70 (10), 69 (10), 59 (28), 58 (12), 57 (14), 55 (16), 43 (23), 41 (13).

*2-{5-*O*-[(*tert*-Butyl)dimethylsilyl]-2,3-*O*-isopropylidene-β-*D*-ribofuranosyl}-*N*-(cyanomethyl)glycinamide* (**29**). A soln. of KCN (126 mg, 1.93 mmol) in H_2O (4 ml) was acidified with AcOH to *ca.* pH 6, treated with a soln. of **28a/b** (632 mg, 1.75 mmol) in dioxane (4 ml), and acidified again with AcOH to pH 6. Formalin (135 µl, *ca.* 1.755 mmol) was slowly added at r.t. within 2 h. After complete consumption of **28a/b** (TLC monitoring), the mixture was added to sat. aq. Na_2CO_3 soln., the aq. layer extracted with CH_2Cl_2, the extract dried (Na_2CO_3) and evaporated, and the residue subjected to FC (silica gel, CH_2Cl_2/MeOH 95:5): **29** (648 mg, 93%). Colorless oil, which became solid upon standing. IR ($CHCl_3$): 3510, 3480, 3390, 3340, 3000, 2960, 2940, 2900, 2860, 1690, 1585, 1560, 1470, 1465, 1385, 1375, 1260, 1160, 1130, 1080, 980, 865, 840, 815. TLC (CH_2Cl_2/MeOH 95:5): R_f 0.35. ^{1}H-NMR ($CDCl_3$): 0.10 (2*s*, Me_2Si); 0.92 (*s*, ^{t}BuSi); 1.34 (*s*, 3 H, Me_2CO_2); 1.53 (*s*, 3 H, Me_2CO_2); 2.78 (*ddd*(=*dt*), *J* = 3.1, 7.4, NH); 3.48 (*dd*, *J* = 3.1, 5.3, H–C(2)); 3.66 (*d*, *J* = 7.4, CH_2CN); 3.77 (*dd*, *J* = 2.9, J_{gem} = 11.4, 1 H, CH_2(5′)); 3.87 (*dd*, *J* = 2.9, J_{gem} = 11.4, 1 H, CH_2(5′)); 4.00 (*ddd*(=*dt*), *J* = 2.9, 4.2, H–C(4′)); 4.13 (*dd*, *J* = 3.4, 5.3, H–C(1′)); 4.64 (*dd*, *J* = 3.4, 6.8, H–C(2′)); 4.68 (*dd*, *J* = 4.2, 6.8, H–C(3′)); 5.48 (br. *s*, 1 H of $CONH_2$); 6.99 (br. *s*, 1 H of $CONH_2$). ^{13}C-NMR ($CDCl_3$): −5.38 (*q*, Me_2Si); 18.48 (*s*, Me_3*C*Si); 25.41 (*q*, *Me*$_2$CO$_2$); 25.96 (*q*, *Me*$_3$CSi); 27.39 (*Me*$_2$CO$_2$); 36.09 (*t*, *C*H$_2$CN); 61.79 (*d*, N*C*HCONH$_2$); 62.54 (*t*, SiO*C*H$_2$); 80.28, 82.09, 83.97, 84.87 (4*d*, C(1′), C(2′), C(3′), C(4′)); 114.59 (*s*, Me$_2$*C*O$_2$); 117.12 (*s*, *C*N); 171.76 (*s*, $CONH_2$). MS: 401 (14); 400 (52, $[M+1]^+$), 399 (6, M^+), 384 (13, $[M-Me]^+$), 373 (35, $[M-HCN]^+$), 357 (12), 356 (22), 355 (63, $[M-CONH_2]^+$), 344 (12), 343 (38), 342 (79, $[M-{}^t\text{Bu}]^+$), 328 (16), 316 (28), 315 (74), 297 (28), 288 (12), 287 (45, $[M-CNCH_2NHCHCONH_2]^+$), 286 (11), 284 (41, $[M-TBDMS]^+$), 270 (35), 258 (14), 257 (34), 229 (13), 223 (13), 221 (11), 187 (13), 183 (15), 171 (64), 155 (17), 143 (29), 130 (18), 129 (64), 117 (62), 115 (26), 112 (22), 109 (19), 101 (29), 99 (18), 97 (39), 89 (51), 85 (41), 81 (20), 75 (74), 73 (75), 69 (42), 67 (20), 59 (61), 58 (21), 57 (46), 56 (22), 55 (27), 45 (24), 44 (21), 43 (72), 42 (35), 41 (61), 39 (28).

*2-{5-*O*-[(*tert*-Butyl)dimethylsilyl]-2,3-*O*-isopropylidene-β-*D*-ribofuranosyl}-*N*-(cyanomethyl)-N-[(2-nitrophenyl)thio]glycinamide* (**30**). To a soln. of 2-nitrobenzenesulfenyl chloride (149 mg, 786 µmol) in THF (2.5 ml) was added amide **29** (100 mg, 250 µmol) and PMP (145 µl, 802 µmol). The mixture was stirred under reflux at 90° for 5.5 h. Sat. aq. Na_2CO_3 soln. (10 ml) was added, the mixture extracted with Et_2O (3 × 10 ml), the combined org. phase dried (Na_2CO_3) and evaporated, and the residue subjected to FC (silica gel, CH_2Cl_2/MeOH 97:3): **30** (100 mg, 72%). Light yellow foam. TLC (CH_2Cl_2/MeOH 95:5): R_f 0.54. ^{1}H- and ^{13}C-NMR ($CDCl_3$): not assigned due to complexity (inversion and rotation isomers).

*6-Amino-3-{5-*O*-[(*tert*-butyl)dimethylsilyl]-2,3-*O*-isopropylidene-β-*D*-ribofuranosyl}pyrazin-2(1*H*)-one* (**1a**). To a soln. of **30** (196 mg, 355 µmol) in MeOH (4 ml) 5.4M MeONa in MeOH (0.1 ml, 540 mmol) was added at 0° (→ immediately deep red). After 3.5 h, the mixture was carefully neutralized at 0° with 2N HCl to pH 7 and evaporated. FC (silica gel, CH_2Cl_2/MeOH 9:1) yielded **1a** (69.7 mg, 49%) as an orange yellow foam. A second FC purification (reversed-phase silica gel (*Merck*, Art. *7719*, silica gel *60* silanized, 70–230 mesh ASTM)) gave **1a** (23 mg, 16%). Colorless sample which on exposure to air became yellow. TLC (CH_2Cl_2/MeOH 9:1): R_f 0.26. ^{1}H-NMR (200 MHz, $CDCl_3$): 0.05 (*s*, Me_2Si); 0.88 (*s*, ^{t}BuSi); 1.38 (*s*, 3 H, Me_2CO_2('*exo*')); 1.57 (*s*, 3 H, Me_2CO_2('*endo*')); 3.63 (*dd*, *J* = 6.5, 11.0, 1 H, CH_2(5′)); 3.75 (*dd*, *J* = 4.2, 10.8, 1 H, CH_2(5′)); 4.12 (*m*, H–C(4′)); 4.56 (*dd*, *J* = 4.1, 6.7, H–C(3′)); 5.11 (*d*, *J* = 4.5, H–C(1′)); 5.29 (*dd*, *J* = 4.7, 6.6, H–C(2′)); 5.6 (br. *s*, NH_2); 6.99 (*s*, H–C(5)). ^{13}C-NMR (50 MHz, $CDCl_3$): −5.12 (*q*, Me_2Si); 18.70 (*s*, Me_3*C*Si); 25.67 (*q*, *Me*$_2$CO$_2$); 26.16 (*q*, *Me*$_3$CSi); 27.54 (*q*, *Me*$_2$CO$_2$); 64.70 (*t*, C(5′)); 81.64, 82.10, 82.26, 85.94 (4*d*, C(1′), C(2′), C(3′), C(4′)); 111.56 (*d*, C(5)*); 114.71 (*s*, Me$_2$*C*O$_2$); 132.61 (*s*, C(6)*); 147.70 (*s*, C(3)**); 156.64 (*s*, C(2)**). GC/MS (P15020): t_R 11.0; 396, 354 (100), 325, 287, 208, 124.

*2-{5-*O*-[(*tert*-Butyl)dimethylsilyl]-2,3-*O*-isopropylidene-β-*D*-ribofuranosyl}-*N*-(1-cyanoethyl)glycinamide* (**31**). A soln. of **28a/b** (721 mg, 2 mmol) and KCN (155 mg, 2.38 mmol) in dioxane/H_2O 2:1 (13.5 ml) was carefully acidified to pH 5 with AcOH. Within 30 min, acetaldehyde (0.16 ml, 2.83 mmol) was added slowly by means of a cooled syringe. The mixture was then stirred at r.t. for *ca.* 1 h (complete consumption of **28a/b**). Sat. aq. Na_2CO_3 soln. (*ca.* 50 ml) was added, the mixture extracted with CH_2Cl_2 (4 × 50 ml), the combined org. phase dried (Na_2SO_4) and evaporated, and the residue subjected to FC (silica gel, CH_2Cl_2/MeOH 95:5): **31** (747.8 mg, 90%), mixture of the four epimers (at C(2) and C(2″)) as a colorless foam. With **28a** as starting material, the epimers **31aa/ab** were obtained partly pure and separately analyzed. With **28b** as starting material, both epimers **31ba/bb** could be better separated *via* FC (AcOEt/hexane/EtOH 50:47:3).

Data of **31aa**: TLC (CH_2Cl_2/MeOH 9:1): R_f 0.49. ^{1}H-NMR ($CDCl_3$): 0.09, 0.10 (2*s*, Me_2Si); 0.91 (*s*, ^{t}BuSi); 1.34 (*s*, 3 H, Me_2CO_2); 1.52, 1.53, 1.55 (1*s*, 1*d*, 6 H, *Me*CHCN, Me_2CO_2); 2.78 (*dd*, *J* = 2.0, 10.3, NH); 3.59 (*dd*, *J* = 2.3, 4.6, H–C(2)); 3.71–3.82 (*m*, 2 H, MeC*H*CN, CH_2(5′)); 3.90–3.94 (*m*, 2 H, H–C(4′), CH_2(5′)); 4.12 (*dd*, *J* = 3.8, 4.4, H–C(1′)); 4.54 (*dd*, *J* = 3.7, 6.8, H–C(2′)); 4.67 (*dd*, *J* = 5.3, 6.8, H–C(3′)); 5.54 (br. *s*, 1 H of $CONH_2$); 7.15 (br. *s*, 1 H of $CONH_2$). ^{13}C-NMR ($CDCl_3$): −5.41 (*q*, MeSi); 18.55 (*s*, Me_3CSi); 19.65 (*q*, *Me*CHCN); 25.51 (*q*, Me_2CO_2); 25.97 (*q*, Me_3CSi); 27.48 (*q*, Me_2CO_2); 44.19 (*d*, *C*HCN); 59.80 (*d*, C(2)); 62.01 (*t*, C(5′)); 79.52, 81.96, 83.59, 84.49 (4*d*, C(1′), C(2′), C(3′), C(4′)); 114.72 (*s*, Me_2CO_2); 120.23 (*s*, CN); 171.93 (*s*, $CONH_2$). FAB-MS ($C_{19}H_{35}N_3O_5Si$, 413.59; pos.): 436 (13, $[M+Na]^+$), 416 (11), 415 (30), 414 (100, $[M+H]^+$), 402 (11), 388 (14), 387 (52), 357 (11), 356 (43), 329 (21), 171 (10), 137 (13), 136 (12), 129 (16), 126 (11), 117 (14), 115 (14), 101 (11), 99 (18), 97 (12), 89 (28), 83 (13), 75 (32), 74 (10), 73 (83), 59 (17), 57 (11), 55 (11).

Data of **31ab**: TLC (CH_2Cl_2/MeOH 9:1): R_f 0.44. ^{1}H-NMR ($CDCl_3$; sample contaminated with *ca.* 15% of **31aa**): 0.10 (2*s*, Me_2Si); 0.92 (*s*, ^{t}BuSi); 1.34 (*s*, 3 H, Me_2CO_2); 1.52 (*s*, 3 H, Me_2CO_2); 1.53 (*d*, *J* = 7.0, *Me*CHCN); 2.50 (*dd*, *J* = 3.4, 8.7, NH); 3.48 (*dd*, *J* = 3.4, 6.4, H–C(2)); 3.66–3.83 (*m*, MeC*H*CN, CH_2(5′)); 4.02 (*m*, H–C(4′)); 4.07 (*dd*, *J* = 3.9, 6.4, H–C(1′)); 4.65 (*dd*, *J* = 4.0, 6.6, H–C(3′)); 4.74 (*dd*, *J* = 3.9, 6.7, H–C(2′)); 5.70 (br. *s*, 1 H of $CONH_2$); 6.85 (br. *s*, 1 H of $CONH_2$). ^{13}C-NMR ($CDCl_3$): −5.40, −5.31 (2*q*, Me_2Si); 18.46 (*s*, Me_3CSi); 20.04 (*q*, *Me*CHCN); 25.41 (*q*, Me_2CO_2); 25.96 (*q*, Me_3CSi); 27.35 (*q*, Me_2CO_2); 44.45 (*d*, *C*HCN); 62.38 (*d*, C(2)); 62.94 (*t*, C(5′)); 81.01, 81.90, 84.98 (3*d*, C(1′), C(2′), C(3′), C(4′)); 114.28 (*s*, Me_2CO_2); 120.10 (*s*, CN); 172.21 (*s*, $CONH_2$).

Data of **31ba**: TLC (AcOEt/hexane/EtOH 50:47:3): R_f 0.32. ^{1}H-NMR (300 MHz, $CDCl_3$): 0.13 (*s*, MeSi); 0.14 (*s*, MeSi); 0.94 (*s*, ^{t}BuSi); 1.35 (*s*, Me_2CO_2); 1.49 (*d*, *J* = 7.0, *Me*CHCN); 1.52 (*s*, 3 H, Me_2CO_2); 2.55 (*dd*, *J* = 6.4, 8.9, NH); 3.36 (*dd*, *J* = 6.4, 7.4, H–C(2)); 3.70–3.80 (*m*, 2 H, MeC*H*CN, CH_2(5′)); 3.85 (*dd*, *J* = 2.7, J_{gem} = 11.4, 1 H, CH_2(5′)); 4.03 (*dd*, *J* = 3.6, 7.4, H–C(1′)); 4.16 (*m*, H–C(4′)); 4.63 (*dd*, *J* = 3.2, 6.4, H–C(3′)); 4.69 (*dd*, *J* = 3.6, 6.4, H–C(2′)); 5.64 (br. *s*, 1 H of $CONH_2$); 6.70 (br. *s*, 1 H of $CONH_2$). ^{13}C-NMR (75 MHz, $CDCl_3$): −5.36, −5.24 (2*q*, Me_2Si); 18.52 (*s*, Me_3CSi); 19.65 (*q*, *Me*CHCN); 25.61 (*q*, Me_2CO_2); 26.02 (*q*, Me_3CSi); 27.42 (*q*, Me_2CO_2); 45.13 (*d*, *C*HCN); 61.28 (*d*, C(2)); 63.90 (*t*, C(5′)); 81.24, 82.84, 85.46, 85.59 (4*d*, C(1′), C(2′), C(3′), C(4′)); 113.89 (*s*, Me_2CO_2); 120.26 (*s*, CN); 172.76 (*s*, $CONH_2$). MS: 387 (2), 371 (14), 330 (20), 329 (100), 271 (14), 171 (27), 129 (24), 117 (10), 101 (11), 75 (13), 73 (11).

Data of **31bb**: TLC (AcOEt/hexane/EtOH 50:47:3): R_f 0.25. ^{1}H-NMR (300 MHz, $CDCl_3$): 0.13 (*s*, MeSi); 0.14 (*s*, MeSi); 0.93 (*s*, ^{t}BuSi); 1.37 (*s*, 3 H, Me_2CO_2); 1.52 (*s*, 3 H, Me_2CO_2); 1.53 (*d*, *J* = 6.9, *Me*CHCN); 2.19 (*dd*, *J* = 5.8, 7.6, NH); 3.39 (*dt*, *J* = 8.0, H–C(2)); 3.68 (*dd*, *J* = 5.8, 6.9, MeC*H*CN); 3.75 (*dd*, *J* = 3.6, J_{gem} = 11.5, 1 H, CH_2(5′)); 3.85 (*dd*, *J* = 2.9, J_{gem} = 11.5, 1 H, CH_2(5′)); 3.97 (*dd*, *J* = 3.5, 8.2, H–C(1′)); 4.16 (*m*, H–C(4′)); 4.66 (*dd*, *J* = 3.3, 6.4, H–C(3′)); 4.80 (*dd*, *J* = 3.5, 6.4, H–C(2′)); 5.67 (br. *s*, 1 H of $CONH_2$); 6.76 (br. *s*, 1 H of $CONH_2$). ^{13}C-NMR (75 MHz, $CDCl_3$): −5.31, −5.24 (2*q*, Me_2Si); 18.52 (*s*, Me_3CSi); 19.88 (*q*, *Me*CHCN); 25.57 (*q*, Me_2CO_2); 26.03 (*q*, Me_3CSi); 27.39 (*q*, Me_2CO_2); 44.64 (*d*, *C*HCN); 61.93 (*d*, C(2)); 63.71 (*t*, C(5′)); 80.99, 83.34, 84.68, 85.85 (4*d*, C(1′), C(2′), C(3′), C(4′)); 113.91 (*s*, Me_2CO_2); 120.39 (*s*, CN); 172.73 (*s*, $CONH_2$). MS: 386 (5), 371 (15), 330 (23), 329 (100), 287 (10), 271 (16), 171 (40), 157 (14), 129 (43), 117 (24), 101 (32), 71 (18), 99 (29), 98 (13), 97 (11), 89 (15), 85 (13), 75 (44), 73 (59), 59 (23), 57 (12), 56 (10), 55 (13).

*2-[5-*O*-[(*tert*-Butyl)dimethylsilyl]-2,3-*O*-isopropylidene-β-*D*-ribofuranosyl]-*N*-(1-cyanoethyl)-*N*-[(2-nitrophenyl)thio]glycinamide* (**32**). Amine **31** (639.7 mg, 1.547 mmol) as a mixture of stereoisomers was dissolved in dry pyridine (5 ml), evaporated under high vacuum, and again dissolved in dry pyridine (5 ml). This soln. was treated with 2-nitrobenzenesulfenyl chloride (440 mg, 2.32 mmol) and a few small crystals of DMAP, and stirred at r.t. After 30 min, a precipitate began to form, and the reaction was complete (TLC monitoring). Following addition of MeOH (0.5 ml), the mixture was evaporated under high vacuum, and the residue subjected to FC (silica gel, CH_2Cl_2/acetone 95:5): **32** (865.3 mg, 99%) as a mixture of stereoisomers. Light yellow foam. TLC (CH_2Cl_2/MeOH 95:5): R_f 0.56 (**32a**); 0.68 and 0.47 (**32b**). IR (identical for **32a** and **32b**; $CHCl_3$): 3480, 3340, 3000, 2960, 2940, 2860, 1695, 1595, 1570, 1515, 1450, 1385, 1340, 1310, 1255, 1160, 1080, 975, 930, 895, 835. ^{1}H-NMR ($CDCl_3$): not assigned due to complexity (rotation and inversion isomers). FAB-MS (pos.): 589 (5, $[M+Na]^+$), 567 (7, $[M+H]^+$); 540 (16, $[M-Me]^+$), 509 (30), 412 (18), 388 (30), 387 (100), 330 (15), 329 (64), 171 (11), 155 (11), 154 (63), 138 (60), 136 (10), 129 (18), 117 (12), 115 (10), 106 (13), 99 (12), 98 (24), 97 (12), 89 (22), 75 (27), 73 (73), 59 (14). Anal. calc. for $C_{25}H_{38}N_4O_7SSi$ (566.74): C 52.98, H 6.76, N 9.89, O 19.76; found (mixture **32a**): C 53.08, H 6.61, N 9.93; found (mixture **32b**): C 53.08, H 6.93, N 9.76.

*6-Amino-3-[5-*O*-[(*tert*-butyl)dimethylsilyl]-2,3-*O*-isopropylidene)-β-*D*-ribofuranosyl]-5-methylpyrazin-2(1*H*)-one* (**1b**). A soln. of **32** (168.4 mg, 297.14 μmol) in THF (1.5 ml) was treated at 0° dropwise with 1.5M LDA (0.4 ml, 600 μmol) and stirred at r.t. After 3 h, traces of **32** were still evident (TLC monitoring). Thus, the mixture was again cooled to 0°, further treated with LDA (0.1 ml, 150 μmol), and further stirred at r.t. for 7 h

(reaction complete). The deep red soln. was taken up in sat. aq. NH_4Cl soln. (5 ml), the pH adjusted to 5–6 with 2N HCl, and the mixture extracted with CH_2Cl_2 (4 × 5 ml). The combined org. phase was dried (Na_2SO_4) and evaporated, and the residue subjected to FC (silica gel, CH_2Cl_2/MeOH 95:5). The obtained brownish foam (111.3 mg, 91%) was precipitated from Et_2O/hexane: **1b** (99.1 mg, 81%). Nearly colorless flakes. M.p. 134–135°. TLC (CH_2Cl_2/MeOH 9:1): R_f 0.42. UV (EtOH): 372 (9257), 240 (11675), 330 (sh). IR ($CHCl_3$): 3500, 3410, 3330, 2990, 2960, 2930, 3360, 1645, 1615, 1535, 1480, 1435, 1385, 1370, 1260, 1185, 1160, 1135, 1075, 1005, 975, 835. ^{1}H-NMR ($CDCl_3$): 0.04, 0.05 (2*s*, Me_2Si); 0.88 (*s*, ^{t}BuSi); 1.38 (*s*, 3 H, Me_2CO_2('*exo*')); 1.59 (*s*, 3 H, Me_2CO_2('*endo*')); 2.16 (*s*, Me–C(5)); 3.71 (*dd*, *J* = 4.7, 11.0, 1 H, CH_2(5′)); 3.77 (*dd*, *J* = 4.4, 11.0, 1 H, CH_2(5′)); 4.14 (*q*-like *m*, H–C(4′)); 4.70 (*dd*, *J* = 3.6, 6.6, H–C(3′)); 5.11 (*d*, *J* = 4.9, H–C(4′)); 5.17–5.20 (*m*, H–C(2′), NH_2); NOE data were used to assign configuration. ^{13}C-NMR ($CDCl_3$): –5.36 (*q*, Me_2Si); 18.00 (*q*, *Me*–C(5)); 18.45 (*s*, Me_3CSi); 25.61 (*q*, Me_2CO_2); 25.99 (*q*, Me_3CSi); 27.52 (*q*, Me_2CO_2); 63.50 (*t*, C(5′)); 81.98, 83.03, 83.45, 85.12 (4*d*, C(1′), C(2′), C(3′), C(4′)); 114.15 (*s*, Me_2CO_2); 122.05 (*s*, C(5)*); 129.19 (*s*, C(6)*); 146.67 (*s*, C(3)**); 155.33 (*s*, C(2)**). GC/MS (P20005): t_R 11.6; 411, 296, 262, 208, 180, 138 (100), 75. FAB-MS (pos.): 434 (37, $[M+Na]^+$), 412 (100, $[M+H]^+$). Anal. calc. for $C_{19}H_{33}N_3O_5Si$ (411.57): C 55.45, H 8.08, N 10.21, O 19.44; found: C 55.63, H 7.89, N 10.04.

N-*{5-{5*-O-*[(*tert-*butyl)dimethylsilyl]-2,3*-O-*isopropylidene-β*-D-*ribofuranosyl}-1,6-dihydro-3-methyl-6-oxo-pyrazin-2-yl}benzamide* (**33**). Pyrazinone **1b** (3.2 g, 7.78 mmol) was co-evaporated twice with pyridine and then dissolved in pyridine (40 ml). DMAP (*ca.* 10 mg) and benzoyl chloride (3 ml, 3.4 equiv.) were added at 0°, and the mixture was stirred at r.t. for *ca.* 2 h until completion (TLC monitoring). Addition of MeOH quenched excess of reagent and dissolved precipitated pyridinium chloride. This mixture of di- and tribenzoylated compounds was hydrolyzed *in situ* with ammonium hydroxide (*ca.* 8 ml) at r.t. After completion of the reaction, the precipitated benzamide was filtered off and washed thoroughly with Et_2O. The supernatant was evaporated and the residue purified by FC (CH_2Cl_2/MeOH 97.5:2.5): **33** (3.78 g, 94%). Colorless oil. TLC (CH_2Cl_2/MeOH 95:5): R_f 0.39. ^{1}H-NMR (300 MHz, $CDCl_3$): 0.04, 0.06 (2*s*, Me_2Si); 0.87 (*s*, ^{t}BuSi); 1.39 (*s*, 3 H, Me_2CO_2('*exo*')); 1.61 (*s*, 3 H, Me_2CO_2('*endo*')); 2.41 (*s*, Me–C(3′)); 3.82 (*m*, CH_2(5″)); 4.27 (*q*-like *m*, H–C(4″)); 4.79 (*dd*, *J* = 3.4, 6.5, H–C(3″)); 5.07 (*dd*, *J* = 4.4, 6.5, H–C(2″)); 5.22 (*d*, *J* = 4.4, H–C(1″)); 7.50, 7.60, 7.91 (3*m*, Ph); 8.3 (1 H, NH). ^{13}C-NMR (75 MHz, $CDCl_3$): –5.45 (*q*, Me_2Si); 18.44 (*s*, Me_3CSi); 19.29 (*q*, *Me*–C(3′)); 25.63 (*q*, Me_2CO_2); 25.94 (*q*, Me_3CSi); 27.52 (*q*, Me_2CO_2); 63.16 (*t*, C(5″)); 81.84, 84.13, 85.68, 85.84 (4*d*, C(1″), C(2″), C(3″), C(4″)); 114.30 (*s*, Me_2CO_2); 127.51, 128.97 (2*d*, C_o and C_m(Ph)); 131.57, 138.97 (2*s*, C(2′), C(3′)); 132.82 (*d*, C_p(Ph)); 133.11 (*s*, C_{ipso}(Ph)); 140.53, 154.21 (2*s*, C(5′), C(6′)); 166.04 (*s*, Ph*C*O). FAB-MS (pos.): 1069 (2, $[2M+Na]^+$), 1031 (1, $[2M]^+$), 538 (10, $[M+Na]^+$), 516 (100, $[M+H]^+$).

N-*{5-{5*-O-*[(*tert-*butyl)dimethylsilyl]-2,3*-O-*isopropylidene-β*-D-*ribofuranosyl}-3-methyl-6-(prop-2-enyloxy)-pyrazin-2-yl}benzamide* (**34**). Prior to use, **33** (104.6 mg, 202.8 μmol) and PPh_3 (79.2 mg, 302 μmol) were dried under high vacuum for at least 1 h at 50°. The flask was then vented with Ar, and the compounds were dissolved in dioxane (5 ml). Allyl alcohol (21 μl, 308.1 μmol) and DEAD (47 μl, 308.3 μmol) were added, and the mixture was stirred for 30 min. When the reaction was not complete, another 0.2 equiv. of DEAD was added. After completion, excess of DEAD was quenched by addition of a few drops of H_2O, and the mixture was evaporated. Chromatography (hexane/Et_2O 7:3) of the residue yielded **34** (74.3 mg, 66%). Colorless foam. TLC (Et_2O/pentane 1:1): R_f 0.49. ^{1}H-NMR (400 MHz, $CDCl_3$): 0.03 (2*s*, Me_2Si); 0.88 (*s*, ^{t}BuSi); 1.39 (*s*, 3 H, Me_2CO_2('*exo*')); 1.60 (*s*, 3 H, Me_2CO_2('*endo*')); 2.45 (*s*, Me–C(3′)); 3.68, 3.70 (2*s*, CH_2(5″)); 4.20 (*m*, H–C(4″)); 4.78 (*m*, CH_2=CHCH_2O); 4.82 (*dd*, *J* = 3.2, 6.4, H–C(3″)); 5.22–5.25 (*m*, H–C(2″), 1 H of CH_2=CHCH_2O); 5.38 (*dq*, 1 H of CH_2=CHCH_2); 5.37 (*d*, *J* = 3.6, H–C(1″)); 5.98–6.08 (*ddt*(=*m*), CH_2=C*H*CH_2O); 7.51, 7.60, 7.93 (3*m*, Ph). ^{13}C-NMR (100 MHz, $CDCl_3$): –5.30 (*q*, Me_2Si); 18.40 (*s*, Me_3CSi); 20.35 (*q*, *Me*–C(3′)); 25.65 (*q*, Me_2CO_2); 25.96 (*q*, Me_3Si); 27.47 (*q*, Me_2CO_2); 63.77 (*t*, C(5″)); 67.16 (*t*, CH_2=CHCH_2O); 81.24, 83.10, 83.42, 86.20 (4*d*, C(1″), C(2″), C(3″), C(4″)); 113.46 (*s*, Me_2CO_2); 117.94 (*t*, CH_2=CHCH_2O); 127.66, 128.86 (2*d*, C_o and C_m(Ph)); 132.48, 132.64 (2*d*, C_p(Ph), CH_2=*C*HCH_2O); 133.70, 138.61, 138.97, 141.40, 154.87 (5*s*, C_{ipso}(Ph), C(2′), C(3′), C(5′), C(6′)); 165.56 (*s*, Ph*C*O). FAB-MS (pos.): 1111.5 (<2, $[2M]^+$), 556 (89, $[M+H]^+$) 105 (100, $PhCO^+$).

N-*[3-Methyl-6-(prop-2-enyloxy)-5-(β*-D-*ribofuranosyl)pyrazin-2-yl]benzamide* (**35**). A soln. of **34** (751 mg, 1.35 mmol) in 1N HCl/MeOH (40 ml) was stirred overnight at r.t. The mixture was neutralized by addition of solid $NaHCO_3$, the salts were filtered off, and the supernatant was evaporated. FC (CH_2Cl_2/MeOH 39:1) of the residue yielded **35** (760 mg, 94%). Colorless oil. TLC (CH_2Cl_2/MeOH 9:1): R_f 0.36. ^{1}H-NMR (300 MHz, CD_3OD): 2.40 (*s*, Me–C(3′)); 3.74 (*dd*, *J* = 3.0, 12.0, 1 H, CH_2(5″)); 3.98 (*dd*, *J* = 2.9, 12.0, 1 H, CH_2(5″)); 4.09 (*m*, H–C(4″)); 4.26–4.31 (*m*, H–C(2″), H–C(3″)); 4.88 (*m*, >2 H, CH_2=CHCH_2O, CD_3OH); 5.23–5.30 (*m*, 2 H, CH_2=CHCH_2O, H–C(1″)); 5.44 (*dq*, 1 H, CH_2=CHCH_2O); 6.05–6.18 (*ddt*(=*m*), CH_2=C*H*CH_2O); 7.53, 7.62, 7.98 (3*m*, Ph). ^{13}C-NMR (75 MHz, CD_3OD): 19.65 (*q*, *Me*–C(3′)); 63.12

(*t*, C(5″)); 68.43 (*t*, CH_2=CHCH_2O); 71.90; 76.88, 81.57, 85.29 (4*d*, C(1″), C(2″), C(3″), C(4″)); 118.41 (*t*, CH_2=CHCH_2O); 129.06, 129.80 (2*d*, C_o and C_m(Ph)); 133.56, 134.11 (2*d*, C_p(Ph), CH_2=CHCH_2O); 134.95, 141.27, 141.85, 143.53, 156.03 (5*s*, C_{ipso}(Ph), C(2′), C(3′), C(5′), C(6′)); 169.17 (*s*, PhCO). FAB-MS (pos.): 825 (<1, $[2M+Na]^+$), 803 (7, $[2M]^+$), 402 (100, $[M+H]^+$). Anal. calc. for $C_{20}H_{23}N_3O_6$ (401.42): C 59.84, H 5.78, N 10.47; found: C 59.33, H 5.82, N 10.21.

*5-Amino-6-methyl-3-(prop-2-enyloxy)pyrazin-2-yl β-*D*-Ribofuranoside* (**36**). Benzamide **35** (745 mg, 1.86 mmol) in a glass autoclave was dissolved in conc. ammonium hydroxide (25 ml) and stirred overnight at 60–70°. After cooling in an ice bath, the mixture was evaporated. FC (CH_2Cl_2/MeOH 95:5) of the residue yielded **36** (506.6 mg, 91.8%). Colorless oil. TLC (CH_2Cl_2/MeOH 9:1): R_f 0.35. ^{1}H-NMR (300 MHz, CD_3OD): 2.24 (*s*, Me–C(6′)); 3.70 (*dd*, *J* = 2.7, 12.0, 1 H, CH_2(5)); 3.93 (*dd*, *J* = 2.9, 12.0, 1 H, CH_2(5)); 4.00 (*m*, H–C(4)); 4.17 (*dd*, *J* = 3.8, 4.9, H–C(2)); 4.28 (*dd*, *J* = 5.0, 6.1, H–C(3)); 4.80 (*m*, CH_2=CHCH_2O); 5.13 (*d*, *J* = 3.8, H–C(1)); 5.21 (*dq*, *J* = 10.5, 1 H, CH_2=CHCH_2O); 5.74 (*dq*, *J* = 17.3, 1 H, CH_2=CHCH_2O); 6.07 (*ddt*, *J* = 5.3, 10.5, 17.3, CH_2=C*H*CH$_2$O). ^{13}C-NMR (75 MHz, CD_3OD): 18.52 (*q*, *Me*–C(6′)); 63.35 (*t*, C(5)); 67.46 (*t*, CH_2=CHCH_2O); 72.09, 77.01, 81.09, 85.28 (4*d*, C(1), C(2), C(3), C(4)); 117.67 (*t*, CH_2=CHCH_2O); 128.12, 129.65, 153.47, 156.44 (4*s*, C(2′), C(3′), C(5′), C(6′)); 134.89 (*d*, CH_2=*C*HCH$_2$O).

*5-{[(Dimethylamino)methylene]amino}-6-methyl-3-(prop-2-enyloxy)pyrazin-2-yl β-*D*-Ribofuranoside* (**37**). Dimethylformamide diethyl acetal (1.7 ml, 9.9 mmol) was added to a soln. of **36** (283 mg, 952 μmol) in DMF (7 ml), and the mixture was stirred at r.t. for 24 h. The solvent was evaporated and the residue purified by FC (CH_2Cl_2/MeOH 9:1): **37** (315 mg, 94.2%). TLC (CH_2Cl_2/MeOH 9:1): R_f 0.38. ^{1}H-NMR (400 MHz, CD_3OD): 2.38 (*s*, Me–C(6′)); 3.10, 3.15 (2*s*, Me_2N); 3.72 (*dd*, *J* = 2.4, 12.0, 1 H, CH_2(5)); 3.97 (*dd*, *J* = 2.7, 12.0, 1 H, CH_2(5)); 4.04 (*m*, H–C(4)); 4.18 (*dd*, *J* = 3.4, 4.8, H–C(2)); 4.30 (*dd*, *J* = 4.9, 6.4, H–C(3)); 4.85 (*m*, >2 H, CH_2=CHCH_2O, CD_3OH); 5.19 (*d*, *J* = 3.4, H–C(1)); 5.23 (*dq*, *J* = 10.5, 1 H, CH_2=CHCH_2O); 5.41 (*dq*, *J* = 17.3, CH_2=CHCH_2O); 6.10 (*ddt*, *J* = 5.3, 10.5, 17.3, CH_2=C*H*CH$_2$O); 8.46 (*s*, NCHN). ^{13}C-NMR (100 MHz, CD_3OD): 18.83 (*q*, *Me*–C(6)); 34.98, 41.10 (2*q*, Me_2N); 63.10 (*t*, C(5)); 67.55 (*t*, CH_2=CHCH_2O); 71.75, 77.21, 81.22, 85.13 (4*d*, C(1), C(2), C(3), C(4)); 117.67 (*t*, CH_2=CHCH_2O); 134.00, 138.81, 154.51, 155.64 (4*s*, C(2′), C(3′), C(5′), C(6′)); 134.90 (*d*, CH_2=*C*HCH$_2$O); 157.46 (*d*, NCHN). FAB-MS (pos.): 705 (7, $[2M+H]^+$), 353 (100, $[M+H]^+$). Anal. calc. for $C_{16}H_{24}N_4O_5$ (352.39): C 54.53, H 6.86, N 15.90; found: C 54.71, H 6.76, N 15.84.

*5-{[(Dimethylamino)methylene]amino}-6-methyl-3-(prop-2-enyloxy)pyrazin-2-yl 3,5-*O*-(1,1,3,3-Tetraisopropyldisiloxane-1,3-diyl)-β-*D*-ribofuranoside* (**38**). Ribofuranoside **37** (118 mg, 294 μmol) was twice co-evaporated with pyridine (2 ml each) and then dissolved in pyridine (3 ml). TIPS-Cl (=1,3-dichloro-1,1,3,3-tetraisopropyldisiloxane; 107 μl, 341 μmol) was added at 0°, and the mixture was slowly warmed to r.t. and stirred overnight. Excess of reagent was quenched by adding a few drops of MeOH. The soln. was then evaporated and the residue subjected to FC (Et_2O/hexane 1:1): **38** (157 mg, 83%). Colorless oil. TLC (CH_2Cl_2/MeOH 95:5): R_f 0.72. ^{1}H-NMR (400 MHz, $CDCl_3$): 0.96–1.18 (*m*, ^{i}PrSi); 2.40 (*s*, Me–C(6′)); 3.00 (*d*, *J* = 2.3, OH–C(2)); 3.11 (*s*, Me_2N); 3.97–4.03 (*m*, CH_2(5), H–C(4)); 4.44–4.47 (*m*, H–C(2)); 4.75–4.86 (*m*, CH_2=CHCH_2O, H–C(3)); 5.21 (*dq*, *J* = 10.4, 1 H, CH_2=CHCH_2O); 5.28 (*d*, *J* = 2.3, H–C(1)); 5.36 (*dq*, *J* = 17.2, 1 H, CH_2=CHCH_2O); 6.06 (*ddt*, *J* = 5.3, 10.4, 17.2, CH_2=C*H*CH$_2$O); 8.36 (*s*, NCHN). FAB-MS (pos.): 1188 (1, $[2M]^+$) 595 (100, $[M+H]^+$).

*5-{[(Dimethylamino)methylene]amino}-6-methyl-3-(prop-2-enyloxy)pyrazin-2-yl 2-*O*-Methyl-3,5-*O*-(1,1,3,3-tetraisopropyldisiloxane-1,3-diyl)-β-*D*-ribofuranoside* (**39**). Methyl iodide (2 ml) and Ag_2O (175 mg, 5.1 equiv.) were added at 0° to a soln. of **38** (175 mg, 294 μmol) in MeCN (4 ml). The flask was closed tightly and the mixture stirred at r.t. overnight in the dark. After addition of MeOH (2 ml), the mixture was filtered through a small bed of silica gel and evaporated. The residue was subjected to FC (hexane/AcOEt 8:2): **39** (107 mg, 59%). Slightly yellow oil. TLC (hexane/AcOEt 8:2): R_f 0.48. ^{1}H-NMR (400 MHz, $CDCl_3$): 1.00–1.17 (*m*, ^{i}PrSi); 2.38 (*s*, Me–C(6′)); 3.11 (*s*, Me_2N); 3.54 (*s*, MeO–C(2)); 3.95–4.08 (*m*, CH_2(5), H–C(4), H–C(2)); 4.74–4.88 (*m*, CH_2=CHCH_2O, H–C(3)); 5.20 (*dq*, *J* = 10.4, 1 H, CH_2=CHCH_2O); 5.36 (*dq*, *J* = 17.2, 1 H, CH_2=CHCH_2O); 6.06 (*ddt*, *J* = 5.3, 10.4, 17.2, CH_2=C*H*CH$_2$O); 8.35 (*s*, NCHN). ^{13}C-NMR (100 MHz, $CDCl_3$): 12.64, 12.74, 13.13, 13.45 (4*d*, 4 Me_2CHSi); 17.09–19.22 (9*q*, 4 *Me*$_2$CHSi, *Me*–C(6)); 34.63, 40.76 (2*q*, Me_2N); 58.94 (*q*, *Me*O–C(2)); 61.23 (*t*, C(5)); 66.33 (*t*, CH_2=CHCH_2O); 73.01, 79.49, 80.49, 84.11 (4*d*, C(1), C(2), C(3), C(4)); 117.16 (*t*, CH_2=CHCH_2O); 133.06, 138.87, 152.36, 154.28 (4*s*, C(2′), C(3′), C(5′), C(6′)); 133.91 (*d*, CH_2=*C*HCH$_2$O); 155.32 (*d*, NCHN). FAB-MS (pos.): 1216 (1, $[2M]^+$), 609 (100, $[M+H]^+$).

*5-{[(Dimethylamino)methylene]amino}-6-methyl-3-(prop-2-enyloxy)pyrazin-2-yl 2-*O*-Methyl-β-*D*-ribofuranoside* (**40**). A soln. of pyridinium fluoride in pyridine (*ca.* 5.4M) was slowly added to a soln. of **39** (251.5 mg, 413 μmol) in pyridine (1.5 ml) in a plastic vial. After the reaction was complete (*ca.* 12 h), it was

quenched with methoxytrimethylsilane (0.5 ml, 1.2 equiv. with respect to F^-) at 0°. The mixture was stirred at r.t. for 15 min, transferred to a glass flask, and evaporated. The residue was subjected to FC (CH_2Cl_2/MeOH 95:5). The product was easily crystallized from CH_2Cl_2/pentane: **40** (141.5 mg, 93.5%). M.p. 190°. TLC (CH_2Cl_2/MeOH 95:5): R_f 0.27. ^{1}H-NMR (400 MHz, $CDCl_3$): 2.44 (*s*, Me–C(6)); 2.72 (*d*, *J* = 8.1, OH); 3.11, 3.13 (2*s*, Me_2N); 3.52 (*s*, MeO–C(2)); 3.79 (*t*, 1 H, CH_2(5)); 3.87 (*dd*, *J* = 2.4, 4.8, H–C(2)); 4.08–4.20 (*m*, 2 H, CH_2(5), H–C(4)); 4.50–4.55 (*m*, H–C(3)); 4.79–4.90 (2*ddt*, CH_2=CHCH_2O); 5.25 (*dq*, *J* = 10.4, CH_2–CHCH_2O); 5.37 (*m*, 2 H, H–C(1), CH_2=CHCH_2O); 6.05 (*ddt*, *J* = 5.3, 10.4, 17.3, CH_2=C*H*CH_2O); 6.23 (*d*, *J* = 10.5, OH); 8.38 (*s*, NCHN). ^{13}C-NMR (100 MHz, $CDCl_3$): 18.71 (*q*, *Me*–C(6)); 34.70, 40.82 (2*q*, Me_2N); 57.80 (*q*, *Me*O–C(2)); 62.13 (*t*, C(5)); 66.54 (*t*, CH_2=CH*C*H_2O); 70.00, 76.43, 84.99, 85.86 (4*d*, C(1), C(2), C(3), C(4)); 117.94 (*t*, *C*H_2=CHCH_2O); 132.99, 138.64, 152.89, 153.34 (4*s*, C(2′), C(3′), C(5′), C(6′)); 133.29 (*d*, CH_2=*C*HCH_2O); 155.26 (*d*, NCHN). FAB-MS (pos.): 367 (100, $[M+H]^+$). Anal. calc. for $C_{17}H_{26}N_4O_5$ (366.42): C 55.73, H 7.15, N 15.29; found: C 56.03, H 6.97, N 15.11.

*5-{[(Dimethylamino)methylene]amino}-6-methyl-3-(prop-2-enyloxy)pyrazin-2-yl 5-*O*-(4,4′-dimethoxytrityl)-2-*O*-methyl-β-*D*-ribofuranoside* (**41**). Ribofuranoside **40** (135.7 g, 370.3 μmol) was co-evaporated twice with pyridine (2–3 ml each) and then dissolved in pyridine (2.5 ml). A soln. of 4,4′-dimethoxytrityl chloride (150 mg, 1.2 equiv.) in pyridine (1 ml) was added, and the mixture was stirred at r.t. overnight. After quenching with MeOH, the solvent was evaporated and the residue purified by FC (hexane/AcOEt 1:1): **41** (182 mg, 73%). Colorless foam. TLC (hexane/AcOEt 1:1): R_f 0.45. ^{1}H-NMR (200 MHz, $CDCl_3$): 2.35 (*s*, Me–C(6)); 2.71 (*d*, *J* = 5.3, OH); 3.12 (*s*, Me_2N); 3.26 (*dd*, *J* = 4.8, 9.9, 1 H, CH_2(5)); 3.35 (*dd*, *J* = 4.3, 9.8, 1 H, CH_2(5)); 3.42 (*s*, MeO–C(2)); 3.77 (*s*, (*Me*O)$_2$Tr); 4.12 (*m*, H–C(4)); 4.36 (*m*, H–C(3), H–C(2)); 4.83 (*dq*, CH_2=CHCH_2O); 5.20 (*dq*, *J* = 10.5, 1 H, CH_2=CHCH_2O); 5.31 (*d*, *J* = 4.3, H–C(1)); 5.35 (*dq*, *J* = 17.3, 1 H, CH_2=CHCH_2O); 6.04 (*ddt*, *J* = 5.3, 10.5, 17.3, CH_2=C*H*CH_2O); 6.75–7.51 (*m*, 13 H, (MeO)$_2$*Tr*); 8.39 (*s*, NCHN).

*5-{[(Dimethylamino)methylene]amino}-6-methyl-3-(prop-2-enyloxy)pyrazin-2-yl 5-*O*-(4,4′-dimethoxytrityl)-2-*O*-methyl-β-*D*-ribofuranoside 3-[2-Cyanoethyl Diisopropylphosphoramidite]* (**42**). Ribofuranoside **41** (130 mg) was co-evaporated with pyridine (3 × with 1 ml) and subsequently dried overnight in a desiccator over P_2O_5 under high vacuum. The desiccator was vented with Ar and the residue dissolved in MeCN (1 ml). DMAP (*ca.* 0.1 equiv.) and *Hünig*'s base (0.15 ml, 4.5 equiv.) were added, followed by 2-cyanoethyl diisopropylphosphoramidochloridite (62 μl, 1.4 equiv.) at 0°, and the reaction was stirred at r.t. After completion, the mixture was diluted with AcOEt (10 ml), the soln. washed with 2M aq. phosphate buffer (pH 7.0; 10 ml) and brine (5 ml), dried (Na_2SO_4), and evaporated, and the residue subjected to FC (hexane/AcOEt/Et_3N 60:40:2): **42** (144 mg, 85%). Slightly yellow oil. TLC (hexane/AcOEt 1:1): R_f 0.28. ^{1}H-NMR (500 MHz, $CDCl_3$; two diastereoisomers): 1.01–1.19 (*m*, 2 *Me*$_2$CH); 2.31–2.39 (2*s* + *m*, Me–C(6), $CNCH_2$); 2.67–2.70 (*m*, 1 H, $CNCH_2$); 3.11 (*s*, Me_2N); 3.25–3.43, 2.52–3.66, 3.89–3.99 (3*m*, $POCH_2$, 2 Me_2CH, CH_2(5)); 3.38, 3.39 (2*s*, MeO–C(2′)); 3.76, 3.77 (3*s*, (*Me*O)$_2$Tr); 4.22–4.28 (*m*, H–C(4)); 4.40–4.67 (3*m*, H–C(2), H–C(3)); 4.81–4.85 (*m*, CH_2=CHCH_2O); 5.19–5.22 (*dm*, 1 H, CH_2=CHCH_2O); 5.34–5.40 (*m*, 2 H, H–C(1), CH_2=CHCH_2); 6.03–6.10 (*m*, CH_2=C*H*CH_2O); 6.74–6.78, 6.97–7.49 (*m*, 13 H, (MeO)$_2$*Tr*), 8.37, 8.38 (2*s*, NCHN). ^{31}P-NMR (200 MHz, $CDCl_3$): 149.6, 150.18. FAB-MS (pos.): 1737 (<1, $2M^+$), 869 (14, $[M+H]^+$).

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Received January 14, 2004