Synthesis of 'Iso or Reversed' *C*-nucleosides Analogues using Push-pull Activated Monosaccharide Derivatives as Precursors Starting from D-furanose

[1,2]IMRAN ALI HASHMI*, [1]FIRDOUS IMRAN ALI, [2]HOLGER FIEST, [2]MANFERED MICHALIK AND [2]KLAUS PESEKE

[1]*Department of Chemistry, University of Karachi, Karachi-75270, Pakistan.*
[2]*Universitat Rostock, Institut für Chemie, Albert-Einstein-Str. 3a, 18059 Rostock, Germany.*

(Received 29th March 2007, revised 6th August 2007)

Summary: Iso or Reversed-*C*-nucleosides constitute a class of nucleoside analogues in which the nucleobase is linked by a carbon-carbon bond to a ribose carbon other than C-1. It is anticipated that iso-*C*-nucleosides may have increased chemical and enzymatic stability under physiological conditions. In this paper we describe the synthesis of ethyl 6- (R,S)-7- (E,Z)-3-*O*-Benzyl-6,8-dicyano-6-deoxy-1,2-*O*-isopropylidene-7-methylsulfanyl-α-D-*xylo*-non-7-en-5-ulofuranuronate (11) as sodium salt and 3-*O*-Benzyl-1,2-*O*-isopropylidene-5-methylsulfanyl-α-D-*xylo*-pentofuran- os-5-ulose (15) from 3-*O*-Benzyl-6-deoxy-1,2-*O*-isopropylidene-α-D-*xylo*-hept-5-ulofuranurononitrile (6) synthesis of 6-Aminomethylene-3-*O*-Benzyl-6-deoxy-1,2-*O*-isopropylidene-α-D-*xylo*-hept-5-ulofuranurononitrile (21) and 7-Amino-ethyl-6- (3-desoxy-1,2-*O*-isopropylidene-α-D-*glycero*-pent-3-enofuran- uronoyl)-2-methylsulfanyl-pyrazolo[1,5-*a*]pyrimidine-3-carboxylate (23) from (*E*)-3-*O*-Benzyl-6-deoxy-6-dimethylaminomethylene-1,2-*O*-isopropylidene-α-D-*xylo*-hept-5-ulofuranurononitrile (7) and synthesis of 3-Amino-4- (3-*O*-Benzyl-1,2-*O*-isopropylidene-α-D-*xylo*pentofuranuronoyl)-5-methylsulfanyl-1-*H*-pyrazole-1-carbamidine (22) from 3-O-Benzyl-6-deoxy-1,2-O-isopropylidene-6-[bis- (methylthio) ethylene]-α-D-xylo-hept-5-ulofuranonitrile (9).

Introduction

Nucleosides are the ribosyl or deoxy ribosyl derivatives of certain pyrimidine and purine bases. They are thus glycosylamines or *N*-glycosides related to nucleotides by the lack of phosphorylation.

C-nucleosides constitute a category of analogues in which the modification occurred at the linkage between the base and the sugar. In *C*-nucleosides, the sugar and the base are linked through a C-C linkage as opposed to *N*-nucleosides where a C-N bond is present. This structural alteration is believed to increase the nucleoside stability towards nucleoside hydrolase enzymes and to inhibit in general the cascade of events leading to DNA or RNA formation [1]. Many naturally occurring *C*-nucleosides have been reported as strong antibiotic agents, which has primed research on *C*-nucleoside analogues. However, despite the large amount of data collected, *C*-nucleosides and especially *C*-nucleosides belonging to the 2-deoxy–D-ribose series have not been explored in an adequate extend [2,3].

Reversed *C*-nucleoside are known with a strong and selective anti HIV and anti HSV activity. Iso ddA is a good representative of this class. Its anti HIV activity is similar to that of ddA and it has no apparent toxicity [4]. The synthesis of iso-*C*-nucleosides [5, 6] has been reported in the literature.

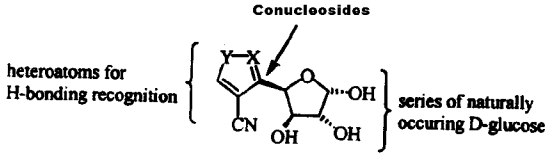
Our research interests are mainly focused on the development of new synthetic methodologies with the aim to generate families of potentially useful drug candidates [7]. As part of this research we set out to develop a synthetic route for a family of 'reverse'-*C*-nucleoside analogues of the D-furanose series. We choose the following key features to define this new nucleoside family: (i) *C*-linkage between the sugar and the heterocyclic moiety through C-5 of the furanose; (ii) the presence of multiple heteroatoms in the aglycon, as potential positions for H-bonding recognition; (iii) the sugar moiety have to derive from the naturally occurring D-glucose (Scheme 1).

Result and Discussion

Synthesis of 'Iso or Reversed'-C-nucleoside Analogues on the Basis of Push-pull Functionalized Monosaccharides

For many years the chemistry of Push-pull compounds is one of the main lines of research in our

*To whom all correspondence should be addressed.



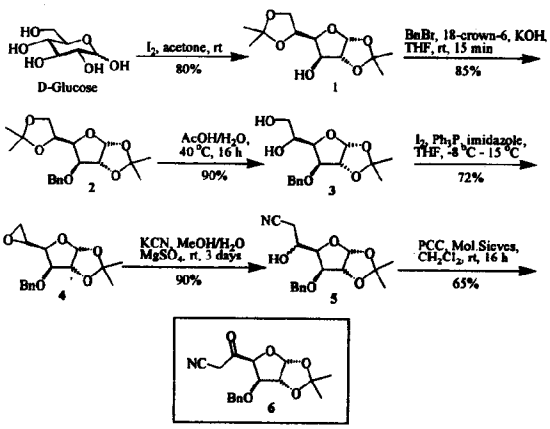
Scheme-1: Target family of 'reversed' *C*-nucleosides.

working group. Push-pull alkenes are compounds having unsymmetrical polarized double bonds. At one end of the alkene system are loaded electron withdrawing substituents and at the other end electron donating groups.

In view of above facts four Push-pull functionalized monosaccharides namely; 3-O-Benzyl-6-deoxy-1,2-O-isopropylidene-α-D-xylo-hept-5-ulofuranurononitrile (6), (*E*)-3-*O*-Benzyl-6-deoxy-6-dimethylaminomethylene-1,2-*O*-isopropylidene-α-D-*xylo*-hept-5-ulofuranurononitrile (7), (*Z*)-3-*O*-Benzyl-6-deoxy-6-dimethylaminomethylene-1,2-*O*-isopropylidene-α-D-*xylo*-hept-5-ulofuranurononitrile (8) and 3-O-Benzyl-6-deoxy-1,2-O-isopropylidene-6-[bis- (methyllthio) ethylene]-α-D-xylo-hept-5-ulofurano-nitrile (9) were prepared to be used as precursors for synthesis of reversed C-nucleoside analogoues. [8].

Synthesis of 3-O-Benzyl-(6)-deoxy-1,2-O-isopropylidene-α-D-xylo-hept-5-ulofuranurononitrile (6)

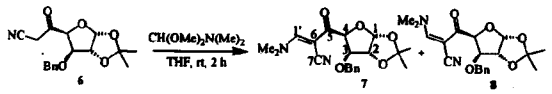
The 3-*O*-benzylated ulose 6 was synthesized starting from D-glucose. Because a protecting group regime was needed, attention was directed towards such groups, which could be cleaved under mild conditions [8] (Scheme 2).



Scheme-2: Synthesis of 3-O-Benzyl-6-deoxy-1,2-O-isopropylidene-a-D-xylohept-5-ulofuranurononitrile (6).

Synthesis of 3-O-Benzyl-6-deoxy-6-dimethylaminome-thylene-1,2-O-isopropylid-ene-α-D-xylo-hept-5-ulo-furanurononitrile (7) and (8)

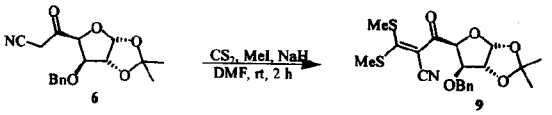
In recent years Push-pull functionalised monosaccharides were prepared in our working group [9, 10]. Bredereck and co-workers reported the reaction of acidic methylene compounds with acetals of amides [11-13]. Similarly, compound 6 was reacted with *N,N*-dimethylformamide dimethylacetal in THF to furnish the crystalline branched chain (*E*)-ulose 7 and the noncrystalline (*Z*)-ulose 8 (Scheme 3).



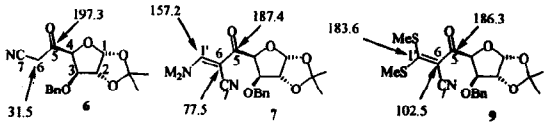
Scheme-3: Synthesis of a-dimethylaminomethylene ulose 7 and 8.

Synthesis of 3-O-Benzyl-6-deoxy-1,2-O-isopropylidene-6-[bis- (methylthio) ethylene]-α-D-xylohept-5-ulofuranonitrile (9)

As it is well known, The α-oxo ketene dithioacetals can be prepared by the reaction of cyclic or acyclic ketones with carbon disulphide in the presence of a base and an alkylating agent [10, 14-16]. In order to synthesize first type of furanosyl substituted Push-pull activated ketene-*S,S*-acetal, compound 6 was reacted with carbon disulphide and methyl iodide in the presence of sodium hydride in dimethylformamide to give 9 as a yellow syrup (Schemes 4 and 5).



Scheme-4: Synthesis of a-oxo ketene dithioacetal 9.

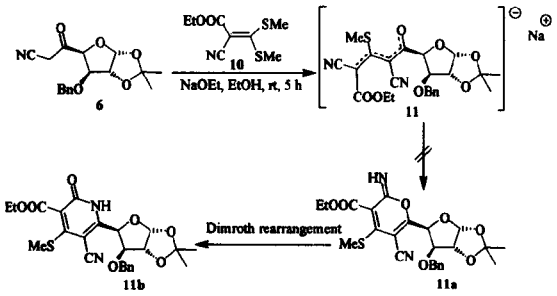


Scheme-5: ^{13}C NMR values of compounds 6, 7 and 9.

Synthesis of Push-pull Activated Chain Elongated Monosaccharide Derivatives

To obtain monosaccharides with a longer carbon chain, we carried out the reaction of 2-cyano-

3,3-bis (methylthio)acrylic acid ethylester (10) with urononitrile (6) in the presence of NaOEt. After a reaction time of 5 h the sodium salt 11 could be isolated in very good yield (Scheme 6).



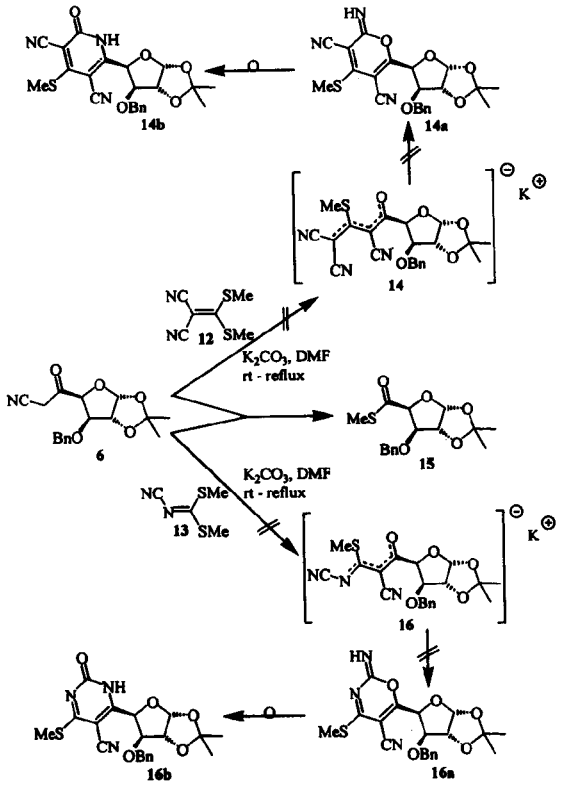
Scheme-6: Synthesis of *C*-branched monosaccharide.

The structure of compound 11 was confirmed by NMR and mass spectrometry. In FAB-positive mass spectrometry molecular ion peak $[M+Na]^+$ appeared at *m/ z* = 509 which proved the presence of compound 11 in salt form.

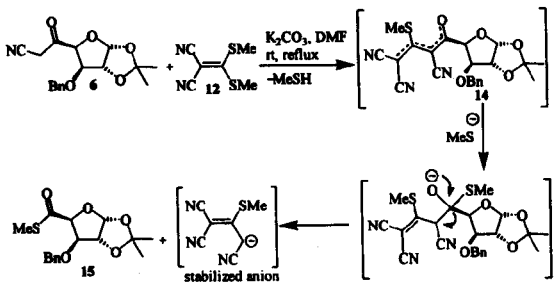
The strong electron delocalisation in (11) also affected ^{13}C NMR data. So the carbonyl carbon resonances were shifted upfield to 180.9 ppm compared with the corresponding values for compound 7 (187.4) and compound 9 (186.). The S-methyl signal was detected at 18.1 ppm and the ester carbonyl atom gave resonance at 163.8 ppm. A cyclization of the sodium salt (11) could yield different 'reversed' *C*-nucleosides for example 11b. Such reactions did not take place due to salt formation.

But on the other hand, when compound 6 was treated with the Push-pull alkenes (12) and azaalkenes (13) respectively, in the presence of NaOEt/ EtOH, we did not succeed to isolate any compound. After that we carried out these reactions in the presence of K_2CO_3/ DMF, neither got the open chain derivatives 14 and 16 nor the cyclized forms 14a or 14b and 16a or 16b were observed. Instead in both cases the respected crystalline compound 15 was formed in good yield (Schemes 7 and 8).

In ^{13}C NMR spectrum the carbonyl carbon resonance appeared at 198.8 ppm and the resonance of the S-methyl group was detected at 10.8 ppm.

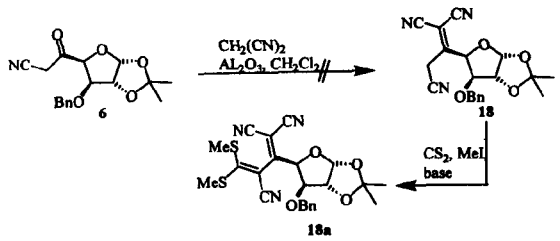


Scheme-7: Unexpected formation of compound 15.



Scheme-8: Possible mechanism for the formation of compound 15.

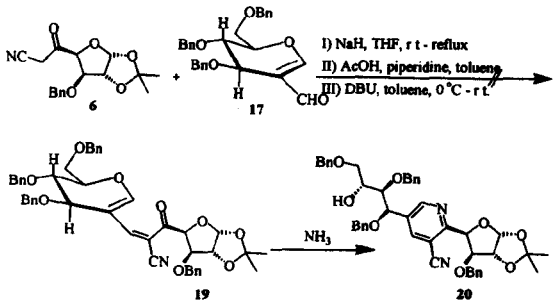
In an effort to synthesize Knoevenagel compound 18, ulose 6 was treated with malononitrile and basic aluminium oxide [17] as a catalyst in dichloromethane at room temperature (Scheme 9). The desired product 18 would open the way for the synthesis of furanosyl substituted Push-pull butadiene derivative, which in further reactions should be used



Scheme-9: Attempt for the synthesis of 18.

for the preparation of *C*-nucleoside derivatives according to known litrature procedures [18-22]. Unfortunately, the Knoevenagel product 18 could not be produced in this way.

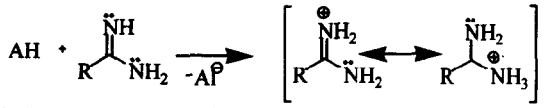
In another attempt we tried to condense the ulose (6) with the 2-formyl-D-glucal (17) to obtain the spacer *C*-disaccharide (19). Different reaction condi-tions were tried (Scheme 10) but, none gave positive results. Corresponding to our strategy disaccharide analogue (19) should be reacted with ammonia to form after a ring transformation reaction developed in our working group [23-25], the unusual pyridine bridged disaccharide (20), having a furanose and an acyclic sugar moiety.



Scheme-10: Attempt for the synthesis of 19 and 20.

Reactions with Amidines

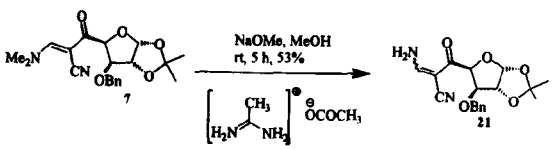
Amidines are much more basic than amides, the pK_a values of amidines are larger than those of amides by about 13. Acetamidine for example has a pK_a value of 12.4. In fact, they are among the strongest neutral bases. An amidine has two nitrogen atoms available for protonation, one is sp^3 hybridized, the other sp^2 hybridized. The sp^3 nitrogen might be expected to be more basic, but protonation occurs at the sp^2 nitrogen atom. This behavior is comparable with an amide in which carbonyl oxygen is protonated first. When protonation occurs at sp^2 hyberdized nitrogen, the positive charge is delocalized over both nitrogen atoms due to presence of lone pairs (Scheme 11) [26].



Scheme-11: Delocalization of positive charge in Amidines.

Amidines represent 1,3-*N*,*N*-dinucleophiles and can be used for the preparation of pyrimidines for example by the reaction with enaminoketones [27-30]. The dimethylaminomethylene ulose with Push-pull functionality should serve for displacement reactions of the dimethylamino group through substituted amidines [31-32]. The higher reactivity of enaminoketones compared with the corresponding α-oxoketene-*S*,*S*-dithioacetals is attributed to the presence of only one donor group in this Push-pull system reflecting the straight forward substitution of the dimethylamino group and ring closure by treatment of dimethylaminomethylene ulose 7 with substituted amidines leading to the formation of pyrimidine derivatives [33].

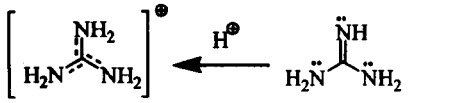
Similarly, ulose (7) was reacted with formamidinium acetate in methanol with sodium methanolate as a base in order to synthesize the corresponding pyrimidine derivative. The spectroscopic data of the isolated compound 21 showed that instead of cyclization, substitution of -N $(Me)_2$ group has taken place by the attack of $-NH_2$ group (Scheme 12). The signals of carbonyl and nitrile function are still present in the IR and NMR spectra and absence of dimethylamino signals in NMR spectrum has confirmed this substitution. The ammonia necessary for this substitution reaction may be formed by an attack of methanol/ methanolate on foramidine.



Scheme-12: Reaction of compound 7 with formamidinium acetate.

Reactions with Guanidine and Substituted Guanidines

Guanidine, pK_a 13.6, is more basic then amidine. On protonation, the positive charge can be delocalised over three nitrogen atoms to give a very stable cation (Scheme 13). All three nitrogen lone pairs cooperate to donate electrons but protonation occurs, again, on the sp^2 nitrogen atom.



guanidine: pK_{aH} 13.6

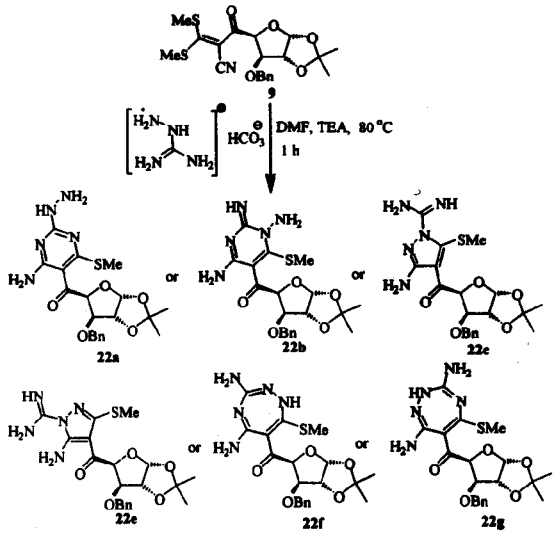
Scheme-13: Guanidine a very strong bases.

Guanidines and substituted guanidines were reacted with enaminoketones to form different pyrimidine derivatives, which represent the class of compounds that exhibits anti-inflammatory and anti-tumor activitives [34-38].

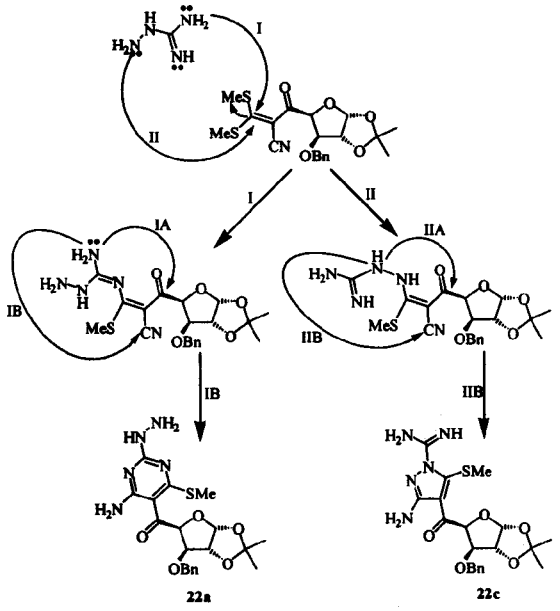
After the successful formation with guanidinium chloride, aminoguanidinium carbonate was utilized for the heterocyclization of α-oxoketene dithioacetal (9) again in DMF in the presence of triethylamine at 80 °C. The reaction was completed after 1 h resulting in the formation of yellowish solid as a pure product.

In EI-MS spectrum the molecular ion peak was observed at *m/ z* 447 corresponding with the substitution of methylthio group by the amino-guanidine. But in the ^{13}C NMR and the IR spectrum no CN signal could be detected. On the other hand, a carbonyl signal appeared at 185.2 ppm in ^{13}C NMR and at 1675 cm^{-1} in IR. That means nitrile function instead of the carbonyl function was involved in this reaction. Furthermore, only one MeS signal was present in ^{13}C NMR (13.1 ppm) and ^{1}H NMR (2.39 ppm) spectrum confirming a cyclization with inclusion of C-1` of the α-oxoketene-dithioacetale 9 and the nitrile group. But, the question which two nitrogen atoms of aminoguanidine were involved in this cyclization is not easy to answer.

Theoretically, six compounds can be formed (Schemes 14 and 15). The amino pyrimidine can act as 1,2-*N,N*, 1,3-*N,N* and 1,4-*N,N* dinucleophile.



Scheme-14: Reaction of compound 9 with amino-guanidine carbonate.



Scheme-15: Postulated mechanism.

Further more, there are two different possibilities to serve as a 1,3-*N,N* dinucleophile. In fact, we were not able to make an exact dicision between the seven formulated structures. Unfortunately the quality of obtained crystals were not good enough for X-ray analysis.

Synthesis of 'Reversed' Pyrazolo[1,5-a]pyrimidine-C-nucleosides

Pyrazolo [1,5-*a*]pyrimidine-*C*-nucleosides shows activity against various cancer cell lines and are also used as probes in biological examinations [39]. Because of their structural analogy with naturally occurring *N*-nucleosides, representative of this class of compounds serve as enzyme substrates or inhibitors [40-45]. There exists only few reports about the synthesis of pyrazolo[1,5-*a*]pyrimidine-*C*-nucleosides. In almost all the cases the sugar part is linked with C-3 or C-7 of the heterocyclic moiety. Usually, the syntheses of these compounds require drastic conditions, long reaction times and/ or complex pathways [46-49]. New reactions that build up C-C bonds and at the same time introduce nitrogen-containing functionalities into the structural framework are especially attractive for the rapid construction of interesting organic molecules with potential biological activity.

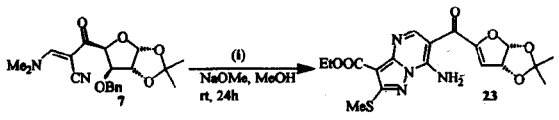
Maeba *et al.,* synthesized pyrazolo[1,5-*a*]pyrimidine-*C*-nucleosides by cyclization of aminopyrazoles with chain extended monosaccharides having an enaminon function in the prolongated chain [50] in acidic medium. In another report given by Robin [51] analogues of pyrazolo-pyrimidine *N*-nucleoside was synthesized.

We chose an entirely different route for the building up of such compounds by utilizing 3 substituted 5-amino-pyrazole-4-carboxylates as 1,3-*N,N* dinucleophiles to furnish novel 'reversed' pyrazolo[1,5-*a*]pyrimidine-*C*-nucleosides. These 5-amino-pyrazoles exhibit a high reactivity towards Push-pull ethene systems especially, the substituted 3,5-diaminopyrazole-4-carboxylates, reported by Gompper *et al.,* [52-53].

Reaction of α-dimethylaminomethylene ulose 1 with 5-Aminopyrazoles

To synthesize 'reversed' furanosyl nucleosides with a more complex heterocyclic unit in the following examinations 5-aminopyrazoles were used as dinucleophiles in the cyclization reaction with the enaminone 7.

When the same reaction was performed in methanol with sodium methanolate as base at room temperature for 24 hours only compound 23 was formed in 62 % yield (Scheme 16).



Scheme-16: Synthesis of pyrozolo[1,5-a]pyrimidine derivative 23.

The spectroscopic data of the heterocyclic part of product 23 are in agreement with those of compound? The absence of the benzyl group and ^{13}C NMR chemical shift of C-3` and C-4` unequivocal displayed elimination of benzyl alcohol. Mass spectrometric values and elemental analysis were in conformity with structure 23.

Experimental

General Procedures

Melting points were determined with a Boĕtius melting point apparatus and are corrected. Optical rotations were measured with a Gyromat-HP (Dr. Kernchen Ltd.) polarimeter. IR spectra were recorded with a Nicolet 205 FT-IR spectrometer. ^1H and ^{13}C NMR spectra were recorded with Bruker spectrometers AC 250 (250.1 MHz and 62.9 MHz, respectively) and ARX 300 (300.1 MHz and 75.5 MHz, respectively), with $CDCl_3$ (compounds **3c** and **7** with DMSO-d_6) as solvent. The calibration of spectra was carried out on solvent signals ($CDCl_3$ δ (^1H) = 7.25; δ (^{13}C) = 77.0; DMSO-d_6: δ (^1H) = 2.50; δ (^{13}C) = 39.7). Mass spectra were obtained with an AMD 402/ 3 spectrometer (AMD Intectra GmbH). Elemental analyses were performed with a Leco CHNS-932. Column chromatography was carried out on silica gel 60 (63 – 200 μm, Merck). Thin-layer chromatography (TLC) was performed on silica gel 60 GF_{254} foils (Merck) with detection by UV-light and by charring with sulphuric acid. Solvents and liquid reagents were purified and dried according to recommended procedures.

Sodium salt of ethyl 6- (R,S)-7- (E,Z)-3-O-Benzyl-6,8-dicyano-6-deoxy-1,2-O-isopropylidene-7-methylsulfanyl-α-D-xylo-non-7-en-5-ulofuranuronate (11)

3-*O*-Benzyl-6-deoxy-1,2-*O*-isopropylidene-α-D-*xylo*-hept-5-ulofuranurononitrile (6) (100 mg,

0.32 mmol) and 2-cyano-3,3-bis (methylthio)acrylic acid ethylester 85.5 mg (0.394 mmol) were dissolved in 5ml anhydrous EtOH. NaOEt (9.12 mg, 0.394 mmol) was added and the mixture was stirred at room temperature for 5 h. After completion of the reaction (monitored by TLC) the solvent was evaporated in vacuum and the residue purified by column chromatography (100 % ethyl acetate) to obtain (11) as a yellow solid. Yield: 127 mg (91 % , yellow solid); R_f: 0.49 (ethyl acetate); $[\alpha]_D^{24}$: + 342.7 (c = 1.0, $CHCl_3$); ^1H NMR (250.133 MHz, $CDCl_3$), δ (ppm): 1.10, 1.55 (2s, 6H, C (CH_3)$_2$); 1.29 (t, 3H, CH_3 (COOEt)); 2.40 (s, 3H, SCH_3); 3.94 (m, 1H, CH_2 (COOEt)); 4.35 (m, 1H, CH_2 (COOEt)); 4.57 (d, 1H, $J_{3,4}$ = 3.6 Hz, H-3); 4.51 (m, 1H, CH*H*Ph); 4.48 (m, 1H, H-2); 4.46 (m, 1H,C*H*HPh); 5.53 (d, 1H, $J_{3,4}$ = 3.6 Hz, H-4); 5.97 (d, 1H, $J_{1,2}$ = 3.9 Hz, H-1); 7.28-7.33 (m, 5H, Ph); ^{13}C NMR (62.89 MHz, $CDCl_3$), δ (ppm): 14.6 (SCH_3); 18.1 (CH_3 (COOEt)); 26.7, 25.2 (C (CH_3)$_2$); 62.0 (CH_2 (COOEt)); 73.5 (CH_2Ph); 78.9 (C-8); 80.7 (C-4); 82.6 (C-2); 85.2 (C-3); 95.1 (C-6); 104.5 (C-1); 112.9 (*C* (CH_3)$_2$); 120.3 (CN); 121.2 (CN); 129.1, 129.3, 130.4 (Ph); 134.6 (*i*-Ph); 163.8 (*CO* (COOEt));178.4 (C-7); 180.9 (CO); MS (FAB pos.), *m/ z* (%): 509 $[M+Na]^+$

3-O-Benzyl-1,2-O-isopropylidene-5-methylsulfanyl-α-D-xylo-pentofuran- os-5-ulose (15)

3-*O*-Benzyl-6-deoxy-1,2-*O*-isopropylidene-α-D-*xylo*-hept-5-ulofuranurononitrile (6) (100 mg, 0.32 mmol) and (bis-methylsulfanylmethylene) malononitrile 81 mg (0.48 mmol) were dissolved in 5ml anhydrous EtOH. NaOEt (9.12 mg, 0.48 mmol) was added and the mixture was stirred at room temperature for 24 h. After completion of the reaction (monitored by TLC) the solvent was evaporated in vacuum and the residue purified by column chromatography (toluene/ ethyl acetate 8:2) to obtain (15) as colourless crystals. Yield: 58 mg (57 % , colourless crystals); M.P: 88 °C; R_f: 0.56 (toluene/ ethyl acetate 9:1); $[\alpha]_D^{24}$: – 127.1 (c = 1.0, $CHCl_3$); ^1H NMR (250.133 MHz, $CDCl_3$), δ (ppm): 1.17, 1.30 (2s, 6H, C (CH_3)$_2$; 2.13 (s, 3H, CH_3S); 4.19 (d, 1H, $J_{3,4}$ = 3.6 Hz, H-3); 4.42 (d, 1H, $J_{CH\ (a),CH\ (b)}$ = 11.9 Hz, CH*H*Ph); 4.56 (d, 1H, $J_{CH\ (a),CH\ (b)}$ = 11.8 Hz, C*H*HPh); 4.64 (d, 1H, $J_{1,2}$ = 3.3 Hz, H-2); 4.71 (d, 1H, $J_{3,4}$ = 3.6 Hz, H-4); 5.92 (d, 1H, $J_{1,2}$ = 3.3 Hz, H-1); 7.20-7.13 (m, 5H, Ph); ^{13}C NMR (62.89 MHz, $CDCl_3$), δ (ppm): 10.8 (CH_3S); 26.3, 27.0 (C (CH_3)$_2$); 72.8 (CH_2Ph); 82.3 (C-4); 82.8 (C-2); 85.5 (C-3); 105.9 (C-1); 112.6 (*C* (CH_3)$_2$); 127.4, 127.8, 128.3 (Ph); 136.9 (*i*-Ph); 198.8 (CO); IR (capillary), ν (cm^{-1}): 1680 (C=O); MS (EI), *m/ z* (%): 325 $[M+1]^+$ $C_{17}H_{19}NO_5$ (324.389) Calcd: C= 59.24, H= 6.21, S=9.88, Found: C=58.80, H= 5.98, S= 9.64

6-Aminomethylene-3-O-Benzyl-6-deoxy-1,2-O-isopropylidene-α-D-xylo-hept-5-ulofuranurononitrile (21)

Sodium (17 mg, 0.74 mmol) was added to methanol (1 ml). After 5 min foramidinium acetate (77 mg, 0.74 mmol) was added and the solution stirred for 20 min. This mixture was then added to another stirred solution of (7) (141 mg, 0.37 mmol) in methanol (2 ml) at room temperature. The resulting mixture was stirred for 5 h. After completion of the reaction (monitored by TLC) the solvent was evaporated in vacuum and the residue purified by column chromatography (100 % ethyl acetate) to obtain (21) as a colourless syrup. Yield: 69 mg (53 % , colourless syrup); R_f: 0.52 (ethyl acetate); ^1H NMR (250.133 MHz, $CDCl_3$), δ (ppm): 1.25, 1.42 (2s, 6H, C (CH_3)$_2$); 4.47 (d, 1H, $J_{CH\ (a),CH\ (b)}$ = 11.9 Hz, CH*H*Ph); 4.56 (m, 2H, H-2, H-3); 4.61 (d, 1H, $J_{CH\ (a),CH\ (b)}$ = 11.9 Hz, C*H*HPh); 5.08 (d, 1H, $J_{3,4}$ = 3.6 Hz, H-4); 6.00 (d, 1H, $J_{1,2}$ = 3.3 Hz, H-1); 7.10-7.37 (m, 6H, H-1`, Ph); 9.82 (d, *J* = 10.6 Hz, $NH_2$); $^{13}C$ NMR (62.89 MHz, $CDCl_3$), δ (ppm): 25.4, 26.0 (C ($CH_3$)$_2$); 71.5 ($CH_2$Ph); 79.8 (C-6); 81.6 (C-2); 81.7 (C-3); 82.2 (C-4); 104.3 (C-1); 111.6 (*C* ($CH_3$)$_2$); 117.9 (CN); 126.6,127.0,127.4 (Ph); 135.9 (*i*-Ph); 158.2 (C-1`); 190.8 (CO); IR (capillary), ν (cm^{-1}): 1690 (C=O), 2235 (CN), 3242, 3100 (NH_2); MS (EI), *m/ z* (%): 344 $[M]^+$ $C_{18}H_{20}N_2O_5$ (344.364) Calcd: C = 62.78, H = 5.85, N = 8.13; Found: C = 62.91, H = 5.68, N = 8.45

3-Amino-4- (3-O-Benzyl-1,2-O-isopropylidene-α-D-xylopentofuranuronoyl)-5-methylsulfanyl-1-H-pyrazole-1-carbamidine (22)

Aminoguanidinium carbonate (48 mg, 0.35 mmol) was added to the solution of (9) (100 mg, 0.24 mmol) in DMF (2 ml) at 80 °C in the presence of triethylamine (0.1 ml, 0.7 mmol). The resulting mixture was stirred for 1 h. After completion of the reaction (monitored by TLC) the solvent was evaporated in vacuum and the residue purified by column chromatography (toluene/ EtOAc 8:2) to furnish compound 22. Yield: 65 mg (61 % , yellow solid); R_f: 0.54 (toluene/ EtOAc 7:3); $[\alpha]_D^{22}$: – 17.1

(c = 1.0, $CHCl_3$); ^1H NMR (250.133 MHz, $CDCl_3$), δ (ppm): 1.28, 1.48 (2s, 6H, C (CH_3)$_2$); 2.35 (s, 3H, SCH_3); 4.26 (d, 1H, $J_{CH\ (a),CH\ (b)}$ = 12.2 Hz, CH*H*Ph); 4.53 (d, 1H, $J_{CH\ (a),CH\ (b)}$ = 13.7 Hz, C*H*HPh); 4.54 (d, 1H, $J_{3,4}$ = 3.6 Hz, H-3`); 4.59 (d, 1H, $J_{1,2}$ = 3.6 Hz, H-2`); 5.34 (d, 1H, $J_{3,4}$ = 3.3 Hz, H-4`); 5.54 (br.s, 2H, $NH_2$); 6.10 (d, 1H, $J_{1,2}$ = 3.6 Hz, H-1`); 7.01-7.04 (m, 2H, Ph); 7.05-7.08 (m, 3H, Ph)); 8.40 (br.s, 1H, NH); ^{13}C NMR (62.89 MHz, $CDCl_3$), δ (ppm): 13.6 (SCH_3); 25.5, 26.1 (C (CH_3)$_2$); 70.9 (CH_2Ph); 80.4 (C-4`); 81.5 (C-2`); 82.0 (C-3`); 101.3 (C-4); 104.3 (C-1`); 111.2 (*C* (CH_3)$_2$); 126.7,127.0,127.1 (Ph); 135.7 (*i*-Ph); 145.8 (C-5); 152.0 (C-3); 153.9 (C-1``); 185.2 (CO); IR (capillary), ν (cm^{-1}): 1675 (CO), 3412, 3463 (NH_2); MS (EI), *m/ z* (%): 447 $[M]^+$ $C_{20}H_{25}N_5O_5S$ (447.506) HR-MS: Calcd: 447.15765 Found: 447.15631

7-Amino-ethyl-6- (3-desoxy-1,2-O-isopropylidene-α-D-glycero-pent-3-eno-furan- uronoyl)-2-methylsulfanyl-pyrazolo[1,5-a]pyrimidine-3-carboxylate (23)

Sodium (13 mg, 0.55 mmol) was added to methanol (1 ml). After 5 min 5-amino-3-methylsulfanyl-1*H*-pyrazole-4-carboxylic acid ethyl ester (106 mg, 0.53 mmol) was added and the solution stirred for 24 min. This mixture was then added to another stirred solution of (7) (100 mg, 0.26 mmol) in methanol (2 ml) at room temperature. The resulting mixture was stirred for 24 h. After completion of the reaction (monitored by TLC) the solvent was evaporated in vacuum and the residue purified by column chromatography (toluene/ EtOAc 7:3) to obtain compound 23. Yield: 69.5 mg (62 % , yellow solid); R_f: 0.50 (toluene/ EtOAc 7:3); $[\alpha]_D^{24}$: + 187.7 (c = 1.0, $CHCl_3$); ^1H NMR (250.133 MHz, $CDCl_3$), δ (ppm): 1.36 (t, 3H, CH_3-COOEt); 1.41, 1.43 (2s, 6H, C (CH_3)$_2$); 2.54 (s, 3H, SCH_3); 4.44 (dd, 2H, *J* = 7.0 Hz, CH_2-COOEt); 5.39 (dd, 1H, $J_{2,3}$ = 2.7 Hz, $J_{1,2}$ = 5.5 Hz, H-2`); 5.97 (d, 1H, $J_{2,3}$ = 2.4 Hz, C-3`); 6.21 (d, 1H, $J_{1,2}$ = 5.5 Hz, H-1`); 9.29 (s, 1H, H-6); 9.41 ($NH_2$); $^{13}C$ NMR (62.89 MHz, $CDCl_3$), δ (ppm): 12.4 (S$CH_3$); 13.5 ($CH_3$-COOEt); 26.6, 27.0 (C ($CH_3$)$_2$); 59.7 ($CH_2$-COOEt); 81.3 (C-2`); 99.5 (C-6); 100.9 (C-3); 106.0 (C-1`); 109.3 (C-3`); 112.1 (*C* (CH_3)$_2$); 147.5 (C-5); 147.8 (C-3a); 154.0 (C-4`); 155.0 (C-7); 160.4 (C-2); 161.6 (*CO*-COOEt); 182.3 (CO); IR (capillary), ν (cm^{-1}): 1669 (*CO*-COOEt), 1590 (CO), 3341, 3390 (NH_2); MS (EI), *m/ z* (%): 420 $[M]^+$ $C_{18}H_{20}N_4O_6S$ (420.437).

References

1. M. L. Sethi, *In Principles of medicinal chemistry*, 5th ed. W. O. Foye, T. L. Lemke, D. A. Williams, Eds., Lippincott. Williams and Wilkins, Philadelphia. **39**, 952 (2002).
2. M. F. A. Adamo, *Tetrahedron*, **60**, 841 (2004).
3. H. J. Schaeffer, L. de Beauchamp; P. Miranda; G. B. Elion; D. J. Bauer and P. Collins, *Nature*, **272**, 583 (1978).
4. Y. Diaz, F. Bravo and S. Castillon, *J. Org. Chem.*, **64**, 6508 (1999).
5. M. Zhang, *Carbohydr. Res.*, **318**, 157 (1999).
6. H. Y. Zhang, *Tetrahedron: Asymmetry*, **9**, 141 (1998).
7. a) D. Michalik and K. Peseke, *J. Carbohydr. Chem.*, **19**, 1049 (2000), b) L. Herrera, Dissertation 2002, Universitat Rostock.
8. I. A. Hashmi, H. Feist, Ma. Michalik, H. Reinke and K. Peseke, *J. Carbohydr. Chem.* **25** (1), 19 (2006).
9. P. Koll, S. Andree, K. Peseke and J. Kopf, *Carbohydr. Res.*, **241**, 273 (1993).
10. K. Peseke, H. D. Ambrosi and M. Michalik, *Carbhydr. Res.*, **194**, 87 (1989).
11. H. Bredereck, G. Simchen and R. Wahl, *Chem. Ber.*, **101**, 4048 (1968).
12. H. Bredereck, F. Effenberger and H. Botsch, *Chem. Ber.*, **97**, 3397 (1964).
13. H. Meerwein, W. Florian, N. Schon and G. Stopp, *Justus Liebigs Ann. Chem.*, **641**, 1 (1961).
14. M. Gomez, J. Quincoces, B. Kuhla, K. Peseke and H. Reinke, *J. Carbohydr. Chem.*, **18**, 57 (1999).
15. K. Peseke, H. Feist and E. Cunny, *Carbhydr. Res.*, **230**, 319 (1992).
16. R. Meisel, K. Peseke and H. Reinke, *J. Carbohydr. Chem.*, **17**, 1083 (1998).
17. F. Texier and A. Foucaud, *Tetrahedron Letters*, **23**, 4927 (1982).
18. K. Peseke, *Z. Chem.*, **17**, 288 (1977).
19. K. Peseke, M. Michalik and U. Schonhusen, *J. Prakt. Chem.*, **328**, 856 (1986).
20. T. Freier, M. Michalik, K. Peseke and H. Reinke, *J. Chem. Soc. Perkin Trans.* **2**, 1265 (1999).
21. M. Lalk, K. Peseke and H. Reinke, *J. Prakt. Chem.*, **341**, 552 (1999).
22. D. Michalik and K. Peseke, *J. Carbohydr. Chem.*, **19**, 141 (2000).
23. I. Rudloff, M. Michalik, A. Montero and K. Peseke, *Synthesis*, 1686 (2001).

24. I. Rudloff, K. Peseke and H. Reinke, *J. Prakt. Chem.*, **340**, 334 (1998).
25. A. Montero, H. Feist, M. Michalik and K. Peseke, *Synthesis*, 664 (2002).
26. J. Clazden, N. Greeves, S. Warren and P. Wothers, *Organic Chemistry*, Oxford University Press Inc., New York, 202 (2001).
27. H. Bredereck, F. Effenberger, H. Botsch and H. Rehn, *Chem. Ber.*, **98**, 1081 (1965).
28. H. Bredereck, F. Effenberger and R. Sell, *Chem. Ber.*, **97**, 3407 (1964).
29. J. Zimmermann, E. Buchdunger, H. Mett, T. Meyer and N. B. Lydon, *Bioorg. Med. Chem. Lett.*, **7**, 187 (1997).
30. J. L. Adams, J. C. Boehm and S. Kassis, *Bioorg. Med. Chem. Lett.*, **8**, 3111 (1998).
31. K. Methling, Dissertation, Universitat Rostock, 2001.
32. T. Lange, Dissertation, Universitat Rostock, 2000.
33. R. Troschutz, T. Dennstedt, *Arch. Pharm. (Weinheim)*, 33 (1994).
34. I. A. Hashmi, F. I. Ali, H. Feist, M. Michalik and K. Peseke, *Synthesis*, Submitted (2007).
35. L. Mosti, G. Menozzi and P. Schenone, *J. Heterocyclic Chem.*, **20**, 649 (1983).
36. G. B. Bennt, R. B. Mason and L. J. Alden, *J. Med. Chem.*, **21**, 623 (1978).
37. A. Kuno, H. Sakai, M. Ohkubo and H. Takasuji, *Chem. Pharm. Bull.*, **41**, 163 (1993).
38. S. Nagai, T. Ueda, A. Nagatsu, K. Nakaoka and N. Murakami, *J. Heterocyclic Chem.*, **35**, 329 (1998).
39. L. B. Townsend and B. L. Cline, *Lectrures in Heterocyclic Chem.*, 1st Ed., *Hetero Corp.*, **4** (1978).
40. K. Gerzon, D. C. Delong and J. C. Cline. *Pure Appl. Chem.*, **28**, 481 (1971).
41. M. I. Lim, R. S. Kline and J. J. Fox. *Tetrahedron Lett*, **21**, 1013 (1980).
42. R. E. Parks, J. D. stoeckler, C. Cambor and S. H. Chu. *Molecular Action and Targets for Cancer Chemotherapeutic Agents*, Academic Press New York, (1981).
43. R. J. Suhadolnik, *Nucleosides as Biological probes*, Wiley Interscience, New York, (1979).
44. R. H. Durland, T. S. Rao, V. Bodepudi and V. Revankar, *Nucleic Acid Res*, **23**, 647 (1995).
45. S. O. Doronina and J. P. Behr, *Chem. Soc. Rev.*, 63 (1997).
46. Y. Ito, M. Wakimura and C. Ito, *Heterocycles*, **36**, 2805 (1993).
47. I. Maeba, Y. Ito and M. Wakimura, *Heterocycles*, **36**, 1617 (1993).
48. J. L. Imbach and B. L. Kam, *J. Carbohydr. Nucleosides and Nucleotides*, **1**, 271 (1974).
49. T. J. Cousineau and J. A. Secrist III, *J. Org. Chem.*, **44**, 4351 (1979).
50. I. Maeba, Y. Nishiyama and A. Sato, *Heterocycles*, **41**, 50 (1995).
51. R. K. Robins and M. P. Schweizer, *J. Org. Chem*, **39**, 3226 (1974).
52. *i.* K. Peseke, *Pharmazie*, **31**, 9 (1976). *ii.* R. Gompper and W. Töpfl, *Chem. Ber.*, **95**, 2881 (1962).
53. T. Sasaki, K. Minamoto, T. Suzuki and S. Yamashita, *Terahedron*, **36**, 865 (1980).