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Synthesis and Antiviral Screening of Some Novel Pyridazine and Triazolopyridazine Nucleosides

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ABSTRACT: *Some novel cyclic and acyclic pyridazine and triazolopyridazine nucleoside derivatives were prepared. Some of the prepared products were selected and tested for antiviral activity against herpes simplex virus type-1 (HSV-1) and hepatitis-A virus (HAV, MBB-cell culture adapted strain). Plaque reduction infectivity assay was used to determine virus count reduction as a result of treatment with test compounds. Compound* **15** *showed the highest effect on the HAV than the other tested compounds.*
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INTRODUCTION

The chemistry of pyridazine and fused heterocyclic pyridazine nucleosides has attracted attention during the last few decades because of its interesting pharmacological activities, especially as potential antiviral, antitumor, and antibiotic agents [1–7]. In connection with our research program for the synthesis of different fused heterocyclic nucleosides with biological interest [8–10], we describe the syntheses of some cyclic and acyclic nucleosides of pyridazine and triazolopyridazine derivatives with promising antiviral activity.

RESULTS AND DISCUSSION

The hydrazone derivatives **2a,b** were prepared by reacting (5,6-diphenylpyridazin-3-yl)-hydrazine (**1**) [11] with some monosaccharides, namely, D-glucose or D-ribose in ethanol and glacial acetic acid (Scheme 1). The products revealed absorption frequencies due to OH + NH and C=N in IR spectra, and their ^{1}H NMR spectra showed the presence of the sugar protons, NH, and azo-methine (CH=N) (cf. the Experimental). Cyclization of hydrazones in acetic anhydride is extensively reported in several literature [12–14]; however, our attempts to cyclize the hydrazone derivatives **2a,b** by heating in acetic anhydride gave two products, as judged by TLC, **3a,b** and **4a,b**. This could be explained by acetylation of hydrazone derivatives **2a,b** giving their corresponding *O*-acetylated sugar hydrazone derivatives **3a,b** followed by oxidative cyclization [14–16] giving the *O*-acetylated cyclic *C*-nucleosides **4a,b** (Scheme 1). The IR spectra of **3a,b** revealed the absence of hydroxyl groups and showed absorption bands due to NH and C=O. The ^{1}H NMR and ^{13}C NMR spectra showed the presence of OAc groups (cf. the Experimental). On the other hand, the absence of NH as well as the azo-methine (CH=N) in the ^{1}H NMR spectra in **4a,b** confirmed their structures (cf. the Experimental).

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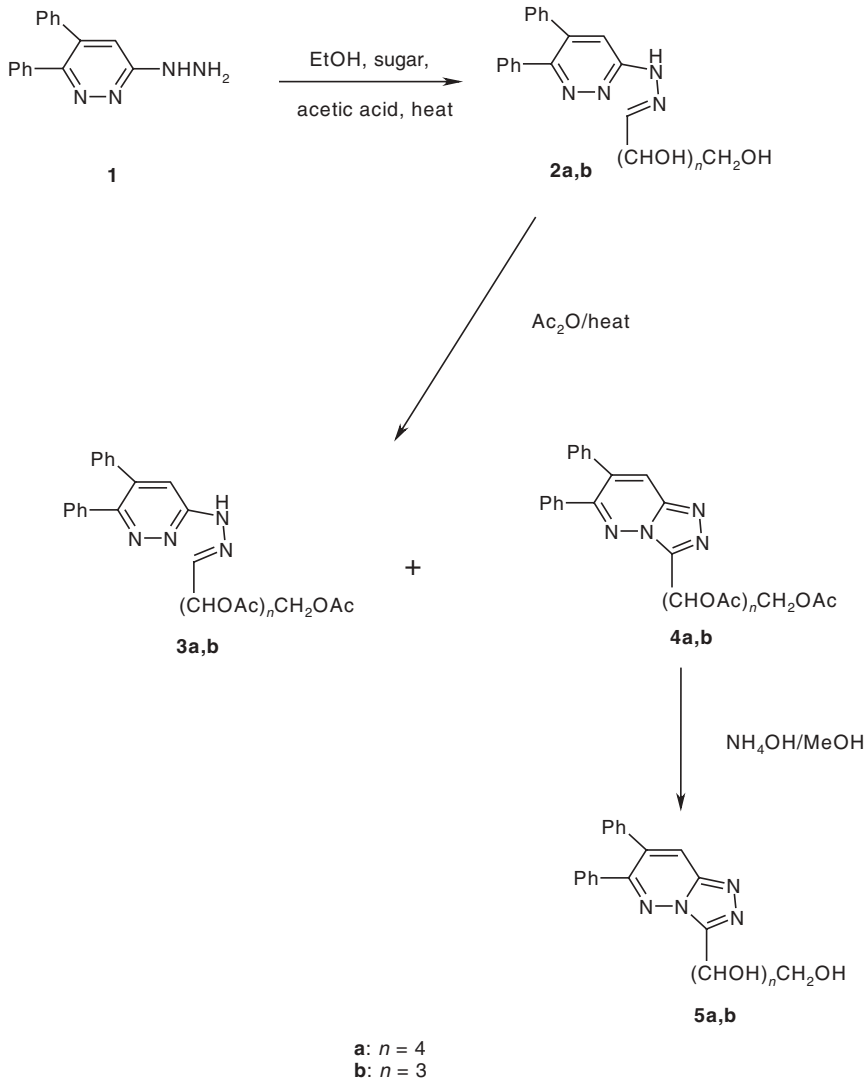
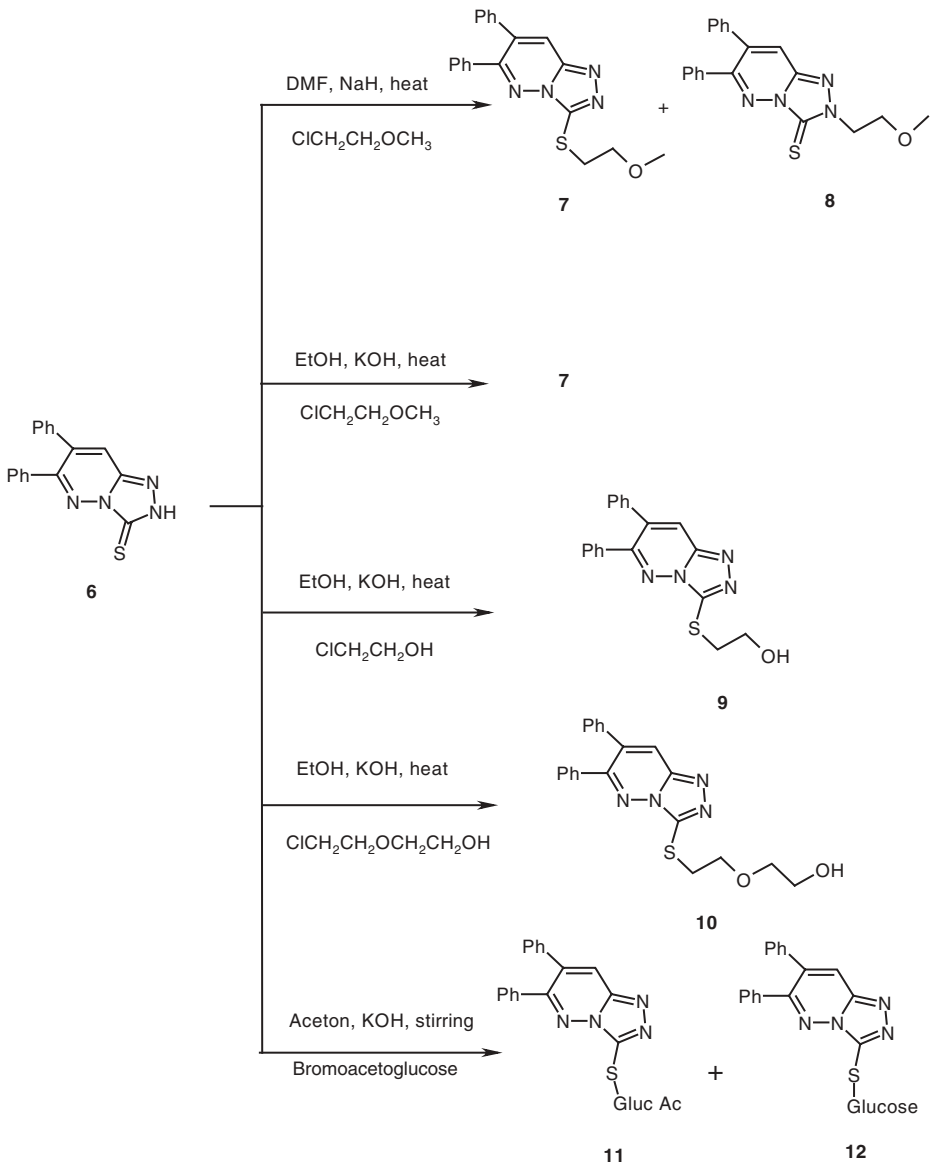


SCHEME 1

Deprotection of **4a,b**, using ammonium hydroxide solution in methanol [12] gave the target free cyclic *C*-nucleosides **5a,b**. The structures of the aforementioned compounds were confirmed on the basis of their elemental and spectral data (cf. the Experimental). Their IR spectra revealed absorption frequencies due to OH and C=N, while their ^{1}H NMR spectra showed signals of alditol protons and of hydroxyl groups (D_2O exchangeable) (cf. the Experimental).

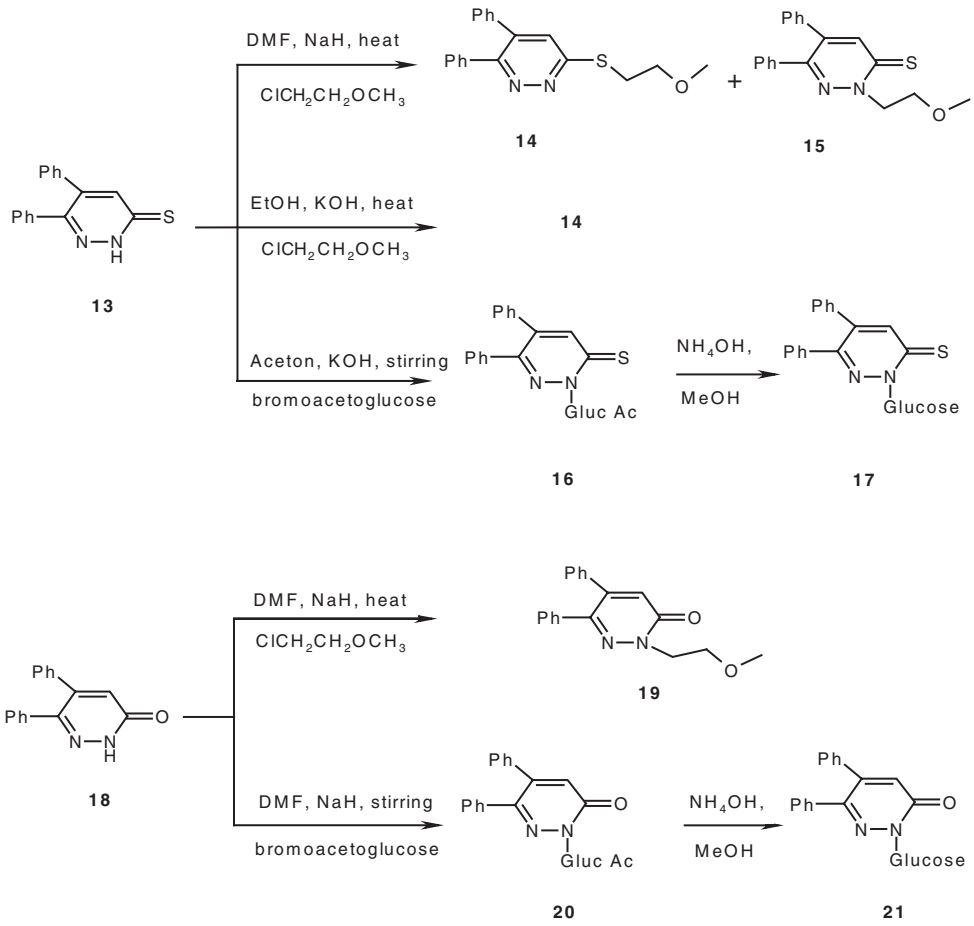
Because of the synthesis of acyclovir, one of the potent antiherpetic drug by Schaffer et al. [17], many attempts have been made by nucleoside chemists to prepare a number of relative compounds with various side chains and glycons [18–20]; however, acyclonucleosides of pyridazine and triazolopyridazine derivatives were rarely reported in the literature. Thus and in continuation of our previous work [8–10] on the preparation of various cyclic and acyclic nucleosides of different heterocyclic compounds, we describe in this report the synthesis of some cyclic and acyclic nucleosides of pyridazine and fused pyridazine derivatives by treating the sodium salt of 6,7-diphenyl-[1,2,4]triazolo[4,3-*b*]pyridazine-3(2*H*)-thione (**6**) [11] (generated in situ, cf., the Experimental) with 2-chloroethyl methyl ether to give the corresponding acyclonucleosides **7** and **8** (Scheme 2). The presence of methoxyethyl protons and the absence of the NH signal in the ^{1}H NMR spectra of **7** and **8** confirmed their structures. Also, the ^{13}C NMR spectra showed the absence of C=S signal of **7**, while that of **8** revealed the presence of C=S signal, which confirmed the difference in the site of attacks between **7** and **8** (cf. the Experimental). On the other hand, treatment of **6** with 2-chloroethyl methyl ether in alcoholic



SCHEME 2

potassium hydroxide solution gave only acyclonucleoside **7** (Scheme 2). Similarly, treatment of **6** with 2-chloroethanol or 2-(2-chloroethoxy)-ethanol in alcoholic potassium hydroxide solution gave compounds assigned to the structures of acyclonucleosides **9** and **10**, respectively (cf. the Experimental, Scheme 2). The treatment of **6** with 2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranosyl bromide in dry acetone in the presence of potassium hydroxide, gave **11** and an unexpected product **12**. The structure of nucleosides **11** and **12** was elucidated on the basis of their elemental and spectral data (cf. the Experimental, Scheme 2). The IR and ^{1}H NMR spectra of **12** revealed signals indicative of the OH groups (cf. the Experimental). Also, the ^{13}C NMR spectrum of **12** showed signals of glucose ring and signals of four acetyl groups were absent (cf. the Experimental). Compound **12** was presumably obtained by the formation of **11** that, on prolonged stirring in the reaction media, was deacetylated to give **12**.

Likewise, treatment of the sodium salt of 5,6-diphenylpyridazine-3-(2*H*)-thione (**13**) [11] or 5,6-diphenylpyridazin-3-(2*H*)-one (**18**) [11] (generated in situ, cf. the Experimental) with 2-chloroethyl methyl ether gave acyclonucleosides **14**, **15,** or **19**, respectively (Scheme 3); while treatment of **13** with 2-chloroethyl methyl ether, in alcoholic potassium hydroxide solution, gave only **14** (cf. the Experimental, Scheme 3).



SCHEME 3

On the other hand, treatment of **13** or **18** with 2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranosyl bromide gave **16** or **20**, respectively (Scheme 3). The structure of nucleosides **16** and **20** was elucidated on the basis of their elemental and spectral data (cf. the Experimental, Scheme 3). The presence of C=S in the ^{13}C NMR spectrum of **16** and C=O groups in the IR spectrum of **20** showed that the site of attack was on *N*-2 (cf. the Experimental, Scheme 3).

Ammonolysis of cyclic nucleosides **16** and **20** afforded deacetylated nucleosides **17** and **21**, respectively. The IR and ^{1}H NMR spectra of the aforementioned products showed signals indicative of the OH groups (cf. the Experimental, Scheme 3).

ANTIVIRAL BIOASSAY

Preparation of Synthetic Compounds for Bioassay

One hundred milligram of each tested compound was dissolved in 1 mL of 10% DMSO in water. The final concentration was 100 μg/μL (the stock solution). The dissolved stock solutions were decontaminated by adding 50 μg/mL antibiotic-antimycotic mixture (10,000 U penicillin G sodium, 10,000 μg streptomycin sulfate, and 250 μg amphotericin B, PAA Laboratories GmbH, Austria).

Cell Culture

African green monkey kidney-derived cells (Vero) and human hepatoma cell line (HepG2) were used. Cells were propagated in Dulbeccos' minimal essential medium (DMEM) supplemented with 10% fetal bovine serum and 1% antibiotic-antimycotic mixture. The *p*H was adjusted to 7.2–7.4 by 7.5% sodium bicarbonate solution. The mixture was sterilized by filtration through 0.2 μm pore size nitrocellulose membrane.

Viruses

Herpes simplex virus type-1 (HSV-1) and hepatitis-A virus (HAV, MBB-cell culture adapted strain) were

obtained from the Environmental Virology Laboratory, Department of Water Pollution Research, National Research Centre, Cairo, Egypt.

Cytotoxicity Assay

Cytotoxicity was assayed for both dimethyl sulfoxide (DMSO) and tested compounds. Serial dilutions were prepared and inoculated on Vero cells grown in 96 well tissue culture plates. The maximum-tolerated concentration (MTC) for each compound was determined by both cell morphology and cell viability by staining with tryban blue dye.

Plaque Reduction Infectivity Assay

On a six-well plate, cell culture (10^5 cell/mL) was cultivated and incubated for 2 days at 37°C. HSV-1 and HAV were diluted to give 10^4 PFU/mL final concentrations for each virus and mixed with the tested compound at the previous concentration and incubated overnight at 4°C. Growth medium was removed from the multiwell plate, and virus-compound mixture was inoculated (100 μL/well). After 1 h contact time, the inoculum was aspirated and 3 mL of MEM with 1% agarose was overlaid with cell sheets. The plates were left to solidify and incubated at 37°C until the development of virus plaques. Cell sheets were fixed in 10% formaline solution for 2 h and stained with crystal violet stain. Control virus and cells were treated identically without any chemical compound. Virus plaques were counted, and the percentage of reduction was calculated [21].

The plaque infectivity assay was carried out to test **2a, 4a, 7, 10, 15**, and **19** for the antiviral activity. The test was performed to include the three possibilities for the antiviral activity: virucidal effect, virus adsorption, and effect on virus replication for both HAV and HSV-1. It was obvious that, at the concentration of 20 μg/10^5 cells, **4a, 7**, and **19** revealed the highest antiviral activity in comparison with Amantadine (C*) as a control. At the concentration of 50 μg/10^5 cells, **7** and **10** revealed the highest antiviral activity in comparison with Amantadine (C*) as a control. In general, **15** showed the highest effect on the HAV than the other five tested compounds and the control (Amantadine), where its antiviral activity increased from 63% at the concentration of 20 μg/10^5 cells to 86% at the concentration of 50 μg/10^5 cells. On the other hand, the six tested compounds did not show any activity against HSV-1.

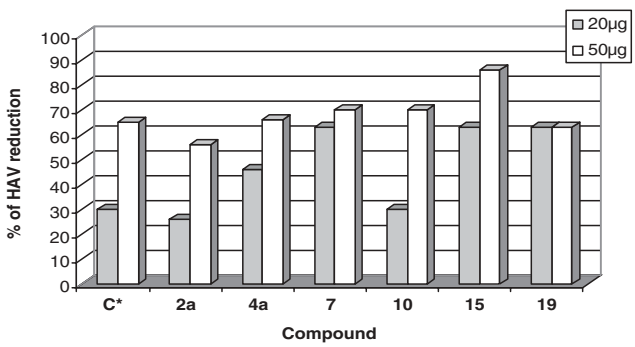


FIGURE 1 Effect of some novel cyclic and acyclic pyridazine and triazolo-pyridazine nucleoside derivatives on HAV (MBB cell culture strain) in comparison with Amantadine (C*) as a control. Compound **15** showed a relatively high effect on the HAV virus (86% at 50 μg/10^5 cells).

EXPERIMENTAL

All melting points were uncorrected and were measured using Electro-thermal IA 9100 apparatus (Shimadzu, Japan). IR spectra were recorded as potassium bromide pellets on a Perkin-Elmer 1650 spectrophotometer (at the National Research Centre, Cairo, Egypt). ^{1}H NMR and ^{13}C NMR spectra were determined on a Jeol-Ex-300 NMR spectrometer, and chemical shifts were expressed as part per million (two values) against TMS as an internal reference (at the Faculty of Science, Cairo University, Cairo, Egypt). Mass spectra were recorded on EI + Q1 MSLMR UPLR (at the National Research Centre, Cairo, Egypt). Microanalyses were done using Mario Elmentar apparatus (at the Organic Microanalysis Unit, National Research Centre, Cairo, Egypt), and the results are within the accepted range (±0.40) of the calculated values. Column chromatography was performed on (Merck) Silica gel 60 (particle size 0.06–0.20 mm).

Compounds **1, 6, 13**, and **18** were prepared according to the published procedure [11].

*Aldehydo-D-sugar-(5,6-diphenylpyridazin-3-yl)-hydrazones (**2a,b**)*

General Procedure: A mixture of **1** (2.62 g, 1 mmol), D-glucose (1.8 g, 1 mmol) or D-ribose (1.4 g, 1 mmol) in 20 mL ethanol, and a catalytic amount of acetic acid (three drops) was heated at 80°C for 2 h. The formed precipitate was filtered when it was hot, washed several times with ethanol, and dried to get **2a,b**.

*Aldehydo-D-glucose-(5,6-diphenylpyridazin-3-yl)-hydrazone (**2a**)*. Yield 80%; mp 184–185°C; IR (KBr): $\nu = 3385$–3218 (broad, OH + NH), 1591 (C=N) cm^{-1}, 1H NMR (DMSO-d_6): $\delta = 3.20$–3.60 (protons of the alditol congregated with the water

signal) [12,13], 3.75–3.80 (m, 2H, CH_2OH), 4.2–4.3 (m, 1H, OH, D_2O exchangeable), 4.42–4.59 (m, 3H, 3OH, D_2O exchangeable), 5.2 (d, $J = 6.4$ Hz, 1H, OH, D_2O exchangeable), 7.20–7.50 (m, 11H, Ar-H, and C*H*=N), 7.80 (s, 1H, C_4-H), 8.40 (s, 1H, NH, D_2O exchangeable). Anal. calcd for $C_{22}H_{24}N_4O_5$ (424.17): C, 62.25; H, 5.70; N, 13.20; found: C, 62.20; H, 5.57; N, 13.28.

*Aldehydo-D-ribose-(5,6-diphenylpyridazin-3-yl)-hydrazone (***2b***)*. Yield 78%; mp 160–162°C; IR (KBr): $\nu = 3390$–3238 (broad, OH + NH), 1598 (C=N) cm^{-1}; ^{1}H NMR (DMSO-d_6): $\delta = 3.25$–3.50 (protons of the alditol congregated with the water signal), 3.55–3.60 (m, 2H, CH_2OH), 4.40–4.50 (m, 1H, OH, D_2O exchangeable), 4.80–4.90 (m, 2H, 2OH, D_2O exchangeable), 6.10 (d, $J = 6.80$ Hz, 1H, OH, D_2O exchangeable), 7.0 (s, 1H, C*H*=N), 7.10–7.35 (m, 11H, Ar-H, and C_4-H), 8.14 (s, 1H, NH, D_2O exchangeable). Anal. calcd for $C_{21}H_{22}N_4O_4$ (394.16): C, 63.95; H, 5.62; N, 14.20; found: C, 64.04; H, 5.60; N, 14.29.

Preparation of **3a,b** *and* **4a,b**

General Procedure: Compounds **2a,b** (1 mmol) were stirred on water-bath in 20 mL acetic anhydride for 3 h. The reaction mixtures were poured onto ice-water with stirring, and the solids that precipitated were collected by filtration, washed with water, dried, and purified on a silica gel column using *n*-hexane: ethyl acetate (4:1) as an eluent to get **3a,b** and **4a,b**, respectively.

*Per-O-acetyl-aldehydo-D-glucose-(5,6-diphenylpyridazin-3-yl)-hydrazone(***3a***)*. Yield 30%; oil; IR (KBr): $\nu = 3228$ (NH), 1728 (CO), 1608 (C=N); ^{1}H NMR ($CDCl_3$, ppm): $\delta = 2.0$–2.2 (m, 15H, 5OAc), 2.89 (m, 2H, CH_2OAc), 3.85–4.20 (m, 2H, 2C*H*OAc), 4.90 (m, 1H, C*H*OAc), 5.10 (m, 1H, C*H*OAc), 7.10–7.35 (m, 10H, Ar-H), 8.18 (s, 1H, C_4-H), 8.61 (s, 1H, C*H*=N), 9.80 (s, 1H, NH, D_2O exchangeable). Anal. calcd for $C_{32}H_{34}N_4O_{10}$ (634.23): C, 60.56; H, 5.40; N, 8.83; found: C, 60.49; H, 5.36; N, 8.76.

*Per-O-acetyl-aldehydo-D-ribose-(5,6-diphenylpyridazin-3-yl)-hydrazone (***3b***)*. Yield 34%; oil; IR (KBr): $\nu = 3230$ (NH), 1692 (CO), 1598 (C=N); ^{1}H NMR ($CDCl_3$, ppm): $\delta = 2.10$–2.22 (m, 12H, 4OAc), 2.90–2.95 (m, 2H, CH_2OAc), 3.82–4.25 (m, 1H, C*H*OAc), 4.92 (m, 1H, C*H*OAc), 5.13 (m, 1H, C*H*OAc), 7.15–7.42 (m, 10H, Ar-H), 8.17 (s, 1H, C_4-H), 8.59 (s, 1H, C*H*=N), 9.80 (s, 1H, NH, D_2O exchangeable). Anal. calcd for $C_{29}H_{30}N_4O_8$ (562.21): C, 61.91; H, 5.38; N, 9.96; found: C, 62.13; H, 5.23; N, 9.91.

*3-(1,2,3,4,5-Penta-O-acetyl-D-glucos-1-yl)-5,6-diphenyl[1,2,4]triazolo[4,3-b]pyridazine (***4a***)*. Yield 70%; mp 160–162°C; IR (KBr): $\nu = 1728$ (CO), 1608 (C=N); ^{1}H NMR ($CDCl_3$, ppm): $\delta = 2.0$–2.2 (m, 15H, 5OAc), 2.80–2.90 (m, 2H, CH_2OAc), 4.3–4.4 (m, 2H, 2C*H*OAc), 5.20–5.30 (m, 1H, C*H*OAc), 5.4–5.5 (m, 1H, C*H*OAc), 7.13–7.40 (m, 10H, Ar-H), 8.06 (s, 1H, C_8-H); ^{13}C NMR ($CDCl_3$, ppm): $\delta = 21.34$–22.21 (4OCH_3), 61.15–71.65 (C-alditol), 124.09–129.78 (Ar-C), 134.76 (C-8), 136.62 (C-7), 144.45 (C-6), 145.30 (C-8a), 155.50 (C-3), 169.80–170.60 (4CO). Anal. calcd for $C_{32}H_{32}N_4O_{10}$ (632.21): C, 60.75; H, 5.10; N, 8.86; found: C, 60.82; H, 5.14; N, 8.72.

*3-(1,2,3,4-Tetra-O-acetyl-D-ribos-1-yl)-5,6-diphenyl-[1,2,4]triazolo[4,3-b]pyridazine(***4b***)*. Yield 66%; mp 154–156°C; IR (KBr): $\nu = 1725$ (CO), 1604 (C=N); ^{1}H NMR ($CDCl_3$, ppm): $\delta = 2.1$–2.2 (m, 12H, 4OAc), 2.84–2.90 (m, 2H, CH_2OAc), 4.3–4.45 (m, 1H, C*H*OAc), 5.22–5.35 (m, 1H, C*H*OAc), 5.4–5.45 (m, 1H, C*H*OAc), 7.2–7.40 (m, 10H, Ar-H), 8.10 (s, 1H, C_8-H). Anal. calcd for $C_{29}H_{28}N_4O_8$ (560.19): C, 62.14; H, 5.03; N, 9.99; found: C, 62.10; H, 5.10; N, 9.91.

*3-(Alditol-1-yl)-6,7-diphenyl-[1,2,4]triazolo[4,3-b]pyridazines (***5a,b***)*

General Procedure: Ammonium hydroxide solution (5 mL, 35%) was added to a solution of **4a,b** (1 mmol) in 20 mL dry methanol, and the reaction mixtures were stirred at room temperature for 2 and 3 h, respectively. The reaction mixtures were evaporated under reduced pressure at 40°C, and the residues were purified on the silica gel column using chloroform: methanol (4:1) as an eluent to get **5a,b**, respectively.

*3-(D-Gluco-pentitol-1-yl)-6,7-diphenyl-[1,2,4]-triazolo[4,3-b]pyridazine(***5a***)*. Yield 45%; oil; IR (KBr): $\nu = 3442$–3320 (OH); ^{1}H NMR (DMSO-d_6): $\delta = 3.3$–3.6 (protons of alditol congregated with the water signal), 3.65–3.9 (m, 2H, CH_2OH), 4.1–4.3 (m, 1H, OH, D_2O exchangeable), 4.9–5.2 (m, 3H, 3OH, D_2O exchangeable), 5.2 (d, $J = 5.4$ Hz, 1H, OH, D_2O exchangeable), 7.1–7.4 (m, 10H, Ar-H), 8.2 (s, 1H, C_8-H). Anal. calcd for $C_{22}H_{22}N_4O_5$ (422.16): C, 62.55; H, 5.25; N, 13.26; found: C, 62.49; H, 5.32; N, 13.29.

*3-(D-Ribo-tetritol-1-yl)-6,7-diphenyl-[1,2,4]triazolo-[4,3-b]pyridazine (***5b***)*. Yield 42%; oil; IR (KBr): $\nu = 3442$–3320 (OH); ^{1}H NMR (DMSO-d_6): $\delta = 3.3$–3.6 (protons of alditol congregated with the water

signal), 3.65–3.9 (m, 2H, CH_2OH), 4.1–4.3 (m, 1H, OH, D_2O exchangeable), 4.9–5.2 (m, 2H, 2OH, D_2O exchangeable), 5.2 (d, $J = 5.4$ Hz, 1H, OH, D_2O exchangeable), 7.1–7.4 (m, 10H, Ar-H), 8.2 (s, 1H, C_8-H). Anal. calcd for $C_{21}H_{20}N_4O_4$ (392.15): C, 64.28; H, 5.14; N, 14.28; found: C, 64.20; H, 5.19; N, 14.20.

Preparation of **7** *and* **8**

1. To a solution of **6** (1 mmol) in 20 mL dry dimethylformamide, sodium hydride (2 mmol) was added, the reaction mixture was stirred at 70°C for 1 h and cooled, and 2-chloroethyl methyl ether (2 mmol) was added with stirring at room temperature for 6 h. The reaction mixture was evaporated under reduced pressure, and the residue was purified on the silica gel column using petroleum ether 40–60°C: ethyl acetate (4:1) as an eluent to get **7** and **8**, respectively.
2. To a solution of 20 mL dry ethanol containing KOH (1 mmol), compound **6** (1 mmol) was added, and the reaction mixture was stirred for 1 h at room temperature. 2-Chloroethyl methyl ether (2 mmol) was added and the reaction mixture was stirred at 70°C for 4 h, then evaporated under reduced pressure, and the residue was recrystallized from ethanol to get a compound identical with **7** in all respects.

*3-(2-Methoxyethylsulfanyl)-6,7-diphenyl-[1,2,4]-triazolo[4,3-b]pyridazine (**7**).* Yields 68% (for method (i)), 77% (for method (ii)); mp 161–162°C; ^{1}H NMR ($CDCl_3$, ppm): $\delta = 3.40$ (s, 3H, OCH_3), 3.63 (t, $J = 11.70$ Hz, 2H, CH_2O), 3.83 (t, $J = 12$ Hz, 2H, CH_2S), 7.15–7.38 (m, 10H, Ar-H), 7.90 (s, 1H, C_8-H); ^{13}C NMR ($CDCl_3$, ppm): $\delta = 31.24$ (OCH_3), 58.84 (CH_2O), 70.96 (CH_2S), 124.09–129.78 (Ar-C), 134.76 (C-8), 136.62 (C-7), 144.45 (C-6), 145.30 (C-8a), 155.50 (C-3). Anal. calcd for $C_{20}H_{18}N_4OS$ (362.12): C, 66.28; H, 5.01; N, 15.46; S, 8.85; found: C, 66.20; H, 5.12; N, 15.40; S, 8.85.

*2-(2-Methoxyethyl)-6,7-diphenyl[1,2,4]triazolo[4,-3-b]pyridazine-3(2H)-thione (**8**).* Yield 32%; oil; ^{1}H NMR ($CDCl_3$, ppm): $\delta = 3.48$ (s, 3H, OCH_3), 3.65 (t, $J = 11.40$ Hz, 2H, CH_2O), 3.80 (t, $J = 11.90$ Hz, 2H, CH_2N), 7.15–7.38 (m, 10H, Ar-H), 8.03 (s, 1H, C_8-H); ^{13}C NMR ($CDCl_3$, ppm): $\delta = 31.87$ (OCH_3), 59.34 (CH_2O), 71.68 (CH_2N), 124.49–129.87 (Ar-C), 134.36 (C-8), 136.96 (C-7), 144.32 (C-6), 144.39 (C-8a), 182.10 (C=S). Anal. calcd for $C_{20}H_{18}N_4OS$ (362.12): C, 66.28; H, 5.01; N, 15.46; S, 8.85; found: C, 66.22; H, 5.11; N, 15.34; S, 8.88.

Preparation of **9** *and* **10**

General Procedure: To a solution of 20 mL dry ethanol containing KOH (1 mmol), **6** (1 mmol) was added, the reaction mixture was stirred for 1 h at room temperature, and then 2-chloroethanol or 2-(2-chloroethoxy)-ethanol (2 mmol) was added and stirred at 70°C for 3 and 4 h, respectively. The reaction mixtures were evaporated under reduced pressure, and the residues were purified on the silica gel column using chloroform: methanol (9:1) as an eluent to get **9** and **10**, respectively.

*3-(2-Hydroxyethylsulfanyl)-6,7-diphenyl-[1,2,4]-triazolo[4,3-b]pyridazine (**9**).* Yield 73%; oil; IR (KBr): $\nu = 3500$–3295 (OH); ^{1}H NMR (DMSO-d_6): $\delta = 3.58$ (m, 2H, CH_2O), 3.68 (t, $J = 6$ Hz, 2H, CH_2S), 5.10 (s, 1H, OH, D_2O exchangeable), 7.22–7.31 (m, 10H, Ar-H), 8.29 (s, 1H, C_8-H). Anal. calcd for $C_{19}H_{16}N_4OS$ (348.10): C, 65.50; H, 4.63; N, 16.08; S, 9.20; found: C, 65.56; H, 4.60; N, 16.10; S, 9.24.

*3-(2-Hydroxyethoxyethylsulfanyl)-6,7-diphenyl-[1,-2,4]triazolo[4,3-b]pyridazine (**10**).* Yield 64%; oil; IR (KBr): $\nu = 3500$–3290 (OH); ^{1}H NMR (DMSO-d_6): $\delta = 3.40$ (m, 4H, $2CH_2$), 3.60 (m, 4H, $2CH_2$), 4.25 (s, 1H, OH, D_2O exchangeable), 7.2–7.4 (m, 10H, Ar-H), 8.25 (s, 1H, C_8-H). Anal. calcd for $C_{21}H_{20}N_4O_2S$ (392.13): C, 64.27; H, 5.14; N, 14.28; S, 8.17; found: C, 64.20; H, 5.12; N, 14.34; S, 8.21.

Preparation of **11** *and* **12**

To a solution of **6** (1 mmol) in 20 mL dry acetone containing KOH (1 mmol), a solution of 2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranosyl bromide (1 mmol) in 10 mL dry acetone was added. The reaction mixture was stirred for 3 h then evaporated under reduced pressure at 40°C, and the residue was purified on the silica gel column using petroleum ether 40–60°C: ethyl acetate (4:1) as an eluent to get **11** and **12**.

*3-(2,3,4,6-Tetra-O-acetyl-α-D-glucopyranosulfanyl)-6,7-diphenyl[1,2,4]triazolo[4,3-b]pyridazine (**11**).* Yield 28%; mp 143–144°C; IR (KBr): $\nu = 1735$ (CO); ^{1}H NMR ($CDCl_3$, ppm): $\delta = 1.90$–2.10 (4s, 12H, $4CH_3CO$), 4.05 (m, 2H, 2H-6′), 4.20–4.35 (m, 2H, H-5′, H-4′), 5.20–5.47 (m, 2H, H-3′, H-2′), 6.03 (d, $J_{1',2'} = 9$ Hz, 1H, H-1′), 7.20–7.32 (m, 10H, Ar-H), 8.30 (s, 1H, C_8-H); ^{13}C NMR ($CDCl_3$, ppm): $\delta = 20.80$–20.95 ($4CH_3$), 61.66 (C-6′), 67.86 (C-4′), 69.40 (C-2′), 74.19 (C-3′), 74.87 (C-5′), 86.12 (C-1′), 124.09–129.78 (Ar-C), 134.76 (C-8), 136.62 (C-7), 144.45 (C-6), 145.30 (C-8a), 155.50 (C-3), 169.80–170.60 (4CO). Anal. calcd for $C_{31}H_{30}N_4O_9S$ (598.63): C, 58.67; H,

4.76; N, 8.83; S, 5.05; found: C, 58.74; H, 4.70; N, 8.89; S, 5.01.

*3-(α-D-Glucopyranosulfanyl)-6,7-diphenyl-[1,2,4]-triazolo[4,3-b]pyridazine (**12**).* Yield 52%; mp 102–104°C; IR (KBr): $\nu = 3500$–3250 (OH); ^{1}H NMR (DMSO-d_6): $\delta = 3.16$–3.50 (m, 6H, 2H-6′, H-5′, H-4′, H-3′, and H-2′), 4.33 (m, 1H, HO-3′, D_2O exchangeable), 5.10 (m, 1H, HO-4′, D_2O exchangeable), 5.18 (m, 2H, HO-2′, D_2O exchangeable), 5.34 (m, 1H, HO-6′, D_2O exchangeable), 6.72 (d, $J_{1',2'} = 9.30$ Hz, 1H, H-1′), 7.0–7.40 (m, 10H, Ar-H), 8.37 (s, 1H, C_8-H); ^{13}C NMR (DMSO-d_6): $\delta = 61.66$ (C-6′), 67.86 (C-4′), 69.40 (C-2′), 74.19 (C-3′), 74.87 (C-5′), 86.12 (C-1′), 124.29–129.58 (Ar-C), 134.71 (C-8), 136.66 (C-7), 144.51 (C-6), 145.36 (C-8a), 155.32 (C-3). Anal. calcd. for $C_{23}H_{22}N_4O_5S$ (466.52): C, 59.22; H, 4.75; N, 12.01; S, 6.87; found: C, 59.28; H, 4.71; N, 12.14; S, 6.84.

Preparation of **14** *and* **15**

Compounds **14** and **15** were prepared from **13** as described for **7** and **8**.

*3-(2-Methoxyethylsulfanyl)-5,6-diphenylpyridazine-(**14**).* Yields 60% (for method (i)), 73% (for method (ii)); mp 161–162°C; ^{1}H NMR ($CDCl_3$, ppm): $\delta = 3.42$ (s, 3H, OCH_3), 3.66 (t, $J = 12$ Hz, 2H, CH_2O), 3.83 (t, $J = 12$Hz, 2H, CH_2S), 7.14–7.40 (m, 10H, Ar-H), 7.93 (s, 1H, C_4-H); ^{13}C NMR ($CDCl_3$, ppm): $\delta = 29.66$ (OCH_3), 58.75 (CH_2O), 70.97 (CH_2S), 126.55–129.75 (Ar-C), 138.47 (C-4), 142.97 (C-5), 156.84 (C-6), 160.19 (C-3). Anal. calcd for $C_{19}H_{18}N_2OS$ (322.11): C, 70.78; H, 5.63; N, 8.69; found: C, 70.70; H, 5.68; N, 8.74.

*2-(2-Methoxyethyl)-5,6-diphenylpyridazine-3(2H)-thione (**15**).* Yield 40%; oil; ^{1}H NMR ($CDCl_3$, ppm): $\delta = 3.42$ (s, 3H, OCH_3), 4.03 (t, $J = 11.40$ Hz, 2H, CH_2O), 5.0 (t, $J = 11.70$ Hz, 2H, CH_2N), 7.10–7.40 (m, 10H, Ar-H), 8.42 (s, 1H, C_4-H); ^{13}C NMR ($CDCl_3$, ppm): $\delta = 30.34$ (OCH_3), 59.50 (CH_2O), 71.28 (CH_2N), 128.18–129.45 (Ar-C), 137.14 (C-4), 142.80 (C-5), 151.45 (C-6), 182.30 (C=S). Anal. calcd for $C_{19}H_{18}N_2OS$ (322.11): C, 70.78; H, 5.63; N, 8.69; found: C, 70.82; H, 5.60; N, 8.65.

*2-(2,3,4,6-Tetra-O-acetyl-α-D-glucopyranosyl)-5,6-diphenylpyridazine-3(2H)-thione (**16**)*

To a solution of **13** (1 mmol) in 20 mL dry acetone containing KOH (1 mmol), a solution of 2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranosyl bromide (1 mmol) in 10 mL dry acetone was added. Then, the reaction mixture was stirred for 1 h, evaporated under reduced pressure at 40°C, and purified on the silica gel column using petroleum ether: ethyl acetate (4:1) at 40–60°C as an eluent to get **16**.

Yield 72%; oil; IR (KBr): $\nu = 1754$ (CO); ^{1}H NMR ($CDCl_3$, ppm): $\delta = 1.96$–2.07 (4s, 12H, $4CH_3CO$), 4.10–4.35 (m, 4H, 2H-6′, 5′-H, H-4′), 5.25 (m, 1H, H-3′), 5.48 (m, 1H, H-2′), 6.02 (d, $J_{1',2'} = 9$ Hz, 1H, H-1′), 7.20–7.38 (m, 10H, Ar-H), 7.80 (s, 1H, C_4-H); ^{13}C NMR ($CDCl_3$, ppm): $\delta = 20.60$–20.73 ($4CH_3$), 61.68 (C-6′), 67.99 (C-4′), 69.49 (C-2′), 74.13 (C-3′), 74.81 (C-5′), 85.98 (C-1′), 128.17–129.50 (Ar-C), 137.14 (C-4), 142.80 (C-5), 151.45 (C-6), 169.80–170.60 (4CO), 182.30 (C=S). Anal. calcd for $C_{30}H_{30}N_2O_9S$ (594.65): C, 60.60; H, 5.09; N, 4.71; S, 5.39; found: C, 60.68; H, 5.14; N, 4.68; S, 5.42.

*2-(α-D-Glucopyranosyl)-5,6-diphenylpyridazine-3(2H)-thione (**17**)*

To a solution of 20 mL dry methanol containing 1 mmol of **16**, ammonium hydroxide solution (3 mL, 35%) was added. Then, the reaction mixture was stirred at room temperature for 3 h, evaporated under reduced pressure at 40°C, and the residue was purified on the silica gel column using chloroform: methanol (4:1) as an eluent to get **17**.

Yield 54%; oil; IR (KBr): $\nu = 3500$–3200 (OH); ^{1}H NMR (DMSO-d_6, ppm): $\delta = 3.20$–3.60 (m, 4H, H-5′, H-4′, H-3′, and H-2′), 4.10 (m, 2H, 2H-6′), 4.52 (m, 1H, HO-3′, D_2O exchangeable), 5.03 (m, 1H, HO-4′, D_2O exchangeable), 5.17 (m, 1H, HO-2′, D_2O exchangeable), 5.34 (m, 1H, HO-6′, D_2O exchangeable), 6.72 (d, $J_{1',2'} = 9.30$ Hz, 1H, H-1′), 7.20–7.35 (m, 10H, Ar-H), 7.77 (s, 1H, C_4-H). Anal. calcd for $C_{22}H_{22}N_2O_5S$ (426.50): C, 61.96; H, 5.20; N, 6.57; S, 7.52; found: C, 62.10; H, 5.12; N, 6.63; S, 7.55.

Preparation of **19**

Compound **19** was prepared from **18** as described for **7** and **8**.

*2-(2-Methoxyethyl)-5,6-diphenylpyridazin-3(2H)-one (**19**).* Yield 32%; oil; IR (KBr): $\nu = 1663$ (CO); ^{1}H NMR ($CDCl_3$, ppm): $\delta = 3.40$ (s, 3H, OCH_3), 3.88 (t, $J = 11.40$ Hz, 2H, CH_2O), 4.47 (t, $J = 11.90$ Hz, 2H, CH_2N), 7.10–7.3 (m, 10H, Ar-H), 7.99 (s, 1H, C_4-H). Anal. calcd for $C_{19}H_{18}N_2O_2$ (306.14): C, 74.49; H, 5.92; N, 9.14; found: C, 74.54; H, 5.99; N, 9.10.

*2-(2,3,4,6-Tetra-O-acetyl-α-D-glucopyranosyl)-5,6-diphenylpyridazin-3(2H)-one (**20**)*

To a solution of 20 mL dry dimethylformamide containing 1 mmol of **19**, sodium hydride (2 mmol) was added. Then, the reaction mixture was stirred at 70°C for 1 h, cooled, and 2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranosyl bromide (1 mmol) was added and stirred at room temperature for 4 h. The reaction mixture was evaporated under reduced pressure, and the residue was purified on the silica gel column using petroleum ether 40–60°C: ethyl acetate (4:1) as an eluent to get **20**.

Yield 75%; oil; IR (KBr): $\nu = 1754$, 1665 (2CO); ^{1}H NMR ($CDCl_3$, ppm): $\delta = 1.95$–2.07 (4s, 12H, $4CH_3CO$), 4.12–4.36 (m, 4H, 2H-6′, 5′-H, H-4′), 5.24 (m, 1H, H-3′), 5.45 (m, 1H, H-2′), 6.0 (d, $J_{1',2'} = 9$ Hz, 1H, H-1′), 7.20–7.35 (m, 10H, Ar-H), 7.9 (s, 1H, C_4-H). Anal. calcd for $C_{30}H_{30}N_2O_{10}$ (578.58): C, 62.28; H, 5.23; N, 4.84; found: C, 62.34; H, 5.20; N, 4.81.

*3-(α-D-Glucopyranosyl)-5,6-diphenylpyridazin-3(2H)-one (**21**)*

To a solution of 20 mL dry methanol containing 1 mmol of **20**, ammonium hydroxide solution (3 mL, 35%) was added, then the reaction mixture was stirred at room temperature for 3 h. The reaction mixture was evaporated under reduced pressure at 40°C, and the residue was purified on the silica gel column using chloroform: methanol (4:1) as an eluent to get **21**.

Yield 54%; oil; IR (KBr): $\nu = 3500$–3200 (OH), 1670 (CO); ^{1}H NMR (DMSO-d_6, ppm):$\delta = 3.25$–3.65 (m, 4H, H-5′, H-4′, H-3′, and H-2′), 4.18 (m, 2H, 2H-6′), 4.50 (m, 1H, HO-3′, D_2O exchangeable), 5.10 (m, 1H, HO-4′, D_2O exchangeable), 5.20 (m, 1H, HO-2′, D_2O exchangeable), 5.35 (m, 1H, HO-6′, D_2O exchangeable), 6.76 (d, $J_{1',2'} = 9.30$ Hz, 1H, H-1′), 7.20–7.40 (m, 10H, Ar-H), 7.70 (s, 1H, C_4-H). Anal. calcd for $C_{22}H_{22}N_2O_6$ (410.43): C, 64.38; H, 5.40; N, 6.83; found: C, 64.32; H, 5.48; N, 6.86.

REFERENCES

[1] Szerkeres, L. G.; Robins, R. K.; Long, R. A. J. Carbohydr Nucleosides Nucleotides 1974, 97, 1.
[2] Bloomer, L. C.; Wotring, L. L.; Townsend, L. B. Cancer Res 1982, 42, 100.
[3] Katz, D. J.; Wise, D. S.; Townsend, L. B. J Med Chem 1982, 25, 813.
[4] Cornia, M.; Casiraghi, G.; Zetta, L. J Org Chem 1991, 56, 5466.
[5] Solomon, M. S.; Hopkins, P. B. Tetrahedron Lett 1991, 32, 5466.
[6] Yokoyama, M.; Tanabe, T.; Toyoshima, A.; Togo, H. Synthesis 1993, 517.
[7] Yokoyama, M.; Toyoshima, A.; Akiba, T.; Togo, H. Chem Lett 1994, 265.
[8] Abdel-Megeid, F. M. E.; Hassan, N. A.; Zahran, M. A.; Rashad, A. E. Sulfur Lett 1998, 21, 269.
[9] Shamroukh, A. H.; Rashad, A. E.; Sayed, H. H. Phosphorus, Sulfur, Silicon, Relat Elem 2005, 180, 2347.
[10] Rashad, A. E.; Ali, M. A. Nucleosides Nucleotides 2006, 25, 17.
[11] Abdel-Motti, F. M.; Shamroukh, A. H.; Sayed, H. H.; Abdel-Megeid, F. M. E. Egypt J Chem 2002, 45, 1123.
[12] Shaban, M. A. E.; Taha, M. A. M.; Nasr, A. Z.; Morgaan, A. E. A. Pharmazie 1995, 50, 784.
[13] Rashed, N.; Shoukry, M.; El-Ashry, E. H. Bull Chem Soc Jpn 1994, 67, 149.
[14] Shaban, M. A. E.; Taha, M. A. M. Bull Chem Soc Jpn 1989, 62, 2701.
[15] El-Kilany, Y.; Rashed, N.; Mackawy, K. Bull Fac Sci Alexandria Uni 1987, 27, 401.
[16] El-Ashry, E. H.; Nassr, M. M. A.; Abdel-Rahman, M. M. A.; Rashed, N.; Mackawy, K. Carbohydr Res 1980, 82, 149.
[17] Schaeffer, H. J.; Beauchamp, L.; de Miranda, P.; G. Elion, G.; Bauer, D. G.; Collins, P. Nature (London) 1978, 272, 583.
[18] Elgemeie, G. E. H.; Mansour, O.; Metwally, N. H. Nucleosides Nucleotides 1999, 18, 113.
[19] Attia, A. M. E.; Ismail, A. A. A. Tetrahedron 2003, 59, 1749.
[20] Zeid, I. F.; Abdel-Rahman, A. A. H.; Abdel-Megeid, A. E. S.; El-Etrawy, A. S. H. Nucleosides Nucleotides 1999, 18, 95.
[21] Farag, R. S.; Shalaby, A. S.; El-Baroty, G. A.; Ibrahim, N. A.; Ali, M. A.; Hassan, E. M. Phytother Res 2004, 18, 30.