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ORIGINAL PAPER

Synthesis of novel seco-acyclo-N-diazolyl-thione nucleosides analogous derived from acetic acid: characterization, complex formation with ions Pb(II), Hg(II) and antibacterial activity

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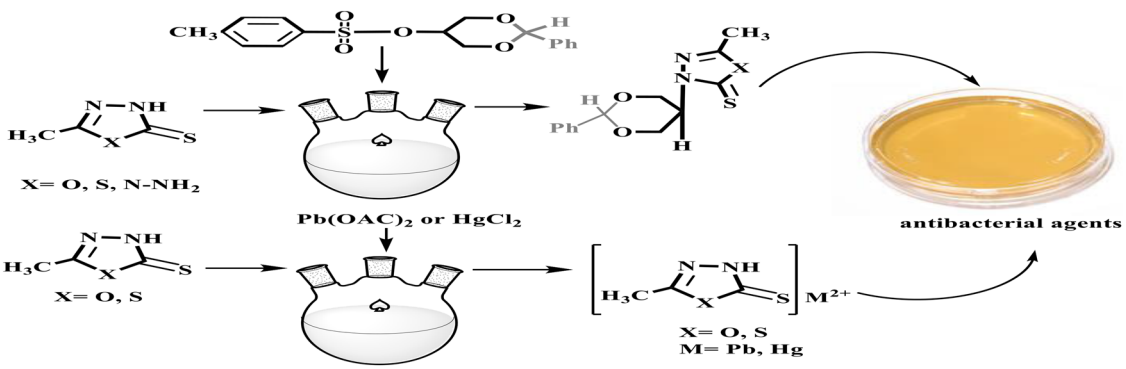
Abstract

Three diazoles namely 5-methyl-1,3,4-oxadiazole -2(3H)-thione, 5-methyl-1,3,4-thiadiazol-2(3H)-thione and 4-amino-5-methyl-2H-1,2,4-triazole-3-thiol were synthesized from acetic acid or ethyl acetate. Seco-acyclo-N-nucleoside analogous was synthesized by condensation of 1,3-benzylidine-glyceryl-2-tosylate with the three diazoles. Structural proof was based upon IR, ^{1}H-NMR, ^{13}C-NMR spectroscopy and MS measurements. The tendency to form complex between 1,3,4-oxadiazole and 1,3,4-thiadiazoles and Pb(II) and Hg(II) ions was achieved, and their structures were assigned by observing some changes in physical properties such as, MP, coloration, R_f (TLC), IR and UV spectroscopy. Most compounds were tested in vitro against Gram-positive and Gram-negative bacteria and showed variable activity. Hg^{2+} complexes of oxadiazole and thiadiazole derivatives exhibited appreciable antibacterial effect at lower MIC, compatible to the reference vancomycin. Similarly, oxadiazole-nucleoside exhibited high effect on Gram-positive bacteria.

Graphic abstract



Keywords 1,3,4-oxadiazole · 1,3,4-thiadiazol · 1,2,4-triazole · Acetic acid · Metallic complexes · Antibacterial activity

Introduction

Interest in acyclic nucleosides started since discovery of acyclovir (A) as a potent anti-herpes drug [1–3]. The unprecedented selectivity of acyclovir as an anti-viral drug and the subsequent clarification of its behavior toward virally coded enzymes instated massive work on acyclovir and its analogues [4–6]. The literature revealed a great number of works published in synthesis

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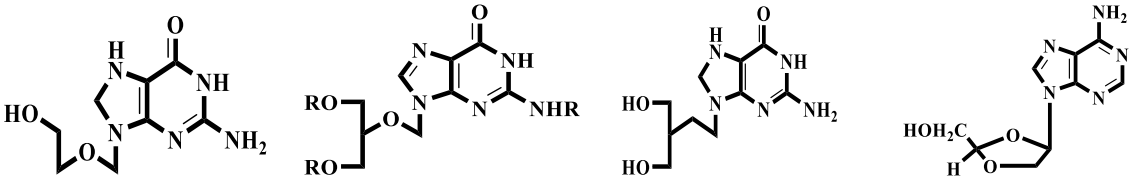
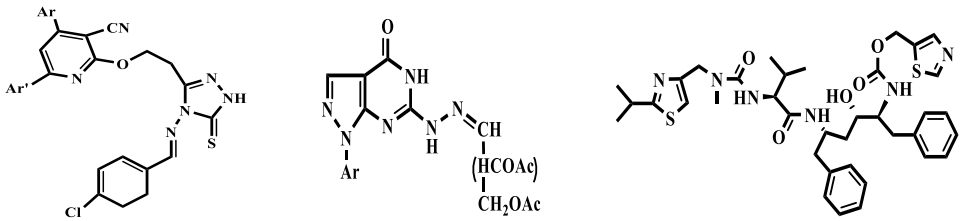
of seco-acyclo nucleosides and glycosides as analogues to acyclovir and its derivatives [7–9].

Several variations to acyclovir basic structure have been reported such as ganciclovir and its derivatives (B) in order to increase their bioavailability, since, like acyclovir, both are poorly absorbed from the alimentary tract and only give rise to low plasma level when they are given by mouth [10–12]. Penciclovir (C) and 2,6-diamino nucleosides similar to DAPD (D) which considered as prodrugs of their corresponding guanine derivatives to which they are converted by enzymic oxidation [13].



(A) R= H or Ac (B) (C) (D)

Many modifications, included replacing of purine with triazole-thione as in (E) [14] and changing the acyclic chains by sugar moiety like glucose in (F), have been exemplified and reviewed [12, 15, 16].



(E) (F) (G)

Some N-acyclo non-glycosides are expected to possess notable pharmacological applications like in ritonavir (G) which is using for treating HIV/hepatitis coinfection and as treatment options for severe acute respiratory syndrome coronavirus 2(SARS-CoV-2), commonly known as COVID-19 [17].

This paper deals with the synthesis of analogues to penciclovir (C) and DAPD (D) by modifying the sugar residue and replacing the guanine heterocyclic by biologically active 5-methyl-1,3,4-oxadiazole-2(3H)-thione, 5-methyl-1,3,4-thiadiazole-2(3H)-thione/thiol and 4-amino-5-methyl-2H-1,2,4-triazole-3-thiol. Moreover, preparation of Pb(II) and Hg(II) complexes with the prepared compounds and testing their bioactivity.

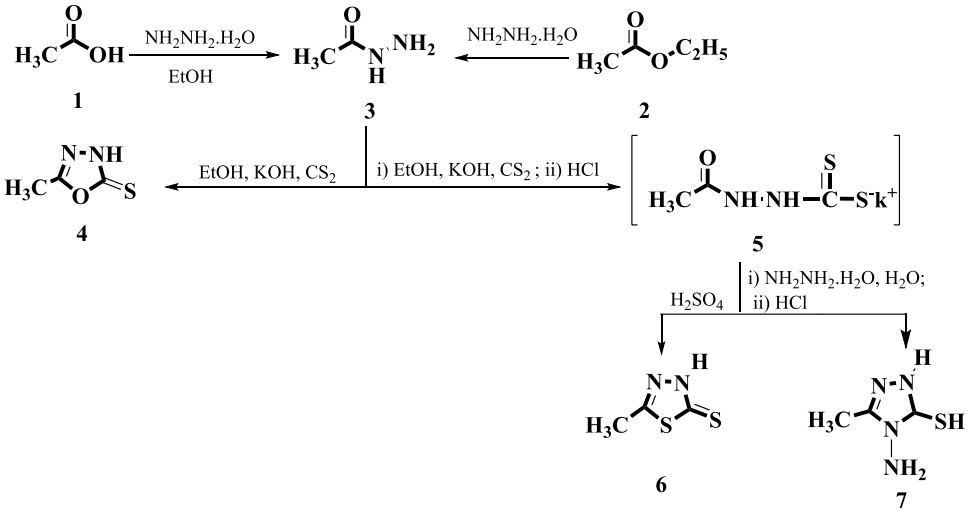
Results and Discussion

Synthesis of 5-methyl-1,3,4-oxadiazole -2(3H)-thione (4), 5-methyl-1,3,4-thiadiazol-2(3H)-thione/thiol (6) and 4-amino-5-methyl-2H-1,2,4-triazole-3-thiol (7)

Nucleobases **4**, **6** and **7** were synthesized by a common route as summarized in Scheme 1.

Acetic acid (**1**) and ethyl acetate (**2**) are both readily available laboratory chemicals, when treated separately with hydrazine hydrate 80%, both resulted acetyl hydrazide (**3**) in corresponding yields 64% and 93%. Oxadiazole (**4**) was obtained in good yield after heating (**3**) with CS_2 in alcoholic KOH at 80 °C for 27 h., followed by addition of HCl. Thiadiazole (**6**) and amino-triazole (7) were obtained after treatment hydrazide (**3**) with CS_2 and alcoholic KOH at 0 °C, then stirred for 2 h at room temperature to yield a solid potassium thionate salt (**5**). The latter without further purification, when treated with H_2SO_4 the result was the thiadiazole (**6**), while by treating the salt with hydrazine hydrate followed by adding HCl, it furnished amino-triazole (**7**). Structural elucidation of **4**, **6** and **7** was proved by IR, ^{1}H-NMR and ^{13}C-NMR (see experimental section).

Scheme 1 Synthetic route of nucleobases **4**, **6** and **7**



Scheme 2 Synthesis of complexes **8–11**

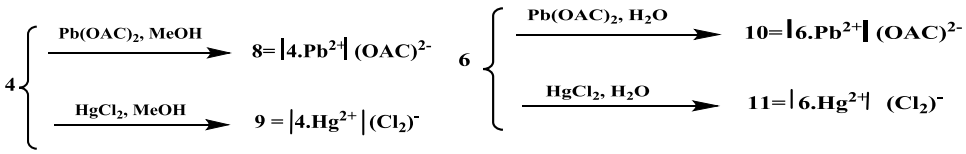


Table 1 Physical properties of ligand **4** and its complexes **8** and **9**

Compound	M.P °C	Color	R_f Acetone / cyclohexane (v/v)	IR: υ (cm^{-1}) C=S	UV: λ_{max} nm (log ε)
4	68	Brown	0.82 (3/2)	1321	205(0.50), 260(1.01)
8	78	Yellow	0.6 (3/2)	1457.92	265(1.35)
9	185	white	0.47(3/2)	1401.03	250(0.31)

Table 2 Physical properties of ligand **6** and its complexes **10** and **11**

Compound	M.P °C	Color	R_f Acetone / cyclohexane (v/v)	IR: υ (cm^{-1}) C=S	UV: λ_{max} nm (log ε)
6	153	White	0.63 (3/2)	1179.26	265(0.56), 310 (2.11)
10	> 300	White	0.59 (3/2)	1196.61	265(1.54)
11	245	White	0.56 (3/2)	1198.54	260(1.18)

Synthesis of Pb(II) and Hg(II) complexes with oxadiazole 4 and thiadiazole 6

Since the diazole-thiones similar to **4** and **6** showed tendency to form complexes with Fe(II), Hg(II) and Pd(II) ions [18–21]. Thus, when alcoholic solution of **4** and **6** was treated with $Pb(OAc)_2$ and $HgCl_2$, (see Scheme 2), gave precipitates with various colors and physical properties as summarized in Scheme 2, Tables 1 and 2.

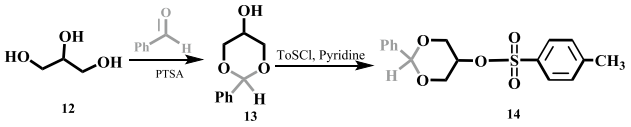
Formation of complexes can be observed by the differences between starting compounds and resulted complexes in: melting points, coloration of mp, Rf's, of TLC, characteristic IR bands of C=S group and UV absorptions, as shown in Tables 1 and 2.

Unfortunately, ^{1}H- and ^{13}C-NMR gave no useful information about the structure of complexes due to the absence of C-H bonds within the areas of complexation.

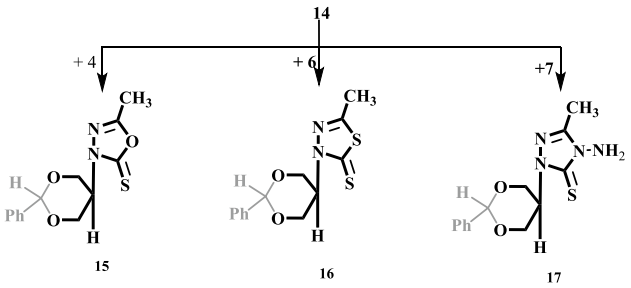
Synthesis of N-nucleosides

Preparation of PTS-glycerol(as sugar analogue)

Glycerol (**12**) as triol resembles the simple acyclic sugar analogues, when treated with benzaldehyde to protect 1, 3-diol groups to give inseparable mixture of stereo isomers of 1,3-O-benzylidenes (**13**) and other traces of 1,2-O-benzylidene isomers [22]. No efforts were applied to separating and identifying that traces since eventually the benzylidene



Scheme 3 Synthesis of 1,3-Benzylidene **13** and PTS-glycerol **14**



Scheme 4 Synthesis of seco-acyclo-N-nucleosides **15–17**

ring will be removed by hydrolysis and end up with symmetrical moiety, 2-O-glyceryl residue (Scheme 3).

Synthesis of seco-acyclo N-Nucleosides 15, 16 and 17

The diazoles **4**, **6** and **7** were coupled smoothly by SN2 reaction with PTS-glycerol **14** to produce seco-acyclo N-Nucleosides **15**, **16** and **17**. Structural proof was assigned by IR, ^{1}H-NMR, ^{13}C-NMR and GC–MS measurements (see experimental section) (Scheme 4).

N1-H exhibited higher nucleophilicity to take part in SN2 substitution reaction as indicated by IR and C^{13} NMR spectra of the products **15–17** by means of existent of C=S bands (IR) and C=S signals (C^{13} NMR), respectively (see experimental section).

Antibacterial evaluation

All synthetic compounds were assayed in vitro using the paper disk diffusion method for their antibacterial and antifungal activities against Gram-positive bacteria, *Staphylococcus aureus*, *Enterococcus faecalis* and Gram-negative bacteria, *Escherichia coli*, *Pseudomonas aeruginosa*. In primary screening, 10 mg mL^{-1} concentration of the synthesized compounds was taken and then, each compound showed an activity was diluted in DMSO for obtaining concentrations: 10/2, 10/4, 10/8, 10/16 mg mL^{-1}, respectively. Vancomycin and colistin were used as positive standards and DMSO as negative standard. The results are tabulated in Table 3.

Compounds **1–3** showed no activity against microorganisms under consideration in concentration 10 mg mL^{-1}.

Heterocyclic compounds **4, 6, 7** exhibited various scales of effect upon microorganisms under consideration. Compounds **4** and **7** showed effect of Gram-positive bacteria *S. aureus* and *E faecalis* only, while **6** showed a negligible effect under experimental conditions.

Hg-metal complexes **9** and **11** showed appreciable effects on Gram-positive bacteria *S. aureus* and Gram-negative bacteria *E. coli*, more than standards vancomycin and colistin.

Among nucleosides, only **15**, which possess oxadiazole ring, showed appreciable effect on Gram-positive bacteria *S. aureus* and *E. faecalis.*

Experimental

General information

All reactions were monitored by TLC, silica gel F254, made by Merck, Germany; iodine was used for visualization. Melting points were determined with a Reichert Thermowar hot plate apparatus and are uncorrected (Universidad de Alicante). IR spectra recorded with a FT-IR 4100LE (JASCO) (PIKE MIRacle ATR) (University of Alicante, Spain) and using KBr disks in a spectrophotometer JASCO FTIR (University of Oran, Es-Senia, Algeria), wavenumbers (υ) are given in cm^{-1}.

UV spectra were recorded by Optizen View 4.2 Spectrometer. ^{1}H NMR and ^{13}C NMR 300 MHz (University of Oran, Es-Senia, Algeria), ^{1}H NMR 300, 400 or 500 MHz and ^{13}C NMR 75 MHz (University of Alicante, Spain), spectra were obtained with a Bruker AC-300 by using TMS as the internal standard. Mass spectra were obtained using a GC–MS Agilent 5973 spectrometer (University of Alicante, Spain), fragment ions in *m/z* in parentheses. The antibacterial tests were carried out in the university hospital of Oran (Algeria). The Mueller Hinton medium was supplied by (Difco).

Synthesis

Acetohydrazide (3) *Method 1*: Ethyl acetate (8.98 g, 102 mmol), hydrazine hydrate 98% (6.76 g, 135 mmol) was added dropwise, and the mixture was refluxed for 6 h. The precipitate was filtered off, washed with hexane and recrystallized from ethanol to give white hygroscopic crystalline **3** (7.01 g, 93%); mp: 58 °C; IR υ (cm^{-1}): 3290.93 (NH, NH_2); 3055.66 (-CH_3); 1662.34 (O=C-N).

Method 2: Acetic acid (4 g, 60 mmol) was refluxed with hydrazine hydrate 80% (3.33 g, 60 mmol) in 10 ml of absolute ethanol for 5 h. The solid obtained was filtered off, washed with cold water, recrystallized from ethanol to give white crystals **3** (3.15 g, 64%); mp. 60 °C.

Table 3 Antibacterial activity of acetic acid 1 and its synthesized derivatives **2–7**, complexes **8–11** and nucleosides **15–17** in mm

Compounds	Gram positive										Gram negative									
	Staphylococcus aureus					*Enterococcus faecalis*					*Escherichia coli*					*Pseudomonas aeruginosa*				
	Inhibition zone					Inhibition zone					Inhibition zone					Inhibition zone				
	a	b	c	d	e	a	b	c	d	e	a	b	c	d	e	a	b	c	d	e
1	–	–	–	–	–	–	–	–	–	–	7	–	–	–	–	–	–	–	–	–
2	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–
3	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–
4	34	26	12	–	–	12	8	–	–	–	–	–	–	–	–	–	–	–	–	–
6	8	–	–	–	–	–	–	–	–	–	–	–	–	–	–	7	–	–	–	–
7	32	18	–	–	–	7	–	–	–	–	–	–	–	–	–	–	–	–	–	–
8	8	–	–	–	–	–	–	–	–	–	9	–	–	–	–	–	–	–	–	–
9	21	18	14	13	–	–	–	–	–	–	24	23	19	18	14	9	8	8	–	–
10	7	–	–	–	–	–	–	–	–	–	9	–	–	–	–	–	–	–	–	–
11	20	18	16	14	14	11	9	8	–	–	21	19	19	18	14	13	–	–	–	–
15	21	14	16	12	8	19	12	10	–	–	7	–	–	–	–	–	–	–	–	–
16	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	12	–	–	–	–
17	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–
*	18	–	–	–	–	17	–	–	–	–	–	–	–	–	–	–	–	–	–	–
**	–	–	–	–	–	–	–	–	–	–	16	–	–	–	–	14	–	–	–	–
DMSO	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–

a,b,c,d,e: concentrations: (10, 10/2, 10/4, 10/8, 10/16) mg/mL.,*respectively*

Key to the inhibition zones activities: (−) for no inhibition or undone, Highly active=Inhibition zone > 15 mm; Moderately active=Inhibition zone 10–15 mm; Slightly active=Inhibition zone 6–8 mm; Inactive=Inhibition zone < 6 mm

*Vancomycin, **Colistin

5-methyl-1,3,4-oxadiazole -2(3H)-thione (4) Acetohydrazide (**3**, 0.32 g, 4.3 mmol) dissolved in ethanol (8 ml) was added to an ethanolic solution of KOH (0.32 g, in ethanol 15 ml). Carbon disulfide (4 ml) was dropped gradually, and the mixture was refluxed for 27 h. The reaction mixture was cooled to room temperature, acidified with concentrated HCl to pH 5–6, filtered off. The solution was extracted several times with ethyl acetate. The combine organic extracts were evaporated and dried. The resulting brown powder was recrystallized from methanol (0.4 g, 80%); mp: 68 °C; IR υ (cm^{-1}): 3436.53 (N–H), 2932.23(C-H), 2506.04 (C-SH), 1629.55 (C=N), 1322.93 (C=S); 1078.01 (C–O–C); ^{1}H NMR (acetone-d_6, σ_H ppm): 12.51 (s, 1H, NH), 1.69 (m, 3H, CH_3); ^{13}C NMR (acetone-d_6, σ_C ppm): 180.68 (C, C=S), 163.22(C, O-C=N), 12.30 (C, CH_3).

5-methyl-1,3,4-thiadiazol-2(3H)-thione/thiol (6) Acetohydrazide **3** (2.66 g, 35 mmol) was added to ethanolic solution of KOH (2.29 g, 40 mmol), and the reaction mixture was cooled in an ice bath. Then, CS_2 (3 g) was added dropwise. After addition, the reaction mixture was stirred for 2 h. The solid obtained **5** was filtered, washed with chilled acetone, dried, and used without further purification for the subsequent reaction.

The above solid was added slowly to concentrate H_2SO_4 (15 ml) with stirring at 0 °C. After addition, the mixture was stirred for further 3 h. The reaction mixture was added to ice cold water, and the solid obtained was filtered and washed with an excess of water till the filtrate became neutral. Then, it was dried and recrystallized from acetone/ water to give (1.66 g, 35%); mp: 153 °C; IR υ (cm^{-1}): 3423.03 (N–H), 2578.36(C-SH), 1632 (C=N), 1179.26(C=S); ^{1}H NMR (DMSO-d_6, σ_H ppm): 12.96 (s, 1H, NH), 9.20 (1H, SH), 1.36 (m, 3H, CH_3); ^{13}C NMR (DMSO, σ_c ppm): 216.03 (C, C=S), 171.82 (C, N=C-S), 20.90 (C, CH_3).

4-amino-5-methyl-2H-1,2,4-triazole-3-thiol (7) A solution of acetohydrazide (2.59 g, 35 mmol) in methanol was treated with a solution of potassium hydroxide (3 g, 53 mmol) in methanol (15 ml) at 0–5 °C with stirring. CS_2 (4 ml) was added slowly, and then, the reaction mixture was stirred overnight at room temperature. The solid product was filtered, washed with diethyl ether, and dried. It was directly used for the next step without further purification.

Hydrazine hydrate (5 ml) was added dropwise to the above solid in water (20 ml) and refluxed for 20 h. The reaction mixture was cooled to room temperature, diluted with water, and acidified with concentrated HCl. The resulting

white precipitate was filtered, washed with cold water, and recrystallized from ethanol/water (2.5 g, 54%), mp: 115 °C; IR υ (cm^{-1}): 3268.19–3098.6 (NH, NH_2), 2933.55 (SH), 1610.28 (C=N); ^{1}H NMR (DMSO-d_6, σ_H ppm): 13.43 (s, H, SH), 5.53 (s, 2H, NH_2), 2.23 (s, 3H, CH_3); ^{13}C NMR (DMSO, σ_C ppm): 165.73 (C, C-SH), 144.67 (C, N–C=N), 10.82 (C, CH_3).

Synthesis of Heterocyclic-Metal Complexes 8–11

Complex 8

Compound **4** (0.3 g, 2.5 mmol) dissolved in methanol (10 ml) was added to a solution of Pb(OAc) (0.4 g, 1.3 mmol) dissolved in the same solvent (10 ml). There was an immediate precipitation of metal complex upon the combination of the two solutions. The resulting yellow powder (0.56 g, 68.5%) was filtered and washed with methanol.

Complex 9

The complex **9** was obtained by treating 0.2 g of (**4**) dissolved in methanol (8 ml) with $HgCl_2$ dissolved in (10 ml) of methanol, molar ratio (2:1). The solution was refluxed for 1 h and half. A white solid was obtained, filtered and washed with methanol to give (0.27 g, 50%).

Complex 10

Compound **6 (**0.17 g**,** 1.2 mmol), dissolved in water (10 ml), was added to a solution of Pb(OAc)(0.24 g, 0.6 mol) in the same solvent. There was an immediate precipitation of metal complex upon the combination of the two solutions. The white powder obtained **10** (0.23 g, 55%) was filtered and washed with water.

Complex 11

Compound **6** (0.15 g, 1 mol), dissolved in water (10 ml), was added to a solution of $HgCl_2$ (0.17 g, 0.5 mmol). The solution was refluxed for 6 h. The white powder complex obtained was filtered and washed with water to give **11** (0.18 g, 57%).

(E)- and (Z)-2-phenyl-[1,3]dioxan-5-ol (13) Glycerol (9 g, 3 mol), benzaldehyde (7 g, 2 mol) in chloroform (25 ml) and ATPS (1.5 g) were mixed and heated under reflux for 6 h, cooled and neutralized by aqueous $NaHCO_3$ to pH=7. The product was extracted two times with dichloromethane, dried over $MgSO_4$, filtered, distilled to dryness to give colorless syrup. (8.5 g, 50%; IR υ (cm^{-1}): 3420.41(OH), 2859.38(CH ar), 1598.71(C=C); ^{1}H NMR ($CDCl_3$, σ_H ppm): 7.69–7.27 (m, 4H, Ar–H), 5.81–5.30 (s, 4H, PhOCH), 4.84–4.15 (m, 2H, 2xCH_2), 3.94–3.40 (m, 1H, CHOH), 2.19 (s, 1H, OH); ^{13}C NMR ($CDCl_3$, $\sigma_{c\ ppm}$): 130.97–127.99(C, CH ar), 105.63–102.71(C, PhOCH), 68.51–64.84 (C, CH_2), 64.58–49.48 (C, CHOH); GC–MS *m/z*: 179.1 (M^+-H^+) for $C_{10}H_{12}0_3$ as in structure **13**.

2-phenyl-1,3-dioxan-5-yl 4-methylbenzenesulfonate (14) 2-Phenyl-[1,3]dioxan-5-ol **13** (2 g, 1.1 mmol) in ethanol, tosyl chloride (2 g, 10 mmol) and some drops of pyridine were mixed and heated under reflux for 6 h, cooled to room temperature and neutralized by aqueous $NaHCO_3$. The product was extracted with dichloromethane, dried over $MgSO_4$, filtered, distilled to dryness to give white syrup **14** (1.42 g, 44.8%); IR υ (cm^{-1}): 1597.81 (C=C), 1351.55 (SO_2), 1094.46(C–O–C); ^{1}H NMR (DMSO-d_6, σ_H ppm): 7.78 (m,4H,Ar–H α to CSO_2O), 7.47(m,2H, Ar–H β to methyl), 7.12 (d,5H, Ar–H), 5.40–5.83 (s, 4H, PhOCH), 4.05 (m,1H, CHOS), 3.29–3.64 (m,4H,2xCH_2 1,3-dioxan ring), 2.41 (s, 3H, CH_3); ^{13}C NMR (DMSO-d_6, σ_C ppm): 145.90–145.29 (C, C- CH_3), 138.22 (C, C^1 phenyl ring), 135.05–133.08 (C, CSO_2O), 130.63- 129.61(2C, CHar β to methyl), 127.95–125.36(C, CHar Phenyl ring), 100.41–104.36(C, PhOCH), 72.93 (C,C^1 1,3-dioxan ring), 63.48–67.96 (2C, CH_2 1.3-dioxan ring), 21.81(C, CH_3); GC–MS *m/z*: 200 ~ fragment of (M-199 for $C_9H_{11}O_3S$ in structure 14).

5-Methyl-3-(2-phenyl-|1,3]-dioxan-5-yl)-3H-1,3,4-oxadiazole-2-thione (15) A solution of **9** (0.21 g, 0.6 mmol) in DMF (2 ml) was added to a solution of (**4**) **(**0.22 g, 1.8 mmol) in DMF (3 ml) and drops of DEA. The mixture was heated under reflux for 48 h. The reaction mixture was cooled, neutralized by water and extracted with dichloromethane. Excess of solvent was evaporated to dryness to give a brown syrup **15** (0.3 g, 43%); IR υ (cm^{-1}): 1634.33(C=N), 1032(C–O–C); ^{1}H NMR ($CDCl_3$, σ_H ppm): 8.02–7.18 (m, 5H, Ar–H), 3.31(m, $2\times CH_{2\ 1,3\text{-dioxan ring}}$), 2.70 (m,1H, HCN), 1.31 (m, 3H, CH_3); ^{13}C NMR ($CDCl_3$, σ_c ppm): 162.63(C, C=S), 140.46 (C, N=C-O oxadiazole ring), 140.29 (C, C^1 phenyl ring), 128.72 (2C, C^3, C^5 phenyl ring), 125.71 (3C, C^2, C^4, C^6 phenyl ring), 42.39 (C, 2xCH_2 1,3-dioxan ring), 36.35 (C, N–C-H), 21.40(C,CH_3); GC–MS *m/z*: 279 (M^+), for $C_{13}H_{14}N_2O_3S$ as in structure **15**.

2-Methyl-5-[2-phenyl-(1,3)-dioxan-5-yl sulfanyl]- (1,3,4) thiadiazole (16) The compound **6** (0.25 g, 1.9 mmol) was dissolved in mixture of DMF and 1,4-dioxane (10 ml), and a solution of **(14)** (0.25 g, 0.74 mmol) in DMF (5 ml) was added. Diethylamine (1 ml) was added, and the reaction mixture was refluxed at for 72 h. The reaction mixture was neutralized with water and extracted with CH_2Cl_2 (3 × 10 ml). The organic layer was evaporated to dryness to give as a brownish syrup **16** (0.44 g, 69%); IR υ (cm^{-1}): 1664.27(C=N), 1178.29(C=S); ^{1}H NMR ($CDCl_3$, σ_H ppm): 7.65 (m,5H,Ar–

H), 5.40 (s,1H,PhOCH), 3.23 (m,2×$CH_{2\ 1.3\text{-dioxan ring}}$), 2.93 (m,1H, HCN), 1.25 (m,3H,CH_3); ^{13}C NMR ($CDCl_3$, σ_c ppm): 207.14 (C,C=S), 162.24 (C, N=C-S thiadiazole ring), 139.43 (C,C^1 phenyl ring), 128.75 (2C, C^3,C^5 phenyl ring), 125.37 (3C,C^2,C^4,C^6 phenyl ring), 100.69 (C, PhCHO), 42.39(C, 2x$CH_2$1,3-dioxan ring), 36.35(C, N–C-H), 21.41 (C,CH_3); GC–MS *m/z*: 295.1 ($M+H^+$), for $C_{13}H_{14}N_2O_2S_2$ as in structure **16**.

4-Amino-5-methyl-2-[(2-phenyl-(1,3)-dioxan-5-yl)-2,4-dihydro-1,2,4-triazol-3-thione (17) Amino triazol **7** (0.3 g, 2.27 mmol) was dissolved in DMF (10 ml), and a compound **14** (0.6 g, 1.79 mmol) in DMF and DEA (1 ml) was mixed at room temperature for 24 h. The reaction mixture was neutralized by water and extracted with dichloromethane. Excess of solvent was evaporated to dryness to give a brown syrup **17** (0.93 g, 78%); IR υ (cm^{-1}): 3107.72 (NH_2), 1663.34(C=N); ^{1}H NMR (DMSO-d_6, σ_H ppm): 7.48 (m, 5H, Ar–H), 5.29 (s, 1H, PhOCH), 3.15 (m,2×$CH_{2\ 1.3\text{-dioxan ring}}$), 2.89 (m, 1H, HCN), 2.69 (s, 2H, NH_2), 1.33 (m,3H,CH_3); ^{13}CNMR (DMSO, σ_c ppm): 170.44 (C, C=S), 154.39 (C, N=C-N triazol ring), 143.29 (C, C^1 Phenyl ring), 133.46 (2C, C^3,C^5 phenyl ring), 130.74 (3C,C^2,C^4,C^6 phenyl ring), 46.58 (C, 2xCH_2 1,3-dioxan ring), 25.92 (C, N–C-H), 15.75 (C,CH_3); GC–MS *m/z*: 292(M^+), for $C_{13}H_{16}O_2N_4S$ as in structure **17**.

Antibacterial test

The respective different strain was spread separately on the Mueller–Hinton for antibacterial activity (CLSI 2004; CLSI 2012). Then, the test organism suspension was added and incubated at 37 ºC for 24 h for bacteria studies. The drugs colistin and vancomycin were taken as standard drug to compare the results, and dimethylsulfoxide (DMSO) was taken as blank [23]. The minimum inhibitory concentration (MIC) was the lowest concentration of test compound that inhibits the visible growth of the organism and was determined in triplicates [24].

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Declarations

Conflict of interest Authors declare there is no conflict of interest.

References

1. H.J. Schaeffer, L. Beauchamp, P. De Miranda, G.B. Elion, D.J. Bauer, P. Collins, Nature **272**, 583–585 (1978)
2. G.B. Elion, P.A. Furman, J.A. Fyfe, P. De Miranda, L. Beauchamp, H.J. Schaeffer, Proc. Natl. Acad. Sci. U S A **74**, 5716–5720 (1977)
3. P.A. Furman, M.H. Clair, J.A. Fyfe, J.L. Rideout, P.M. Keller, G.B. Elion, J. Virol. **32**, 72–77 (1979)
4. C.K. Chu, S.J. Cutler, J. Hetrocycl. Chem. **23**, 289–319 (1986)
5. M. Baba, S. Mori, S. Shigeta, E. De Clercq, Antimicrob. Agents. Chemother. **31**, 337–339 (1987)
6. L. Naesens, R. Snoeck, G. Andrei, J. Balzarini, J. Neyts, E. De Clercq, Antiviral. Chem. Chemother. **8**, 1–23 (1997)
7. T. Agnieszka, K. Ludmiła, S. Joanna, T. Ostrowski, A. Mikołaj, I. Ilgiz, J. Vet. Res. **63**, 513–518 (2019)
8. T. Goslinski, B. Golankiewicz, E. De Clercq, J. Balzarini, J. Med. Chem. **45**, 5052–5057 (2002)
9. M.V. Avcioglu, A. Golcu, Synthesis and reactivity in inorganic, metal-organic, and nano-metal. Chemistry **45**, 581–590 (2015)
10. K.K. Ogilvie, H.R. Hanna, N. Nguyen-ba, K.O. Smith, Nucleosides Nucleotides **4**, 507–513 (1985)
11. N. Ghal, B. Johnston, L. Beauchamp, T. Naseree, T. Scott, R. Flanagan, M. Rodriguez, Nucleosides Nucleotides **14**, 1591–1600 (1995)
12. A.H. Shamroukh, A.E. Rashad, S.T. Gaballah, D.A.A. Osman, A.I. Hashem, F.M.E. Abdel-Megeid, Chem. Res. J. **5**(2020), 89–96 (2020)
13. H.O. Kim, R.F. Schinazi, S. Nampalli, K. Shanmuganathan, D.L. Cannon, A.J. Alves, L.S. Jeong, J.W. Beach, C.K. Chu, J. Med. Chem. **36**, 30–37 (1993)
14. R.M. Abdelhameed, H.A. El-Sayed, M. El-Shahat, A.A. El-Sayed, O.M. Darwe, Curr. Bioact. Compd **14**, 169–179 (2018)
15. D. Wawrzyniak, G. Framski, P. Januszczyk, T. Ostrowski, D. Baraniak, Z.J. Wechmann, J. Fogt, A. Manikowski, D. Baranowski, K. Rolle, J. Boryski, Chem. Select **5**, 13370–13375 (2020)
16. T. Cheviet, S. Wein, G. Bourchenin, M. Lagacherie, C. Périgaud, R. Cerdan, S. Peyrottes, J. Med. Chem **63**, 8069–8087 (2020)
17. D. Ramesh, B.G. Vijayakumar, T. Kannan, ChemMedChem **16**, 1403–1419 (2021)
18. J. Boulerba, A.A. Othman, M.B. Taouti, Chem. Sci. Int. J. **18**, 1–9 (2017)
19. M. Chehrouri, A.A. Othman, S.J. Cecilia, C.M. Cabrerizo, J.M. Sansano, Synth. Commun. **49**(10), 1301–1307 (2019)
20. M. Chehrouri, A.A. Othman, C.M. Cabrerizo, M. Gholinejad, J.M. Sansano, Monatsh. Chem. **151**, 173–180 (2020)
21. M. Chehrouri, A.A. Othman, J.M. Sansano, J. Mar. Chim. Heterocycl. **19**, 51–62 (2020)
22. J. Deutsch, A. Martin, H. Lieske, Jour. Catal. **245**, 428–435 (2007)
23. M. Balouiri, M. Sadiki, S.K. Ibnsouda, J. Pharm. Anal. **6**, 71–79 (2016)
24. M.S. Desai, A.M. Seekatz, N.M. Koropatkin, N. Kamada, C.A. Hickey, M. Wolter, N.A. Pudlo, S. Kitamoto, N. Terrapon, A. Muller, V.B. Young, B. Henrissat, P. Wilmes, T.S. Stappenbeck, G. Núñez, E.C. Martens, Cell **167**, 1339–1353 (2016)