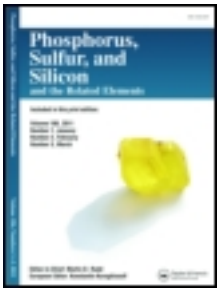

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The Synthesis of Triazolothiadiazines and Thiadiazoles From 1,2-Bis-(4-amino-5-mercapto-1,2,4-triazol-3-yl)- Ethanol and Ethane

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The Synthesis of Triazolothiadiazines and Thiadiazoles From 1,2-Bis-(4-amino-5-mercapto-1,2,4-triazol-3-yl)-Ethanol and Ethane

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*The reaction of DL-malic and succinic acids with thiocarbohydrazide afforded 1,2-bis[4-amino-5-mercapto-1,2,4-triazol-3-yl]-ethane derivatives **3a** and **3b**. The reaction of **3a,b** with phenacyl bromide and benzoin afforded 1,2-bis-1,2,4-triazolo [3,4-b][1,3,4]thiadiazine derivatives **4** and **5**. The carboethoxymethylation of **3a** and **3b** gave **6a** and **6b**, respectively, and their reactions with carbon disulfide and benzoylisothiocyanate gave the 1,2-bis-1,2,4-triazolo[3,4-b][1,3,4]thiadiazole **7** and **9**, and with* p*-nitrobenzaldehyde gave a Schiff's base and dihydrothiadiazole **8**. The structures were confirmed by using ^{1}H and ^{13}C NMR spectra. Selected members of these compounds were screened for antimicrobial activity.*

Keywords 1,2,4-triazole; antimicrobial activity; Malic acid; *seco C*-nucleosides; succinic acid; triazolo[3,4-*b*][1,3,4]thiadiazine; triazolo[3,4-*b*][1,3,4]thiadiazole

INTRODUCTION

The biological activity of acyclic nucleosides has stimulated extensive research for the synthesis of their analogues.[1,2] The synthetic nucleoside ribavirin containing a 1,2,4-triazole ring has a broad spectrum of activity against RNA and DNA viruses,[3] and it has become a unique drug, when combined with the pegylated interferon-α, for

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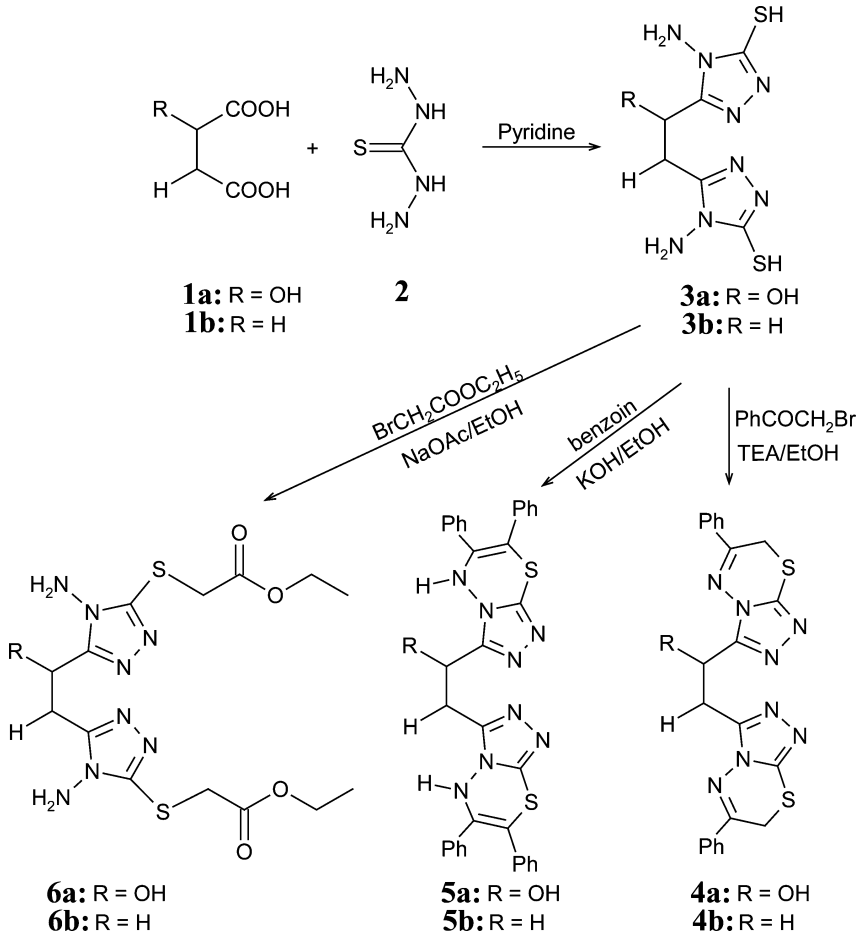
the treatment of hepatitis C virus infections.[4] Consequently, various approaches have been developed for the synthesis of its analogues and for evaluating the biological activities of various 1,2,4-triazoles and their fused ring systems.[5,6] Those functionalized with amino and mercapto groups have found a wide interest as precursors for fused heterocyclic compounds and/or candidates for biological activities.[7] We have been interested in the acyclic nucleosides and their *C*-nucleoside analogues.[2] Recently, the syntheses of the *seco C*-nucleosides 4-amino-3-(D-*gluco*- or D-*galacto*pentitol-1-yl)-5-mercapto-1,2,4-triazoles and the double headed acyclic nucleoside analogues[8–11] have been achieved. Surprisingly, the activity of the D-*gluco* analogue as an inhibitor for glycosidase enzymes[12–14] was found to be less than the simple analogue 4-amino-5-mercapto-3-methyl-1,2,4-triazole.[15] Such results attracted our attention to synthesise the 4-amino-5-mercapto-1,2,4-triazole ring linked to alkyl chains possessing a varied number of hydroxy groups. In a former publication,[16] we prepared 1,2-bis(1,2,4-triazol-3-yl)ethane-1,2-diol. Continuing our work in this project, the synthesis of 1,2-bis(triazolothiadiazinyl and thiadiazolyl)ethane and ethanol analogues were prepared, and their reactivities toward various cyclizing agents have been studied. Selected examples were screened for antimicrobial activity.

INVESTIGATION, RESULTS, AND DISCUSSION

The synthesis of 1,2-bis-(4-amino-5-mercapto-4*H*-1,2,4-triazol-3-yl) ethan-1-ol (**3a**) has been achieved by the dehydrative cyclization of dl-malic acid (**1a**) with thiocarbohydrazide (**2**) in dry pyridine (Scheme 1). Similarly, 1,2-bis-(4-amino-5-mercapto-4*H*-1,2,4-triazol-3-yl)ethane (**3b**) was prepared from succinic acid (**1b**). Since compound **3b** has a difference in m.p. from that reported in the literature,[8] the structure which we obtained was confirmed by IR, ^{1}H NMR, and microanalysis.

The ^{1}H NMR spectrum of **3a** exhibited two multiplets at δ5.26 ppm for CH, 3.28 ppm for CH_2 and a doublet at δ5.96 ppm for the OH group, respectively. This assignment was confirmed by the irradiation of the CH signal at δ 5.26 ppm, whereby the multiplet at δ3.28 ppm became two triplets, and the doublet of the OH became a singlet. Its ^{13}C NMR spectrum showed signals at δ29.4 and 60.8 ppm for CH_2 and CHOH. The ^{1}H NMR spectrum of **3b** showed at δ3.12 ppm a singlet due to the $2CH_2$ groups as a consequence of the symmetry in **3b**.

The amino mercapto triazoles **3a** and **3b** were reacted with phenacyl bromide under basic conditions to afford the triazolothiadiazine derivatives **4a** and **4b**, respectively, in good yields. The ^{1}H NMR spectra of



SCHEME 1

each of **4a** and **4b** showed a singlet in the range δ 4.31–4.33 ppm due to the $2CH_2S$ groups, but the ^{13}C NMR spectra of **4a** showed their carbon signals at δ 28.9 and 30.2 ppm for the $2CH_2S$, whereas those of **4b** appeared as a signal at δ 22.9 ppm for **4b**.

The cycloaddition of compounds **3a** and **3b** with benzoin afforded the triazolothiadiazine derivatives **5a** and **5b**, respectively. The IR spectrum of **5a** showed bands at 3379 and 3415 cm^{-1} for NH and OH stretching frequencies, and the ^1H NMR spectra of **5a** and **5b** showed, in addition to the expected signals, the presence of the aromatic protons in the range δ 7.24–7.97 ppm and 2NH protons at δ8.01 and 8.03 ppm, respectively.

Heating compounds **3a** and **3b** with ethyl bromoacetate under basic conditions gave the carboethoxymethylated **6a** and **6b** without detecting their cyclized derivatives. The absorption bands in their IR spectra showed bands at 1729 and 3325 cm^{-1} indicating the presence of the ester carbonyl and NH_2 groups, respectively. The ^1H NMR spectrum of **6a** showed the presence of the ethyl group at δ 1.17 (t) and 4.13 (q) ppm. Irradiation of the signal at δ1.17 ppm changed the quartet at 4.13 ppm to a singlet, and the irradiation at δ 3.34 ppm changed the signal at 5.21 ppm to a singlet confirming the assigned. The ^{13}C NMR spectrum of **6a** showed signals at δ 13.8, 13.9, 29.1, 61.0, 61.1, and 61.8 ppm for the unsymmetrical $2CH_3$, CH_2, CH, and $2CH_2O$ groups (Scheme 1).

The reaction of **3a** and **3b** with carbon disulfide in the presence of alcoholic KOH gave the corresponding triazolothiadiazole **7a** and **7b**. Each of their ^1H NMR spectra showed a singlet at δ 13.61 and 13.47 ppm for the SH group and compound **7a** showed a doublet at δ 5.95 ppm for the OH group. The irradiation of the signal at δ 5.24 ppm changed the multiplet at δ 3.19–3.32 ppm to a doublet of a doublet, thus confirming the assigned structure. The ^{13}C NMR spectrum of **7a** showed signals at δ 29.5 and 60.8 ppm characteristic for CH_2 and CH groups, respectively.

Treatment of triazoles **3a** with *p*-nitrobenzaldehyde in acetic acid or DMF gave the two geometric isomers of the schiff base **8a**. Its ^1H NMR spectrum showed two signals at δ 10.29 and 10.41 ppm for CH=N of the *syn* and *anti* isomer and a broad signal at δ 13.9 ppm for the two SH groups. The ^1H NMR spectrum of the similar product **8b** from **3b** indicated its existence as dihydrothiadiazole as the major form in addition to the Schiff base as a minor one in the ratio 1:0.25.

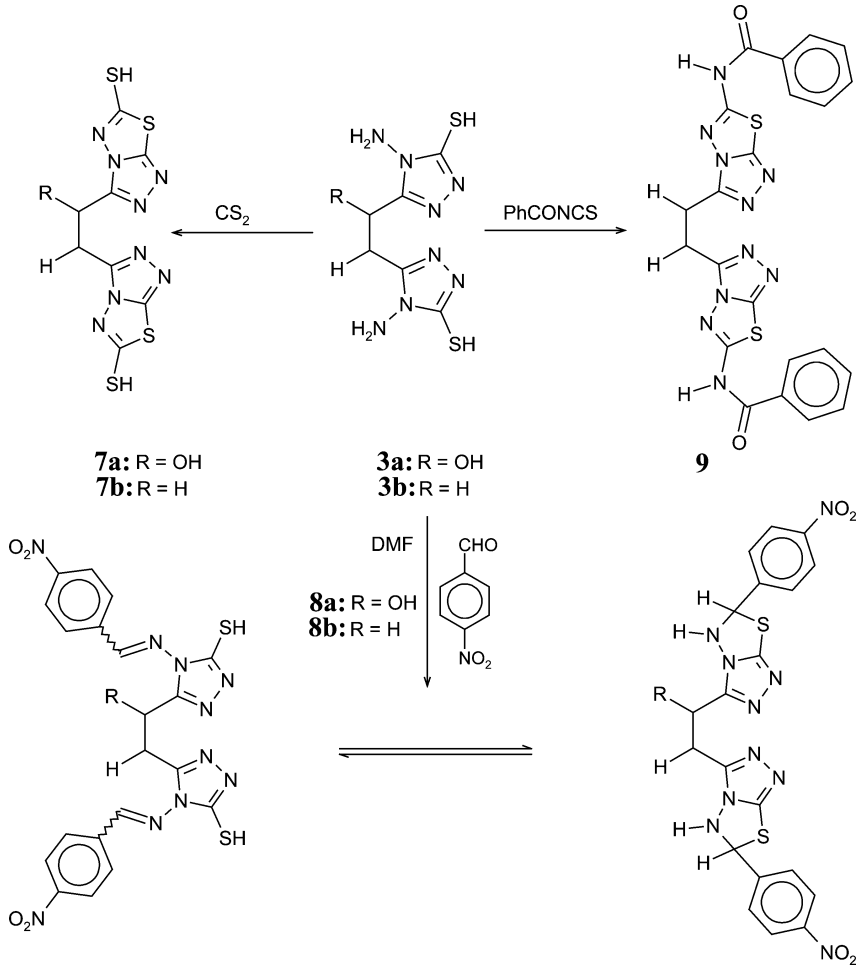
Thus, the spectrum showed signals at δ 5.56, 10.14, 11.27, and 13.49 ppm characteristic for SCHAr, CH=N, NH, and SH groups (Scheme 2).

The reaction of **3b** with benzoylisothiocyanate in DMF afforded triazolothiadiazole **9**. The reaction proceeded via the attack of aminotriazole to the electrophilic carbon of benzoylisothiocyanate to afford the thiourea, which underwent cyclization with the elimination of the H_2S molecule. Its IR spectrum showed a band at 1684 cm^{-1} for the amidic carbonyl group, and its ^1H NMR spectrum showed a singlet at δ 2.96 and 11.8 ppm for the CH_2 and NH group and the disappearance of the SH group (Scheme 2).

The results of the antimicrobial activity of compounds **4a** and **4b** showed interesting degrees of antibacterial activity. Pencillin was used as a reference to evaluate the potency of the tested compounds. Compounds **4a** and **4b** showed higher antibacterial activity than the standard drug (pencillin). Compound **6a** did not show any activity against

TABLE I Antimicrobial Activity of Compounds 4 and 6 (Inhibition Zones mm, Minimum Inhibitory Concentration μg/mL)

Compound No.	*S. aureus*	*S. lutea*	*B. subtilis*	*P. aeruginosa*	*K. peneumonie*	*E. coli*	*C. albicans*
4a	—	25	20	20	15	16	—
4b	10	25	18	20	15	17	—
6a	—	—	—	—	—	—	—
6b	—	—	—	—	—	—	20
Pencillin	15	17	20	13	14	15	—
Nystatin	—	—	—	—	—	—	15



SCHEME 2

the tested microorganisms, while compound **6b** showed higher activity against *C. albicans* than the standard drug (Nystatin) and was inactive against Gram +ve, and Gram −ve bacteria. The results of biological activities encourage further work on such a ring system.

EXPERIMENTAL

Melting points were determined with a Melt-Temp apparatus and are uncorrected. IR spectra were recorded with a Perkin-Elmer model 1600 FTIR spectrometer as KBr discs. ^{1}H and ^{13}C NMR spectra were determined with a JEOL-JNM-LA 400 and 100 MHz Spectrometer. Chemical shifts are expressed on the δ (ppm) scale using TMS as an internal standard. Elemental analyses were determined on a Perkin-Elmer 240 (microanalysis).

1,2-Bis-(4-amino-5-mercapto-4H-1,2,4-triazol-3-yl)-ethane derivatives (3): General Procedure

A mixture of **1a** and **1b** (10 mmol) and **2** (20 mmol) in dry pyridine (10 mL) was heated under reflux for 6 h. The reaction mixture was cooled and poured into cold water. The product was filtered, washed with water, and crystallized from hot water to give colorless crystals.

1,2-Bis-(4-amino-5-mercapto-4H-1,2,4-triazol-3-yl)-ethan-1-ol (3a)

56% yield; m.p. 224–225°C. IR (cm^{-1}): 1633 (C=N), 3166 (NH_2), 3257 (OH). ^{1}H NMR (400 MHz. DMSO-d_6), (δ ppm): δ = 3.28 (m, 2 H, CH_2), 5.26 (t, 1 H, J = 6.1 Hz, CH), 5.52, 5.55 (2 s, 4 H, 2 NH_2), 5.96 (d, 1 H, J = 6.1 Hz, OH), 13.48, 13.61 (2 br s, 2 H, 2 SH). Irradiation at δ 5.26 changed the multiplet at δ 3.20–3.35 to two triplets and the doublet at δ 5.96 to a singlet. ^{13}C NMR (100 MHz. DMSO-d_6), (δ ppm): δ = 29.4 (CH_2), 60.8 (CH), 148.8, 152.1, 165.6, 166.1 (4 C=N). Anal. calcd. for $C_6H_{10}N_8OS_2$ (274.3): C, 26.27; H, 3.67; N, 40.85. Found: C, 26.23; H, 3.53; N, 40.75.

1,2-Bis-(4-amino-5-mercapto-4H-1,2,4-triazol-3-yl)-ethane (3b)

60% yield; m.p. 234–235°C (lit.[8] m.p. 242–244, 220–222°C). IR (cm^{-1}): 1614 (C=N), 3156 (NH_2). ^{1}H NMR (400 MHz. DMSO-d_6), (δ ppm): δ = 3.12 (s, 4 H, 2 CH_2), 5.33 (s, 4 H, 2 NH_2), 13.12 (br s, 2 H, 2 SH). Anal. calcd. for $C_6H_{10}N_8S_2$ (258.3): C, 27.90; H, 3.88; N, 43.38. Found: C, 27.92; H, 3.72; N, 43.30.

1,2-Bis-[6-phenyl-1,2,4-triazolo[3,4-b][1,3,4]thiadiazin-3-yl]-ethane derivatives (4): General Procedure

A solution of phenacyl bromide (20 mmol) in ethanol (10 mL) was added dropwise to a solution of **3a** or **3b** (10 mmol) and three drops of triethylamine in ethanol (10 mL). The mixture was heated under reflux for 6 h and then cooled. The precipitate was crystallized from DMF.

1,2-Bis-[6-phenyl-1,2,4-triazolo[3,4-b][1,3,4]thiadiazin-3-yl]-ethan-1-ol (4a)

80% yield; m.p. 265–266°C. ^{1}H NMR (400 MHz. DMSO-d_6), (δ ppm): δ = 3.65 (d, 2 H, J = 6.7 Hz, CH_2), 4.33 (s, 4 H, 2 CH_2S), 4.35 (d, 1 H, J = 6.1 Hz, OH), 5.59 (t, 1 H, J = 6.7 Hz, CH), 7.49–7.80 (m, 6 H, Ar-H), 8.01 (d, 4 H, J = 7.2 Hz, Ar-H). ^{13}C NMR (100 MHz. DMSO-d_6), (δ ppm): δ = 22.8 (CH_2), 28.9, 30.2 (2 CH_2S), 61.6 (CH), 127.3, 127.4, 128.8, 128.9, 131.7, 131.8, 133.3, 133.4, (Ar-C), 151.0, 151.1, 153.2, 153.3, 154.5, 164.6, (6 C=N). Anal. calcd. for $C_{22}H_{18}N_8OS_2$ n(474.6): C, 55.68; H, 3.82; N, 23.61. Found: C, 55.60; H, 3.79; N, 23.50.

1,2-Bis-[6-phenyl-1,2,4-triazolo[3,4-b][1,3,4]thiadiazin-3-yl]-ethane (4b)

83% yield; m.p. 244–245°C. IR (cm^{-1}): 1620 (C=N). ^{1}H NMR (400 MHz. DMSO-d_6), (δ ppm): δ = 3.41 (s, 4 H, 2 CH_2), 4.31 (s, 4 H, 2 CH_2S), 7.48–7.97 (m, 6 H, Ar-H), 8.17 (d, 4 H, J = 7.2 Hz, Ar-H). ^{13}C NMR (100 MHz. DMSO-d_6), (δ ppm): δ = 21.7, 22.9 (CH_2 and CH_2S), 127.3 128.9, 131.8, 133.3 (Ar-C), 140.3, 152.1, 154.6 (3 C=N). Anal. calcd. for $C_{22}H_{18}N_8S_2$(458.6): C, 57.62; H, 3.96; N, 24.43. Found: C, 57.14; H, 3.98; N, 24.09.

1,2-Bis-[6,7-diphenyl-1,2,4-triazolo[3,4-b][1,3,4]thiadiazin-3-yl]-ethane derivatives (5): General Procedure

A mixture of **3a** or **3b** (10 mmol), benzoin (20 mmol), and 1N aqueous potassium hydroxide (5 mL) in ethanol (10 mL) was boiled under reflux for 4 h. The reaction mixture was cooled and poured into ice-cold water. The precipitate was filtered off, washed with water, and crystallized from ethanol.

1,2-Bis-[6,7-diphenyl-1,2,4-triazolo[3,4-b][1,3,4]thiadiazin-3-yl]-ethan-1-ol (5a)

50% yield; m.p. 274–275°C. IR (cm^{-1}): 3379 (NH), 3415 (OH). ^{1}H NMR (400 MHz. DMSO-d_6), (δ ppm): δ = 3.38 (d, 2 H, J = 6.7 Hz, CH_2), 5.31 (d, 1 H, J = 6.1 Hz, OH), 6.02 (s, 2 H, 2NH), 6.09 (t, 1 H, J = 6.7 Hz,

CH), 7.24–7.59 (m, 12 H, Ar-H), 8.01 (d, 8 H, $J = 7.2$ Hz, Ar-H). Anal. calcd. for $C_{34}H_{26}N_8OS_2$ (626.8): C, 65.15; H, 4.18; N, 17.88. Found: C, 65.02; H, 4.10; N, 17.60.

1,2-Bis-[6,7-diphenyl-1,2,4-triazolo[3,4-b][1,3,4]thiadiazin-3-yl]-ethane (5b)

66% yield; m.p. 259–260°C. IR (cm^{-1}): 3416 (NH). ^{1}H NMR (400 MHz. DMSO-d_6), (δ ppm): $\delta = 3.54$ (s, 4 H, $2CH_2$), 5.95 (br s, 2 H, 2 NH), 7.26–7.97 (m, 20 H, Ar-H). Anal. calcd. for $C_{34}H_{26}N_8S_2$ (610.8): C, 66.86; H, 4.29; N, 18.35. Found: C, 66.80; H, 4.24; N, 18.15.

1,2-Bis-[4-amino-5-carboethoxymethylthio-4H-(1,2,4-triazol-3-yl)]-ethane Derivatives (6): General Procedure

A solution of **3a** or **3b** (10 mmol) in anhydrous ethanol (10 mL) was treated with anhydrous sodium acetate (2 mmol) and ethyl bromoacetate (40 mmol). The reaction mixture was heated under reflux for 8 h and then cooled. The product was filtered, washed with water and ethanol, and then crystallized from ethanol, to give a colorless crystal.

1,2-Bis-[4-amino-5-carboethoxymethylthio-4H-(1,2,4-triazol-3-yl)]-ethan-1-ol (6a)

90% yield; m.p. 150–151°C. IR (cm^{-1}): 1729 (C=O, ester), 3323 (NH_2), 3439 (OH). ^{1}H NMR (400 MHz. DMSO-d_6), (δ ppm): $\delta = 1.17$ (t, 6 H, $J = 2.1$ Hz, 2 CH_3), 3.34 (dd, 2 H, $J = 11.6$, 11.7 Hz, CH_2), 4.00 (2 s, 4 H, 2 CH_2S), 4.08 (d, 1 H, $J = 6.1$ Hz, OH), 4.13 (q, 4 H, $J = 2.1$ Hz, 2 CH_2O), 5.21 (m, 1 H, $J = 6.7$ Hz, CH), 6.96, 6.04 (2 s, 4 H, 2 NH_2). ^{13}C NMR (100 MHz. DMSO-d_6), (δ ppm): $\delta = 13.8$, 13.9 (2 CH_3), 29.1 (CH_2), 32.5, 32.6 (2 CH_2S), 61.0,61.1 (2 CH_2O), 61.8 (CH), 150.4, 151.6, 153.5, 156.8 (4 C=N), 168.3, 168.4 (2 C=O). Anal. calcd. for $C_{14}H_{22}N_8O_5S_2$ (446.5): C, 37.66; H, 4.97; N, 25.09. Found: C, 37.38; H, 4.58; N, 24.89.

1,2-Bis-[4-amino-5-carboethoxymethylthio-4H-(1,2,4-triazol-3-yl)]-ethane (6b)

85% yield; m.p. 175–176°C. IR (cm^{-1}): 1614 (C=N), 1729 (C=O, ester), 3325 (NH_2). ^{1}H NMR (400 MHz. DMSO-d_6), (δ ppm): δ = 1.14 (t, 6 H, $J = 2.1$ Hz, 2 CH_3), 3.35 (s, 4 H, 2 CH_2), 3.98 (s, 4 H, CH_2S), 4.15 (q, 4 H, $J = 2.1$ Hz, 2 CH_2O), 6.08 (s, 4 H, 2 NH_2). Anal. calcd. for $C_{14}H_{22}N_8O_4S_2$ (430.5): C, 39.06; H, 5.15; N, 26.03. Found: C, 38.95; H, 5.01; N, 25.89.

1,2-Bis-[6-mercapto-4H-1,2,4-triazolo[3,4-b][1,3,4]thiadiazol-3-yl]-ethane Derivatives (7): General Procedure

To a stirred solution of potassium hydroxide (15 mmol) in ethanol (20 mL) was added **3a** or **3b** (10 mmol) followed by carbon disulfide

(50 mL). The reaction mixture was refluxed for 24 h, cooled, poured into ice-cold water, and then acidified with acetic acid. The precipitate was collected by filtration and crystallized from aqueous ethanol.

1,2-Bis-[6-mercapto-4H-1,2,4-triazolo[3,4-b][1,3,4]thiadiazol-3-yl]-ethan-1-ol (7a)

60% yield; m.p. 245–246°C. ^{1}H NMR (400 MHz. DMSO-d_6), (δ ppm): $\delta = 3.19$–3.32 (m, 2 H, CH_2), 5.24 (t, 1 H, $J = 6.6$ Hz, CH), 5.95 (d, 1 H, $J = 6.1$ Hz, OH), 13.61 (s, 2 H, 2 SH). ^{13}C NMR (100 MHz. DMSO-d_6), (δ ppm): δ = 29.5 (CH_2), 60.8 (CH), 148.8, 148.9, 152.1, 152.2, 165.6, 166.2 (6 C=N). Anal. calcd. for $C_8H_6N_8OS_4$ (358.5): C, 26.80; H, 1.69; N, 31.26. Found: C, 26.56; H, 1.82; N, 31.52.

1,2-Bis-[6-mercapto-4H-1,2,4-triazolo[3,4-b][1,3,4]thiadiazol-3-yl]-ethane (7b)

59% yield; m.p. 248–249°C. ^{1}H NMR (400 MHz. DMSO-d_6), (δ ppm): δ = 3.06 (s, 4 H, 2 CH_2), 13.47 (s, 2 H, 2 SH). ^{13}C NMR (100 MHz. DMSO-d_6), (δ ppm): $\delta = 20.7$ (CH_2), 150.3, 150.5, 165.9 (3 C=N). Anal. calcd. for $C_8H_6N_8S_4$ (342.5): C, 28.05; H, 1.77; N, 32.72. Found: C, 27.87; H, 1.68; N, 32.82.

The Reaction of p-nitrobenzaldehyde with 3a: (1,2-Bis-[4-p-nitrobenzylideneamino-5-mercapto-1,2,4-triazol-3-yl]-ethan-1-ol)

A mixture of **3a** (10 mmol) and *p*-nitrobenzaldehyde (20 mmol) in DMF (20 mL) was refluxed for 8 h, cooled, and then poured into ice-cold water. The yellow precipitate was crystallized from DMF/ethanol; 75% yield; m.p. 254–256°C. IR (cm^{-1}): 1638 (C=N), 3427 (OH). ^{1}H NMR (400 MHz. DMSO-d_6), (δ ppm): $\delta = 3.46$ (d, 2 H, $J = 6.6$ Hz, CH_2), 5.38 (t, 1 H, $J = 6.7$ Hz, CH), 6.29 (d, 1 H, $J = 6.1$ Hz, OH), 7.90–8.47 (m, 8 H, Ar-H), 10.29, 10.41 (2 s, 2 H, 2 HC=N, *syn* and *anti*), 13.9 (br s, 2 H, 2 SH). Anal. calcd. for $C_{20}H_{16}N_{10}O_5S_2$ (540.5): C, 44.44; H, 2.99; N, 25.91. Found: C, 44.14; H, 2.95; N, 26.12.

The Reaction of p-Nitrobenzaldehyde with 3b: (1,2-Bis-[5,6-dihydro-p-nitrophenyl-1,2,4-triazolo[3,4-b][1,3,4]thiadiazol-3-yl]-ethane)

A mixture of **3b** (10 mmol) and *p*-nitrobenzaldehyde (20 mmol) in DMF (20 mL) was refluxed for 8 h, cooled, and then poured into ice-cold water. The yellow precipitate was crystallized from DMF/ethanol; 85% yield in ratio 1:0.25%; m.p. 258–259°C. IR (cm^{-1}): 1607 (C=N), 3157–3220

(br, NH). ^{1}H NMR (400 MHz. DMSO-d_6), (δ ppm): δ = 3.34 (s, 4 H, 2 CH_2), 5.56 (s, 1.5 H, 2 HCNS), 7.89–8.50 (m, 8 H, Ar-H), 10.14 (s, 0.5 H, 2 HC=N), 11.27 (br s, 1.5 H, 2 NH), 13.49 (s, 0.5 H, 2 SH). Anal. calcd. for $C_{20}H_{16}N_{10}O_4S_2$ (524.5): C, 45.80; H, 3.07; N, 26.71. Found: C, 45.50; H, 3.00; N, 26.50.

1,2-Bis-[6-benzoylamino-1,2,4-triazolo[3,4-b][1,3,4]thiadiazol-3-yl]-ethane (9)

A mixture of benzoyl chloride (10 mmol) and ammonium thiocyanate (10 mmol) in dry DMF (20 mL) was refluxed for 30 min. The mixture was filtered off, and the filtrate was collected. Compound **3b** (10 mmol) in DMF (10 mL) was added to the filtrate, and the mixture was refluxed 4 h; the reaction mixture was concentrated and then cooled. The solid product was filtered and crystallized from DMF to give yellow crystals; 65% yield; m.p. 284–285°C. IR (cm^{-1}): 1684 (C=O), 3163 (NH). ^{1}H NMR (400 MHz. DMSO-d_6), (δ ppm): δ = 2.96 (s, 4 H, 2 CH_2), 7.05–7.98 (m, 6 H, Ar-H), 7.97 (d, 4 H, J = 7.6 Hz, Ar-H), 11.8 (s, 2 H, 2 NH). ^{13}C NMR (100 MHz. DMSO-d_6), (δ ppm): δ = 20.4 (CH_2), 127.8, 128.5, 128.6, 130.7, 132.9, 151.1, 165.5, 167.4 (Ar-C, 3 C=N and C=O). Anal. calcd. for $C_{22}H_{16}N_{10}O_2S_2$ (516.6): C, 51.15; H, 3.12; N, 27.11. Found: C, 50.92; H, 3.00; N, 26.95.

ANTIMICROBIAL ACTIVITY

The antimicrobial activities of some newly synthesized compounds were screened for their antibacterial and antifungal activity against six species of bacteria and one fungi, namely *Stophylococcus aureus, Sarcina lutea*, and *Bacillus subtilis* as Gramm +ve; *Pseudomonas arreuginosa, Klebseilla pneumonie*, and *Eschericia coli* as Gram −ve; and *Candida albicans* as fungi, using a cup plate agar diffusion method.[17] The tested compounds were dissolved in DMF to get a solution of 1 mg/mL concentration. The inhibition zones were measured in mm at the end of an incubation period of 48 h at 37°C. Dimethylformamide showed no inhibition zones. The fungi cultures were maintained on a dextrose agar medium. Penicillin and Nystatin were used as a reference.

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