Synthesis of New 1,2,4-Triazole[3,4-*b*][1,3,4]thiadiazoles Bearing Pyrazole as Potent Antimicrobial Agents

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A new series of 6-(aryl/heteryl)-3-(5-methyl-1-phenyl-1*H*-4-pyrazolyl)[1,2,4]triazolo[3,4-*b*][1,3,4]thiadiazoles (7a—j) has been synthesized by the reaction of 4-amino-5-(5-methyl-1-phenyl-1*H*-4-pyrazolyl)-4*H*-1,2,4-triazol-3-yl-hydrosulfide (6) with $POCl_3$ and the corresponding aryl/heteryl carboxylic acid, in ethanol at reflux temperature for 12 h. All the synthesized compounds were tested for *in vitro* activities against certain strains of bacteria such as *Staphylococcus aureus*, *Bacillus subtilis*, *Escherichia coli* and fungi such as *Aspergillus niger*, *Aspergillus nodulans*, *Alternaria alternate*. Compounds having 4-chlorophenyl (7d), 4-aminophenyl (7f), 4-nitrophenyl (7h) and 3-pyridyl (7i) substituents at 6-position of thiadiazole ring, showed marked inhibition of bacterial and fungal growth nearly equal to the standards. The other new compounds also showed appreciable activity against the test bacteria and fungi.

Key words fused-triazole; antibacterial agent; antifungal agent; anfimicrobial agent

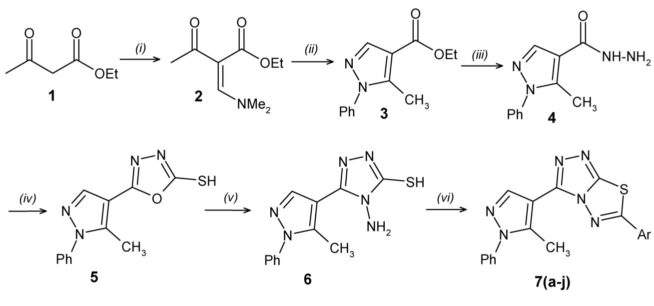
Pyrazole and its derivatives represent one of the most active classes of compounds possessing a wide spectrum of biological activities, including antibacterial,[1] antifungal,[2] herbicidal,[3] insecticidal[4] and other biological activities.[5—7] Similarly, the biological activities of various 1,2,4-triazole derivatives and their *N*-bridged heterocyclic analogs have been widely investigated as antitumor,[8] antiviral,[9] anti-inflammatory,[10] analgesic[11] and antidepressant.[12] 1,2,4-Triazole system is also an important starting material in the synthesis of biologically active heterocycles, which constitute an important class of organic compounds with diverse biological activities, including antiparasitic, analgesic, antibacterial and anti-inflammatory activities.[13—18] Further, triazole fused with other heterocyclic rings is also found to possess diverse applications in the field of medicine.[19—22] The commonly known systems are triazolo-pyridines,[19] triazolo-pyridazines,[20] triazolo-pyrimidines,[21] triazolo-pyrazines,[22] triazolo-triazines[21] and triazolo-thiadiazines.[23] In addition, it has been reported that thiadiazoles exhibit a broad spectrum of biological effectiveness such as anti-parkinsonism,[24] hypoglycaemic,[25] anticancer,[26] anti-inflammatory,[27] anti-asthmatic[28] and anti-hypertensive[29] activities.

Inspired by the biological profile of pyrazole, triazole, thiadiazole, and in continuation of our research on biologically active heterocycles,[30—35] it was thought worthwhile to synthesize some new heterocyclic compounds containing pyrazole, triazole and thiadiazol rings in one molecular frame work. We report herein the synthesis of a new series of 1,2,4-triazole[3,4-*b*][1,3,4]thiadiazoles bearing pyrazole and their antimicrobial activity.

Results and Discussion

Synthesis The starting material, (*E*)-2-acetyl-3-(dimethylamino)-2-propenoate (**2**), was obtained *via* condensation of ethyl acetoacetate (**1**) with *N*,*N*-dimethyldimethoxymethanamine,[36] which on cyclo-condensation with phenyl hydrazine resulted 5-methyl-1-phenyl-1*H*-4-pyrazolecarboxylate (**3**).[36] The 5-methyl-1-phenyl-1*H*-4-pyrazolecarbohydrazide (**4**) was obtained in 72% yield *via* hydrazinolysis of compound **3** with hydrazine hydrate. The hydrazide **4** on reaction with carbon disulfide and potassium hydroxide, in ethanol, followed by acidification gave 5-(5-methyl-1-phenyl-1*H*-4-pyrazolyl)-1,3,4-oxadiazol-2-yl-hydrosulfide (**5**) in 70% yield. Reaction of **5** with hydrazine hydrate under reflux for 6 h resulted 4-amino-5-(5-methyl-1-phenyl-1*H*-4-pyrazolyl)-4*H*-1,2,4-triazol-3-yl-hydrosulfide (**6**) in 69% yield. The final compounds, 6-(aryl/heteryl)-3-(5-methyl-1-phenyl-1*H*-4-pyrazolyl)[1,2,4]triazolo[3,4-*b*][1,3,4]thiadiazole (**7a**—**j**), were synthesized in 66—78% *via* the reaction of **6** with $POCl_3$ and the corresponding aryl/heteryl carboxylic acids, in ethanol at reflux temperature for 12 h (Chart 1). The structures of all the newly synthesized compounds were confirmed by their elemental analyses, electron ionization (EI) mass, IR, ^{1}H- and ^{13}C-NMR spectral data.

In the IR spectra of **7a**—**j**, appearance of bands at 1610 (C=C), 1520 (C=N) cm^{-1} and the absence of NH_2 and SH stretching vibrations provided the evidence of ring closure, involving $–NH_2$ and SH groups. Similarly, the absence of signals for the –SH and $–NH_2$ protons in the ^{1}H-NMR spectra



7: Ar=**a**) phenyl; **b**) 4-methylphenyl; **c**) 4-methoxyphenyl; **d**) 4-chlorophenyl; **e**) 2-chloropheyl; **f**) 4-aminophenyl; **g**) 4-hydroxyphenyl; **h**) 4-nitrophenyl; **i**) 3-pyridyl; **j**) 4-pyridyl.

Reagents and conditions: (i) $Me_2NCH(OMe)_2$; (ii) Ph–NH–NH_2; (iii) NH_2–NH_2H_2O/EtOH, reflux, 72%; (iv) CS_2/KOH/EtOH, reflux, 12 h, 70%; (v) NH_2–NH_2H_2O/EtOH, reflux 6 h, 69%; (vi) Ar-COOH/$POCl_3$/EtOH, reflux 12 h, 66—78%.

Chart 1

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followed by the presence of aromatic protons in the region of δ 7.60 and 7.92 ppm well support the structures. In the ^{13}C-NMR spectra, the prominent signals corresponding to the carbons of [1,2,4]triazolo[3,4-*b*][1,3,4]thiadiazole ring for all the compounds, observed nearly at δ 152.1, 144.3 and 134.9 ppm are proof of further evidence of their structures. In summary all the newly synthesized compounds exhibited satisfactory spectral data consistent with their molecular structures.

Biological Properties. Antibacterial Activity The newly synthesized compounds **7a**—**j** were tested for their *in vitro* antibacterial activity against *Staphylococcus aureus*, *Bacillus subtilis* and *Escherichia coli* by using the agar disc-diffusion method.[37] The results of the preliminary antimicrobial testing of the prepared compounds, the typical broad spectrum of antibacterial drug ciprofloxacin are shown in Table 1.

Among the tested compounds, four compounds showed considerable activity almost equal to the activity of ciprofloxacin. The other compounds were found to be moderate or least effective. In order to get some meaningful results, the structure–activity relationship was carried out. From the bacterial screening results it has been observed that the compounds having 4-chlorophenyl (**7d**), 4-aminophenyl (**7f**), 4-nitrophenyl (**7h**) and 3-pyridyl (**7i**) substituents at 6-position of thiadiazole ring, showed marked inhibition of bacterial growth. Compounds having phenyl (**7a**), 4-methylphenyl (**7b**) and 4-methoxyphenyl (**7c**) substituents at 6-position of thiadiazole ring showed least activity: whereas the compounds having 2-chlorophenyl (**7e**), 4-hydroxyphenyl (**7g**) and 4-pyridyl (**7j**) substituents at 6-position of thiadiazole ring showed moderate effect on the growth of bacteria. The comparison of growth inhibition zone diameter (mm) for the selected compound **7** and standard drug against different bacteria is presented in Fig. 1.

Antifungal Activity All the newly synthesized compounds **7a**—**j** were also screened for their antifungal activity against *Aspergillus niger*, *Aspergillus nodulans* and *Alternaria alternate* by food poison technique.[38] The results of the preliminary antifungal testing of the prepared compounds, the typical broad spectrum of the potent antifungal drug amphotericin B are shown in Table 1. The antifungal activity data reveal that compounds containing 4-chlorophenyl (**7d**), 4-aminophenyl (**7f**) and 3-pyridyl (**7i**) substitutents at 6-position of thiadiazole ring, are showing excellent activity against the test fungi and nearly equal to the standard amphotericin B.

Table 1. Antimicrobial Activity of Compounds (**7a**—**j**)

Compound	Growth inhibition zone diameter (mm)					
	Antibacterial activity			Antifungal activity		
	S. aureus	*B. subtilis*	*E. coli*	*A. niger*	*A. nodulans*	*A. alternata*
7a	12	8	9	6	6	6
7b	9	8	8	—	9	8
7c	16	10	12	11	10	12
7d	20	21	19	18	19	19
7e	15	14	13	14	7	8
7f	23	24	23	21	24	22
7g	12	14	15	13	13	16
7h	23	23	19	20	14	16
7i	22	24	27	18	20	22
7j	18	13	16	16	15	16
Ciprofloxacin	22	22	25	—	—	—
Amphotericin B	—	—	—	20	23	20

The compounds **7a**—**j** and the standards used were of 100 μg/8 mm discs.

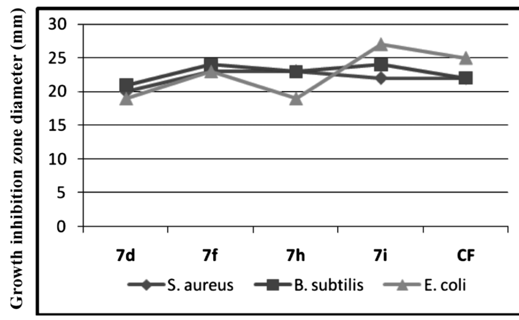

Fig. 1. Comparison of Antibacterial Activity (Growth Inhibition Zone Diameter, mm) of Selected Compounds with Ciprofloxacin (CF)

Conclusion

In conclusion, a new series of 1,2,4-triazole[3,4-*b*][1,3,4]-thiadiazoles bearing pyrazoles **7a**—**j** has been synthesized and evaluated for their antimicrobial activity. Most of the new compounds showed appreciable antimicrobial activity. Among them, the compounds having 4-chlorophenyl (**7d**), 4-aminophenyl (**7f**), 4-nitrophenyl (**7h**) substituents at 6-position of thiadiazole ring showed marked inhibition of bacterial and fungal growth nearly equal to the standards.

Experimental

General All the reagents used were of commercial grade. Reactions were monitored by thin-layer chromatography (TLC) on pre-coated silica gel F_{254} plates obtained from Merck and the compounds visualized either by exposure to UV light or dipping in 1% aqueous potassium permanganate solution. Melting points were determined through a Fisher–Johns apparatus and are uncorrected. IR spectra were recorded using KBr disk on a Perkin–Elmer FTIR spectrometer. The ^{1}H- and ^{13}C-NMR spectra were recorded on a Varian Gemini spectrometer (300 MHz for ^{1}H- and 75 MHz for ^{13}C-NMR). Chemical shifts are reported in δ ppm units with respect to tetramethyl silane (TMS) as internal standard and coupling constants (J) are reported in Hz units. Mass spectra were recorded on a VG micro mass 7070H spectrometer. Elemental analyses (C, H, N), determined by means of a Perkin–Elmer 240 CHN elemental analyzer, were within $\pm$0.4% of theory.

Typical Procedure 5-Methyl-1-phenyl-1*H*-4-pyrazolecarbohydrazide (4) A mixture of compound **3** (5 mmol) and hydrazine hydrate (5 mmol) in ethanol (15 ml) was refluxed for 4 h, cooled to room temperature and filtered. The crude product was recrystallized from ethanol to give new intermediate **4** as white crystal. Yield 72%, mp 162—164 °C. IR (KBr) cm^{-1}: 3269, 3062, 2933, 1660, 1612, 1501. ^{1}H-NMR (CDCl$_3$) δ: 2.61 (s, 3H, CH$_3$), 5.49 (s, 2H, NH$_2$), 7.15—7.25 (m, 5H, Ar-H), 8.26 (s, 1H, Ar-H), 8.96 (s, 1H, NH). ^{13}C-NMR (CDCl$_3$) δ: 15.6, 110.7, 124.9, 127.7, 128.3, 138.5, 139.6, 141.0, 165.1. MS *m/z*: 217 (M^{+}+1). *Anal.* Calcd for $C_{11}H_{12}N_4O$: C, 61.10; H, 5.59; N, 25.91. Found: C, 61.06; H, 5.65; N, 25.95.

Typical Procedure 5-(5-Methyl-1-phenyl-1*H*-4-pyrazolyl)-1,3,4-oxadiazol-2-yl-hydrosulfide (5) A mixture of compound **4** (5 mmol), potassium hydroxide (5 mmol) and carbon disulfide (7.5 mmol), in ethanol (100 ml) was heated under reflux with stirring for 12 h and the solvent was distilled *in vacuo*, the residual mass was poured over crushed ice and neutralized the alkaline solution with 10% hydrochloric acid. The precipitated crude product was filtered, washed with water, dried and recrystallized from ethanol to give pure compound **5**. Yield 70%, mp 136—138 °C. IR (KBr) cm^{-1}: 3162, 2914, 2845, 1612, 1604, 1504, 1343, 1266. ^{1}H-NMR (DMSO-d_6) δ: 2.52 (s, 3H, CH$_3$), 7.15—7.28 (m, 5H, Ar-H), 8.21 (s, 1H, Ar-H). 11.61 (s, 1H, NH/SH). ^{13}C-NMR (DMSO-d_6) δ: 13.7, 124.0, 125.1, 127.5, 128.7, 136.7, 138.5, 139.7, 157.8, 170.0. MS *m/z*: 258 (M^{+}). *Anal.* Calcd for $C_{12}H_{10}N_4O_5$:

C, 55.80; H, 3.90; N, 21.69. Found: C, 55.86; H, 3.82; N, 21.64.

Typical Procedure 4-Amino-5-(5-methyl-1-phenyl-1*H*-4-pyrazolyl)-4*H*-1,2,4-triazol-3-yl-hydrosulfide (6) To a warm solution of compound **5** (5 mmol) in ethanol (50 ml), 80% hydrazine hydrate (7.5 mmol) was added drop wise and the reaction mixture was heated under reflux for 6 h. The solvent was distilled *in vacuo*, cooled and the crystals separated were filtered, washed with cold ethanol and recrystallized from chloroform to give pure compound **6**. Yield 69%, mp 142—143 °C. IR (KBr) cm^{-1}: 3300, 3145, 2849, 2915, 1614, 1510, 1340. ^{1}H-NMR ($CDCl_3$) δ: 1.30 (s, 2H, NH_2), 2.49 (s, 3H, CH_3), 7.15—7.25 (m, 5H, Ar-H), 8.05 (s, 1H, Ar-H), 12.20 (s, 1H, NH/SH). ^{13}C-NMR ($CDCl_3$) δ: 14.7, 119.8, 125.1, 127.9, 129.0, 134.2, 137.6, 138.6, 142.0, 146.9. MS *m/z*: 273 (M^+). *Anal.* Calcd for $C_{12}H_{12}N_6S_2$: C, 52.93; H, 4.44; N, 30.86. Found: C, 52.90; H, 4.50; N, 30.80.

Typical Procedure 6-(Aryl/Heteryl)-3-(5-methyl-1-phenyl-1*H*-4-pyrazolyl)[1,2,4]triazolo[3,4-*b*][1,3,4]thiadiazole (7a—j) To a solution of compound **6** (5 mmol) in $POCl_3$ (10 ml), a solution of aryl/heteryl carboxylic acid (5 mmol) in ethanol (15 ml) was added and the reaction mixture was heated under reflux for 12 h under anhydrous conditions. The solvent was distilled *in vacuo*, the residual mass was poured over crushed ice and the excess $POCl_3$ was neutralized with 10% sodium bicarbonate solution. The solid thus separated was filtered, washed with 10% sodium bicarbonate solution and finally with water, dried and recrystallized from ethanol to give pure compounds.

3-(5-Methyl-1-phenyl-1*H*-4-pyrazolyl)-6-phenyl[1,2,4]triazolo[3,4-*b*][1,3,4]thiadiazole (7a) This was obtained by reacting compound **6** (1.365 g) with benzoic acid (0.61 g) as described in the typical procedure. Yield 66%, mp 137—139 °C. IR (KBr) cm^{-1}: 3035, 2922, 1610, 1520, 1515. ^{1}H-NMR (DMSO-d_6) δ: 2.51 (s, 3H, CH_3), 7.20—7.30 (m, 7H, Ar-H), 7.60 (d, J=7.7 Hz, 1H, Ar-H), 7.92 (d, J=7.6 Hz, 2H, ArH), 8.06 (s, 1H, Ar-H). ^{13}C-NMR (DMSO-d_6) δ: 14.1, 113.0, 124.6, 126.8, 128.0, 129.7, 131.2, 132.0, 133.2, 134.9, 135.8, 137.4, 140.5, 144.3, 152.1. MS *m/z*: 358 (M^+). *Anal.* Calcd for $C_{19}H_{14}N_6S$: C, 63.67; H, 3.94; N, 23.45. Found: C, 63.70; H, 3.88; N, 23.47.

6-(4-Methylphenyl)-3-(5-methyl-1-phenyl-1*H*-4-pyrazolyl)[1,2,4]triazolo[3,4-*b*][1,3,4]thiadiazol (7b) This was obtained by reacting **6** (1.365 g) with 4-methylbenzoic acid (0.68 g) as described in the typical procedure. Yield 68%, mp 126—128 °C. IR (KBr) cm^{-1}: 3032, 2928, 1615, 1520, 1510. ^{1}H-NMR (DMSO-d_6) δ: 2.37 (s, 3H, CH_3), 2.54 (s, 3H, CH_3), 7.20—7.30 (m, 7H, Ar-H), 8.00—8.05 (m, 2H, Ar-H). ^{13}C-NMR (DMSO-d_6) δ: 14.2, 23.1, 113.4, 124.5, 128.0, 129.6, 130.9, 132.9, 133.7, 134.6, 135.5, 137.3, 139.0, 140.1, 144.2, 151.9. MS *m/z*: 373 (M^+). *Anal.* Calcd for $C_{20}H_{16}N_6S$: C, 64.50; H, 4.33; N, 22.56. Found: C, 64.42; H, 4.30; N, 22.52.

6-(4-Methoxyphenyl)-3-(5-methyl-1-phenyl-1*H*-4-pyrazolyl)[1,2,4]triazolo[3,4-*b*][1,3,4]thiadizole (7c) This was obtained by reacting **6** (1.365 g) with 4-methoxybenzoic acid (0.76 g) as described in the typical procedure. Yield 67%, mp 130—132 °C. IR (KBr) cm^{-1}: 3035, 2930, 1612, 1522, 1510, 1070. ^{1}H-NMR (DMSO-d_6) δ: 2.52 (s, 3H, CH_3), 3.67 (s, 3H, OCH_3), 6.82 (d, J=8.4 Hz, 2H, Ar-H), 7.15—7.20 (m, 5H, Ar-H), 7.40 (d, J=8.4 Hz, 2H, Ar-H), 8.05 (s, 1H, Ar-H). ^{13}C-NMR (DMSO-d_6) δ: 14.3, 56.7, 113.4, 116.0, 124.5, 128.1, 128.5, 129.4, 132.9, 134.3, 135.6, 137.2, 140.0, 143.9, 152.1, 157.4. MS *m/z*: 388 (M^+). *Anal.* Calcd for $C_{20}H_{16}N_6OS$: C, 61.84; H, 4.15; N, 21.63. Found: C, 61.80; H, 4.20; N, 21.58.

6-(4-Chlorophenyl)-3-(5-methyl-1-phenyl-1*H*-4-pyrazolyl)[1,2,4]triazolo[3,4-*b*][1,3,4]thiadiazole (7d) This was obtained by reacting **6** (1.365 g) with 4-chlorobenzoid acid (0.775 g) as described in the typical procedure. Yield 70%, mp 141—143 °C. IR (KBr) cm^{-1}: 3062, 2942, 1610, 1515, 1512, 685. ^{1}H-NMR (DMSO-d_6) δ: 2.52 (s, 3H, CH_3), 7.15—7.20 (m, 5H, Ar-H), 7.37 (d, J=8.5 Hz, 2H, Ar-H), 7.80 (d, J=8.5 Hz, 2H, Ar-H), 8.06 (s, 1H, ArH). ^{13}C-NMR (DMSO-d_6) δ: 14.0, 113.1, 124.5, 128.1, 128.9, 129.2, 130.9, 134.0, 134.7, 135.0, 135.4, 137.1, 140.2, 143.8, 152.3. MS *m/z*: 392 (M^+). *Anal.* Calcd for $C_{19}H_{13}ClN_6S$: C, 58.09; H, 3.34; N, 21.39. Found: C, 58.12; H, 3.30; N, 21.44.

6-(2-Chlorophenyl)-3-(5-methyl-1-phenyl-1*H*-4-pyrazolyl)[1,2,4]triazolo[3,4-*b*][1,3,4]thiadiazole (7e) This was obtained by reacting **6** (1.365 g) with 2-chlorobenzoic acid (0.775 g) as described in the typical procedure. Yield 72%, mp 136—138 °C. IR (KBr) cm^{-1}: 3061, 2939, 1611, 1515, 1510, 686. ^{1}H-NMR (DMSO-d_6) δ: 2.53 (s, 3H, CH_3), 7.00—7.10 (m, 2H, Ar-H), 7.20—7.30 (m, 5H, ArH), 7.40—7.50 (m, 2H, ArH), 8.03 (s, 1H, Ar-H). ^{13}C-NMR (DMSO-d_6) δ: 14.5, 113.2, 124.3, 127.3, 128.4, 129.3, 130.4, 132.0, 132.4, 134.1, 134.6, 135.0, 135.5, 137.2, 140.1, 142.7, 152.2. MS *m/z*: 392 (M^+). *Anal.* Calcd for $C_{19}H_{13}ClN_6S$: C, 58.09; H, 3.34; N, 21.39. Found: C, 58.05; H, 3.30; N, 21.33.

4-[3-(5-Methyl-1-phenyl-1*H*-4-pyrazolyl)[1,2,4]triazolo[3,4-*b*][1,3,4]thiadiazol-6-yl]aniline (7f) This was obtained by reacting **6** (1.365 g) with 4-aminobenzoic acid (0.685 g) as described in the typical procedure. Yield 68%, mp 142—144 °C. IR (KBr) cm^{-1}: 3320, 3034, 2928, 1612, 1514, 1510. ^{1}H-NMR (DMSO-d_6) δ: 2.51 (s, 3H, CH_3), 6.63 (d, J=8.4 Hz, 2H, Ar-H), 7.20—7.30 (m, 5H, Ar-H), 7.80 (d, J=8.4 Hz, 2H, Ar-H), 8.04 (s, 1H, Ar-H). ^{13}C-NMR (DMSO-d_6) δ: 14.3, 113.5, 116.2, 124.7, 126.9, 128.0, 128.9, 129.5, 134.5, 135.4, 137.4, 140.3, 144.0, 152.0, 152.7. MS *m/z*: 374 (M^++1). *Anal.* Calcd for $C_{19}H_{15}N_7S$: C, 61.11; H, 4.05; N, 26.26. Found: C, 61.15; H, 4.00; N, 26.27.

4-[3-(5-Methyl-1-phenyl-1*H*-4-pyrazolyl)[1,2,4]triazolo[3,4-*b*][1,3,4]thiadiazol-6-yl]phenol (7g) This was obtained by reacting **6** (1.365 g) with 4-hydroxy acid (0.69 g) as described in the typical procedure. Yield 76%, mp 140—142 °C. IR (KBr) cm^{-1}: 3400, 3030, 2960, 1615, 1520, 1512. ^{1}H-NMR (DMSO-d_6) δ: 2.52 (s, 3H, CH_3), 5.20 (s, 1H, OH), 6.76 (d, J=8.4 Hz, 2H, Ar-H), 7.20—7.30 (m, 7H, Ar-H), 8.04 (s, 1H, Ar-H). ^{13}C-NMR (DMSO-d_6) δ: 14.2, 113.7, 117.0, 124.6, 127.5, 128.3, 129.4, 134.4, 134.9, 135.4, 137.3, 140.1, 144.3, 152.0, 161.1. MS *m/z*: 374 (M^+). *Anal.* Calcd for $C_{19}H_{14}N_6OS$: C, 60.95; H, 3.77; N, 22.43. Found: C, 60.89; H, 3.72; N, 22.41.

3-(5-Methyl-1-phenyl-1*H*-4-pyrazolyl)-6-(4-nitrophenyl)[1,2,4]triazolo[3,4-*b*][1,3,4]thiadiazole (7h) This was obtained by reacting **6** (1.365 g) with 4-nitrobenzoic acid (0.835 g) as described in the typical procedure. Yield 71%, mp 167—169 °C. IR (KBr) cm^{-1}: 3062, 2971, 1620, 1555, 1520, 1510, 1370. ^{1}H-NMR (DMSO-d_6) δ: 2.54 (s, 3H, CH_3), 7.20—7.30 (m, 5H, Ar-H), 8.00—8.10 (m, 3H, Ar-H), 8.27 (d, J=8.4 Hz, 2H, Ar-H). ^{13}C-NMR (DMSO-d_6) δ: 14.3, 113.5, 120.1, 124.3, 124.8, 128.3, 129.4, 134.6, 135.4, 137.2, 140.4, 140.9, 144.4, 149.0, 152.1. MS *m/z*: 403 (M^+). *Anal.* Calcd for $C_{18}H_{13}N_7O_2S$: C, 56.57; H, 3.25; N, 24.30. Found: C, 56.53; H, 3.21; N, 24.26.

3-(5-Methyl-1-phenyl-1*H*-4-pyrazolyl)-6-(3-pyridyl)[1,2,4]triazolo[3,4-*b*][1,3,4]thiadiazole (7i) This was obtained by reacting **6** (1.365 g) with nicotinic acid (0.615 g) as described in the typical procedure. Yield 74%, mp 153—155 °C. IR (KBr) cm^{-1}: 3034, 2961, 1618, 1522, 1520, 1510. ^{1}H-NMR (DMSO-d_6) δ: 2.52 (s, 3H, CH_3), 7.20—7.30 (m, 6H, Ar-H), 7.90 (d, J=5.7 Hz, 2H, Ar-H), 8.05 (s, 1H, Ar-H), 8.86 (d, J=5.7 Hz, 2H, Ar-H). ^{13}C-NMR (DMSO-d_6) δ: 14.3, 113.4, 124.6, 125.9, 128.0, 129.5, 134.5, 135.2, 135.8, 136.5, 137.3, 140.0, 144.3, 150.3, 152.3, 153.1. MS *m/z*: 360 (M^++1). *Anal.* Calcd for $C_{18}H_{13}N_7S$: C, 60.15; H, 3.65; N, 2728. Found: C, 60.10; H, 3.62; N, 27.32.

3-(5-Methyl-1-phenyl-1*H*-4-pyrazolyl)-6-(4-pyridyl)[1,2,4]triazolo[3,4-*b*][1,3,4]thiadiazole (7j) This was obtained by reacting **6** (1.365 g) with isonicotinic acid (0.615 g) as described in the typical procedure. Yield 70%, mp 157—159 °C. IR (KBr) cm^{-1}: 3036, 2961, 1621, 1520, 1515, 1510. ^{1}H-NMR (DMSO-d_6) δ: 2.52 (s, 3H, CH_3), 7.20—7.30 (m, 5H, Ar-H), 7.90 (d, J=6.1 Hz, 2H, Ar-H), 8.04 (s, 1H, Ar-H), 8.87 (d, J=6.1 Hz, 2H, Ar-H). ^{13}C-NMR (DMSO-d_6) δ: 14.3, 113.5, 123.4, 124.5, 128.1, 129.4, 134.5, 135.3, 137.1, 140.1, 141.7, 144.4, 152.4, 154.1. MS *m/z*: 360 (M^++1). *Anal.* Calcd for $C_{18}H_{13}N_7S$: C, 60.15; H, 3.65; N, 27.28 Found: C, 60.20; H, 3.60; N, 27.29.

Antibacterial Assay For the antimicrobial assay standard inoculums were introduced on to the surface of sterile agar plates, and a sterile glass spreader was used for even distribution of the inoculums. The discs measuring 8.0 mm in diameter were prepared from Whattman no. 1 filter paper and sterilized by dry heat at 120 °C for 1 h. The sterile discs previously soaked in a known concentration (100 μg/8 mm disc) of the test compounds were placed in nutrient agar medium. The plates were inverted and incubated for 24 h at 37 °C. The inhibition zones were measured and compared with the standard. The antimicrobial activity data of the test compounds are presented in Table 1.

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