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Synthesis, characterization and biological activity of some 1,3,4-oxadiazole derivatives with benzothiazole moiety

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ABSTRACT

Synthesis of 1,3,4-oxadiazole derivatives has attracted considerable attention in view of therapeutic applications. In the presented research work, a series of novel 5-substituted-1,3,4-oxadiazole-2-thiol containing piperazinyl benzothiazole compounds by integrating piperazinyl benzothiazoles with 5-substituted-1,3,4-oxadiazole-2-thiol. All the synthesized compounds have been characterized by using elemental analysis, FT-IR, ^{1}H NMR, ^{13}C NMR spectroscopy and further supported by mass spectroscopy. Purity of all the compounds has been checked on thin layer chromatographic plate and HPLC technique. All the synthesized compounds were screened for their antibacterial activities against Staphylococcus aureus, Staphylococcus pyogenus, Escherichia coli and Pseudomonas aeruginosa and for antifungal activity against Candida albicans and Asperigillus niger . The biological activities of the synthesized compounds have been compared with standard drugs like Ampicillin and Greseofulvin. The compounds exhibited significant antibacterial and moderate antifungal activities. These compounds can be further exploited to get the potent lead compounds. The detailed synthesis and the antimicrobial screening of the new compounds are reported.

Keywords: 1,3,4-Oxadiazole, Benzothiazole, Piperazine, Antibacterial activity, Antifungal activity.

INTRODUCTION

Oxadiazoles belong to an important group of heterocyclic compounds having –N=C-O- linkage. 1,3,4-oxadiazole is a thermally stable aromatic heterocycle and exist in two partially reduced forms; 2,3-dihydro-1,3,4-oxadiazole(1,3,4-oxadiazoline) and 2,5-dihydro-1,3,4-oxadiazole(1,3,4-oxadiazoline) depending on the position of the double bond. A large number of heterocyclic compounds containing the 1, 3, 4-Oxadiazoles ring are associated with diverse pharmacological properties such as antitubercular [1-3], antimicrobial [4], anti-inflammatory [5-7], anticonvulsant [8, 9], anticancer [10-12], antioxidant [13-15], CNS depressant[16], antihypertensive[17], sedative-hypnotic[18], anti-diabetic[19], anthelminthic [20], analgesic [21], Anti-ulcerogenic activity [22] and genotoxic [23] activities etc.

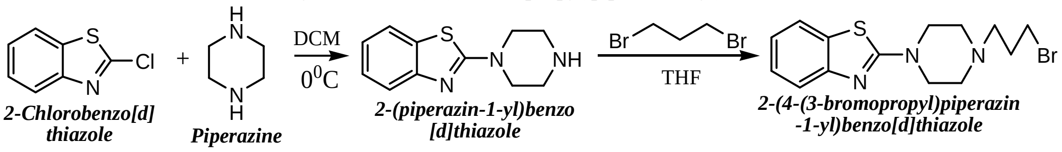
The pharmacological potency of 1,3,4-oxadiazole as well as benzothiazole analogues has drawn our attention to synthesize the compounds containing benzothiazole moiety in a single molecular frame work. As a part of our continuous search for potential bioactive molecules for antimicrobial activity, a series of hybrid compounds were synthesized that comprise the piperazine, benzothiazoles and 1,3,4-oxadiazole heterocyclic ring system in a single molecule. Therefore, this work deals with the synthesis of a series of novel 5-substituted-1,3,4-oxadiazole-2-thiol containing piperazinyl benzothiazole compounds by integrating piperazinyl benzothiazoles with 5-substituted-1,3,4-oxadiazole-2-thiol and screening their antimicrobial activities.

MATERIALS AND METHODS

Melting points were determined in open capillary tubes and are uncorrected. Formation of the compounds was checked by TLC on silica gel-G plates of 0.5 mm thickness and spots were located by iodine and UV light. All compounds were purified by recrystallization with suitable organic solvents. IR spectra were recorded on Brooker-ALPHA FT-IR instrument using KBr pellet method. Mass spectra were recorded on Shimadzu GC-MS-QP-2010 model using direct inlet probe technique. ^{1}H NMR and ^{13}C NMR was determined in $CDCl_3$ solution on a Bruker Ac 400 MHz spectrometer. Purity of the synthesized compounds was checked by HPLC Agilent. The results are in agreements with the structures assigned.

Synthesis of 2-(4-(3-bromopropyl)piperazin-1-yl)benzo[*d*]thiazole:
Activated zinc powder (10 m.mol) is added to a solution of piperazine (10 m.mol), 1,3- dibromopropane (10 m.mol) in THF (10 ml) and stirred at room temperature for 2 h. After completion of the reaction, the mixture is filtered and the solid is washed with solvent ether (30 ml). The combined filtrate was treated with 10 % $NaHCO_3$ (10 ml), water (20 ml), dried (Na_2SO_4) and evaporated. The crude product was purified by column chromatography to yield pure 2-(4-(3-bromopropyl)piperazin-1-yl)benzo[*d*]thiazole. Colourless solid; yield 78 %; M.P: 125-127^{0}C; ^{1}H NMR (300 MHz, $CDCl_3$) δ : 2.08–2.23 (qn, 2H), 2.58–2.80 (m, 6H, 3 × N–CH_2), 3.30–3.51 (m, 4H, 2 × N–CH_2), 3.64 (t, 2H, *J* = 6.8 Hz, Br–CH_2), 7.32 (m, 2H, Ar–H,), 7.61 (m, 2H, Ar–H); ^{13}C NMR (75 MHz, $CDCl_3$) δ : 45.4, 51.5, 117.3, 118.5, 120.3, 123.6, 133.4, 152.9, 169.2; MS (m/z): 340 $(M+1)^+$.

Scheme 1: Synthesis of 2-(4-(3-bromopropyl)piperazin-1-yl)benzo[*d*]thiazole



Synthesis of substituted-1,3,4-oxadiazole-2-thiol: Synthesis of 1,3,4-oxadiazole-2-thiol was achieved by starting with different substituted aromatic carboxylic acids. Substituted aromatic acids (1 m.mol) were first converted into corresponding esters adding catalytic amount of concentrated H_2SO_4 in methanol. The resulting solution is refluxed for 2-4 h, after completion of reaction (confirmed by TLC) evaporate the solvent to dryness. Add water and DCM and shake well. The organic layer is separated and proceeds for the next reaction. To a solution of appropriate ester (1 m.mol) in methanol a solution of hydrazine hydrate (3.0 m.mol) was added, and the mixture is refluxed for about 2-4 h; evaporate the solvent to dryness and recrystallize the solid with ethanol. To the solution of hydrazide derivative in CH_3OH, CS_2 (3.0 m.mol), and KOH (1.5 m.mol) was added. The resulting mixture was refluxed for 4-6 h, after completion of reaction the reaction mixture was quenched by the addition of dil.HCl and then poured into water containing ice, a solid substituted-1,3,4-oxadiazole-2-thiol derivatives were obtained. Recrystallize the crude product in ethanol.

5-(4-methoxyphenyl)-1,3,4-oxadiazole-2-thiol: White solid; M.P: 169-171^{0}C; IR (KBr): 1245 (C=S), 1618 (C=N), 3073 cm^{-1} (=C–H of Ar); ^{1}H NMR (300 MHz, $CDCl_3$) δ : 3.75 (s, 3H, CH_3), 7.59–7.64 (m, 1H, Ar–H), 7.70 (d, 1H, *J* = 7.0 Hz, Ar–H), 7.82 (d, 1H, *J* =7.0 Hz, Ar–H), 7.95 (s, 1H, Ar–H); ^{13}C NMR (75 MHz, $CDCl_3$) δ : 58.2, 113.3, 115.5, 156.4, 160.4, 174.3; MS (m/z): 208 (M^+).

5-(4-nitrophenyl)-1,3,4-oxadiazole-2-thiol: Yellow solid; M.P: 223-225°C; ^{1}H NMR (300 MHz, $CDCl_3$) δ : 7.12–7.20 (m, 2H, Ar–H), 7.95 (d, 1H, *J* = 7.0 Hz, Ar–H), 8.11 (s, 1H, Ar–H); MS (m/z): 223 (M^+).

5-(p-tolyl)-1,3,4-oxadiazole-2-thiol: White solid; M.P: 237-239°C; ^{1}H NMR (300 MHz, $CDCl_3$) δ : 2.65 (s, 3H, Ar–CH_3), 7.27–2.48 (m, 2H, Ar–H), 7.90–7.99 (m, 2H, Ar–H); MS (m/z): 193$(M+1)^+$.

5-(4-chlorophenyl)-1,3,4-oxadiazole-2-thiol: White solid; M.P: 240-242^{0}C; ^{1}H NMR (300 MHz, $CDCl_3$) δ : 7.41 (d, 2H, *J* = 8.6 Hz, Ar–H), 7.92 (d, 2H, *J* = 8.6 Hz, Ar–H); MS (m/z): 213 (M^+).

5-(4-fluorophenyl)-1,3,4-oxadiazole-2-thiol: White solid; M.P: 259-261°C; ^{1}H NMR (300 MHz, $CDCl_3$) δ : 8.17–7.86 (m, 2H, Ar–H); 7.27–6.99 (m, 2H, Ar–H); ESI-MS (m/z): 196 (M^+).

5-(4-bromophenyl)-1,3,4-oxadiazole-2-thiol: Pale yellow solid; mp: 154-156°C; ^{1}H NMR (300 MHz, $CDCl_3$) δ : 7.12–7.20 (m, 2H, Ar–H), 7.95 (d, 1H, *J* = 7.0, Ar–H), 8.11 (s, 1H, Ar–H); MS (m/z): 256 (M^+).

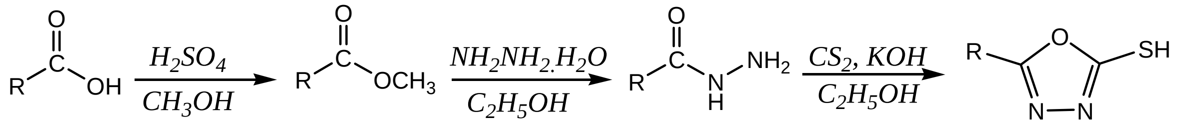
5-(4-(tert-butoxy)phenyl)-1,3,4-oxadiazole-2-thiol: White solid; M.P: 157-159°C; ^{1}H NMR (400 MHz, $CDCl_3$) δ : 1.36 (s, 9H, 3 x CH_3), 7.26–7.32 (m, 2H, Ar–H), 7.71–7.79 (d, 1H, *J* = 5.0 Hz, Ar–H), 8.05 (s, 1H, Ar–H); MS (m/z): 250 (M^+).

4-(5-mercapto-1,3,4-oxadiazol-2-yl)phenol: White solid; M.P: 227-229°C; ^{1}H NMR (400 MHz, $CDCl_3$) δ : 7.14–7.22 (m, 2H, Ar–H), 7.71 – 7.79(d, 1H, *J* = 5.0 Hz, Ar–H), 8.05 (s, 1H, Ar–H); MS (m/z): 194 (M^+).

5-phenyl-1,3,4-oxadiazole-2-thiol: White solid; M.P: 185-187^{0}C; ^{1}H NMR (400 MHz, $CDCl_3$) δ : 7.16–7.20 (m, 1H, Ar–H), 7.28–7.36 (m, 2H, Ar–H), 7.59–7.72 (m, 2H, Ar–H); MS (m/z): 178 (M^+).

N-(4-(5-mercapto-1,3,4-oxadiazol-2-yl)benzyl)benzamide: White solid; M.P: 216-218^{0}C; ^{1}H NMR (300 MHz, $CDCl_3$) δ : 4.73 (d, 2H, *J* = 4.8 Hz, NH–CH_2), 7.20 (t, 1H, *J* = 6.8 Hz, Ar–H), 7.28–7.36 (m, 2H, Ar–H), 7.45–7.52 (m, 2H, Ar–H); MS (m/z): 235 (M^+).

Scheme 2: Synthesis of Substituted-1,3,4-oxadiazole-2-thiols



Synthesis of Substituted phenyl-1,3,4-oxadiazol-2-ylthiopropylpiperazinylbenzo[*d*]-thiazole ((BT-1 to BT-10): A mixture of 5-substituted-1,3,4-oxadiazole-2-thiol (3.0 m.mol) and KF-Al_2O_3 (4.5 m.mol) in dry CH_3CN (15 ml) was stirred for 20 min under N_2 atmosphere. Bromopropylpiperazin-yl-benzo[*d*]thiazole (3.2 m.mol) was added to the above mixture and stirred for about 4 h. After the completion of reaction (confirmed by TLC), the solvent was evaporated and cold water was added to the reaction mixture and stirred for 30 min. Extract the organic compound with ethyl acetate. The ethyl acetate layer is dried over anhydrous sodium sulphate. The compound was purified by column chromatography on silica eluting with ethyl acetate and *n*-hexane.

2-(3-(4-(benzo[d]thiazol-2-yl)piperazin-1-yl)propylthio)-5-(4-methoxyphenyl)-1,3,4-oxadiazole (BT-1): White solid; M.P: 132-134^{0}C; IR (KBr, cm^{-1}): 1462, 1578, 1652, 2817, 2925, 3049; ^{1}H NMR (400 MHz, $CDCl_3$) δ : 2.04–2.12 (qn, 2H, $CH_2CH_2CH_2$), 2.54–2.64 (m, 6H, 3 × N–CH_2), 3.37 (t, 2H, *J* = 6.7 Hz, S–CH_2), 3.63–3.69 (m, 4H, 2 × N–CH_2), 3.87 (s, 3H, –OCH_3), 6.98–7.00 (m, 2H, Ar–H), 7.07 (t, 1H, *J* = 7.7 Hz, Ar–H), 7.29 (t, 1H, *J* = 7.7 Hz, Ar–H), 7.55 (d, 1H, *J* = 7.7 Hz, Ar–H), 7.60 (d, 1H, *J* = 7.7 Hz, Ar–H), 7.94 (d, 2H, *J* = 6.0 Hz, Ar–H); ^{13}C NMR (75 MHz, $CDCl_3$) δ : 26.2, 30.4, 48.2, 52.2, 55.3, 56.4, 114.4, 116.1, 119.0, 120.6, 121.3, 125.3, 125.9, 128.3, 130.6, 152.9, 162.1, 163.6, 165.6, 168.6; MS (m/z): 468 $(M+1)^+$. Anal. calcd for $C_{23}H_{25}N_5O_2S_2$; C, 59.08; H, 5.39; N, 14.98; O, 6.84; S, 13.71.

2-(3-(4-(benzo[d]thiazol-2-yl)piperazin-1-yl)propylthio)-5-(4-nitrophenyl)-1,3,4-oxadiazole (BT-2): Yellow solid; M.P: 144-146^{0}C; ^{1}H NMR (300 MHz, $CDCl_3$) δ : 2.01–2.15 (qn, 2H, $CH_2CH_2CH_2$), 2.52–2.67 (m, 6H, 3 × N–CH_2), 3.44 (t, 2H, *J* = 6.5 Hz, S–CH_2), 3.59–3.71 (m, 4H, 2 × N–CH_2), 7.03–7.11 (m, 1H, Ar–H), 7.24–7.31 (m, 1H, Ar–H), 7.45–7.64 (m, 2H, Ar–H), 8.21 (s, 1H, Ar–H), 8.22 (s, 1H, Ar–H), 8.37 (s, 1H, Ar–H), 8.39 (s, 1H, Ar–H); ^{13}C NMR (75 MHz, $CDCl_3$) δ : 26.2, 30.4, 45.3, 52.1, 55.2, 109.2, 113.6, 115.5, 119.2, 123.4, 124.3, 142.9, 147.8, 161.1, 163.8, 166.7; MS (m/z): 483 $(M+1)^+$. Anal. calcd for $C_{22}H_{22}N_6O_3S_2$; C, 54.75; H, 4.60; N, 17.41; O, 9.95; S, 13.29

2-(3-(4-(benzo[d]thiazol-2-yl)piperazin-1-yl)propylthio)-5-p-tolyl-1,3,4-oxadiazole (BT-3): White solid; M.P: 159-161^{0}C; IR (KBr, cm^{-1}): 1462, 1576, 1640, 2818, 2922; ^{1}H NMR (200 MHz, $CDCl_3$) δ : 2.05–2.13 (qn, 2H, $CH_2CH_2CH_2$), 2.42 (s, 3H, Ph–CH_3), 2.56–2.65 (m, 6H, 3 × N–CH_2), 3.38 (t, 2H, *J* = 7.2 Hz, S–CH_2), 3.63–3.70 (m, 4H, 2 × N–CH_2), 7.07 (t, 1H, *J* = 7.6 Hz, Ar–H), 7.27–7.32 (m, 3H, *J* = 7.5 Hz, Ar–H), 7.55 (d, 1H, *J* = 8.6 Hz, Ar–H), 7.59 (d, 1H, *J* = 7.6 Hz, Ar–H), 7.94 (d, 2H, *J* = 7.6 Hz, Ar–H); ^{13}C NMR (75 MHz, $CDCl_3$) δ : 21.6, 26.3, 30.4, 48.2, 52.3, 56.4, 119.0, 120.6, 121.3, 125.9, 126.5, 129.7, 130.7, 142.1, 152.6, 164.0, 165.8, 168.7; MS (m/z): 451$(M+1)^+$. Anal. calcd for $C_{23}H_{25}N_5OS_2$; C, 61.17; H, 5.58; N, 15.51; O, 3.54; S, 14.20.

2-(3-(4-(benzo[d]thiazol-2-yl)piperazin-1-yl)propylthio)-5-(4-chlorophenyl)-1,3,4-oxadiazole (BT-4): White solid; M.P: 155-157^{0}C; ^{1}H NMR (200 MHz, $CDCl_3$) δ : 2.03–2.13 (qn, 2H, $CH_2CH_2CH_2$), 2.54–2.69 (m, 6H, 3 × N–CH_2), 3.40 (t, 2H, *J* = 6.7 Hz, S–CH_2), 3.62–3.71 (m, 4H, 2 × N–CH_2), 7.05–7.12 (m, 1H, Ar–H), 7.27–7.34 (m, 2H, Ar–H), 7.47 (s, 1H, Ar–H), 7.50 (s, 1H, Ar–H), 7.60 (d, 1H, *J* = 7.5 Hz, Ar–H), 7.93 (s, 1H, Ar–H) 7.96 (s, 1H, Ar–H); ^{13}C NMR (75 MHz, $CDCl_3$) δ : 25.9, 30.1, 47.2, 55.3, 56.5, 108.7, 116.2, 116.5, 119.0, 120.6, 121.4, 126.0, 128.8, 128.9, 130.7, 152.7, 164.5, 166.3, 168.7; MS (m/z): 472 $(M+1)^+$. Anal. calcd for $C_{22}H_{22}ClN_5OS_2$; C, 55.98; H, 4.70; Cl, 7.51; N, 14.84; O, 3.39; S, 13.59

2-(3-(4-(benzo[d]thiazol-2-yl)piperazin-1-yl)propylthio)-5-(4-fluorophenyl)-1,3,4-oxadiazole (BT-5): White solid; M.P: 133-135^{0}C; ^{1}H NMR (300 MHz, $CDCl_3$) δ : 2.06–2.16 (qn, 2H, $CH_2CH_2CH_2$), 2.54–2.64 (m, 6H, 3 × N–CH_2), 3.39 (t, 2H, *J* = 7.2 Hz, S–CH_2), 3.62–3.69 (m, 4H, 2 × N–CH_2), 6.99–7.07 (m, 1H, Ar–H), 7.14–7.30 (m, 3H, Ar–H), 7.55 (d, 1H, *J* = 8.3 Hz, Ar–H), 7.60 (d, 1H, *J* = 8.3 Hz, Ar–H), 7.98–8.06 (m, 2H, Ar–H); ^{13}C NMR (75 MHz, $CDCl_3$) δ : 26.3, 30.4, 48.3, 52.3, 56.4, 116.2, 116.5, 119.0, 120.6, 121.4, 125.9, 128.8, 128.9, 130.7, 152.7, 164.5, 164.8, 166.3, 168.7; MS (m/z): 456 $(M+1)^+$. Anal. calcd for $C_{22}H_{22}FN_5OS_2$; C, 58.00; H, 4.87; F, 4.17; N, 15.37; O, 3.51; S, 14.08. Anal. calcd for $C_{22}H_{22}FN_5OS_2$; C, 58.00; H, 4.87; F, 4.17; N, 15.37; O, 3.51; S, 14.08.

2-((3-(4-(benzo[d]thiazol-2-yl)piperazin-1-yl)propyl)thio)-5-(4-bromophenyl)-1,3,4-oxadiazole (BT-6): White solid; M.P: 143-145^{0}C; ^{1}H NMR (300 MHz, $CDCl_3$) δ : 2.05–2.16 (qn, 2H, $CH_2CH_2CH_2$), 2.53–2.64 (m, 6H, 3 × N–CH_2), 3.51 (t, 2H, *J* = 7.6 Hz, S– CH_2), 3.66–3.75 (m, 4H, 2 × N–CH_2), 6.95–7.03 (m, 1H, Ar–H), 7.12 (t, 1H, *J* = 7.5 Hz, Ar–H), 7.20 (d, 1H, *J* = 8.3 Hz, Ar–H), 7.30 (d, 1H, *J* = 7.5 Hz, Ar–H), 7.44 (s, 1H, Ar–H), 7.56 (s, 1H, Ar–H), 7.72 (s, 1H, Ar–H), 7.78 (s, 1H, Ar–H); ^{13}C NMR (75 MHz, $CDCl_3$) δ : 26.0, 30.9, 45.5, 52.3, 56.5, 108.7, 118.2, 121.6, 123.4, 125.2, 128.6, 131.7, 142.9, 148.6, 161.6, 163.8; MS (m/z): 516 $(M+1)^+$. Anal. calcd for $C_{22}H_{22}BrN_5OS_2$; C, 51.16; H, 4.29; Br, 15.47; N, 13.56; O, 3.10; S, 12.42.

2-(3-(4-(benzo[d]thiazol-2-yl)piperazin-1-yl)propylthio)-5-(4-tert-butoxyphenyl)-1,3,4-oxadiazole (BT-7): White solid; M.P: 135-137^{0}C; ^{1}H NMR (300 MHz, $CDCl_3$) δ : 1.46 (s, 9H, 3 ×CH_3), 2.04–2.17 (qn, 2H, $CH_2CH_2CH_2$), 2.55–2.72 (m, 6H, 3 × N–CH_2), 3.40 (t, 2H, *J* = 7.2 Hz, S–CH_2), 3.64–3.71 (m, 4H, 2 × N–CH_2), 6.90–7.01 (m, 3H, Ar–H), 7.09 (t, 1H, *J* = 7.5 Hz, Ar–H), 7.18 (d, 1H, *J* = 6.7 Hz, Ar–H), 7.32 (d, 1H, *J* = 7.5 Hz, Ar–H), 7.78 (s, 1H, Ar–H) 7.80 (s, 1H, Ar–H); ^{13}C NMR (75 MHz, $CDCl_3$) δ : 26.2, 26.8, 51.3, 52.0, 60.4, 86.0, 114.9, 115.5, 117.7, 118.3, 121.8, 124.5, 125.3, 130.8, 153.2, 157.4, 164.5, 168.0; MS (m/z): 510 $(M+1)^+$. Anal. calcd for $C_{26}H_{31}N_5OS_2$; C, 63.25; H, 6.33; N, 14.19; O, 3.24; S, 12.99

4-(5-(3-(4-(benzo[d]thiazol-2-yl)piperazin-1-yl)propylthio)-1,3,4-oxadiazol-2-yl)phenol (BT-8): White solid; M.P: 155–157^{0}C;^{1}H NMR (400 MHz, $CDCl_3$) δ : 1.99–2.13 (qn, 2H, $CH_2CH_2CH_2$), 2.55–2.64 (m, 6H, 3 × N–CH_2), 2.86 (s, 1H), 3.37 (t, 2H, *J* = 7.5 Hz, S–CH_2), 3.62–3.69 (m, 4H, 2 × N–CH_2), 6.92 (s, 1H, Ar–H), 6.95 (s, 1H, Ar–H), 7.08 (m, 2H, Ar–H), 7.56 (d, 1H, *J* = 7.7Hz, Ar–H), 7.60 (d, 1H, *J* = 8.6 Hz, Ar–H), 7.87 (s, 1H, Ar–H), 7.97 (s, 1H, Ar–H); ^{13}C NMR (75 MHz, $CDCl_3$) δ : 28.0, 34.1, 51.3, 52.0, 60.4, 116.3, 116.4, 118.3, 118.7, 121.8, 124.5, 125.3, 130.8, 153.2, 158.5, 164.5, 168.0; MS (m/z): 454 $(M+1)^+$. Anal. calcd for $C_{22}H_{23}N_5O_2S_2$; C, 58.26; H, 5.11; N, 15.44; O, 7.05; S, 14.14.

2-(3-(4-(benzo[d]thiazol-2-yl)piperazin-1-yl)propylthio)-5-phenyl-1,3,4-oxadiazole (BT-9): White solid; M.P: 121-123^{0}C; ^{1}H NMR (200 MHz, $CDCl_3$) δ : 2.03–2.15 (qn, 2H, $CH_2CH_2CH_2$), 2.54–2.64 (m, 6H, 3 × N–CH_2), 3.40 (t, 2H, *J* = 6.9 Hz, S–CH_2), 3.62–3.70 (m, 4H, 2 × N–CH_2), 7.04–7.12 (m, 2H, Ar–H), 7.46–7.63 (m, 5H, Ar–H), 7.98–8.05 (m, 2H, Ar–H); ^{13}C NMR (75 MHz, $CDCl_3$) δ : 26.3, 30.4, 48.3, 52.3, 56.4, 119.0, 120.6, 121.3, 125.9, 128.9, 131.5, 152.6, 164.4, 165.6, 168.6; MS (m/z): 438 $(M+1)^+$. Anal. calcd for $C_{22}H_{23}N_5OS_2$; C, 60.39; H, 5.30; N, 16.00; O, 3.66; S, 14.66

N-((5-((3-(4-(benzo[d]thiazol-2-yl)piperazin-1-yl)propyl)thio)-1,3,4-oxadiazol-2-yl)methyl)benzamide (BT-10): Pale yellow solid; M.P: 184-186^{0}C; ^{1}H NMR (200 MHz, $CDCl_3$) δ : 2.06– 2.15 (qn, 2H, $CH_2CH_2CH_2$), 2.54–2.69 (m, 6H, 3 × N–CH_2), 3.40 (t, 2H, *J* = 6.7 Hz, S–CH_2), 3.62–3.71 (m, 4H, 2 × N–CH_2), 4.63 (s, 2H, CO–CH_2), 7.05–7.12 (m, 2H, Ar–H), 7.27–7.34 (m, 2H, Ar–H), 7.47 (s, 1H, Ar–H), 7.50 (s, 1H, Ar–H), 7.60 (d, 1H, *J* = 7.5 Hz, Ar–H), 8.23–8.26 (m, 2H, Ar–H); ^{13}C NMR (75 MHz, $CDCl_3$) δ : 26.0, 29.5, 30.3, 34.8, 48.0, 52.2, 56.3, 118.9, 120.6, 121.4, 125.9, 127.1, 128.5, 129.6, 130.6, 132.0, 133.0, 152.5, 164.6, 165.4, 167.4, 168.6; MS (m/z): 494 $(M+1)^+$. Anal. calcd for $C_{24}H_{26}N_6O_2S_2$; C, 58.28; H, 5.30; N, 16.99; O, 6.47; S, 12.97.

Scheme-3: Synthesis of Substituted phenyl-1,3,4-oxadiazol-2-ylthiopropylpiperazinylbenzo[*d*]-thiazole ((BT-1 to BT-10)

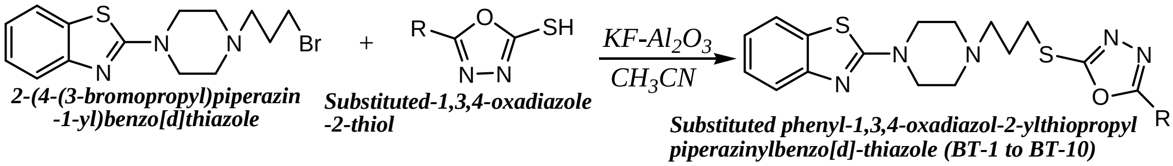
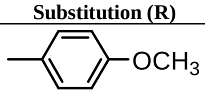
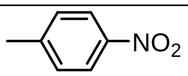
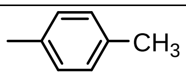
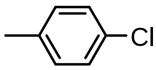
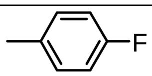
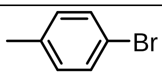
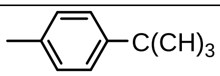
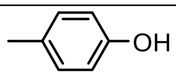
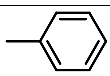
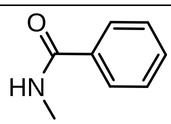


Table 1: Physical constants of Substituted phenyl-1,3,4-oxadiazol-2-ylthiopropylpiperazinylbenzo[*d*]-thiazole ((BT-1 to BT-10)

Compound	Substitution (R)	M.F	M.W	M.P (^{0}C)	Yield (%)
BT-1		$C_{23}H_{25}N_5O_2S_2$	467.6	132-134	69
BT-2		$C_{22}H_{22}N_6O_3S_2$	482.6	144-146	70
BT-3		$C_{23}H_{25}N_5OS_2$	451.6	159-160	85
BT-4		$C_{22}H_{22}ClN_5OS_2$	472	155-157	90
BT-5		$C_{22}H_{22}FN_5OS_2$	455.6	133-135	93
BT-6		$C_{22}H_{22}BrN_5OS_2$	516.5	143-145	82
BT-7		$C_{26}H_{31}N_5OS_2$	493.7	135-137	79
BT-8		$C_{22}H_{23}N_5O_2S_2$	453.6	155-157	74
BT-9		$C_{22}H_{23}N_5OS_2$	437.6	121-123	87
BT-10		$C_{24}H_{26}N_6O_2S_2$	494.6	184-186	80

BIOLOGICAL EVALUATION:

Preparation of Culture Media: Nutrient broth was used as growth medium for bacteria and Saubouraud dextrose broth for fungi. Nutrient broth was prepared by dissolving 13gm of dehydrated powder (HI-media) in 100ml of distilled water. Saubouraud dextrose broth was prepared by dissolving 4gm of dextrose and 1gm of peptone in 100ml of distilled water. The media were sterilized by autoclaving at 15lbs pressure for 20 minutes.

Preparation of Stock Culture: Stock cultures were obtained by aseptically transferring a loopful of test organisms to 100ml of sterile broth and incubated for 24 hours at 37^0C.

Standardization of Stock Culture: Stock cultures were placed in the incubator (37^0C for bacteria and 24^0C for fungi) and shaken well. One ml of stock cultures was aseptically transferred to 9 ml of sterile water containing 0.05% tween 80. This was mixed with using a cyclomixer and serially diluted from 10^{-1} to 10^{-10}. From each dilution, 0.2ml was taken and spread on sterile nutrient agar plates for bacteria and Sabouraud dextrose agar plates for fungi, which were incubated for 18 hours. After incubation, the numbers of colonies in the plate were counted. The number of colonies for a plate that was formed from the maximum dilute tube was noted. The number of microorganisms in stock were then calculated and expressed as colony forming units per ml (cfu/ml). By back calculation the stock culture was found to contain 15 × 10^8 cfu/ml.

Preparation of Working Stock Culture: Stock culture (0.1ml) was diluted with nutrient broth (100ml) and Sabouraud dextrose broth (100ml) respectively to obtain 10^5 cfu/ml. This was then used for further *in vitro* screening.

Preparation of Drug Dilutions: Solutions of the title compounds in DMSO (1mg/ml) were prepared and used for screening their antimicrobial activity.

Antimicrobial Screening: In the search of new antimicrobial agents, all the twelve synthesized compounds were subjected to antimicrobial screening by estimating the minimum inhibitory concentration (MIC) by adopting serial dilution technique. Test was carried out on four bacterial strains, namely *Bacillus subtilis* (MTCC 441), *Staphylococcus aureus* (MTCC 96), *Pseudomonas aeruginosa* (MTCC 741), *Escherichia coli* (MTCC 443) and two fungal strains, namely *Candida albicans* (MTCC 227) and *Aspergilla niger* (MTCC 282).

Table 2: Antimicrobial activity of Substituted phenyl-1,3,4-oxadiazol-2-ylthiopropylpiperazinylbenzo[*d*]-thiazole ((BT-1 to BT-10)

Compound	Minimal Inhibitory Concentration (µg/ml)					
	Antibacterial Activity				Antifungal activity	
	B. subtilis	*S.aureus*	*E.coli*	*P.aeruginosa*	*C.albicans*	*A.niger*
BT-1	250	250	100	100	500	500
BT-2	100	100	62.5	500	200	200
BT-3	62.5	100	100	62.5	500	250
BT-4	250	500	250	250	500	500
BT-5	100	62.5	100	250	100	500
BT-6	500	200	200	100	250	250
BT-7	200	100	62.5	100	500	500
BT-8	50	100	100	200	100	500
BT-9	100	200	200	500	250	500
BT-10	100	250	100	200	500	500
Ampicillin	250	100	100	100	NT	NT
Greseofulvin	NT	NT	NT	NT	500	500

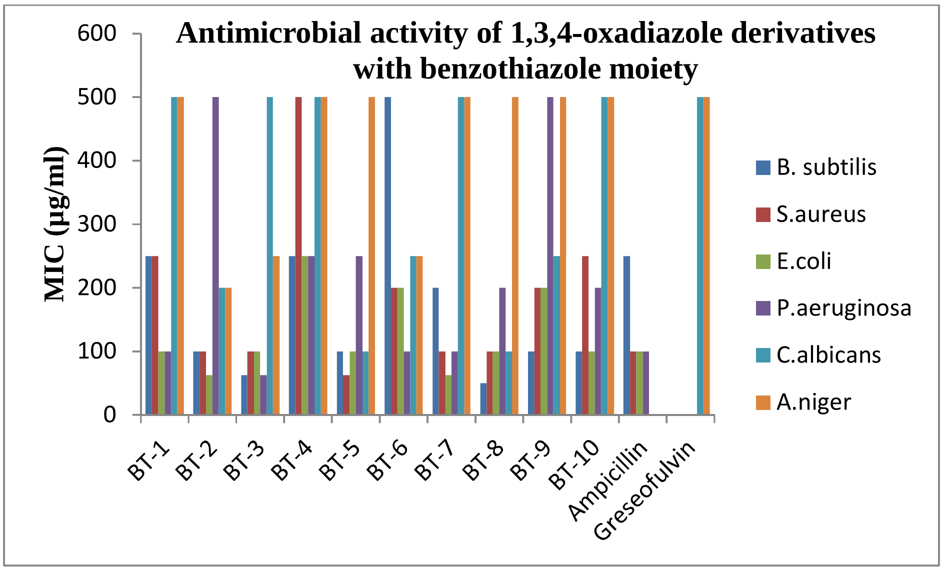


Figure 1: Antimicrobial activity of 1,3,4-oxadiazole derivatives with benzothiazole moiety

Determination of MIC: The study involved a series of six assay tubes for each title compound against each microorganism. The entire test was done in duplicate. To the first assay tube, 1.8ml of seeded broth and 0.2ml of title compound (1mg/ml) was added and mixed thoroughly and the two fold serial dilution was done up to the sixth tube containing 1 ml of seeded broth. The additions of the drug solution and serial dilution were done under strict aseptic conditions. Solvent control, negative control (growth control) and drug control were maintained during the experiment. The assay tubes were incubated at 37^0C and 25^0C respectively for 24 hours for bacteria and fungi. The lowest concentration, which apparently caused complete inhibition of growth of microorganisms, was considered as the minimum inhibitory concentration (MIC). The MIC values of the test compounds are recorded in Table 3.

RESULTS AND DISCUSSION

2-Chlorobenzoxazole with piperazine in the presence of DCM and treated with 1,3-dibromopropane forms 2-(4-(3-bromopropyl)piperazin-1-yl)benzo[*d*]thiazole. Substituted aromatic acids were first converted into corresponding esters. To a solution of appropriate ester in methanol a solution of hydrazine hydrate was added. To the solution of hydrazide derivative in CH_3OH, CS_2, and KOH forms the substituted-1,3,4-oxadiazole-2-thiol derivatives. A mixture of 5-substituted-1,3,4-oxadiazole-2-thiol and KF-Al_2O_3 in dry CH_3CN added to the Bromopropylpiperazin-yl-benzo[*d*]thiazole gives Substituted phenyl-1,3,4-oxadiazol-2-ylthiopropylpiperazinylbenzo[*d*]-thiazole ((BT-1 to BT-10). All the synthesized compounds were subjected to antimicrobial screening by estimating the minimum inhibitory concentration (MIC) by adopting serial dilution technique.

The data recorded in Table 2 indicated that compounds BT-2, BT-3, BT-5, BT-8, BT-9 and BT-10 are more potent towards *Bacillus subtilis.* The compounds BT-1, BT-4 and BT-7 are moderately potent towards the *Bacillus subtilis*. Compound BT-5 is more potent towards *Staphylococcus aureus.* Compounds BT-2, BT-3, BT-7 and BT-8 are moderately potent towards the *Staphylococcus aureus.* Compounds BT-2 and BT-7 more potent towards the *Escherichia coli* and compounds BT-1, BT-3, BT-5, BT-8 and BT-10 are moderately potent towards the *Escherichia coli*. Compound BT-3 is more potent and compounds BT-1, BT-6 and BT-7 are moderately potent towards the *Pseudomonas aeruginosa.* All these compounds are compared with the standard reference (Ampicillin) for their antibacterial activities. Compounds BT-2, BT-5 and BT-8 are more potent and Compounds BT-6 and BT-9 are moderately potent towards the *Candida albicans.* Compounds BT-2, BT-3 and BT-6 are more potent towards the *Aspergilla niger.* All these compounds are compared with the standard reference (Greseofulvin) for their antifungal activities.

CONCLUSION

In conclusion, the synthesis of various Substituted phenyl-1,3,4-oxadiazol-2-ylthiopropylpiperazinylbenzo[*d*]-thiazole ((BT-1 to BT-10) were achieved by reacting the mixture of 5-substituted-1,3,4-oxadiazole-2-thiol and KF-Al_2O_3 in dry CH_3CN added to the Bromopropylpiperazin-yl-benzo[*d*]thiazole. All the ten synthesized 1,3,4-oxadiazole derivatives with benzothiazole moiety were evaluated for their antimicrobial activities. Results revealed that the compounds BT-2, BT-3, BT-7 and BT-8 showed good antibacterial activity while BT-5 and BT-8 showed good antifungal activity. The remaining compounds were shown moderate antimicrobial activity. The study would be a fruitful matrix for the development of 1,3,4-oxadiazole derivatives with benzothiazole moiety for further bio-evaluation.

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