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MEDICINAL CHEMISTRY RESEARCH

ORIGINAL RESEARCH

Synthesis and in vitro antiproliferative activity of 2,5-disubstituted-1,3,4-oxadiazoles containing trifluoromethyl benzenesulfonamide moiety

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Abstract A series of new 2,5-disubstituted-1,3,4-oxadiazoles **6(a–j)** have been conveniently synthesized by intermolecular oxidative cyclization of (*E*)-2-(arylbenzylidene)-2-[(4-methoxyphenyl)amino]acetohydrazides promoted by iodobenzene diacetate as an oxidant. The synthesized compounds were characterized by elemental analyses, FT-IR, LCMS, ^{1}H NMR, and ^{13}C NMR spectral studies. All the compounds were evaluated for their in vitro antiproliferative effect using the 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl tetrazolium bromide assay method against four human cancer cell lines (K562, Colo-205, MDA-MB 231, IMR-32) for the time period of 24 h. Among the analogs, compounds **6h** and **6i** showed good activity on all cell lines, whereas the other compounds in the series exhibited moderate activity.

Keywords 2,5-Disubstituted-1,3,4-oxadiazoles · MTT assay · Antiproliferative activity · Human cancer cell lines

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Introduction

Cancer treatment has been a major endeavor of research and development in academia and pharmaceutical industry for the last many years as it is one of the leading causes of death (Jermal *et al*., 2008). A variety of anticancer drugs are currently in clinical use and some of these drugs can be applied successfully for the treatment of several neoplastic diseases such as leukemia or testicular cancer. Still cancer remains the leading cause of death in developing and developed countries. Many of the available anticancer agents exhibit undesirable side effects such as reduced bioavailability, toxicity, and drug-resistance (Eckhatdt, 2002; Altmann, 2001; O'Dwyer and Druker, 2001). Therefore, the search for novel and selective anticancer agents is urgently required due to problems associated with currently available anticancer drugs.

1,3,4-Oxadiazoles are important class of heterocyclic compounds with a broad range of biological activities such as analgesic (Gilani *et al*., 2010), antimicrobial (Sahin *et al*., 2002), antiviral (Wang *et al*., 2012), anticonvulsant (Bondock *et al*., 2012), antihypertensive (Bankar *et al*., 2009), antiproliferative (Liu *et al*., 2012), enzyme Inhibitors (Gosselin *et al*., 2010), and 5-HT-receptor antagonists (Liao *et al*., 2000). The five-member 1,3,4-oxadiazole heterocycles are also useful intermediates in organic synthesis (Warrener, 2000). 2,5-Disubstituted-1,3,4-oxadiazole derivatives possess broad spectrum of pharmacological activities such as anticancer (Rahman, 2013), antimicrobial, and antiviral (Somani *et al*., 2011).

The chemistry of fluorine-containing compounds has been tremendously developed owing to their unique properties such as high-thermal stability and lipophilicity. Their biological studies clearly indicate that the presence of trifluoromethyl group gives useful biological activity, and the

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subject is of considerable growing interest (Plant, 2004). The increased activity is attributed to the presence of fluorine atoms (highly electronegative) in the molecules which increase the lipophilicity and affects the partitioning of molecules into membranes and facilitates hydrophobic interactions of the molecules with specific binding sites on either receptor or enzymes. Meanwhile, sulfonamide has attracted the increasing attention in the supramolecular chemistry and supramolecular medicinal chemistry (Zhou *et al*., 2009). The sulfonamides constitute an important class of drugs, possessing a host of several types of biological properties including antifungal (Sisik *et al*., 2009), antiviral (Gawin *et al*., 2008), and antitumor (Bouissane *et al*., 2006) in recent years. In connection with such studies, the synthesis and antiproliferative activity of new 2,5-disubstituted-1,3,4-oxadiazoles against human cancer cell lines has been reported in this article.

Experimental

Chemistry

All solvents and reagents were purchased from Merck chemicals, India. Melting range was determined using Veego Melting Point VMP III apparatus. Elemental analyses were recorded on Costech ECS 4010 CHNS-O Elemental Analyzer. The FT-IR spectra were recorded using KBr disks on FT-IR Jasco 4100 infrared spectrophotometer. ^{1}H and ^{13}C NMR spectra were recorded on Bruker DRX-500 spectrometer at 400 MHz using DMSO-d_6 as solvent. The mass spectra of the samples were recorded using the instrument LC-MSD-Trap-XCT.

*Ethyl((4-trifluoromethyl)phenyl)sulfonylglycinate (**3**)*

To a stirred solution of glycine ethylester hydrochloride **1** (0.1 mmol), triethylamine (0.25 mmol) was added in 50 ml of dichloromethane at room temperature. This mixture was stirred for 5 min, and then 4-(trifluoromethyl)benzenesulfonyl chloride **2** (0.1 mmol) was added at this temperature. The reaction mixture was stirred for 1 h at room temperature and monitored by TLC. After completion of the reaction, the reaction mixture was washed with water and then dried with anhydrous sodium sulfate. The solvent was evaporated to dryness, and the pure product was crystallized from petroleum ether. It was obtained as a white solid, yield 82 %; mp 121–123 °C; ^{1}H NMR (DMSO-d_6, 400 MHz): δ = 7.75 (2H, dd, J = 2.16, 6.88, H-3, H-5), 7.12 (2H, dd, J = 2.04, 6.8 Hz, H-2, H-6), 4.59 (2H, s, –CH_2), 4.41 (2H, m, CH_2), 1.16 (3H, t, J = 7.2 Hz, –CH_3); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 170.0 (C, C=O), 147.6 (C, C-1), 134.0 (C, J_{CF} = 270 Hz, C-4), 127.5 (CH, C-2, C-6), 126.2 (CH, J_{CF} = 270 Hz, C-3, C-5), 124.6 (J_{CF} = 270 Hz, CF_3), 63.2 (CH_2, O–CH_2CH_3), 43.4 (NH–CH_2), 14.5 (CH_3); Anal. Calcd. for $C_{11}H_{11}N$: C, 42.58; H, 3.57; N, 4.51. Found: C, 42.71; H, 3.84; N, 4.87.

*N-(2-Hydrazinyl-2-oxoethyl)-4-(trifluoromethyl)benzenesulfonamide (**4**)*

Compound **4** was prepared by refluxing a mixture of ethyl ((4-trifluoromethyl)phenyl)sulfonylglycinate (**3**) (0.1 mmol) and hydrazine hydrate (5 ml) in 20 ml of absolute ethanol for 3 h. The reaction mixture was cooled to room temperature, and the solid obtained was filtered, washed with ethanol. It was obtained as a white solid, yield 80 %; mp 143–145 °C; ^{1}H NMR (DMSO-d_6, 400 MHz): δ = 7.79 (2H, dd, J = 2.12, 6.93, H-3, H-5), 7.10 (2H, dd, J = 2.18, 6.92 Hz, H-2, H-6), 4.60 (2H, s, –CH_2), 4.51 (2H, s, NH_2); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 170.2 (C, C=O), 140.6 (C, C-1). 134.0 (C, J_{CF} = 30 Hz, C-4), 128.4 (C-2, C-6), 125.8 (J_{CF} = 4.8 Hz, C-3, C-5), 124.4 (J_{CF} = 270 Hz, CF_3), 45.4 (NH–CH_2); Anal. Calcd. for $C_9H_{10}N_3$: C, 36.36; H, 3.39; N, 14.14. Found: C, 36.51; H, 3.52; N, 14.32.

*General procedure for the synthesis of (E)-N-(2-(2-benzylidenhydrazinyl)-2-oxoethyl)-4-(trifluoromethyl)benzenesulfonamides (**5a–j**)*

An equimolar mixture of **4** (0.01 mmol) and substituted benzaldehydes was refluxed in ethanol in the presence of few drops of acetic acid for 2 h. The progress of the reaction was monitored by TLC. After completion of the reaction, the reaction mass was cooled, and the solid formed was filtered to give corresponding hydrazones.

*(E)-N-(2-(2-Benzylidenhydrazinyl)-2-oxoethyl)-4-(trifluoromethyl)benzenesulfonamide (**5a**)*

White needles (MeOH) (this compound was prepared by the reaction of compound **4** and benzaldehyde. It was obtained as a white solid); mp 150–151 °C; IR (KBr) ν_{max} 3300, 3150, 3073, 1681, 1631, 1156, 1061 cm^{-1}; ^{1}H NMR (DMSO-d_6, 400 MHz,): δ = 9.35 (1H, s, NH). 8.62 (1H, s, N=CH), 8.14 (2H, d, J = 8.02 Hz, H-2′, H-6′), 8.01 (1H, s, NH), 7.62 (3H, m, H-3, H-5), 7.40 (2H, d, J = 8.53 Hz, H-3′, H-5′), 7.03 (2H, d, J = 8.51 Hz, H-2, H-6), 4.58 (2H, d, –CH_2); ^{13}C NMR (DMSO-d_6, 400 MHz): δ = 170.8 (C, C=O), 145.6 (C, C-1), 144.8 (CH, C=N), 134.2 (C, J_{CF} = 34.2 Hz, C-4), 131.0 (C, C-1′), 129.5 (CH, C-2, C-6), 128.2 (CH, C-2′, C-6′), 124.6 (CH, J_{CF} = 4.2 Hz, C-3, C-5), 124.5 (J_{CF} = 272 Hz, CF_3), 116.2 (CH, C-3′, C-5′), 115.7 (CH, C-4′), 45.4 (NH–CH_2); Anal. Calcd. for $C_{16}H_{14}N_3$: C, 49.87; H, 3.66; N, 10.90. Found: C, 49.33; H, 3.78; N, 10.70.

*(E)-N-(2-(2-(4-Methylbenzylidene)hydrazinyl)-2-oxoethyl)-4-(trifluoromethyl)benzene sulfonamide (**5b**)*

White needles (EtOH) (this compound was prepared by the reaction of compound **4** and 4-methylbenzaldehyde. It was obtained as a white solid); mp 161–163 °C. IR (KBr) ν_{max} 3320, 3155, 3071, 1671, 1638, 1159, 1060 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz): $\delta = 9.28$ (1H, s, NH), 8.51 (1H, s, N=CH), 8.03 (1H, s, NH), 7.85–7.98 (4H, m, H-3, H-5, H-2′, H-6′), 7.23–7.56 (4H, m, H-2, H-6, H-3′, H-5′), 4.59 (2H, s, $–CH_2$), 18 (3H, s, $–CH_3$); ^{13}C NMR (DMSO-d_6, 100 MHz): $\delta = 171.0$ (C, C=O) ppm, 145.6 (C, C-1), 144.7 (CH, C=N), 134.0 (C, $J_{CF} = 33.1$ Hz, C-4), 130.1 (C-1), 129.2 (C-2, C-6), 126.6 (C-2′, C-6′), 124.6 ($J_{CF} = 4.8$ Hz, C-3, C-5), 124.5 ($J_{CF} = 271.0$ Hz, CF_3), 116.2 (C-3′, C-5′), 115.6 (C-4′), 45.3 (NH–CH_2), 20.5 (CH_3, Ar-CH_3); Anal. Calcd. for $C_{17}H_{16}N_3$: C, 51.12; H, 4.04; N, 10.52. Found: C, 51.32; H, 4.21; N, 10.43.

*(E)-N-(2-(2-(4-Methoxybenzylidene)hydrazinyl)-2-oxoethyl)-4-(trifluoromethyl)benzene sulfonamide (**5c**)*

White needles (EtOH) (this compound was prepared by the reaction of compound **4** and 4-methoxybenzaldehyde. It was obtained as a white solid); mp 163–165 °C; IR (KBr) ν_{max} 3312, 3158, 3070, 1680, 1634, 1151, 1064 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz): $\delta = 9.39$ (1H, s, NH), 8.47 (1H, s, N=CH), 8.04 (1H, s, NH), 7.98 (2H, d, H-2′, H-6′), 7.51–7.72 (4H, m, H-3, H-5, H-3′, H-5′), 7.06 (2H, dd, H-2, H-6), 4.58 (2H, s, $–CH_2$), 3.88 (3H, s, $–OCH_3$); ^{13}C NMR (DMSO-d_6, 100 MHz): $\delta = 171.1$ (C, C=O), 160.3 (C, C-4′), 149.8 (C, C-1), 144.2 (CH, C=N), 134.2 ($J_{CF} = 32.0$ Hz, C-4), 130.5 (CH, C-2′, C-6′), 129.6 (CH, C-2, C-6), 127.2 (C, C-1′), 126.1 (CH, $J_{CF} = 34.0$ Hz, C-3, C-5), 124.4 ($J_{CF} = 274.2$ Hz, CF_3), 116.4 (CH, C-3′, C-5′), 55.7 (O–CH_3), 45.7 (NH–CH_2); Anal. Calcd. for $C_{17}H_{16}N_3$: C, 49.15; H, 3.88; N, 10.12. Found: C, 49.25; H, 3.64; N, 10.32.

*(E)-N-(2-(2-(2-Chlorobenzylidene)hydrazinyl)-2-oxoethyl)-4-(trifluoromethyl)benzene sulfonamide (**5d**)*

Yellow needles (EtOH) (this compound was prepared by the reaction of compound **4** and 2-chlorobenzaldehyde. It was obtained as a yellow solid); mp 154–156 °C; IR (KBr) ν_{max} 3303, 3145, 3073, 1681, 1631, 1159, 1050 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz): $\delta = 9.32$ (1H, s, NH), 8.51 (1H, s, N=CH), 8.05 (1H, s, NH), 7.66–7.78 (3H, m, H-3, H-5, H-6′), 7.59 (1H, s, H-3′), 7.04–7.12 (4H, m, H-2, H-6, H-4′, H-5′), 4.59 (2H, s, $–CH_2$); ^{13}C NMR (DMSO-d_6, 100 MHz): $\delta = 171.0$ (C, C=O), 144.6 (C, C-1), 140.1 (CH, C=N), 134.4 (C, $J_{CF} = 32.6$ Hz, C-4), 133.5 (C, C-2′), 132.7 (CH, C-1′), 130.4 (CH, C-2, C-6), 127.7 (CH, C-3′), 126.6 (CH, $J_{CF} = 4.0$ Hz, C-3, C-5), 124.8 ($J_{CF} = 272.0$ Hz, CF_3), 124.0 (CH, C-6′), 45.8 (NH–CH_2); Anal. Calcd. for $C_{16}H_{13}N_3$: C, 45.78; H, 3.12; N, 10.01. Found: C, 45.47; H, 3.31; N, 10.32.

*(E)-N-(2-(2-(4-Chlorobenzylidene)hydrazinyl)-2-oxoethyl)-4-(trifluoromethyl)benzene sulfonamide (**5e**)*

Yellow needles (EtOH) (this compound was prepared by the reaction of compound **4** and 4-chlorobenzaldehyde. It was obtained as a yellow solid); mp 167–169 °C; IR (KBr) ν_{max} 3300, 3143, 3070, 1665, 1621, 1158, 1061 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz): δ = 9.37 (1H, s, NH), 8.62 (1H, s, N=CH), 8.03 (1H, s, NH), 7.97 (2H, d, $J = 8.0$ Hz, H-2′, H-6′), 7.68 (2H, d, $J = 8.08$ Hz, H-3, H-5), 7.42 (2H, d, $J = 8.51$ Hz, H-3′, H-5′), 7.05 (2H, d, $J = 8.51$ Hz, H-2, H-6), 6.36 (s, NH), 4.58 (2H, s, $–CH_2$); ^{13}C NMR (DMSO-d_6, 100 MHz); δ = 171.0 (C, C=O), 148.5 (C, C-1), 144.1 (CH, C=N), 134.4 (C, $J_{CF} = 31.5$ Hz, C-4), 134.0 (C, C-4′), 132.2 (C, C-1′), 131.0 (CH, C-2′, C-6′), 129.4 (CH, C-2, C-6), 128.2 (CH, C-2′, C-6′), 127.3 (CH, $J_{CF} = 4.0$ Hz, C-3, C-5), 124.8 ($J_{CF} = 275$ Hz, CF_3), 116.2 (CH, C-3′, C-5′), 45.8 (NH–CH_2). Anal. Calcd. for $C_{16}H_{13}N_3$: C, 45.78; H, 3.12; N, 10.01. Found: C, 45.65; H, 3.32; N, 10.30.

*(E)-N-(2-(2-(4-Bromobenzylidene)hydrazinyl)-2-oxoethyl)-4-(trifluoromethyl)benzene sulfonamide (**5f**)*

White needles (EtOH) (this compound was prepared by the reaction of compound **4** and 4-bromobenzaldehyde. It was obtained as a white solid); mp 148–150 °C; IR (KBr) ν_{max} 3310, 3154, 3070, 1660, 1625, 1150, 1068 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz) δ = 9.29 (1H, s, NH), 8.48 (1H, s, N=CH), 8.06 (1H, s, NH), 7.96 (2H, d, $J = 8.56$ Hz, H-3′, H-5′), 7.45 (2H, d, $J = 8.5$ Hz, H-3, H-5), 7.04–7.19 (4H, m, H-2, H-6, H-2′, H-6′), 4.59 (2H, s, $–CH_2$); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 171.0 (C, C=O), 149.8 (C, C-1), 144.0 (CH, C=N), 134.4 (C, $J_{CF} = 33.7$ Hz, C-4), 132.8 (C, C-1′), 131.7 (CH, C-2′, C-6′), 129.6 (CH, C-2, C-6), 128.5 (C, C-4′), 127.0 (CH, $J_{CF} = 4.8$ Hz, C-3, C-5), 124.4 ($J_{CF} = 271.0$ Hz, CF_3), 116.9 (CH, C3′, C-5′), 46.1 (NH–CH_2); Anal. Calcd. for $C_{16}H_{13}N_3$: C, 41.39; H, 2.82; N, 9.05. Found: C, 41.43; H, 2.93; N, 9.24.

*(E)-N-(2-(2-(2-Nitrobenzylidene)hydrazinyl)-2-oxoethyl)-4-(trifluoromethyl)benzene sulfonamide (**5g**)*

Yellow needles (EtOH) (this compound was prepared by the reaction of compound **4** and 2-nitrobenzaldehyde. It was obtained as a yellow solid); mp 164–166 °C; IR (KBr) ν_{max} 3300, 3150, 3070, 1680, 1630, 1160, 1060 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz): δ = 9.34 (1H, s, NH), 8.46

(1H, s, N=CH), 8.10–8.15 (1H, m, H-3′), 8.08 (1H, s, NH), 7.72–7.85 (5H, m, H-3, H-5, H-4′, H-5′, H-6′), 7.08 (2H, dd, $J = 2.14$, 6.8 Hz, H-2, H-6), 4.60 (2H, s, $–CH_2$); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 171.0 (C, C=O), 149.5 (C, C-1), 143.3 (CH, C=N), 135.4 (C, C-2′), 134.0 (C, $J_{CF} = 32.6$ Hz, C-4), 130.1 (CH, C-5′), 128.4 (CH, C-2, C-6), 128.2 (C, C-1′), 128.0 (CH, C-3′), 127.0 (CH, $J_{CF} = 4.4$ Hz, C-3, C-5), 124.7 (CH, C-6′), 124.0 ($J_{CF} = 275$ Hz, CF_3), 45.5 (NH–CH_2); Anal. Calcd. for $C_{16}H_{13}N_4$: C, 44.65; H, 3.04; N, 13.02. Found: C, 44.84; H, 3.14; N, 13.32.

*(E)-N-(2-(2-(4-Fluorobenzylidene)hydrazinyl)-2-oxoethyl)-4-(trifluoromethyl)benzene sulfonamide (**5h**)*

White needles (EtOH) (this compound was prepared by the reaction of compound **4** and 4-fluorobenzaldehyde. It was obtained as a white solid); mp 159–161 °C; IR (KBr) ν_{max} 3306, 3162, 3055, 1670, 1630, 1151, 1061 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz): δ = 9.29 (1H, s, NH). 8.48 (1H, s, N=CH), 8.09 (1H, s, NH), 7.89 (2H, dd, $J = 1.7$, 8.70 Hz, H-2′, H-6′), 7.22–7.38 (4H, m, H-3, H-5, H-3′, H-5′), 6.97 (2H, dd, $J = 2.11$, 6.9 Hz, H-2, H-6), 4.59 (2H, s, $–CH_2$); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 171.0 (C, C=O), 166.1 (C, $J_{CF} = 238.20$ Hz, C-4′), 149.2 (C, C-1), 144.0 (CH, C=N), 134.4 (C, $J_{CF} = 32.0$ Hz, C-4), 130.3 (CH, C-2′, C-6′), 130.2 (C, C-1′), 128.6 (CH, C-2, C-6), 127.6 (CH, $J_{CF} = 4.4$ Hz, C-3, C-5), 124.7 (CH, C-6′), 124.0 ($J_{CF} = 272.0$ Hz, CF_3), 116.2 (CH, $J_{CF} = 9.5$ Hz C-3′, C-5′), 45.7 (NH–CH_2); Anal. Calcd. for $C_{16}H_{13}N_3$: C, 47.64; H, 3.25; N, 10.42. Found: C, 47.81; H, 3.43; N, 10.62.

*(E)-N-(2-(2-(3,4-Dichlorobenzylidene)hydrazinyl)-2-oxoethyl)-4-(trifluoromethyl)benzene.3, sulfonamide (**5i**)*

Off white needles (EtOH) (this compound was prepared by the reaction of compound **4** and 3,4-dichlorobenzaldehyde. It was obtained as an off white solid); mp 159–161 °C; IR (KBr) ν_{max} 3325, 3140, 3062, 1664, 1644, 1158, 1058 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz): δ = 9.26 (1H, s, NH). 8.43 (1H, s, N=CH), 8.09 (1H, s, NH), 7.31–7.34 (1H, t, $J = 8.5$ Hz, H-5′), 7.35–7.42 (4H, m, H-3, H-5, H-2′, H-6′), 7.06 (2H, d, $J = 8.90$ Hz, H-2, H-6), 4.59 (2H, s, $–CH_2$); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 171.1 (C, C=O), 149.6 (C, C-1), 145.8 (CH, C=N), 134.1 (C, $J_{CF} = 33.0$ Hz, C-4), 133.4 (C, C-1′), 130.7 (C, C-3′, C-4′), 128.8 (CH, C-2, C-6), 127.6 (CH, $J_{CF} = 4.1$ Hz, C-3, C-5), 127.0 (CH, C-5′), 125.6 (CH, C-6′), 124.0 ($J_{CF} = 274.6$ Hz, CF_3), 45.5 (NH–CH_2); Anal. Calcd. for $C_{16}H_{12}N_3$: C, 42.31; H, 2.66; N, 9.25. Found: C, 42.52; H, 2.51; N, 9.35.

*(E)-N-(2-(2-(2,4-Dimethoxybenzylidene)hydrazinyl)-2-oxoethyl)-4-(trifluoromethyl)benzene sulfonamide (**5j**)*

White needles (EtOH) (this compound was prepared by the reaction of compound **4** and 2,4-dimethoxybenzaldehyde. It was obtained as a white solid); mp 173–175 °C; IR (KBr) ν_{max} 3310, 3144, 3063, 168, 1631, 1156, 1061 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz): δ = 9.39 (1H, s, NH), 8.47 (1H, s, N=CH), 8.04 (1H, s, NH), 7.98 (2H, d, $J = 8.64$ Hz, H-1, H-6′), 7.51–7.72 (4H, m, H-3, H-5, H-3′, H-5′), 7.15 (2H, d, $J = 5.1$ Hz, H-2, H-6), 4.58 (2H, s, $–CH_2$), 3.88 (3H, s, $–OCH_3$); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 171.0 (C, C=O), 160.2 (C, C-4′), 150.0 (C, C-2′), 149.2 (C, C-1), 149.0 (CH, C-6′), 146.0 (CH, C=N), 134.3 (C, $J_{CF} = 32.7$ Hz, C-4), 128.9 (CH, C-2, C-6), 127.2 (CH, $J_{CF} = 4.45$ Hz, C-3, C-5), 124.2 ($J_{CF} = 273.4$ Hz, CF_3), 118.2 (CH, C-3′), 110.0 (CH, C-5′), 52.3 (O-CH_3), 45.5 (NH–CH_2); Anal. Calcd. for. $C_{18}H_{18}N_3$: C, 48.54; H, 4.07; N, 9.43. Found: C, 48.74; H, 4.32; N, 9.63.

*General procedure for the synthesis of N-((5-phenyl-1,3,4-oxadiazole-2-yl)methyl)-4-(trifluoromethyl)benzenesulfonamide (**6a–j**)*

To a stirred solution of **5a–j** (0.01 mmol) in dichloromethane (10 ml) at room temperature, iodobenzenediacetate (0.011 mmol) was added in single lot, and the mixture was stirred for 1 h. The progress of the reaction was monitored by TLC. After completion of the reaction, the reaction mass was concentrated under reduced pressure, and the solid was isolated with pet ether. The isolated solid was recrystallized from methanol and dried to yield 2,5-disubstituted 1,3,4-oxadiazoles **6a–j** in 70–92 % yield.

*N-((5-Phenyl-1,3,4-oxadiazole-2-yl)methyl)-4-(trifluoromethyl)benzenesulfonamide (**6a**)*

White needles (MeOH) (this compound was prepared by the reaction of compound **5a** and iodobenzenediacetate. It was obtained as a white solid); yield 74 %; mp 153–155 °C; IR (KBr) ν_{max} 3300, 3063, 1680, 1557, 1160, 1061 cm^{-1}; 1H NMR (DMSO-d_6, 400 MHz): δ = 8.15 (1H, s, NH), 8.02 (2H, d, $J = 8.8$ Hz, H-2′, H-6′), 7.74–7.78 (3H, m, H-3′, H-4′, H-5′), 7.57 (2H, d, $J = 8.46$ Hz, H-3, H-5), 7.08 (2H, d, $J = 8.2$ Hz, H-2, H-6), 4.60 (2H, s, $–CH_2$); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 164.7 (oxadiazole C-5), 157.8 (oxadiazole C-2), 149.6 (C, C-1), 134.0 (C, $J_{CF} = 31.8$ Hz, C-4), 129.5 (CH, C-2, C-6), 128.5 (CH, $J_{CF} = 4.44$ Hz C-3, C-5), 128.2 (CH, C-2′, C-6′), 124.6 ($J_{CF} = 270.6$ Hz, CF_3), 119.4 (C, C-1′), 116.2 (CH, C-3′, C-5′), 115.7 (CH, C-4′), 42.1 (CH_2, NH–CH_2); MS (ESI) *m/z*: 384.2 $[M+1]^+$; Anal. Calcd. for $C_{16}H_{12}N_3$: C, 50.13; H, 3.16; N, 10.96. Found: C, 50.00; H, 3.14; N, 10.92.

*N-((5-(p-Tolyl)-1,3,4-oxadiazole-2-yl)methyl)-4-(trifluoromethyl)benzenesulfonamide (**6b**)*

White needles (MeOH) (this compound was prepared by the reaction of compound **5b** and iodobenzenediacetate. It was obtained as a white solid); yield 72 %; mp 171–173 °C; IR (KBr) ν_{max} 3310, 3068, 1670, 1552, 1164, 1066 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz): δ = 8.14 (1H, s, NH), 8.0–8.15 (2H, d, J = 8.5 Hz, H-2′, H-6′), 7.58 (2H, d, J = 8.15 Hz, H-3, H-5), 7.35 (4H, m, H2, H-6, H-3′, H-5′), 4.60 (2H, s, $–CH_2$), 2.18 (3H, s, Ar-CH_3); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 164.3 (oxadiazole C-5), 158.8 (oxadiazole C-2), 149.6 (C, C-1), 134.0 (C, J_{CF} = 32.3 Hz, C-4), 129.2 (CH, C-2, C-6), 128.9 (CH, J_{CF} = 4.4 Hz, C-3, C-5), 127.0 (CH, C-2′, C-6′), 124.6 (J_{CF} = 273.6 Hz, CF_3), 42.4 (CH_2, NH–CH_2), 15.3 (CH_3, Ar-CH_3); MS (ESI) *m/z*: 398.2 $[M+1]^+$; Anal. Calcd. for $C_{17}H_{14}N_3$: C, 51.38; H, 3.55; N, 10.57. Found: C, 51.35; H, 3.50; N, 10.61.

*N-((5-(4-Methoxyphenyl)-1,3,4-oxadiazole-2-yl)methyl)-4-(trifluoromethyl)benzene sulfonamide (**6c**)*

White needles (MeOH) (this compound was prepared by the reaction of compound **5c** and iodobenzenediacetate. It was obtained as a white solid); yield 79 %; mp 144–146 °C; IR (KBr) ν_{max} 3290, 3048, 1661, 1545, 1154, 1057 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz): δ = 8.15 (1H, s, NH), 7.82 (2H, dd, J = 2.0, 8.34 Hz, H-2′, H-6′), 7.19–7.32 (4H, m, H-3, H-5, H-3′, H-5′), 7.06 (2H, dd, J = 1.84, 8.40 Hz, H-2, H-6), 4.58 (2H, s, $–CH_2$), 3.86 (3H, s, $–OCH_3$); ^{13}C NMR (DMSO-d_6 100 MHz): δ = 163.9 (oxadiazole C-5), 161.4 (oxadiazole C-2), 160.2 (C, C-4′), 149.8 (C, C-1), 134.0 (C, J_{CF} = 32.5 Hz, C-4), 129.6 (CH, C-2, C-6), 127.9 (CH, J_{CF} = 4.13 Hz, C-3, C-5), 124.6 (J_{CF} = 271.8 Hz, CF_3), 116.4 (C, C-1′), 115.8 (CH, C-2′, C-6′), 114.4 (CH, C-3′, C-5′), 55.6 (CH_3, O–CH_3), 41.6 (CH_2, NH–CH_2); MS (ESI) *m/z*: 416.2 $[M+1]^+$; Anal. Calcd. for $C_{17}H_{14}N_3$: C, 49.40; H, 3.41; N, 10.17. Found: C, 49.45; H, 3.40; N, 10.10.

*N-((5-(2-Chlorophenyl)-1,3,4-oxadiazole-2-yl)methyl)-4-(trifluoromethyl)benzenesulfonamide (**6d**)*

White needles (MeOH) (this compound was prepared by the reaction of **5d** and iodobenzenediacetate. It was obtained as a white solid); yield 73 %; mp 155–157 °C; IR (KBr) ν_{max} 3300, 3063, 1680, 1557, 1160, 1061 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz): δ = 8.12 (1H, s, NH), 7.76 (2H, d, J = 8.34 Hz, H-3, H-5), 7.66–7.74 (2H, m, H-3′, H-6′), 7.04 (4H, m, H-2, H-6, H-4′, H-5′), 4.59 (2H, s, $–CH_2$); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 164.1 (oxadiazole C_5), 163.4 (oxadiazole C_2), 149.7 (C, C-1), 134.2 (C, J_{CF} = 32.4 Hz, C-4), 133.7 (CH, C-2′), 130.0 (C, C-4′), 129.9 (CH, C-3′), 128.6 (CH, C-2, C-6), 127.5 (CH, J_{CF} = 31.7 Hz, C-3, C-5), 124.9 (J_{CF} = 270.7 Hz, CF_3), 124.0 (CH, C-6′), 116.2 (C, C-1′), 107.0 (CH, C-5′), 41.1 (CH_2, NH–CH_2); MS (ESI) *m/z*: 419.2 $[M+2]^+$; Anal. Calcd. for $C_{16}H_{11}N_3$ (in %): C, 46.00; H, 2.65; N, 10.06. Found: C, 46.02; H, 2.69; N, 9.90.

*N-((5-(4-Chlorophenyl)-1,3,4-oxadiazole-2-yl)methyl)-4-(trifluoromethyl)benzenesulfonamide (**6e**)*

White needles (MeOH) (this compound was prepared by the reaction of **5e** and iodobenzenediacetate. It was obtained as a white solid); yield 76 %; mp 181–183 °C; IR (KBr) ν_{max} 3310, 3060, 1680, 1557, 1160, 1061 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz): δ = 8.14 (s, NH), 8.02 (2H, d, J = 7.82 Hz, H-2′, H-6′), 7.74 (2H, d, J = 7.8 Hz, H-3′, H-5′), 7.57 (2H, d, J = 8.0 Hz, H-3, H-5), 7.09 (2H, d, J = 8.1 Hz, H-2, H-6), 4.60 (2H, s, $–CH_2$); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 166.2 (oxadiazole C_5), 162.4 (oxadiazole C_2), 148.5 (C, C-1), 134.4 (C, J_{CF} = 32.0 Hz, C-4), 134.0 (C, C-1′), 129.4 (CH, C-2, C-6), 128.2 (CH, J_{CF} = 32.6 Hz, C-3, C-5), 127.3 (CH, C-2′, C-6′), 124.9 (J_{CF} = 272.8 Hz, CF_3), 116.2 (CH, C-3′, C-5′), 115.6 (C, C-4′), 42.2 (CH_2, NH–CH_2); MS (ESI) *m/z*: 419.0 $[M+2]^+$; Anal. Calcd. for $C_{16}H_{11}N_3$ (in %): C, 46.00; H, 2.65; N, 10.06. Found: C, 46.08; H, 2.62; N, 9.99.

*N-((5-(4-Bromophenyl)-1,3,4-oxadiazole-2-yl)methyl)-4-(trifluoromethyl)benzenesulfonamide (**6f**)*

White needles (MeOH) (this compound was prepared by the reaction of **5f** and iodobenzenediacetate. It was obtained as a white solid); yield 78 %; mp 170–172 °C; IR (KBr) ν_{max} 3320, 3055, 1670, 1550, 1160, 1060 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz) δ: 8.15 (1H, s, NH), 7.94 (2H, d, J = 8.61 Hz, H-2′, H-6′), 7.44–7.72 (4H, m, H-3, H-5, H-3′, H-5′), 7.05 (2H, d, J = 8.42 Hz, H-2, H-6), 4.60 (2H, s, $–CH_2$); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 165.1 (oxadiazole C_5), 163.1 (oxadiazole C_2), 149.8 (C, C-1), 134.3 (C, J_{CF} = 32.2 Hz, C-4), 131.7 (CH, C-2′, C-6′), 129.6 (CH, C-2, C-6), 128.6 (CH, J_{CF} = 4.12 Hz, C-3, C-5), 127.0 (C, C-1′), 123.9 (J_{CF} = 272.2 Hz, CF_3), 116.5 (CH, C-3′, C-5′), 115.2 (C. C-4′), 42.2 (CH_2, NH–CH_2); MS (ESI) *m/z*: 461.0 $[M+1]^+$; Anal. Calcd. for $C_{16}H_{11}BrF_3N_3O_3S$ (in %): C, 41.57; H, 2.40; N, 10.38. Found: C, 41.50; H, 2.43; N, 10.32.

*N-((5-(2-Nitrophenyl)-1,3,4-oxadiazole-2-yl)methyl)-4-(trifluoromethyl)benzenesulfonamide (**6g**)*

White needles (MeOH) (this compound was prepared by the reaction of **5g** and iodobenzenediacetate. It was obtained as a white solid); yield 73 %; mp 163–165 °C; IR (KBr) ν_{max} 3290, 3050, 1668, 1564, 1168, 1066 cm^{-1}; 1H NMR (DMSO-d_6, 400 MHz): δ = 8.14 (1H, s, NH),

8.11–8.14 (2H, m, H-3′, H-6′), 7.76–7.82 (4H, m, H-3, H-5, H-4′, H–5′), 7.15 (2H, dd, $J = 2.12$, 6.90 Hz, H-2, H6), 4.59 (2H, s, $–CH_2$); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 165.1 (oxadiazole C_5), 162.5 (oxadiazole C_2), 149.5 (C, C-1), 135.4 (C, C-2′), 134.0 (C, $J_{CF} = 32.2$ Hz, C-4), 130.1 (CH, C-5′), 129.0 (CH, C-2, C-6), 128.5 (CH, $J_{CF} = 4.11$ Hz, C-3, C-5), 124.1 ($J_{CF} = 270.9$ Hz, CF_3), 116.8 (CH, C-4′), 115.7, (CH, C-6′), 42.4 (CH_2, NH–CH_2); MS (ESI) *m/z*: 429.1 $[M+1]^+$; Anal. Calcd. for $C_{16}H_{11}N_4$ (in %): C, 44.86; H, 2.59; N, 13.08. Found: C, 44.80; H, 2.55; N, 13.00.

*N-((5-(4-Fluorophenyl)-1,3,4-oxadiazole-2-yl)methyl)-4-(trifluoromethyl)benzenesulfonamide (**6h**)*

White needles (MeOH) (this compound was prepared by the reaction of **5h** and iodobenzenediacetate. It was obtained as a white solid); yield 84 %; mp 186–188 °C; IR (KBr) ν_{max} 3306, 3061, 1670, 1556, 1163, 1062 $cm^{-1;}$ ^1H NMR (DMSO-d_6, 400 MHz): δ = 8.14 (1H, s, NH), 7.98 (2H, d, $J = 8.55$ Hz, H-2′, H-6′), 7.50 (4H, m, H-3, H-5, H-3′, H-5′), 7.11 (2H, dd, $J = 2.06$, 7.4 Hz, H-2, H-6), 4.60 (2H, s, $–CH_2$); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 165.9 (oxadiazole C_5), 163.1 (oxadiazole C_2), 166.1 (C, $J_{CF} = 239.59$ Hz C-4′), 149.2 (C, C-1), 134.4 (C, $J_{CF} = 32.0$ Hz, C-4), 130.3 (C, C-2′, C-6′), 129.6 (CH, C-2, C-6), 129.0 (CH, $J_{CF} = 4.0$ Hz, C-3, C-5), 124.1 ($J_{CF} = 271.8$ Hz, CF_3), 116.0 (CH, $J_{CF} = 9.70$ Hz, C-3′, C-5′), 115.1 (C, C-1′), 42.3 (CH_2, NH–CH_2); MS (ESI) *m/z*: 402.1 $[M+1]^+$; Anal. Calcd. for $C_{16}H_{11}N_3$ (in %): C, 47.88; H, 2.76; N, 10.47. Found: C, 47.89; H, 2.72; N, 10.51.

*N-((5-(3,4-Dichlorophenyl)-1,3,4-oxadiazole-2-yl)methyl)-4-(trifluoromethyl)benzene sulfonamide (**6i**)*

White needles (MeOH) (this compound was prepared by the reaction of **5i** and iodobenzenediacetate. It was obtained as a white solid); yield 80 %; mp 173–175 °C; IR (KBr) ν_{max} 3309, 3066, 1674, 1560, 1154, 1069 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz) δ: 8.15 (1H, s, NH), 7.82–7.84 (2H, m, H-2′, H-6′), 7.39 (2H, d, $J = 8.8$ Hz, H-3, H-5), 7.31 (1H, s, H-5′), 7.12 (2H, dd, $J = 1.90$, 6.80 Hz, H-2, H-6), 4.59 (2H, s, $–CH_2$); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 165.8 (oxadiazole C_5), 162.8 (oxadiazole C_2), 149.6 (C, C-1), 134.1 (C, $J_{CF} = 32.47$ Hz, C-4), 133.4 (C, C-4′), 130.7 (C, C-3′), 128.8 (CH, C-2, C-6), 128.5 (CH, $J_{CF} = 4.41$ Hz, C-3, C-5), 127.0 (CH, C-6′), 1256 (C, C-1′), 124.2 ($J_{CF} = 274.3$ Hz, CF_3), 116.4 (CH, C-2′), 115.6 (CH, C-5′), 42.1 (CH_2, NH–CH_2); MS (ESI) *m/z*: 354.2 $[M+2]^+$; Anal. Calcd. for $C_{16}H_{10}N_3$ (in %): C, 42.49; H, 2.23; N, 9.29. Found: C, 42.45; H, 2.20; N, 9.28.

*N-((5-(2,4-Dimethoxyphenyl)-1,3,4-oxadiazole-2-yl)methyl)-4-(trifluoromethyl)benzene sulfonamide (**6j**)*

White needles (MeOH) (this compound was prepared by the reaction of **5j** and iodobenzenediacetate. It was obtained as a white solid); yield 92 %; mp 158–160 °C; IR (KBr) ν_{max} 3960, 3060, 1680, 1555, 1160, 1060 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz): δ = 8.15 (1H, s, NH), 7.50–7.66 (3H, m, H-3, H-5, H-6′), 7.14–7.20 (4H, m, H-2, H-6, H-3′, H-5′), 4.59 (2H, s, $–CH_2$), 3.89 (3H, s, $–OCH_3$); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 166.6 (oxadiazole C_5), 163.5 (oxadiazole C_2), 150.0 (C, C-4′), 149.2 (C, C-2′), 149.0 (C, C-1), 134.4 (C, $J_{CF} = 32.0$ Hz, C-4), 129.0 (CH, C-2, C-6), 128.4 (CH, $J_{CF} = 4.12$ Hz, C-3, C-5), 124.1 ($J_{CF} = 270.0$ Hz, CF_3), 118.2 (CH, C-6′), 116.2 (C, C-1′), 110.0 (CH, C-3′), 108.2 (CH, C-5′), 52.3 (CH_3, O-CH_3), 42.1 (CH_2, NH–CH_2); MS (ESI) *m/z*: 446.2 $[M+1]^+$; Anal. Calcd. for $C_{18}H_{16}N_3$ (in %): C, 48.76; H, 3.64; N, 9.48. Found: C, 48.79; H, 3.60; N, 9.42.

Antiproliferative activity

The 3-(4,5-dimethyl-thiazol-2-yl)-2,5-diphenyl tetrazolium bromide (MTT) was dissolved (5 mg/ml) in a phosphate buffer saline (pH 7.2) and filtered before use. The RPMI 1640 cell culture medium, MTT, and fetal bovine serum (FBS) were purchased from Merck chemicals.

Human metastatic breast cancer (MDA-MB 231), human chronic myeloid leukemia (K562), human colon carcinoma (Colo-205), and human neuroblastoma (IMR-32) cell lines were procured from the National Center for Cell Sciences in Pune, India. All cells were grown in RPMI-1640 supplemented with 10 % heat inactivated FBS, 100 IU/ml penicillin, 100 mg/ml streptomycin and 2 mM-glutamine. The cultures were maintained in a humidified atmosphere with 5 % CO_2 at 37 °C.

The potential effects on cell viability were investigated using the MTT assay (Mallesha *et al.*, 2012). The MTT assay is an indicator of metabolically active cells. A known number of MDA-MB 231, K562, Colo-205, and IMR-32 cells were transferred into 96 well plates in a volume of 200 µl of culture medium and incubated for 48 h before the addition of the test compound. Cells were then exposed to known concentrations of the compound to be tested (10 µM expressed as final concentration) for 24 h at 37 °C. After drug exposure, the culture medium was removed, and 20 µl of MTT reagent (diluted in culture medium, 5 mg/ml) was added. After incubating for 4 h, the MTT/medium was removed, and DMSO (100 µl) was added to each well; the plates were agitated for 1 min. Absorbance of the colored solution was measured on a multiwell plate reader (Victor3, Perkin Emler) using a test wavelength of 570 nm.

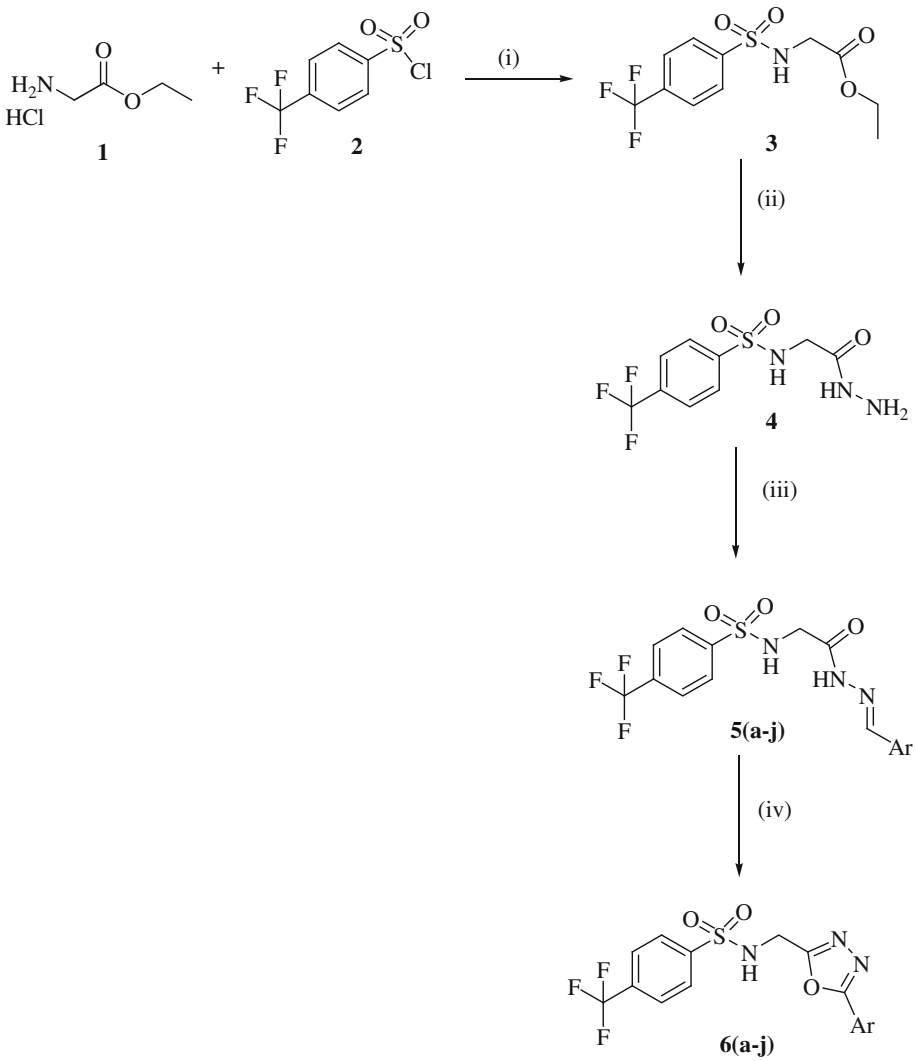
Results were evaluated by comparing the absorbance of the wells containing compound-treated cells with the absorbance of wells containing 0.1 % DMSO alone (solvent control). Conventionally, cell viability was estimated to be 100 % in the solvent control, and the assay was performed in triplicate.

Results and discussion

Chemistry

The title compounds were prepared using the synthetic strategy described in Scheme 1. The chemical structures of the new compounds are given in Table 1. Compound **3** was synthesized by the reaction of glycine ethyl ester hydrochloride **1** with 4-(trifluoromethyl)benzenesulfonyl chloride **2** in the presence of a base. Compound **4** was synthesized by the reaction of **3** with hydrazine hydrate in ethanol as per the reported procedure (Jayashankar *et al*., 2009). Compounds **5a–j** was prepared by refluxing a mixture of aldehydes in absolute ethanol for about 2 h. A series of new compounds (**6a–j**) have been accomplished in excellent yields by the oxidation of **5a–j** with one equivalents of iodobenzenediacetate (IBD) in dichloromethane. Compounds **5a–j** and **6a–j** were synthesized based on reported procedure (Prakash *et al*., 2010).

The FT-IR spectra were recorded using KBr pellets in the range of 4,000–400 cm^{-1}. The structure of compounds **5a–j** was confirmed using IR and ^{1}H NMR. The IR spectra of the compound **5a** exhibited characteristic absorption bands at 1,631 and 3,150 cm^{-1} due to carbonyl and NH groups, respectively. The ^{1}H NMR spectra of **5a** showed two singlet due to N=CH and NH at δ 8.62 and δ 9.35, respectively. The structures of all compounds **6a–j** were confirmed by their spectral (IR, LCMS, ^{1}H NMR, and ^{13}C NMR) and elemental analyses. The characterization of product **6a–j** was based upon a careful comparison of their



Scheme 1 Reagent and reaction conditions: *i* DCM, TEA, r.t., 1 h; *ii* hydrazine hydrate, reflux, 3 h; *iii* substituted benzaldehydes, EtOH, H^{+}, r.t., 2 h; and *iv* iodobenzene diacetate, DCM, r.t., 1 h

Table 1 Chemical structure of **6(a–j)**

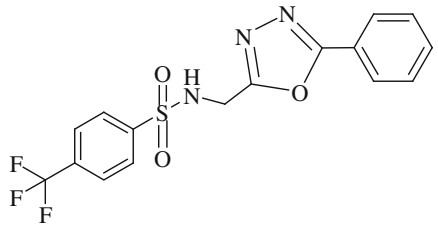
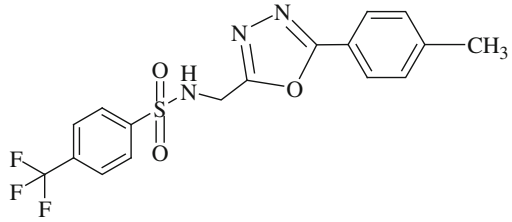
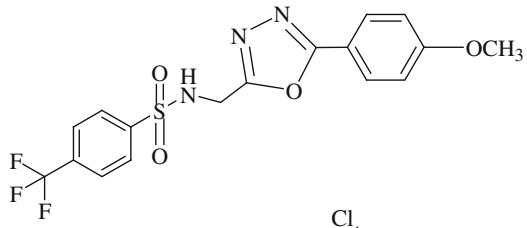
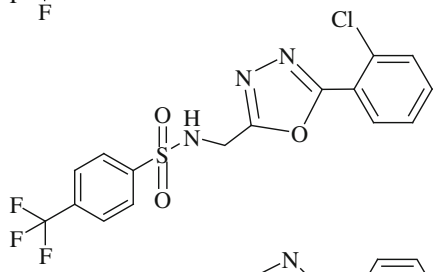
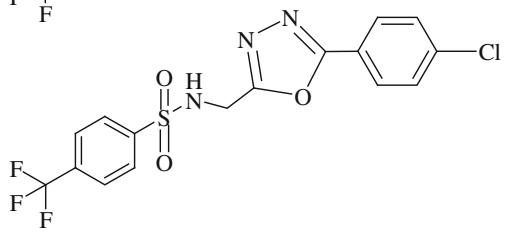
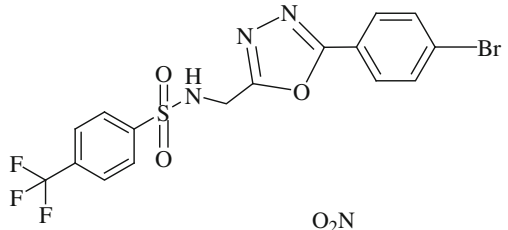
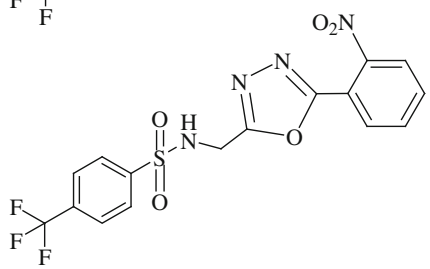
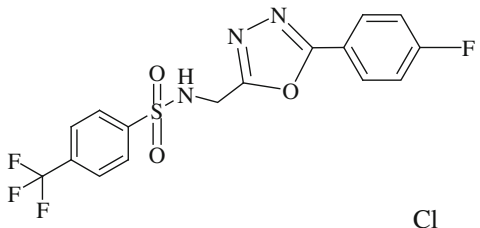
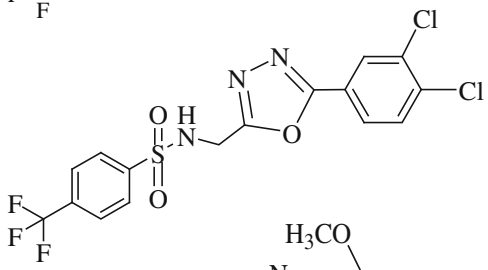
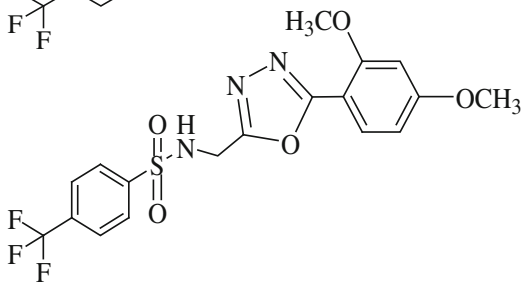
Compound	Ar	Structure
6a	Phenyl	
6b	4-Methylphenyl	
6c	4-Methoxylphenyl	
6d	2-Chlorophenyl	
6e	4-Chlorophenyl	
6f	4-Bromophenyl	
6g	2-Nitrophenyl	

Table 1 continued

Compound	Ar	Structure
6h	4-Fluorophenyl	
6i	3,4-Dichlorophenyl	
6j	2,4-Methoxyphenyl	

IR and ^{1}H NMR spectra with those of **5a**–**j**. For example, IR spectra of **6a** were found to be transparent in the region of NH stretch and CO stretch. In ^{1}H NMR spectra of **6a**–**j**, the disappearance of their singlet due to N=CH around δ 8.4–8.6 and NH proton around δ 9.3–9.5, thus, confirming the oxidation of **5a**–**j** into **6a**–**j**. ^{13}C NMR spectra present the correct number of carbon atoms at the appropriate chemical shift values. The mass spectra of **6a** showed molecular ion peak at *m*/*z* 384.2 which is in agreement with the molecular formula $C_{16}H_{12}F_3N_3O_3S$. The elemental analyses data showed good agreement between the experimentally determined values and the theoretically calculated values within ±0.4 %.

In vitro antiproliferative activity

Antiproliferative activity of 1,3,4-oxadiazoles against human neuroblastoma (IMR-32) cell lines (Gudipati *et al*., 2011) has been reported. A new class of sulfonamides has been used in the treatment of diseases arising from abnormal cell growth and proliferation (Shan *et al*., 1999). The antiproliferative action of the synthesized compounds **6a**–**j** was tested against four different cell lines. The activity was evaluated by measuring the levels of surviving cells after incubation for 24 h with the test samples using the MTT colorimetric assay based on the ability of metabolically active cells to convert the pale yellow MTT to a blue formazan product which is quantifiable spectrophotometrically. The percentage cell survival for tested compounds against MDA-MB 231, K562, Colo-205, and IMR-32 cells is tabulated in Table 2. The extent of inhibition of cell lines by **6h** and **6i** is schematically presented in Fig. 1. The results were expressed as percentage of cell proliferation compared with cells in control (cells treated with vehicle, 0.1 % DMSO).

Table 2 Antiproliferative activity of **6a**–**j** against human cancer cells determined using MTT test

Compound	% Cell survival at 10 μM			
	K562	Colo-205	MDA-MB 231	IMR-32
6a	–[a]	50.28	50.04	48.70
6b	–[a]	48.50	47.50	47.33
6c	–[a]	47.10	46.20	47.15
6d	45.51	60.01	57.50	55.40
6e	45.50	60.10	57.47	54.39
6f	42.00	52.20	51.40	50.31
6g	43.22	55.32	53.00	51.20
6h	59.31	64.00	62.40	68.22
6i	48.20	61.70	60.05	60.81
6j	–[a]	42.52	40.68	41.50
Control (0.1 % DMSO)	100	100	100	100

[a] Represents <30 % cell survival

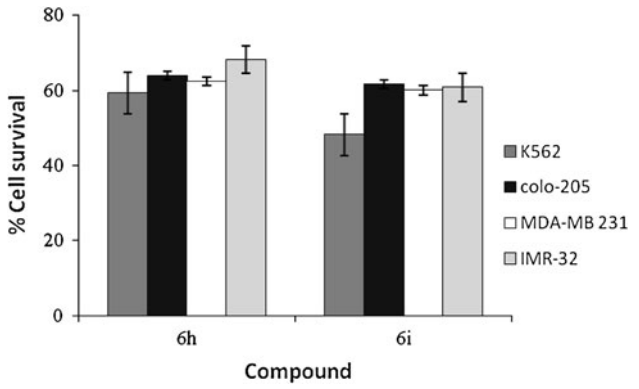


Fig. 1 MTT assay for **6h** and **6i** at 10 μM

Structure activity relationship can be drawn for the derivatives **6(d–i)** containing electronegative atoms, which reveals that, compound **6h** has more electronegative fluorine atom compared to **6(d–g)** and **6i** having other halogen atoms. On correlating the structures of the synthesized compounds with their antiproliferative activity, it was observed that compounds bearing groups like fluoro group and chloro groups on phenyl ring possess high potency in MTT. The SAR study of these compounds indicate that the introduction of fluoro group in **6i** and chloro substituents at the para and meta positions of the phenyl moiety in **6j** showed the best antiproliferative activity. In the same aspect, compound **6i** has disubstituted chlorine atom that exhibits relatively good inhibition compared to **6(d–g)** which are having mono-substituted halogen atom. Introducing electron-donating methoxy groups to **6(a–c)** and **6j** of the phenyl ring of the substituent at ortho, meta, and para positions resulted in the loss of activity. However, the presence of electron-withdrawing groups in the phenyl ring increased the antiproliferative efficacy. On the basis of these observations, we thought of synthesizing a new class of heterocycles, wherein potent 1,3,4-oxadiazole moiety is linked to biological active 4-(trifluoromethyl)benzenesulfonamide moiety at C-5 position and different substituted aryl groups at C-2 position on the basis of combinatorial synthesis, which is the current trend being practiced in most of the drug discoveries.

Conclusion

In conclusion, a series of new 2,5-disubstituted-1,3,4-oxadiazoles **6(a–j)** were synthesized and their antiproliferative activity has been evaluated. Compound **6h** containing fluoro group and **6i** with chloro groups seems to be the most active against all the four cell lines. From this study, we were able to identify a few active molecules which are capable of inhibiting the growth of human cancer cell lines in vitro. The additional modification and diversification of functional groups in order to improve the anticancer activity is currently in progress.

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