Med Chem Res
DOI 10.1007/s00044-016-1543-9

MEDICINAL CHEMISTRY RESEARCH

ORIGINAL RESEARCH

Synthesis and antimicrobial evaluation of hydrazones derived from 4-methylbenzenesulfonohydrazide in aqueous medium

Sunita Ghiya[1] · **Yogesh C. Joshi**[1]

Received: 23 January 2015 / Accepted: 18 February 2016

Abstract A series of biologically active N′-substituted-4-methylbenzenesulfonohydrazide derivatives were synthesized by condensation of 4-methylbenzenesulfonohydrazide and aromatic carbonyl compounds in the presence of polystyrene sulfonic acid in aqueous medium. The synthesized compounds were characterized by IR, NMR and mass spectra. The compounds have been evaluated for antimycobacterial, antibacterial and antifungal activities.

Keywords Hydrazones · Condensation · Antimycobacterial activity · Antibacterial activity · Antifungal activity · Polystyrene sulfonic acid

Introduction

Tuberculosis is one of the infectious diseases caused by *Mycobacterium tuberculosis* which is responsible for nearly 3–4 million deaths every year worldwide. *Mycobacterium tuberculosis* strain develops resistance to front-line antimycobacterial drugs viz. isoniazid, *rifampicin* and second-line drugs such as aminosalicylic acid, *kanamycin* and *capreomycin*. Therefore, there is a need to develop new compounds with better antitubercular activity, less toxicity and safer therapeutic profile (Scior *et al.*, 2002; Tripathi *et al.*, 2005). Hydrazones are biologically important molecules and act as antitubercular agents (Kucukguzel *et al.*, 1999; Koçyigit and Rollas, 2002; Patole *et al.*, 2003; Maccari *et al.*, 2005; Cocco *et al.*, 1999; Karah *et al.*, 2002; Rando *et al.*, 2002) along with wide range of pharmaceutical properties (Savini *et al.*, 1995) viz. antimicrobial, anticancer (Savini *et al.*, 2004), antipyretic (Sridhar and Ramesh, 2001), antinociceptive (Chunha *et al.*, 2002), analgesic, anti-inflammatory (Sondhi *et al.*, 2006) and anticonvulsant activities (Cates and Rasheed, 1984). Various biological activities associated with hydrazone were attributed due to the presence of –CONHN=CH– linkage (Negi *et al.*, 2012). Sulfonyl hydrazones and sulfonamide (Oleveria *et al.*, 2011) having (–SO_2–NHN=CH–) linkage must possess antitubercular properties. In our earlier work, we have synthesized few sulfonylated heterocyclic compounds which exhibit anthelmintic and antimicrobial activities (Saingar *et al.*, 2011).

Hydrazones are generally synthesized in MeOH/EtOH under reflux conditions using additives (Joshi *et al.*, 2008; Bedia *et al.*, 2006). Some important methods for the synthesis of hydrazones involve use of $ZnCl_2$ (Billman and Tai, 1958), $TiCl_2$ (White and Weingart, 1967), $MgSO_4$-PPTL (Branchaud, 1983), $Mg(ClO_4)_2$ (Chakraborti *et al.*, 2004), K-10 (Vaas *et al.*, 1999; Landge *et al.*, 2007) and SiO_2–$NaHSO_4$ (Bazgir 2006) as catalyst. In recent years, organic reactions under solvent-free conditions have gained popularity (Metger, 1998), since the majority of solvents are either toxic or inflammable and considerably enhance the cost of overall synthesis. Solvent-free conditions reduce reaction time and make the isolation easier (Cave *et al.*, 2001; Imrie *et al.*, 2007). Improved yields of hydrazone derivatives have been achieved under microwave irradiation or ultrasound irradiation under solvent-free conditions using polystyrene sulfonic acid (Polshettiwar and Verma, 2007) or SiO_2 (Liete *et al.*, 2008),

✉ Yogesh C. Joshi
drycj_16@yahoo.com

Sunita Ghiya
ghiyasunita.16@gmail.com

[1] Department of Chemistry, University of Rajasthan, Jaipur 302004, India

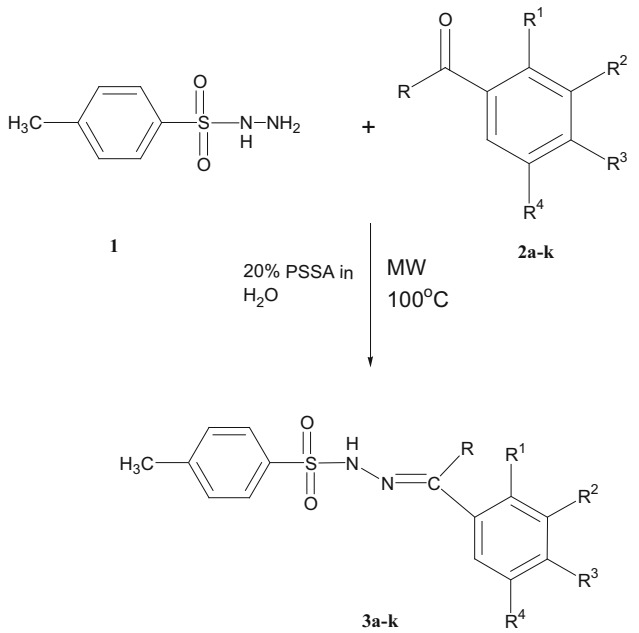
Published online: 03 March 2016

respectively. It is desirable to use water instead of organic solvent as a reaction medium as it is environmentally benign. The efficiency of polystyrene sulfonic acid (PSSA) in aqueous medium has prompted us to explore the possibility of using this reagent for the synthesis of N′-substituted-4-methylbenzenesulfonohydrazide derivatives by condensation of aromatic carbonyls with 4-methylbenzenesulfonohydrazide under microwave irradiation.

Results and discussion

Chemistry

The synthetic method is depicted in Scheme 1. Hydrazones (**3a–k**) were prepared by condensation of equimolar amounts of 4-methylbenzenesulfonohydrazide and substituted benzaldehydes/ketones in aqueous polystyrene sulfonic acid solution under microwave irradiation at 100 °C. Ketones require longer reaction time when compared to aldehydes. All the reactions proceeded smoothly within 4–8 min with excellent yields (Table 1). Reaction time and yields in conventional refluxing method for the preparation of the hydrazones are also incorporated in Table 1. The synthesis of N′-substituted-4-methylbenzenesulfonohydrazide derivatives using polystyrene sulfonic acid (PSSA) in aqueous medium under microwave irradiation is reported for first time. The chemical structures of the compounds were confirmed by elemental analysis, IR, ^{1}H & ^{13}C NMR and mass spectrometry. In compound **3a** ^{1}H NMR signal for –CH= proton and –NH– proton appeared as singlet at 7.69 and 8.27 ppm, respectively. Aromatic and aliphatic protons of all the compounds were with their ^{1}H NMR spectra. All compounds showed satisfactory elemental analysis data. The mass spectra of compounds showed molecular ion peaks.



Scheme 1 Synthesis of N′-substituted-4-methylbenzenesulfonohydrazides

Biology

Antimicrobial activity

The antimicrobial activities of synthesized compounds were evaluated by determining minimum inhibitory concentration (MIC, μM) values and zone of inhibition (mm). The Gram-positive (*Staphylococcus aureus*; MTCC 96) and Gram-negative (*Escherichia coli*; MTCC 1650) bacteria and fungi (*Aspergillus niger*; MTCC 1781 and *Candida albicans*; MTCC 227) were tested by agar diffusion method. Ampicillin and fluconazole were used as reference antibacterial and antifungal drugs, respectively. The results of antimicrobial activities and MIC's of compound **3a–k** against various microbial strains are summarized in Table 2. From activity data, it is clear that compounds of chloro and methoxy group led to increase in antimicrobial activity. Compounds **3a, 3b, 3c, 3d** and **3k** showed good antibacterial activity against all bacterial strains, having more than 12 mm zone of inhibition at MIC values of 17.7–20.2 μM. These compounds also possessed good antifungal activity having more than 10 mm zone of inhibition at MIC 35.4–81.1 μM.

Antimycobacterial activity

Antimycobacterial evaluation was performed according to agar diffusion method in vitro. Isoniazid was used as reference antitubercular drug. The results of antimycobacterial activities and MIC's of compound **3a–k** against *M. tuberculosis* $H_{37}Rv$ are summarized in Table 3. The tested compounds showed activities against *M. tuberculosis* $H_{37}Rv$ with MIC values ranging from 88.5 to 411.1 μM. Compound **3a** containing bromo group at meta position showed inhibition at MIC = 88.5 μM; compounds **3b**, **3c** and **3d** containing chloro group at para, meta and ortho positions showed good inhibition at MIC = 101.4 μM. Compounds **3e** and **3f** containing methoxy group at para and meta positions, showed inhibition at MIC = 411.1 μM. Compounds **3g** and **3i** containing two methoxy group at meta, para and ortho, para positions showed inhibition with MIC = 187.1 μM. Compound **3k** containing nitro group at para position showed excellent inhibition with MIC = 48.04 μM. Compound **3h** containing three methoxy groups at 3,4,5 positions showed inhibition at MIC = 85.8 μM. Compound **3j** containing $–NH_2$ group at

Table 1 Synthesis of N′-substituted-4-methylbenzenesulfonohydrazide (**3a–k**)

Entry	Compd.	R	R^1	R^2	R^3	R^4	Time	Yield (%)
a	**3a**	H	H	Br	H	H	5 min (5 h)	92 (65)
b	**3b**	H	H	H	Cl	H	4 min (5 h)	93 (65)
c	**3c**	H	H	Cl	H	H	5 min (5 h)	92 (60)
d	**3d**	H	Cl	H	H	H	5 min (4.5 h)	92 (65)
e	**3e**	H	H	H	OMe	H	4 min (3 h)	96 (70)
f	**3f**	H	H	OMe	H	H	5 min (4 h)	92 (60)
g	**3g**	H	H	OMe	OMe	H	5 min (3.5 h)	94 (65)
h	**3h**	H	H	OMe	OMe	OMe	6 min (6 h)	90 (60)
i	**3i**	H	OMe	H	OMe	H	5 min (5 h)	94 (70)
j	**3j**	Me	H	H	NH_2	H	8 min (6 h)	90 (60)
k	**3k**	Me	H	H	NO_2	H	8 min (6 h)	90 (60)

Values in parenthesis indicate required time and yield obtained under conventional method

Table 2 Antimicrobial activity of compounds **3a–k**

Compd.	MIC values in μM (zone of inhibition, mm)			
	Antibacterial activity		Antifungal activity	
	S. aureus	E. coli	A. niger	C. albicans
3a	17.7 (14)	17.7 (12)	35.4 (10)	35.4 (12)
3b	20.2 (14)	40.5 (14)	40.5 (12)	40.5 (12)
3c	20.2 (12)	40.5 (12)	81.1 (10)	81.1 (10)
3d	20.2 (12)	20.2 (14)	40.5 (12)	40.5 (12)
3e	41.0 (12)	41.0 (12)	82.2 (12)	82.2 (10)
3f	82.1 (10)	82.1 (12)	164.4 (10)	164.4 (10)
3g	74.7 (10)	36.6 (14)	74.7 (12)	36.6 (14)
3h	137.2 (10)	137.2 (10)	137.2 (12)	137.2 (12)
3i	74.7 (12)	37.4 (12)	74.7 (12)	74.7 (12)
3j	165 (12)	165 (10)	165 (10)	165 (10)
3k	18.7 (14)	18.7 (14)	75.0 (12)	37.5 (10)
Ampicillin	16.8 (20)	16.8 (18)	–	–
Fluconazole	–	–	40.8 (18)	20.4 (16)

para position showed inhibition at MIC = 103 μM. In general, compounds containing electron-releasing groups, such as methoxy groups and electronegative group such as chloro led to increase in activity against *M. tuberculosis* (Pandit and Dodiya, 2013).

Experimental

Materials and methods

All the chemicals required for the synthesis were purchased from Sigma-Aldrich, E-Merck and HIMEDIA. Melting points were recorded in open capillaries. IR spectra were recorded on an FTIR-8400S SHIMADZU spectrophotometer using KBr pellets. The absorption bands were reported in cm^{-1}. ^1H NMR and ^{13}C NMR spectra were recorded on Bruker's NMR spectrometer at 300 MHz and 75 MHz, respectively, in $CDCl_3$/DMSO-d_6 using tetramethylsilane (TMS) as an internal standard (s = singlet, d = doublet, t = triplet, m = multiplates and br = broad). Chemical shifts were expressed in δ (ppm) relative to the TMS. Coupling constant (J) is written in (Hz). Mass Spectra were measured on Water's mass spectrometer. Elemental analysis was carried out on a Perkin Elmer CHNS/O Analyzer 2400. The progress of the reaction was monitored by TLC analysis using precoated silica aluminum plates (Merck Silica gel $^{60}F_{254}$), and spots were visualized under ultraviolet light irradiation (254 nm) or by exposure to iodine vapors. Column chromatography was performed over silica gel (Merck, particle size 60–120 mesh) using gradient mixture of ethyl acetate and hexanes as eluent.

Table 3 Antitubercular activity of compounds **3a–k**

Compound	MIC values (μM) of M. tuberculosis $H_{37}Rv$
3a	88.5
3b	101.4
3c	101.4
3d	101.4
3e	411.1
3f	411.1
3g	187.1
3h	85.8
3i	187.1
3j	103.0
3k	48.04
Isoniazid	3.6

*General procedure for preparation of **3a–k***

Conventional method

A mixture of 4-methylbenzenesulfonohydrazide **1** (1 mmol), aromatic carbonyls **2** (1 mmol) and AcOH (0.1 ml) in ethanol (10 ml) was heated under reflux for 3–6 h with continuous stirring. The progress of the reaction was monitored by TLC. After completion of the reaction, the reaction mixture was concentrated in vacuo. The crude product was purified by silica gel column chromatography using gradient mixture of ethyl acetate and hexanes as eluent. All the compounds were recrystallized from ethanol.

Green method

A mixture of 4-methylbenzenesulfonohydrazide **1** (1 mmol) and carbonyl compound **2** (1 mmol) in 20 % aqueous PSSA solution (1 ml) was exposed to microwave at 100 °C in a closed vessel for 4–6 min for aldehydes (and 8 min for ketones). The progress of the reaction was monitored by TLC analysis. After completion of reaction (as indicated by TLC), products were isolated by filtration and then recrystallized from ethanol (Fig. 1).

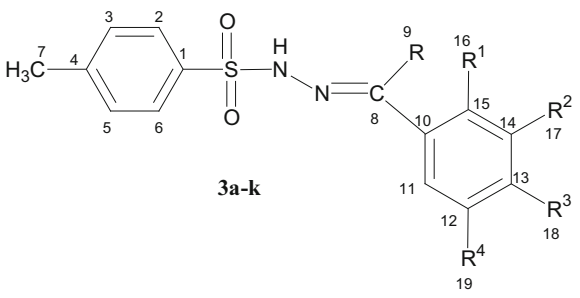


Fig. 1 Numbering for Carbon atoms in hydrazones

*N'-(3-bromobenzylidene)-4-methylbenzenesulfonohydrazide **(3a)*** Yellow powder; mp 135 °C; IR (KBr) υ_{max}, cm^{-1}: 3280 (NH), 3095 (Ar–H), 2955 (=CH), 1635 (C=N), 1345, 1150 (SO_2); ^{1}H NMR (300 MHz, $CDCl_3$): δ ppm 2.41 (s, 3H, CH_3), 7.19–7.24 (m, 1H, Ar–H), 7.32 (d, 2H, Ar–H, *J* 8.1), 7.45–7.49 (m, 2H, Ar–H), 7.72–7.73 (m, 1H, Ar–H), 7.86 (d, 2H, Ar–H, *J* 8.1), 7.69 (s, 1H, –N=CH–Ar), 8.27 (s, 1H, NH); ^{13}C NMR (75 MHz, $CDCl_3$): δ ppm 21.6 (CH_3, C-7), 122.8 (C, C-4), 126.0 (C, C-11), 127.9 (C, C-3, C-5), 129.8 (C, C-2, C-6), 129.8 (C, C-13), 130.1 (C, C-15), 133.2 (C, C-12), 135.1 (C, C-14), 135.2 (C, C-10), 144.4 (C, C-1), 145.9 (HC=N, C-8); MS (70 eV, *m/z*): 355.18 (M + 2). Anal. calcd for $C_{14}H_{13}BrN_2O_2S$: C, 47.60; H, 3.71; N, 7.93. Found: C, 47.15; H, 3.85; N, 8.02.

*N'-(4-chlorobenzylidene)-4-methylbenzenesulfonohydrazide **(3b)*** Yellow powder; mp 150 °C; IR (KBr) υ_{max}, cm^{-1}: 3270 (NH), 3105 (Ar–H), 2952 (=CH), 1645 (C=N), 1360, 1150 (SO_2); ^{1}H NMR (300 MHz, $CDCl_3$): δ ppm 2.35 (s, 3H, CH_3), 7.17–7.87 (m, 8H, Ar–H), 7.64 (s, 1H, –N=CH–Ar), 7.87 (s, 1H, NH); ^{13}C NMR (75 MHz, $CDCl_3$): δ ppm 20.9 (CH_3, C-7), 124.5 (C, C-3, C-5), 128.9 (C, C-2, C-6), 129.3 (C, C-4), 130.2 (C, C-14), 131.5 (C, C-11, C-15), 136.1 (C, C-10), 136.7 (C, C-13), 141.1 (C, C-1), 155.2 (HC=N, C-8); MS (70 eV, *m/z*): 309.78 (M + 1). Anal. calcd for $C_{14}H_{13}ClN_2O_2S$: C, 54.46; H, 4.24; N, 9.07. Found: C, 54.25; H, 4.29; N, 8.93.

*N'-(3-chlorobenzylidene)-4-methylbenzenesulfonohydrazide **(3c)*** Yellow powder; mp 144 °C; IR (KBr) υ_{max}, cm^{-1}: 3275 (NH), 3100 (Ar–H), 2960 (=CH), 1640 (C=N), 1340, 1140 (SO_2); ^{1}H NMR (300 MHz, $CDCl_3$): δ ppm 2.18 (s, 3H, CH_3), 7.29–7.86 (m, 8H, Ar–H), 7.55 (s, 1H, –N=CH–Ar), 8.21 (s, 1H, NH); ^{13}C NMR (75 MHz, $CDCl_3$): δ ppm 20.2 (CH_3, C-7), 129.4 (C, C-15), 131.5 (C, C-13), 127.5 (C, C-4), 124.9 (C, C-3, C-4), 128.8 (C, C-2, C-6), 130.2 (C, C-11), 132.6 (C, C-12), 133.8 (C, C-14), 135.3 (C, C-10), 142.1 (C, C-1), 154.7 (HC=N, C-8); MS (70 eV, *m/z*): 309.75 (M + 1). Anal. calcd for $C_{14}H_{13}ClN_2O_2S$: C, 54.46; H, 4.24; N, 9.07. Found: C, 54.25; H, 4.35; N, 8.99.

*N'-(2-chlorobenzylidene)-4-methylbenzenesulfonohydrazide **(3d)*** Yellow powder; mp 132 °C; IR (KBr) υ_{max}, cm^{-1}: 3270 (NH), 3100 (Ar–H), 2965 (=CH), 1648 (C=N), 1355, 1160 (SO_2); ^{1}H NMR (300 MHz, $CDCl_3$): δ ppm 2.42 (s, 3H, CH_3), 7.23–7.35 (m, 5H, Ar–H), 7.86–7.93(m, 3H, Ar–H), 7.96 (s, 1H, –N=CH–Ar), 8.16 (s, 1H, NH); ^{13}C NMR (75 MHz, $CDCl_3$): δ ppm 20.1 (CH_3, C-7), 124.5 (C, C-3, C-5), 126.8 (C, C-4), 128.2 (C, C-2, C-6), 129.2 (C, C-14), 130.5 (C, C-11), 131.6 (C, C-12), 132.2 (C, C-13), 135.1 (C, C-15), 136.1 (C, C-10), 141.2 (C, C-1), 156.1 (HC=N, C-8); MS (70 eV, *m/z*): 309.79 (M + 1). Anal. calcd for $C_{14}H_{13}ClN_2O_2S$: C, 54.46; H, 4.24; N, 9.07. Found: C, 54.98; H, 4.45; N, 9.39.

N'-(4-methoxybenzylidene)-4-methylbenzenesulfonohydrazide ***(3e)*** Red crystal; mp 105 °C; IR (KBr) υ_{max}, cm^{-1}: 3265 (NH), 3045 (Ar–H), 2868 (=CH), 1628 (C=N), 1370, 1160 (SO_2); ^1H NMR (300 MHz, $CDCl_3$): δ ppm 2.39 (s, 3H, CH_3), 3.81 (s, 3H, OCH_3), 6.82–6.89 (m, 2H, Ar–H), 7.29 (d, 2H, Ar–H, *J* 8.1), 7.48–7.54 (m, 2H, Ar–H), 7.86 (d, 2H, Ar–H, *J* 8.1), 7.73 (s, 1H, –N=CH–Ar), 8.10 (s, 1H, NH); ^{13}C NMR (75 MHz, $CDCl_3$): δ ppm 21.5 (CH_3, C-7), 55.3 (OCH_3, C-18), 114.0 (C, C-12, C-14), 122.4 (C, C-4), 127.9 (C, C-3, C-5), 129.0 (C, C-11, C-15), 129.6 (C, C-2, C-6), 135.2 (C, C-10), 144.1 (C, C-1), 148.5 (HC=N, C-8), 162.5 (C, C-13); MS (70 eV, *m/z*): 305.39 (M + 1). Anal. calcd for $C_{15}H_{16}N_2O_3S$: C, 59.19; H, 5.30; N, 9.20. Found: C, 58.90; H, 5.21; N, 8.95.

N'-(3-methoxybenzylidene)-4-methylbenzenesulfonohydrazide ***(3f)*** Yellow powder; mp 130 °C; IR (KBr) υ_{max}, cm^{-1}: 3265 (NH), 3060 (Ar–H), 2980 (=CH), 1620 (C=N), 1365, 1155 (SO_2); ^1H NMR (300 MHz, $CDCl_3$): δ ppm 2.33 (s, 3H, CH_3), 3.74 (s, 3H, OCH_3), 6.82–6.86 (m, 1H, Ar–H), 7.02–7.08 (m, 2H, Ar–H), 7.16 (s, 1H, Ar–H), 7.23 (d, 2H, Ar–H, *J* 8.4), 7.65 (s, 1H, –N=CH–Ar), 7.80 (d, 2H, Ar–H, *J* 8.4), 7.98 (s, 1H, NH); ^{13}C NMR (75 MHz, $CDCl_3$): δ ppm 21.1 (CH_3, C-7), 56.3 (OCH_3, C-17), 114.2 (C, C-13), 116.2 (C, C-15), 120.8 (C, C-11), 122.1 (C, C-4), 126.2 (C, C-3, C-5), 128.3 (C, C-12), 129.7 (C, C-2, C-6), 136.4 (C, C-10), 141.8 (C, C-1), 156.0 (HC=N,C-8), 162.1 (C, C-14); MS (70 eV, *m/z*): 305.12 (M + 1). Anal. calcd for $C_{15}H_{16}N_2O_3S$: C, 59.19; H, 5.30; N, 9.20. Found: C, 59.25; H, 5.15; N 9.32.

N'-(3,4-dimethoxybenzylidene)-4-methylbenzenesulfonohydrazide ***(3g)*** Yellow solid; mp 129 °C; IR (KBr) υ_{max}, cm^{-1}: 3260 (NH), 3058 (Ar–H), 2945 (=CH), 1625 (C=N), 1375, 1173 (SO_2); ^1H NMR (300 MHz, $CDCl_3$): δ ppm 2.23 (s, 3H, CH_3), 3.43 (s, 6H, OCH_3), 6.75–7.87 (m, 7H, Ar–H), 7.75 (s, 1H, –N=CH–Ar), 8.02 (s, 1H, NH); ^{13}C NMR (75 MHz, $CDCl_3$): δ ppm 20.0 (CH_3, C-7), 56.4 (OCH_3, C-17, C-18), 108.9 (C, C-12), 111.1 (C, C-15), 115.2 (C, C-11), 122.2 (C, C-4), 128.1 (C, C-3, C-5), 129.9 (C, C-2, C-6), 136.8 (C, C-10), 143.9 (C, C-1), 147.8 (C, C-14), 149.6 (C, C-13), 151.3 (HC=N, C-8); MS (70 eV, *m/z*): 335.41 (M + 1). Anal. calcd for $C_{16}H_{18}N_2O_4S$: C, 57.47; H, 5.43; N, 8.38. Found: C, 58.23; H, 5.65; N, 8.45.

N'-(3,4,5-trimethoxybenzylidene)-4-methylbenzenesulfonohydrazide ***(3h)*** Red powder; mp 142 °C; IR (KBr) υ_{max}, cm^{-1}: 3268 (NH), 3085 (Ar–H), 2940 (=CH), 1630 (C=N), 1360, 1160 (SO_2); ^1H NMR (300 MHz, $CDCl_3$): δ ppm 2.25 (s, 3H, CH_3), 3.74 (s, 9H, OCH_3), 6.76–7.81 (m, 6H, Ar–H), 7.76 (s, 1H, –N=CH–Ar), 8.05 (s, 1H, NH); ^{13}C NMR (75 MHz, $CDCl_3$): δ ppm 20.6 (CH_3, C-7), 56.7 (OCH_3, C-17, C-18, C-19), 107.9 (C, C-11, C-15), 125.5 (C, C-4), 125.8 (C, C-3, C-5), 129.6 (C, C-2, C-6), 135.5 (C, C-13), 137.3 (C, C-10), 141.4 (C, C-1), 148.8 (C, C-12, C-14), 154.9 (HC=N, C-8); MS (70 eV, *m/z*): 365.51 (M + 1). Anal. calcd for $C_{17}H_{20}N_2O_5S$: C, 56.03; H, 5.53; N, 7.69. Found: C, 55.90; H, 5.60; N, 7.75.

N'-(2,4-dimethoxybenzylidene)-4-methylbenzenesulfonohydrazide ***(3i)*** Yellow powder; mp 125 °C; IR (KBr) υ_{max}, cm^{-1}: 3270 (NH), 3040 (Ar–H), 2970 (=CH), 1620 (C=N), 1340, 1125 (SO_2); ^1H NMR (300 MHz, $CDCl_3$): δ ppm 2.35 (s, 3H, CH_3), 3.40 (s, 3H, OCH_3), 3.78 (s, 3H, OCH_3), 6.72–7.85 (m, 7H, Ar–H), 7.80 (s, 1H, =CH), 8.08 (s, 1H, NH); ^{13}C NMR (75 MHz, $CDCl_3$): δ ppm 20.1 (CH_3, C-7), 58.3 (-OCH_3, C-18), 64.5 (-OCH_3, C-16), 99.9 (C, C-14), 106.1 (C, C-12), 110.5 (C, C-11), 122.3 (C, C-4), 126.4 (C, C-3, C-5), 129.9 (C, C-2, C-6), 135.9 (C, C-10), 141.8 (C, C-1), 154.9 (HC=N, C-8), 164.2 (C, C-13), 168.2 (C, C-15); MS (70 eV, *m/z*): 335.42 (M + 1). Anal. calcd for $C_{16}H_{18}N_2O_4S$: C, 57.47; H, 5.43; N, 8.38. Found: C, 57.55; H, 5.30; N, 8.47.

N'-(1-(4-aminophenyl)ethylidene)-4-methylbenzenesulfonohydrazide ***(3j)*** Yellow powder; mp 188 °C; IR (KBr) υ_{max}, cm^{-1}: 3260 (NH), 3010 (Ar–H), 3050, 3450 (NH_2) 1615 (C=N), 1360, 1165 (SO_2); ^1H NMR (300 MHz, $CDCl_3$): δ ppm 2.10 (s, 3H, CH_3), 2.32 (s, 3H, CH_3), 2.5 (s, 2H, NH_2), 6.70–7.77 (m, 8H, Ar–H), 10.2 (s, 1H, NH); ^{13}C NMR (75 MHz, $CDCl_3$): δ ppm 17.7 (CH_3, C-9), 21.1 (CH_3, C-7), 126.2 (C, C-3, C-5), 121.2 (C, C-4), 115.3 (C, C-11, C-15), 130.7 (C-2, C-6), 129.9 (C, C-12, C-14), 136.5 (C, C-10), 141.5 (C, C-1), 149.5 (C, C-13), 155.6 (H_3C-C=N, C-8); MS (70 eV, *m/z*): 334.41 (M + 1). Anal. calcd for $C_{15}H_{17}N_3O_2S$: C, 59.38; H, 5.65; N, 13.85. Found: C, 59.48; H, 5.58; N, 13.94.

N'-(1-(4-nitrophenyl)ethylidene)-4-methylbenzenesulfonohydrazide ***(3k)*** Orange powder; mp 173 °C; IR (KBr) υ_{max}, cm^{-1}: 3282 (NH), 3120 (Ar–H), 1645 (C=N), 1350, 1145 (SO_2), 1520, 1325 (NO_2); ^1H NMR (300 MHz, DMSO-d_6): δ ppm 2.01 (s, 3H, CH_3), 2.35 (s, 3H, CH_3), 7.20–7.99 (m, 8H, Ar–H), 9.75 (s, 1H, NH); ^{13}C NMR (75 MHz, DMSO-d_6): δ ppm 17.8 (CH_3, C-9), 21.2 (CH_3, C-7), 121.5 (C, C-4), 123.8 (C, C-3, C-5), 124.7 (C, C-11, C-15), 128.3 (C, C-2, C-6), 129.9 (C, C-12, C-14), 137.5 (C, C-10), 142.0 (C, C-1), 150.8 (C, C-13), 157.1 (H_3C-C=N, C-8); MS (70 eV, *m/z*): 304.21 (M + 1). Anal. calcd for $C_{15}H_{15}N_3O_4S$: C, 54.04; H, 4.54; N, 12.60. Found: C, 54.15; H, 4.85; N, 12.32.

Antibacterial activity

The antibacterial activities of the synthesized compounds **3a–k** were determined by agar diffusion method as

recommended by the National Committee for Clinical Laboratory Standards (NCCLS) (NCCLS, 1992, 1997, 1998) against selected Gram-positive bacteria, *Staphylococcus aureus* (MTCC 3160), and Gram-negative bacteria, *Escherichia coli* (MTCC 1650) strains. One hundred microliters of broth culture containing test strain was spread over the nutrient agar plates. The wells of 6 mm diameter were dug on the nutrient agar plates and filled with respective test compounds separately. For comparison, DMSO and ampicillin were used as a solvent control and as a reference antibacterial agent, respectively. Triplicate plates of each microorganism were prepared, and inoculated plates were incubated at 37 ± 2 °C for 24 h, and the resulting zones of inhibition (in mm) were measured. The minimum inhibitory concentrations, at which no growth was observed, were taken as the MIC value.

Antifungal activity

The compounds were screened for their antifungal activity on the fungal strains *Aspergillus niger* (MTCC 1781) and *Candida albicans* (MTCC 227). One hundred microliters of fungal suspension was spread on potato dextrose agar (PDA) plates. The wells of 6 mm diameter were dug on the inoculated plates and filled with respective test compounds separately. For comparison, DMSO and Fluconazole were used as solvent control and reference antifungal agent, respectively. Inoculated plates were incubated at 28 ± 2 °C for 48 h, and the resulting zones of inhibition (in mm) were measured. The minimum inhibitory concentrations at which no fungal growth was observed were recorded as the MIC value.

Antitubercular activity

The primary screening was conducted at 8 μg/mL against *M. tuberculosis* $H_{37}Rv$ (ATCC 27294) by agar diffusion method (Gautam *et al.*, 2007). Stock solutions of individual compounds were prepared in DMSO. A sterile 6-mm borer was used to bore holes into the inoculums-seeded solidified nutrient agar. Twenty-five microliters of individual compounds was loaded into the labeled well in the prepared media plate using sterile pipette. The test was performed in three parallel experiments. The plates were kept in refrigerator for prediffusion of the sample and incubated at 37 °C for 48 h. Growth of test organism was observed after 48 h, and the zone of inhibition was measured in mm. The antimycobacterial activity of isoniazid was also simultaneously demonstrated. The MIC values of the synthesized compounds were determined using the broth microdilution assay against the test *Mycobacterium* strains.

Conclusion

In summary, a rapid and green method for the synthesis of tosylhydrazone derivatives has been developed. The synthesized compounds were screened for antituberculosis and antimicrobial activities. Compounds **3a–d** and **3k** showed good activity against *M. tuberculosis* $H_{37}Rv$ and for all microbial strains tested. The results suggest that N′-substituted-4-methylbenzenesulfonohydrazides might be interesting templates for generating effective antimycobacterial and antimicrobial lead molecules.

Acknowledgments Authors thank the Head, Department of Chemistry, University of Rajasthan, for her support. Sunita Ghiya thanks UGC, New Delhi, for a Ph.D. fellowship.

References

Bazgir A (2006) Microwave-assisted efficient synthesis of diimines in dry media using silica gel supported sodium hydrogen sulfate as reusable solid support. J Chem Res 1:1–2

Bedia KK, Elcin O, Seda U, Fatma K, Nathaly S, Sevim R, Dimoglo A (2006) Synthesis and characterization of novel hydrazide-hydrazones and the study of their structure-antituberculosis activity. Eur J Med Chem 41:1253–1261

Billman JH, Tai KM (1958) Reduction of Schiff Bases. II. Benzhydrylamines and structurally related compounds. J Org Chem 23(4):535–553

Branchaud BP (1983) Studies on the preparation and reactions of tritylsulfenimines. J Org Chem 48(20):3531–3538

Cates LA, Rasheed MS (1984) Phosphorus GABA analogues as potential prodrugs. Pharm Res 1(6):271–274

Cave GWV, Raston CL, Scott JL (2001) Recent advances in solventless organic reactions: towards benign synthesis with remarkable versatility. Chem Commun 21:2159–2169

Chakraborti AK, Bhagat S, Rudrawar S (2004) Magnesium perchlorate as an efficient catalyst for the synthesis of imines and phenylhydrazones. Tetrahedron Lett 45(41):7641–7644

Chunha AC, Tributino JLM, Miranda ALP, Fraga CAM, Barreiro EJ (2002) Synthesis and pharmacological evaluation of novel antinociceptive N-substituted-phenylimidazolyl-4-acylhydrazone derivatives. Farmaco 57(12):999–1007

Cocco MT, Congiu C, Onnis V, Pusceddu MC, Schivo ML, Logu A (1999) Synthesis and antimycobacterial activity of some isonicotinoylhydrazones. Eur J Med Chem 34(12):1071–1076

Gautam RA, Saklani A, Jachak SM (2007) Indian medicinal plants as a source of antimycobacterial agents. J Ethnopharmacol 110:200–223

Imrie C, Kleyi P, Nyamori VO, Gerber TIA, Levendis DC, Look J (2007) Further solvent-free reactions of ferrocenylaldehydes: synthesis of 1,1′-ferrocenyldiimines and ferrocenylacrylonitriles. J Organomet Chem 692(16):3443

Joshi SD, Vagdevi HM, Vaidya VP, Gadaginamath GS (2008) Synthesis of new 4-pyrrol-1-yl benzoic acid hydrazide analogs and some derived oxadiazole, triazole and pyrrole ring systems: a novel class of potential antibacterial and antitubercular agents. Eur J Med Chem 43:1989–1996

Karah N, Kocabalkanh A, Gursoy A, Ates O (2002) Synthesis and antitubercular activity of 4-(3-coumarinyl)-3-cyclohexyl-4-thiazolin-2-one benzylidenehydrazones. Farmaco 57(7):589–593

Koçyiğit KB, Rollas S (2002) Synthesis, characterization and evaluation of antituberculosis activity of some hydrazones. Farmaco 57(7):595–599

Kucukguzel SG, Rollas S, Kucukguzel I, Kiraz M (1999) Synthesis and antimycobacterial activity of some coupling products from 4-aminobenzoic acid hydrazones. Eur J Med Chem 34(12): 1093–1100

Landge SM, Atanassova V, Thimmaih M, Torok B (2007) Microwave-assisted oxidative coupling of amines to imines on solid acid catalysts. Tetrahedron Lett 48(29):5161–5164

Liete ACL, Moreira DRM, Coelho LCD, Menezes FD, Brondani DJ (2008) Synthesis of aryl-hydrazones via ultrasound irradiation in aqueous medium. Tetrahedron Lett 49(9):1538–1541

Maccari R, Ottana R, Vigorita MG (2005) In vitro advanced antimycobacterial screening of isoniazid-related hydrazones, hydrazides and cyanoboranes: part 14. Bioorg Med Chem Lett 15(10):2509–2513

Metger JO (1998) Solvent-free organic syntheses. Angew Chem Int Ed 37(21):2975

National Committee for Clinical Laboratory Standards (1992) Methods for determining bactericidal activity of antimicrobial agents: tentative Guidelines, Villanova. NCCLS (Publication no NCCLS M 26-T)

National Committee for Clinical Laboratory Standards (1997) Methods for antimicrobial susceptibility testing anaerobic bacteria, Approved Standard-Fourth Edition. Villanova, NCCLS, (Publication no NCCLS M 11-A4)

National Committee for Clinical Laboratory Standards (1998) Performance standards for antimicrobial susceptibility testing. Eighth information supplement, Villanova. NCCLS (Publication no NCCLS M 100-58)

Negi VJ, Sharma AK, Negi JS, Ram V (2012) Biological activities of hydrazone derivatives in the new millennium. IJPC 2(4):100–109

Oleveria KN, Chiaradia LD, Martins PGA, Mascarello A, Cordeiro MNS, Guido RVC, Andricopulo AD, Yunes RA, Nunes RJ, Vernal J, Terenzi H (2011) Sulfonyl-hydrazones of cyclic imides derivatives as potent inhibitors of the *Mycobacterium tuberculosis* protein tyrosine phosphatase B (PtpB). Med Chem Commun 2:500–504

Pandit U, Dodiya A (2013) Synthesis and antitubercular activity of novel pyrazole-quinazolinone hybrid analogs. Med Chem Res 22:3364–3371

Patole J, Sandbhor U, Padhye S, Deobagkar DN, Anson CE, Powell A (2003) Structural chemistry and in vitro antitubercular activity of acetylpyridine benzoyl hydrazone and its copper complex against Mycobacterium smegmatis. Bioorg Med Chem Lett 13(1):51–55

Polshettiwar V, Verma RS (2007) Polystyrene sulfonic acid catalyzed greener synthesis of hydrazones in aqueous medium using microwaves. Tetrahedron Lett 48(32):5649–5652

Rando DG, Sato DN, Siqueira L, Malvezzi A, Leite CQF, Amaral AT, Ferreira EI, Tavares LC (2002) Potential tuberculostatic agents. Topliss application on benzoic acid [(5-nitro-thiophen-2-yl)-methylene]-hydrazide series. Bioorg Med Chem 10(3):557–560

Saingar S, Kumar R, Joshi YC (2011) Synthesis and biological activity of novel 1*H*-1,4-diazepines containing benzene sulfonyl piperazine moiety. Med Chem Res 20:975–980

Savini L, Massarelli P, Chiasserini L, Sega A, Pellerano C, Brazi A, Nocentini G (1995) Chelating agents as potential antitumorals 2-quinolylhydrazones and bis-2-quinolylhydrazones. Eur J Med Chem 30:547–552

Savini L, Massarelli P, Travagli V, Pellerano C, Novellino E, Cosentino S, Pisano MB (2004) New alpha-(N)-heterocyclichydrazones: evaluation of anticancer, anti-HIV and antimicrobial activity. Eur J Med Chem 39(2):113–122

Scior T, Meneses Morales I, Garces Eisele SJ, Domeyer D, Laufer S (2002) Antitubercular isoniazid and drug resistance of *Mycobacterium tuberculosis*—a review. Arch Pharm (Weinheim) 335:511–525

Sondhi SM, Dinodia M, Kumar A (2006) Synthesis, anti-inflammatory and analgesic activity evaluation of some amidine and hydrazone derivatives. Bioorg Med Chem 14(13):4657–4663

Sridhar SK, Ramesh A (2001) Synthesis and pharmacological activities of hydrazones, Schiff and Mannich bases of isatin derivatives. Bio Pharm Bull 24(10):1149–1152

Tripathi RP, Tewari N, Dwivedi N, Tiwari VK (2005) Fighting tuberculosis: an old disease with new challenges. Med Res Rev 25:93–131

Vaas RS, Dudas J, Varma RS (1999) Solvent-free synthesis of *N*-sulfonylimines using microwave irradiation. Tetrahedron Lett 40(27):4951–4954

White WA, Weingart H (1967) A versatile new enamine synthesis. J Org Chem 32(1):213–214