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Der ChemicaSinica, 2015, 6(2):29-34

Der Chemica Sinica
Pelagia Research Library
ISSN: 0976-8505
CODEN (USA) CSHIA5

Synthesis, characterization and antimicrobial evaluation of some novel benzenesulfonylhydrazone derivatives of benzimidazole

Prakash Mehta[1], Pankaj Chovatiya[2] and Hitendra S. Joshi[2]*

[1]*Home Science Department, Dr.SubhashMahilaArts,Commerce& Home Science College, Junagadh-362001, Gujarat, India.*
[2]*Department of Chemistry, Saurashtra University, Rajkot-360005, Gujarat,India.*

ABSTRACT

*In the current research work, the synthesis of some new 2-(1H-benzo[d]imidazol-2-yl)-N-(4-(N'-substituted-phenylsulfonyl)benzohydrazide)benzamide derivatives(**5a-5m**) is described.The novel compounds were characterized using different spectroscopic methods. The new compounds were evaluated for antibacterial and antifungal activity. Assessment of the antibacterial andantifungal activities of these novel compounds against a different cell cultures revealed that compound **5d, 5k** and **5m** are the active ones.*

Keywords: benzenesulfonamide, benzimidazole,antibacterial activity, antifungal activity

INTRODUCTION

In recent years, attention has increasingly been given to the synthesis of benzimidazole derivatives. The synthesis of novel benzimidazole derivatives remains a main focus of medicinal research. Benzimidazoles are regarded as a promising class of bioactive heterocyclic compounds that exhibit a range of biological activities. The benzimidazole ring is an important pharmacophore in modern drug discovery. Indeed, a number of important drugs used in differenttherapeutic areas contain the benzimidazole ring,as proton pumpinhibitors (omeprazole and lansoprazole), antihistaminic (astemizole), antihypertensives (candesartan, telmisartan),antihistaminics (astemizole), antihelmintics (albendazole, mebendazole and flubendazole).Many derivatives of benzimidazole show antiparasitic and antiprotozoal activities. In recent years, benzimidazole derivatives have attracted particular interest due to their anticancer activity. Benzimidazoles showed anticancer activity against DNA topoisomerase I and colon cancer cell lines[1].benzimidazole and its derivatives have received much attention due to their chemotherapeutic values. benzimidazole is a structural isosters of naturally occurring nucleotide, due to which it interact easily with the biopolymers of living system. Benzimidazole has been used as a lead structure and part of central scaffold in some important drugs. Benzimidazole derivatives have found the appreciation in diverse therapeutic areas including antimicrobial[2], antioxidant[3], anthelmintic[4], anticancer[5], antihypertensive[6], anti-inflammatory[7], analgesic[8], antiprotozoal[9], anti-hepatitis B virus[10], antiulcer[11], antiviral[12], antifungal[13] and anticonvulsant[14] activity.

They are of wide interest because of their diversified biological activity and clinical applications. Thus, the aim of the current study was to synthesize novel benzimidazole derivatives that incorporated different heterocycles, such as different compounds with the backbone of sulphonamide linkage.sulphonamide linkage in such compounds strongly affects their properties. Aimed at the synthesis of benzimidazolescontaining sulphonamide, we developed a method for preparing 2-(1H-benzo[d]imidazol-2-yl)-N-(4-(N'-substituted-phenylsulfonyl)benzohydrazide) benzamide (see Table I for physical data).A variety of biological activities regarding benzimidazole core have led us to explore benzimidazole chemistry by synthesizing its derivatives bearing different sulphonamide system.

MATERIALS AND METHODS

Melting points were taken in open capillary method and are uncorrected. IR spectra were recorded on FTIR-8400 spectrophotometer (Shimadzu, Kyoto, Japan), using DRS prob. KBr pallet. ^{1}H-NMR spectra of the synthesized compounds were recorded on a Bruker-Avance-II (400 MHz) DMSO-d_6 solvent. Chemical shifts are expressed in δ ppm downfield from TMS as an internal standard. Mass spectra were determined using direct inlet probe on a GCMS-QP 2010 mass spectrometer (Shimadzu, Kyoto, Japan). The purity of the compounds were checked by thin layer chromatography (TLC) on silica gel plates usingHexane: Ethyl acetate as eluent and spots were developed in UV. Physical constants of the synthesized compounds **5a-5m** are shown in Table 1.

2.1Experimental

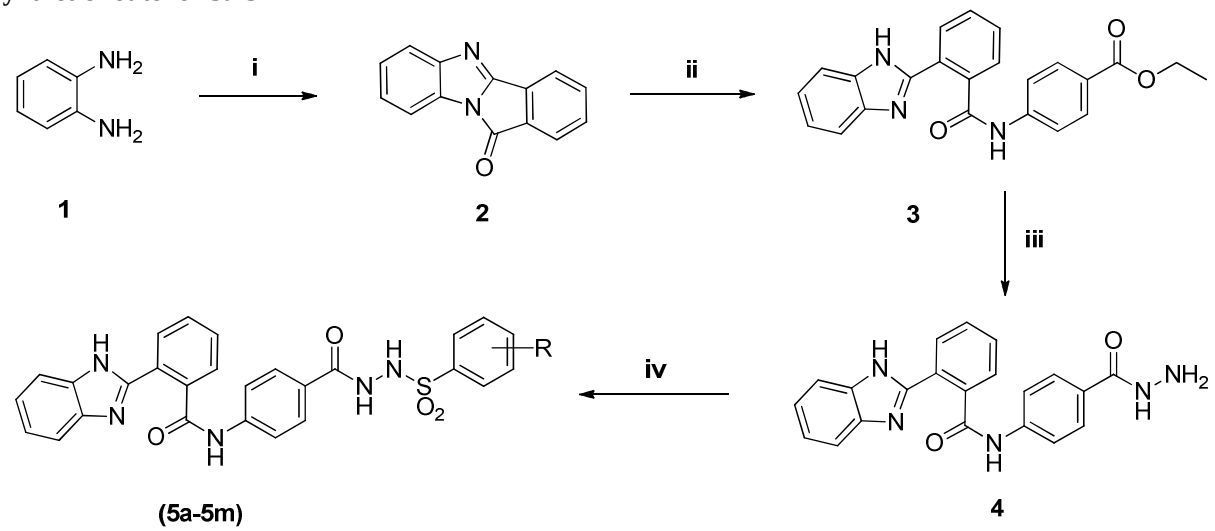
2.1.1Procedure for synthesis of 11H-benzo[4,5]imidazo[2,1-a]isoindol-11-one (2)*[15-17]*:An equimolar amount of pthalic anhydride and o-phenylenediamine(**1)** were taken in RBF. Fusion reaction was carried out at 140-150 °C to obtained o-Benzoylene 2-1-benzimidazole. Reaction mass was poured in chilled water and solid obtained was collected and dried in vacuo. Yield 79%

2.1.2 Procedure for synthesis of ethyl 4-(2-(1H-benzo[d]imidazol-2-yl)benzamido)benzoate (3) *[15-17]:*A mixture of o-benzoylene 2-1-benzimidazole(**2**) (1 mmol) and benzocaine (1 mmol) were refluxed for 4-5 hours in DMF at 150 °C. Completion of reaction was monitored by TLC. The reaction was poured in chilled water and solid was filtered filter out the precipitate of crude product and dried in vacuo. Crystalline from DMSO to obtained analytical grade pure 2-o-(4'-carbethoxyphenyl amino carbonyl phenyl) benzimidazole. Yield 85 %

2.1.3 Procedure for synthesis of 2-(1H-benzo[d]imidazol-2-yl)-N-(4-(hydrazinecarbonyl)phenyl)benzamide (4) *[15-17]*:In ethanolic solution of 2-o-(4'-carbethoxyphenyl amino carbonyl phenyl)-benzimidazole(**3**) (1 mmol), hydrazine hydrate (10 mmol) was added and reflux overnight. Cool down the reaction and filter the precipitate product. Wash the product with chilled ethanol to collect the analytical pure grade 2-(1H-Benzimidazol-2-yl)-N-(4-hydrazinocarbonyl-phenyl) benzamide.Yield 80 %

2.1.4General preparation of 2-(1H-benzo[d]imidazol-2-yl)-N-(4-(N'-substitutedphenylsulfonyl)benzohydrazide)benzamide derivatives(5a-5m):A mixture of o-Benzimidazol-2-'yl-benzamido-p-benzoyl hydrazine (0.01M) and various benzene sulphonyl chloride (0.01M) was refluxed in dry pyridine for 4-5 hours. The solvent was distilled off and product was isolated and recrystallized from DMSO. Physical constantsof the synthesized compounds are shown in Table 1.

Synthetic route for 5a-5m



(i) Pthalic anhydride, reflux, 140-150 °C,5h; (ii) Benzocaine, DMF, 150 °C, 4-5h; (iii) NH_2NH_2, ethanol, reflux, overnight; (iv) Substituted Benzenesulphonyl chloride, dry pyridine, 4-5h.

Table 1: Physical Constant of 2-(1H-benzo[d]imidazol-2-yl)-N-(4-(N'-substituted-phenylsulfonyl)benzohydrazide) benzamide derivatives (5a-5m).

Entry	R	Molecular Formula	M.P. °C	Yield %
5a	H	$C_{27}H_{21}N_5O_4S$	208	63
5b	4-Methoxy	$C_{28}H_{23}N_5O_5S$	189	67
5c	3-Methoxy	$C_{28}H_{23}N_5O_5S$	135	59
5d	2-Nitro,4-trifluoromethyl	$C_{28}H_{19}F_3N_6O_6S$	217	62
5e	3,4-Dimethoxy	$C_{29}H_{25}N_5O_6S$	247	69
5f	4-Cyno	$C_{28}H_{20}N_6O_4S$	193	73
5g	4-Fluoro	$C_{27}H_{20}FN_5O_4S$	215	58
5h	4-Chloro	$C_{27}H_{20}ClN_5O_4S$	174	67
5i	4-Trifluoromethyl	$C_{28}H_{20}F_3N_5O_4S$	169	61
5j	4-Bromo	$C_{27}H_{20}BrN_5O_4S$	194	63
5k	4-Trifluoromethoxy	$C_{28}H_{20}F_3N_5O_5S$	105	67
5l	3,4-Dichloro	$C_{27}H_{19}Cl_2N_5O_4S$	212	59
5m	4-Fluoro,3-trifluoromethyl	$C_{28}H_{19}F_4N_5O_4S$	249	65

2.2 Analytical data

2.2.1 2-(1H-benzo[d]imidazol-2-yl)-N-(4-(2-(phenylsulfonyl)hydrazinecarbonyl)phenyl) benzamide (5a): IR (ν_{max} cm^{-1}, KBr): 3430 (NH-N str. (asym.)), 3320 (N-H str.), 3030 (C-H str.), 1640 (C=O str.), 1590 (C-N str.), 1330 (S=O str. (sym.)), 1190 (S=O str. (asym.)), 720 (NH-N str. (sym.)). ^{1}H NMR (400 Hz, DMSO-d_6) δ ppm: 9.15(s, 1H), 8.06(s, 1H), 7.22-7.86(m, 17H), 5.11(s, 1H), 2.18(s, 1H); EI^+ m/z: 511; Anal. Calcd.for $C_{27}H_{21}N_5O_4S$; C (62.10%), H (4.28%), N (12.93%), S (6.27%). Found: C (63.39%), H (4.14%), N (13.69%), S (6.27%).

2.2.2 2-(1H-benzo[d]imidazol-2-yl)-N-(4-(2-((4-methoxyphenyl)sulfonyl)hydrazinecarbonyl)phenyl)benzamide (5b): IR (ν_{max} cm^{-1}, KBr): 3430 (NH-N str. (asym.)), 3320 (N-H str.), 3030 (C-H str.), 2985 (C-H str. (asym.)), 2870 (C-H str. (sym.)), 1640 (C=O str.), 1590 (C-N str.), 1330 (S=O str. (sym.)), 1190 (S=O str. (asym.)), 720 (NH-N str. (sym.)). ^{1}H NMR (400 Hz, DMSO-d_6) δ ppm: 9.15(s, 1H), 8.09(s, 1H), 7.22-7.97(m, 16H), 5.24(s, 1H), 3.67(s, 3H), 1.89(s, 1H); EI^+ m/z: 541; Anal. Calcd.for$C_{28}H_{23}N_5O_5S$;C (62.10%), H (4.28%), N (12.93%), S (5.92%). Found: C (62.23%), H (4.02%), N (12.65%), S (5.12%).

2.2.3 2-(1H-benzo[d]imidazol-2-yl)-N-(4-(2-((3-methoxyphenyl)sulfonyl)hydrazinecarbonyl)phenyl)benzamide (5c): IR (ν_{max} cm^{-1}, KBr): 3430 (NH-N str. (asym.)), 3320 (N-H str.), 3030 (C-H str.), 1640 (C=O str.), 1590 (C-N str.), 1330 (S=O str. (sym.)), 1190 (S=O str. (asym.)), 720 (NH-N str. (sym.)). ^{1}H NMR (400 Hz, DMSO-d_6) δ ppm: 9.21(s, 1H), 8.04(s, 1H), 7.28-7.89(m, 16H), 5.16(s, 1H), 3.78(s, 3H), 2.23(s, 1H); EI^+ m/z: 541; Anal. Calcd.for$C_{28}H_{23}N_5O_5S$; C (61.35%), H (4.20%), N (13.25%), S (6.07%). Found: C (61.23%), H (4.19%), N (13.26%), S (6.12%).

2.2.4 2-(1H-benzo[d]imidazol-2-yl)-N-(4-(2-((2-nitro-4-(trifluoromethyl)phenyl)sulfonyl)hydrazinecarbonyl)phenyl)benzamide (5d): IR (ν_{max} cm^{-1}, KBr): 3430 (NH-N str. (asym.)), 3320 (N-H str.), 3150(O-H str.), 3030 (C-H str.), 2985 (C-H str. (asym.)), 2870 (C-H str. (sym.)), 1640 (C=O str.), 1590 (C-N str.), 1330 (S=O str. (sym.)), 1190 (S=O str. (asym.)), 720 (NH-N str. (sym.)). ^{1}H NMR (400 Hz, DMSO-d_6) δ ppm: 9.09(s, 1H), 8.78(s, 1H), 8.16(s, 1H), 7.22-7.86(m, 14H), 5.11(s, 1H), 2.26(s, 1H); EI^+ m/z: 624; Anal. Calcd.for$C_{28}H_{19}F_3N_6O_6S$; C (53.85%), H (3.07%), N (13.46%), S (5.13%). Found: C (53.12%), H (3.45%), N (13.32%), S (5.28%).

2.2.5 2-(1H-benzo[d]imidazol-2-yl)-N-(4-(2-((3,4-dimethoxyphenyl)sulfonyl)hydrazinecarbonyl)phenyl)benzamide (5e): IR (ν_{max} cm^{-1}, KBr): 3430 (NH-N str. (asym.)), 3320 (N-H str.), 3150(O-H str.), 3030 (C-H str.), 1640 (C=O str.), 1590 (C-N str.), 1330 (S=O str. (sym.)), 1190 (S=O str. (asym.)), 720 (NH-N str. (sym.)). ^{1}H NMR (400 Hz, DMSO-d_6) δ ppm: 9.36(s, 1H), 8.25(s, 1H), 7.22-7.86(m, 15H), 5.28(s, 1H), 3.89(s, 3H), 3.68(s, 3H), 1.89(s, 1H); EI^+ m/z: 571; Anal. Calcd.for$C_{29}H_{25}N_5O_6S$; C (60.94%), H (4.41%), N (12.25%), S (5.61%). Found: C (60.36%), H (4.46%), N (12.30%), S (5.59%).

2.2.6 2-(1H-benzo[d]imidazol-2-yl)-N-(4-(2-((4-cyanophenyl)sulfonyl)hydrazinecarbonyl)phenyl)benzamide (5f): IR (ν_{max} cm^{-1}, KBr): 3430 (NH-N str. (asym.)), 3320 (N-H str.), 3150(O-H str.), 3030 (C-H str.), 2985 (C-H str. (asym.)), 2870 (C-H str. (sym.)), 1640 (C=O str.), 1590 (C-N str.), 1330 (S=O str. (sym.)), 1190 (S=O str. (asym.)), 720 (NH-N str. (sym.)). ^{1}H NMR (400 Hz, DMSO-d_6) δ ppm: 9.32(s, 1H), 8.05(s, 1H), 7.22-7.97(m, 16H), 5.88(s, 1H), 2.73(s, 1H); EI^+ m/z: 536; Anal. Calcd.for$C_{28}H_{20}N_6O_4S$; C (62.68%), H (3.76%), N (15.66%), S (5.98%). Found: C (62.59%), H (3.71%), N (15.49%), S (5.87%).

2.2.7 2-(1H-benzo[d]imidazol-2-yl)-N-(4-(2-((4-fluorophenyl)sulfonyl)hydrazinecarbonyl)phenyl)benzamide (5g): IR (ν_{max} cm^{-1}, KBr): 3430 (NH-N str. (asym.)), 3320 (N-H str.), 3150(O-H str.), 3030 (C-H str.), 1640 (C=O

str.), 1590 (C-N str.), 1330 (S=O str. (sym.)), 1190 (S=O str. (asym.)), 720 (NH-N str. (sym.)). ^{1}H NMR (400 Hz, DMSO-d_6) δ ppm: 9.27(s, 1H), 8.31(s, 1H), 7.22-7.86(m, 16H), 5.31(s, 1H), 2.45(s, 1H); EI$^+$ m/z: 529; Anal. Calcd.for$C_{27}H_{20}FN_5O_4S$ l; C (61.24%), H (3.81%), N (13.23%), S (6.06%). Found: C (61.31%), H (3.78%), N (13.14%), S (6.16%).

2.2.8 2-(1H-benzo[d]imidazol-2-yl)-N-(4-(2-((4-chlorophenyl)sulfonyl)hydrazinecarbonyl)phenyl)benzamide (5h): IR (ν_{max} cm^{-1}, KBr): 3540(Ar-OH str.), 3430 (NH-N str. (asym.)), 3320 (N-H str.), 3150(O-H str.), 3030 (C-H str.), 1640 (C=O str.), 1590 (C-N str.), 1330 (S=O str. (sym.)), 1190 (S=O str. (asym.)), 720 (NH-N str. (sym.)). ^{1}H NMR (400 Hz, DMSO-d_6) δ ppm: 9.74(s, 1H), 8.58(s, 1H), 7.22-7.86(m, 16H), 4.86(s, 1H), 2.28(s, 1H); EI$^+$ m/z: 545; Anal. Calcd.for$C_{27}H_{20}ClN_5O_4S$; C (59.39%), H (3.69%), N (12.83%), S (5.87%). Found: C (59.41%), H (3.56%), N (12.88%), S (5.67%).

2.2.9 2-(1H-benzo[d]imidazol-2-yl)-N-(4-(2-((4-(trifluoromethyl)phenyl)sulfonyl)hydrazinecarbonyl)phenyl)benzamide (5i): IR (ν_{max} cm^{-1}, KBr): 3430 (NH-N str. (asym.)), 3320 (N-H str.), 3150(O-H str.), 3030 (C-H str.), 1640 (C=O str.), 1590 (C-N str.), 1450(-CH_3 bend.), 1330 (S=O str. (sym.)), 1190 (S=O str. (asym.)), 720 (NH-N str. (sym.)). ^{1}H NMR (400 Hz, DMSO-d_6) δ ppm: 10.9(s, 1H), 9.74(s, 1H), 8.65(s, 1H), 7.22-7.86(m, 16H), 5.84(s, 1H), 2.55(s, 3H), 2.48(s, 1H); EI$^+$ m/z: 579; Anal. Calcd.for$C_{28}H_{20}F_3N_5O_4S$; C (58.03%), H (3.48%), N (12.08%), S (5.53%). Found: C (57.79%), H (3.39%), N (11.89%), S (5.61%).

2.2.10 2-(1H-benzo[d]imidazol-2-yl)-N-(4-(2-((4-bromophenyl)sulfonyl)hydrazinecarbonyl)phenyl)benzamide (5j):IR (ν_{max} cm^{-1}, KBr): 3430 (NH-N str. (asym.)), 3320 (N-H str.), 3150(O-H str.), 3030 (C-H str.), 1640 (C=O str.), 1590 (C-N str.), 1450(-CH_3 bend.), 1330 (S=O str. (sym.)), 1190 (S=O str. (asym.)), 720 (NH-N str. (sym.)). ^{1}H NMR (400 Hz, DMSO-d_6) δ ppm: 9.84(s, 1H), 8.11(s, 1H), 7.22-7.86(m, 16H), 5.08(s, 1H), 2.35(s, 1H); EI$^+$ m/z: 589; Anal. Calcd.for$C_{27}H_{20}BrN_5O_4S$; C (54.92%), H (3.41%), N (11.86%), S (5.43%). Found: C (54.97%), H (3.36%), N (11.90%), S (5.36%).

2.2.11 2-(1H-benzo[d]imidazol-2-yl)-N-(4-(2-((3-(trifluoromethoxy)phenyl)sulfonyl)hydrazinecarbonyl)phenyl)benzamide (5k): IR (ν_{max} cm^{-1}, KBr): 3430 (NH-N str. (asym.)), 3320 (N-H str.), 3150(O-H str.), 3030 (C-H str.), 1640 (C=O str.), 1590 (C-N str.), 1450(-CH_3 bend.), 1330 (S=O str. (sym.)), 1190 (S=O str. (asym.)), 720 (NH-N str. (sym.)). ^{1}H NMR (400 Hz, DMSO-d_6) δ ppm: 9.15(s, 1H), 8.71(s, 1H), 7.22-7.86(m, 16H), 5.21(s, 1H), 2.48(s, 3H), 2.07(s, 1H); EI$^+$ m/z: 595; Anal. Calcd.for$C_{28}H_{20}F_3N_5O_5S$; C (56.47%), H (3.38%), N (11.76%), S (5.38%). Found: C (56.39%), H (3.41%), N (11.68%), S (5.42%).

2.2.12 2-(1H-benzo[d]imidazol-2-yl)-N-(4-(2-((3,4-dichlorophenyl)sulfonyl)hydrazinecarbonyl)phenyl)benzamide (5l): IR (ν_{max} cm^{-1}, KBr): 3430 (NH-N str. (asym.)), 3320 (N-H str.), 3030 (C-H str.), 1640 (C=O str.), 1590 (C-N str.), 1330 (S=O str. (sym.)), 1190 (S=O str. (asym.)), 720 (NH-N str. (sym.)). ^{1}H NMR (400 Hz, DMSO-d_6) δ ppm: 9.26(s, 1H), 8.17(s, 1H), 8.0(s, 1H), 7.33-7.98(m, 14H), 5.12(s, 1H), 2.63(s, 1H); EI$^+$ m/z: 579; Anal. Calcd.for$C_{27}H_{19}Cl_2N_5O_4S$; C (55.87%), H (3.30%), N (12.07%), S (5.52%). Found: C (55.78%), H (3.23%), N (12.02%), S (5.47%).

2.2.13 2-(1H-benzo[d]imidazol-2-yl)-N-(4-(2-((3-fluoro-4-(trifluoromethyl)phenyl)sulfonyl)hydrazinecarbonyl)phenyl)benzamide (5m): IR (ν_{max} cm^{-1}, KBr): 3430 (NH-N str. (asym.)), 3320 (N-H str.), 3030 (C-H str.), 1640 (C=O str.), 1590 (C-N str.), 1450(-CH_3 bend.), 1330 (S=O str. (sym.)), 1190 (S=O str. (asym.)), 720 (NH-N str. (sym.)). ^{1}H NMR (400 Hz, DMSO-d_6) δ ppm: 9.34(s, 1H), 8.19(s, 1H), 7.22-7.86(m, 15H), 5.25(s, 1H), 2.81(s, 1H); EI$^+$ m/z: 597; Anal. Calcd.for$C_{28}H_{19}F_4N_5O_4S$; C (56.28%), H (3.20%), N (11.72%), S (5.37%). Found: C (56.32%), H (3.17%), N (11.65%), S (5.41%).

RESULTS AND DISCUSSION

Antimicrobial evaluation

The purified products were screened for its antimicrobial activity. The nutrient agar borth was prepared by the usual method and was inoculated aseptically with 0.5 ml of 24hold subculture of *Bacillus megaterium, Stsphylococus citrus, Escherichia coli* and *Salmonella typhosa*in separate conical flask at 40-50^{o}C and mixed well by gentle shaking. About 25 ml of content of the flask were poured and evenly spread in petridish (13 cm in diameter) and allowed to set for two hrs. The cups (10 mm in diameter) were formed by the help of borer in agar medium and filled with 0.05 ml (0.5 mg/ml) solution of sample in DMF.

The plates were incubated at 37°C for 24h and the control was also maintained with 0.05 ml of DMF in similar manner. The zone of inhibition of the bacterial growth was measured in mm.*Aspergilliusniger* was employed for testing antifungal activity using cup-plate method. The culture was maintained in Sabouraud's agar slants. Sterilised Sabouraud's agar medium was inoculated with 72h old 0.5 ml suspension of fungal spores in a separate flask. About 25 ml of the inoculated medium was evenly spreaded on a sterilised petridish and allowed to set for 2h. The cups (10 mm in diameter) were punched in petridish and loaded with 0.5 ml of (0.5 mg/mL) solution of sample in DMF. The plates were inoculated at 30°C for 48h. After the completion of inoculation period the zone of inhibition of growth in form of diameter was measured in mm. along the test solution in each petridish one cup was filled with solvent which acts as control. The zone of inhibition was recorded in **Table 2.**

Table 2: Anti-microbial activity 2-(1H-benzo[d]imidazol-2-yl)-N-(4-(N'-substituted-phenylsulfonyl)benzohydrazide) benzamide derivatives (5a-5m).

Entry	Antibacterial Activity (Zone of inhibition in m.m.)				Antifungal Activity (Zone of inhibition in m.m.)
	Gram +ve Bacteria		Gram –ve Bacteria		
	B. Megaterium	S. Citrus	E. Coli	S. Typhosa	A. Niger
5a	12	14	12	12	16
5a	13	11	10	10	10
5b	11	12	11	14	14
5c	16	13	15	13	13
5d	15	10	14	11	12
5e	10	10	12	10	13
5f	11	13	15	12	16
5g	10	11	12	14	10
5h	10	16	10	13	10
5i	14	12	11	12	11
5j	13	16	13	13	10
5k	12	12	16	14	14
5l	16	14	13	10	13
5m	24	-	23	24	-
Chloramphenicol	26	-	-	23	-
Norfloxacin	23	24	21	-	-
Griseofulvin	-	-	-	-	24

CONCLUSION

In this present work, we have described the synthesis of a series of 2-(1H-benzo[d]imidazol-2-yl)-N-(4-(N'-substituted-phenylsulfonyl)benzohydrazide) benzamide derivatives. The synthesized compounds were characterized by ^{1}H NMR, Mass and IR spectroscopy and the obtained results are showing good agreement with the synthesized structures.Synthesized compounds screened for their biological study. Indeed, compound 5d, 5k & 5m proved good active and rest are moderately active as compare to standard dugs.

Acknowledgment
Authors are thankful to Department of Chemistry for providing laboratory facilities. The authors are also thankful for the facilities and grants given under UGC-SAP for Department Research Support (DRS) and Department of Science and Technology (DST) New Delhi for Fund for Improvement of Science and Technology (FIST). Authors are also thankful to RSIC Chandigarh for providing ^{1}H NMR &^{13}C NMR spectral analysis of the compounds.

Authors have no financial conflicts with anyone.

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