STUDIES ON NOVEL AZETIDINONES BASED ON BENZENE SULFONYLHYDRAZIDE

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Abstract

Benzene sulfonyl hydrazide (**1**) undergoes facile condensation with aromatic aldehydes **2(a-h)** afforded the corresponding N-arylidene benzene sulfonyl hydrazides (**3a-h**) in good yield. Cyclo condensation of these hydroxides (**3a-h**) with chloro acetyl chloride yielded N-(3-chloro-2-oxo-4-arylazetidin-1-yl) benzene sulfonamide (**4a-h**). The reaction of Sulfapyridine with N-(3-chloro-2-oxo-4-aryl azetidin-1-yl) benzene sulfonamide (**4a-h**) produce. 4-((2-aryl-4-oxo-1- (phenylsulfonamido)azetidin-3-yl) amino)-N-(pyridin-2-yl) benzenesulfonamide **(5a-h)**. The structure of these compounds were established on the basis of analytical and spectral data. All the produce compounds **(4a-h)** and **(5a-h)** were evaluated for their antibacterial and antifungal activities by agar cup method.

Keywords: Benzene sulfonyl hydrazine, Azetidinone, Schiff bases, Antibacterial activity, Antifungal activity and Spectral study.

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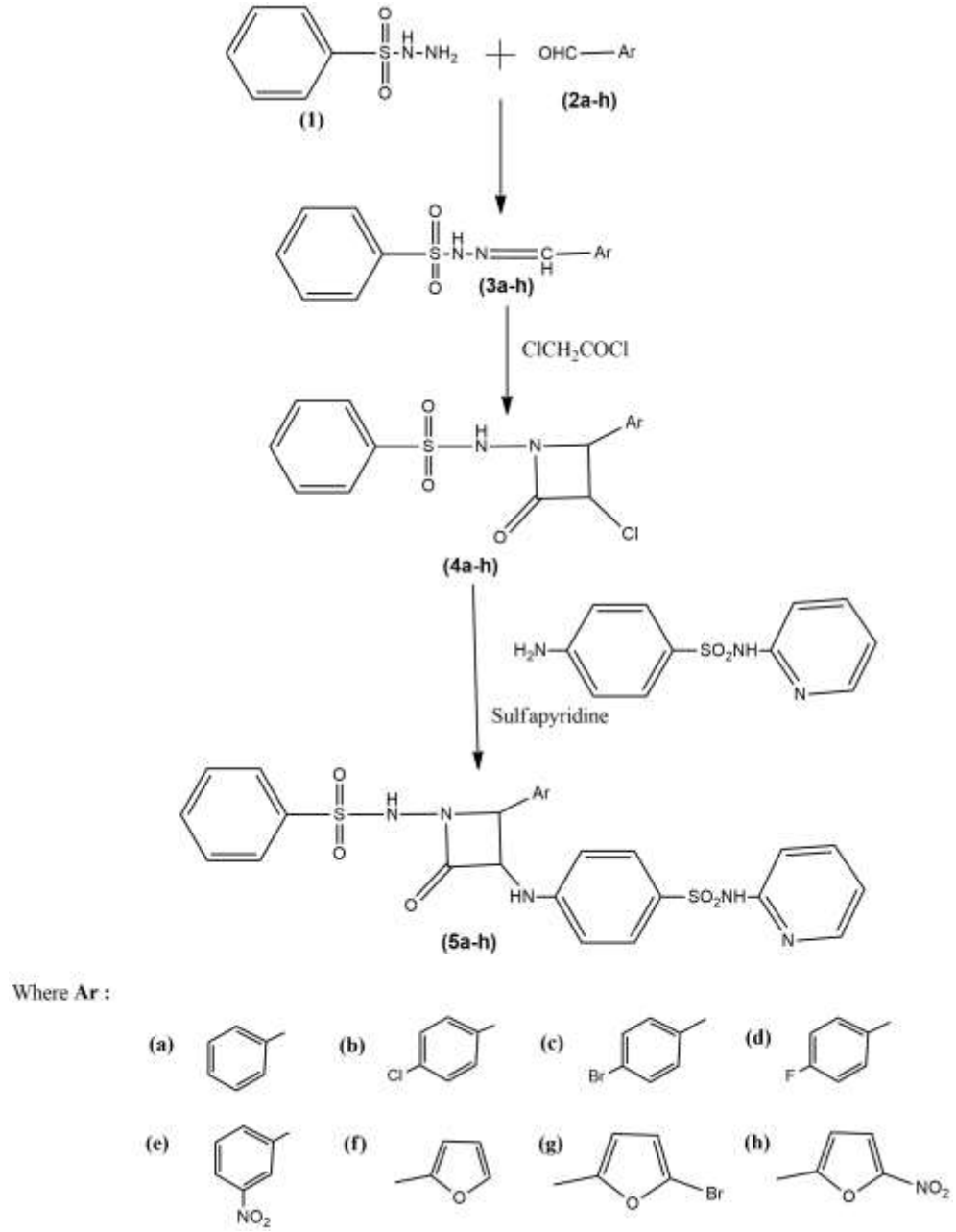
INTRODUCTION

The sulphonyl-hydrazide i.e. $–SO_2NHNH_2$ is a class of organic compound. Its chemistry received more interest in both synthetic organic chemistry and biological field. The aliphatic or aromatic sulfonyl hydrazides exhibit many pharmaceutical activities [1-3]. The aryl sulfonyl hydrazides are reported in literature [4-6]. The post reactions of aryl sulfonyl hydrazide towards various heterocyclic compounds have been reported for biological activity [7]. More particularly the hydrazone of aryl sulfonyl hydrazide having pharmaceutical activities like, anti T.B., anti alzhemer, antimicrobial, antineoplastic etc [8-10]. Other applications like metal chelation [11-15] also reported. The pyrrole derivatives based on sulfonyl hydrazide have been reported recently [16] as an anti T.B.

2-Azetidinones i.e. β-lactams are well known heterocyclic compounds for medicinal value [17-23]. These derivatives can be synthesized from Schiff bases. The Schiff bases of aryl sulfonyl hydrazide are reported [16]. But these transformation into β-lactams has not been reported. Also, post reaction of these β-lactams with sulfa drugs is not reported as sulfa drugs are well known antibacterial drugs. [24] Thus with the view of enhance biological properties present communication comprises the studies on 2-azitidinone derivatives based on benzene sulfonyl hydrazides and post reaction with sulfa pyridine (sulfa drugs). The research work is drawn as follow (Scheme-1).



EXPERIMENTAL

Melting points were determined in open capillary tubes and were uncorrected. The IR spectra were recorded in KBr pellets on a Nicolet 400D spectrometer and ^{1}H NMR and ^{13}CNMR spectra were recorded in DMSO with TMS as internal standard on a Bruker spectrometer at 400 MHz and 100 MHz, respectively. LC-MS of selected samples taken on LC-MSD-Trap-SL_01046. Elemental content of all compounds determined by Thermofinigun Flash EA (Italy).The sulfur and halogen determined by Carious method.

Preparation of N-arylidene benzene sulfon ohydrazide (3a-h):

The Schiff Bases (or Hydrazones) (3a-h) were prepared (Scheme-1) by refluxing solution of benzene sulfonyl hydrazide (1) and aromatic aldehyde derivatives (2a-h) at stochiometric ratio in THF solvent for 8 hr. The solvent THF was distilled under pressure. The solid was washed by water and used for post reaction. The detail analysis of 3a-h are furnished in Table -1.

Table: 1 Analytical Data and Elemental Analysis of Schiff bases (3a-h)

Compd.	Molecular formula (Mol.wt.)	Yield (%)	M.P.* ^{0}C	Elemental Analysis									
				%C		% H		%N		%S		%X (X=Cl,Br,F)	
				Found	Calcd.	Found	Calcd.	Found	Calcd.	Found	Calcd.	Found	Calcd.
3a	$C_{13}H_{12}N_2O_2S$ (260)	84	252	59.9	59.98	4.6	4.65	10.7	10.76	12.3	12.32	-	-
3b	$C_{13}H_{11}N_2O_2SCl$ (294.5)	76	237	52.9	52.97	3.7	3.76	12.0	12.03	10.8	10.88	11.9	12.05
3c	$C_{13}H_{11}N_2O_2SBr$ (339)	79	246	46.0	46.03	3.2	3.27	8.2	8.26	9.4	9.45	23.5	23.59
3d	$C_{13}H_{11}N_2O_2SF$ (278)	72	243	56.0	56.10	3.9	3.98	10.0	10.07	11.5	11.52	6.7	6.83
3e	$C_{13}H_{11}N_3O_4S$ (305)	70	244	51.1	51.14	3.6	3.63	13.7	13.76	10.4	10.50	-	-
3f	$C_{11}H_{10}N_2O_3S$ (250)	79	238	52.7	52.79	4.0	4.03	11.1	11.19	12.8	12.81	-	-
3g	$C_{11}H_9N_2O_3SBr$ (329)	81	242	40.1	40.14	2.7	2.76	8.5	8.51	9.7	9.74	24.2	24.27
3h	$C_{11}H_9N_3O_5S$ (295)	65	248	44.7	44.74	3.0	3.07	14.2	14.23	10.8	10.86	-	-

***** Uncorrected

Preparation of N-(3- chloro- 2- oxo-4- arylazet idin-1-yl)benzenesulfonamide (4a-h) :

A N-arylidene benzene sulfonohydrazide (**3a-h**) (0.002 mole) and triethyl amine (TEA) (0.004 mole) were dissolved in 1,4-dioxane (50 ml), cooled, and stirred. To this well-stirred cooled solution chloro acetyl chloride (0.004 mole) was added dropwise. The reaction mixture was stirred for 2-3 hours and left at room temperature for 3 days. The resultant mixture was concentrated, cooled, poured into ice water. Then filtered, washed by ethanol and air-dried. Dissolved in THF solvent and reprecipitated by rectified spirit. Yield white puffy powered of N-(3-chloro-2-oxo-4-arylazetidin-1-yl) benzene sulfonamide (**4a-h**). All the compounds were characterized by elemental contents and data are shown in Table-2.

Table:2 Analysis of 2-Azetidinone derivatives (4a-h)

Compd.	Molecular formula (Mol.wt.)	Yield (%)	M.P.* ^{0}C	Elemental Analysis									
				%C		% H		%N		%S		%X(X=Cl,Br,F)	
				Found	Calcd.	Found	Calcd.	Found	Calcd.	Found	Calcd.	Found	Calcd.
4a	$C_{15}H_{13}N_2O_3SCl$ (336)	66	285	53.4	53.49	3.8	3.89	8.3	8.32	9.5	9.52	10.5	10.53
4b	$C_{15}H_{12}N_2O_3SCl_2$ (371.5)	66	272	48.5	48.53	3.2	3.26	7.5	7.55	8.6	8.64	19.1	19.10
4c	$C_{15}H_{12}N_2O_3SClBr$ (415)	68	276	43.3	43.34	2.9	2.91	8.5	8.53	7.7	7.71	8.5 19.2	8.53 19.22
4d	$C_{15}H_{12}N_2O_3SFCl$ (354)	60	271	50.7	50.78	3.4	3.41	5.3	5.35	9.0	9.04	9.9 5.3	9.99 5.35
4e	$C_{15}H_{12}N_3O_5SCl$ (381)	62	274	47.1	47.19	3.1	3.17	11.0	11.01	8.3	8.40	9.2	9.29
4f	$C_{13}H_{11}N_2O_4SCl$ (413)	66	272	47.7	47.78	3.3	3.39	8.5	8.57	9.8	9.81	10.8	10.85
4g	$C_{13}H_{10}N_2O_4SClBr$ (403)	63	270	38.4	38.49	2.4	2.48	6.9	6.91	7.8	7.90	8.7 19.6	8.74 19.70
4h	$C_{13}H_{10}N_3O_6SCl$ (371)	61	268	41.9	42.00	2.7	2.71	11.2	11.30	8.6	8.63	9.5	9.54

***** Uncorrected

Preparation of 4-(2-substituted phenyl)-4-oxo-1-(phenyl sulfonamido) azetidin-3-ylamino)-N-(pyridin-2-yl) benzene sulfonamide (5a-h):

In a round bottom flask, add N-(3-chloro-2-oxo-4-arylazetidin-1-yl) benzene sulfonamide **(4a-h)** (0.02 mol), Sulfapyridine (0.02mol) in THF stirred until a clear solution resulted. Then K_2CO_3 (1.15 g) was added. The reaction mixture was refluxed with constant stirring. After 3 hours solid precipitate separates out in the reaction mixture; the reaction was allowed to continue for another hour after which the solid product was filtered, washed, and dried. Compounds were recrystallized with a mixture of acetone/acetonitrile. All the compounds were characterized by elemental contents and data are shown in Table-3.

The IR, NMR, LC-MS spectral features of compound 3a-h, 4a-h and 5a-h are interpreted commonly for their structural assignments. The details are given in results and discussion.

Table: 3 Analytical Data and Elemental Analysis of derivatives (5a-h)

Compd.	Molecular formula (Mol.wt.)	Yield (%)	M.P.* ^{0}C	Elemental Analysis									
				%C		% H		%N		%S		%X (X=Cl,Br,F)	
				Found	Calcd.	Found	Calcd.	Found	Calcd.	Found	Calcd.	Found	Calcd.
5a	$C_{26}H_{23}N_5O_5S_2$ (549)	64	252	56.8	56.82	4.2	4.22	12.7	12.74	11.6	11.67	-	-
5b	$C_{26}H_{22}N_5O_5S_2Cl$ (584.5)	62	257	53.4	53.47	3.7	3.80	11.9	11.99	10.9	10.98	6.0	6.07
5c	$C_{26}H_{22}N_5O_5S_2Br$ (628)	65	264	49.6	49.68	3.5	3.53	11.1	11.14	10.1	10.20	12.7	12.71
5d	$C_{26}H_{22}N_5O_5S_2F$ (567)	61	260	55.0	55.02	3.9	3.91	12.3	12.34	11.2	11.30	3.3	3.35
5e	$C_{26}H_{22}N_6O_7S_2$ (594)	58	248	52.5	52.52	3.7	3.73	14.1	14.13	10.7	10.79	-	-
5f	$C_{24}H_{21}N_5O_6S_2$ (539)	55	252	53.4	53.42	3.9	3.92	12.9	12.98	11.8	11.89	-	-
5g	$C_{24}H_{20}N_5O_6S_2Br$ (618)	66	266	46.6	46.61	3.2	3.26	11.3	11.32	10.3	10.37	12.9	12.92
5h	$C_{24}H_{20}N_6O_8S_2$ (584)	63	263	49.3	49.31	3.4	3.45	14.3	14.38	10.9	10.97	-	-

***** Uncorrected

RESULTS AND DISCUSSION

It was observed that benzenesulfonyl hydrazide (**2**) on condensation with aromatic aldehydes to yield N-arylidene benzene sulfonyl hydrazide (**3a-h**) [16].The structures of **(3a-h)** were confirmed by elemental analysis and IR spectra showing absorption band at 3030-3040 cm^{-1} (C-H, of Ar.), 3115 cm^{-1} (NH), 1630-1635 cm^{-1} (C=N),1235-1075,1380-1370 cm^{-1} (SO_2NH), 1085cm^{-1} (C-Cl), 710 cm^{-1} (C-Br), 1260 cm^{-1} (C-F) and 1550,1370 cm^{-1} (-NO_2). ^{1}HNMR :7.64–7.88(m,5H, Ar-H), 8.40(s,1H, NH),8.38(s,1H,CH), (a) 7.55-7.86(m,5H,Ar-H), (b) 7.55-7.80(m,4H,Ar-H), (c) 7.60-7.75(m,4H,Ar-H), (d) 7.39-7.82(m,4H,Ar-H), (e) 8.12 -8.35 (m,4H,Ar-H), (f) 6.55-7.78(m,3H,Furan-H), (g) 6.82-7.10(m,2H,Furan-H) ,(h) 7.12 -7.60 (m,2H,Furan-H).The C, H, N analysis data of all compounds are presented in Table -1.

The cyclocondensation of **(3a-h)** with chloro acetyl chloride resulted in formation of N-(3-chloro-2-oxo-4-aryl azetidin-1-yl) benzene sulfonamide (**4a-h**). The structures assigned to **(4a-h)** were supported by the elemental analysis and IR spectra showing absorption bands at 1740-1730 cm^{-1} (C=O of monocyclic β-lactam),3035-3030cm^{-1} (C-H, of Ar.), 3115 cm^{-1} (NH), 1235-1075,1380-1370 cm^{-1} (SO_2NH), 1342(C-N),1085cm^{-1} (C-Cl),710 cm^{-1} (C-Br),1260 cm^{-1} (C-F) and 1550.1370 cm^{-1} (C-NO_2). The ^{1}HNMR : 7.65–7.90 (m,5H,Ar-H), 8.40(s,1H,NH), 5.32 (d,1H,C_2-H),5.70(d,1H,C_3-H),(a)7.29-7.42 (m,5H, Ar-H),(b) 7.45-7.50 (m,4H,Ar-H), (c) 7.20-7.94 (m,4H,Ar-H), (d) 7.21-7.29(m,4H,Ar-H), (e) 7.68 -8.20 (m,4H,Ar-H), (f) 6.45-7.68 (m,3H,Furan-H), (g) 6.30-6.39(m,2H,Furan-H) ,(h) 6.82 -7.55 (m,2H,Furan-H).The C, H, N analysis data of all compounds are presented in Table -2.

The reaction of **(4a-h)** with Sulfapyridine resulted in formation of 4-((2-aryl-4-oxo-1-(phenyl sulfonamido) azetidin-3-yl)amino)-N-(pyridin-2-yl)benzene sulfonamide **(5a-h)**. The post reaction of chlorine of azetidinone has been reported in literature [25]. The structures assigned to **(5a-h)** were supported by the elemental analysis and IR spectra showing absorption bands at 1740-1730 cm^{-1} (C=O of mono cyclic β-lactam), 3035-3030cm^{-1}(C-H, of Ar.), 3115 cm^{-1} (NH), 1630-1635 cm^{-1} (C=N), 1235-1075, 1380-1370 cm^{-1} (SO_2NH), 1085cm^{-1} (C-Cl),710 cm^{-1} (C-Br),1260 cm^{-1} (C-F) and 1550,1370 cm^{-1} (C-NO_2). ^{1}HNMR :6.65–8.10(m,13H,Ar-H),8.40-8.42(s,2H,NH),4.25(s,1H,NH),5.32(d,1H,C_2-H), 5.70 (d,1H,C_3-H), (a) 7.29-7.42(m,5H,Ar-H),(b) 7.45-7.50(m,4H,Ar-H), (c) 7.20-7.94(m,4H,Ar-H),(d)7.21-7.29 (m,4H,Ar-H), (e) 7.68 -8.20 (m,4H,Ar-H), (f) 6.45-7.68(m,3H,Furan-H), (g) 6.30 -6.39 (m,2H,Furan-H) ,(h) 6.82 -7.55 (m,2H,Furan-H). The C, H, N analysis data of all compounds are presented in Table -3.

The examination of data reveals that the elemental contents are consistent with the predicted structure shown in scheme-1. IR/NMR spectral data assigned the predicted structure of all three series of compounds 3(a-h), 4(a-h), and 5(a-h).

The LC-MS of selected compounds 3b, 4b and 5b shows the peak of M+ ion peak at 295.1, 371.8 and 584.9 which is consistent of their corresponding molecular weight 294.5, 371.5 and 584.5 respectively. All these facts confirm the structures 3a-h, 4a-h and 5a-h. (Scheme-1)

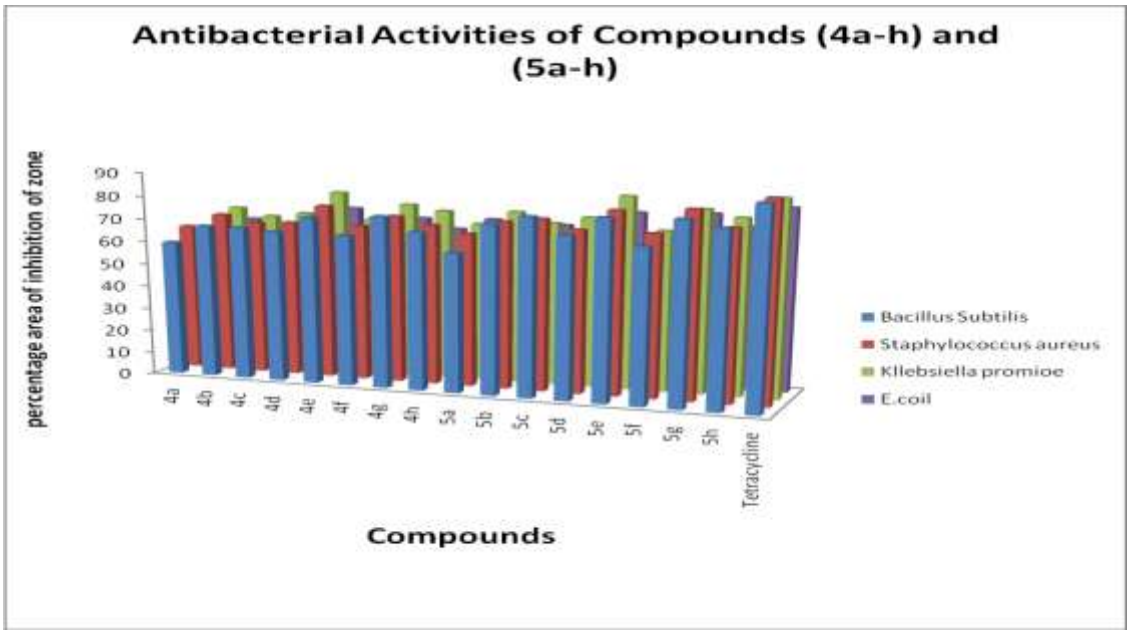
BIOLOGICAL SCREENING

Antibacterial Activities

Antibacterial activities of all the compounds were studied against gram-positive bacteria (*Staphylococcus aureus and Bacillus subtilis*) and gram-negative bacteria (*E.coli, and klebsiella promioe*) at a concentration of 50µg/ml by agar cup plate method [26- 28]. Methanol system was used as control in this method. Under similar condition using tetracycline as a standard for comparison carried out control experiment. The percentage area of inhibition of zone measured. All the compounds show good toxicity against bacteria. However, compounds 5e and 5g were found more active against the above microbes. Other compounds found to be less or moderate active than tetracycline (Table–4). The compounds **(5a-h)** are more toxic than **(4a-h).**

Table: 4 Antibacterial Activity of derivatives (4a-h) and (5a-h)

Compounds	Gram +Ve		Gram -Ve	
	Bacillus subtilis	***Staphylococcs aureus***	***Klebsiella promioe***	***E.coli***
4a	59	64	55	57
4b	67	70	71	64
4c	67	67	68	62
4d	66	68	70	63
4e	73	76	80	71
4f	66	68	69	60
4g	75	73	76	68
4h	69	70	74	64
5a	61	67	69	69
5b	74	73	75	70
5c	78	75	71	68
5d	71	71	74	70
5e	79	80	84	75
5f	68	71	70	62
5g	80	82	80	76
5h	77	75	77	71
Tetracycline	88	88	86	80



The results show that all the compounds display similar trend as studied for antibacterial activity.

Antifungal Activities

The fungicidal activity of all the compounds was studied by (agar cup method) at 1000 ppm concentration in vitro. Plant pathogenic organisms used were *Rhizopus nigricum***,** *Aspergillus niger, Fusarium oxyporium and Botrydepladia thiobromine*. The antifungal activity of all the compounds was measured on each of these plant pathogenic strains on a potato dextrose agar (PDA) medium. Such a PDA medium contained potato 200gm, dextrose 20gm, agar 20gm and water one liter. Five days old cultures were employed. The compounds to be tested were suspended (1000ppm) in a PDA medium and autoclaved at 120^{o} C for 15 min. at 15atm.pressure. These medium were poured into sterile Petri plates and the organisms were inoculated after cooling the Petri plates. The

percentage inhibition for fungi was calculated after five days using the formula given below:

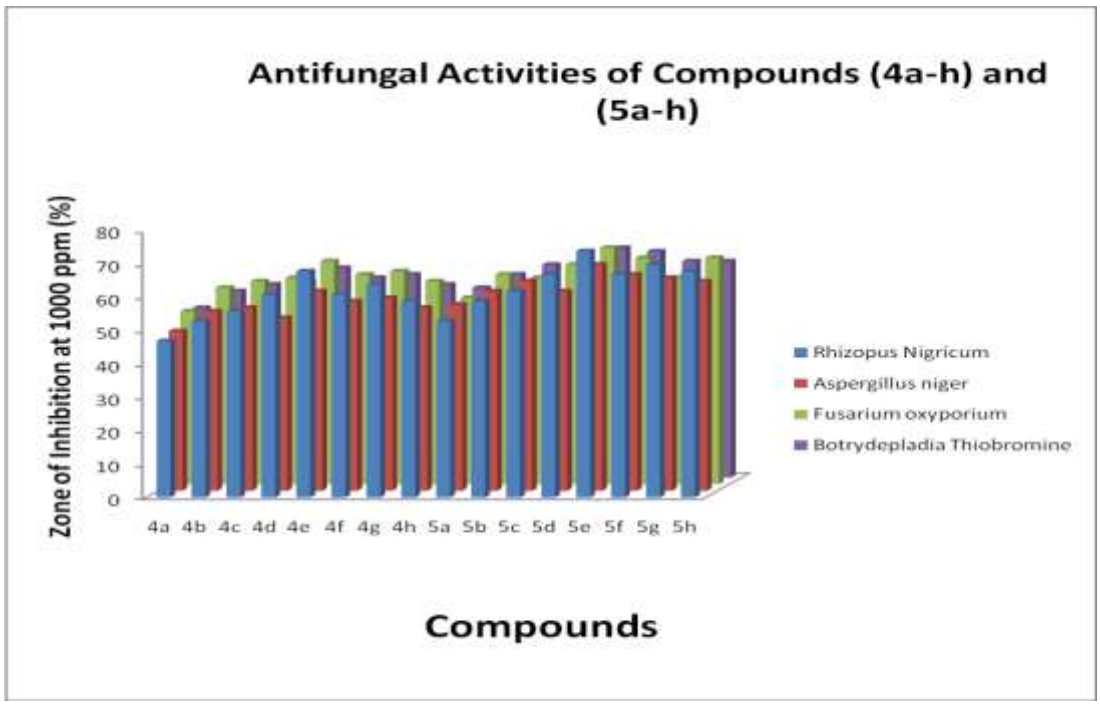
Percentage of inhibition = 100(X-Y) / X

Where, X = Area of colony in control plate
Y = Area of colony in test plate

The fungicidal activity displayed by various compounds (4a-h) and (5a-h) is shown in Table-5.

Table: 5 Antifungal Activities of Compounds (4a-h) and (5a-h)

Zone of Inhibition at 1000 ppm (%)				
Compounds	***Rhizopus Nigricum***	***Aspergillus niger***	***Fusarium oxyporium***	***Botrydepladia Thiobromine***
4a	47	48	52	51
4b	53	54	59	56
4c	56	55	61	58
4d	61	52	62	59
4e	68	60	67	63
4f	61	57	63	60
4g	64	58	64	61
4h	59	55	61	58
5a	53	56	56	57
5b	59	60	63	61
5c	62	63	62	64
5d	67	60	66	63
5e	74	68	71	69
5f	67	65	68	68
5g	70	64	62	65
5h	68	63	68	65



ACKNOWLEDGMENTS

The Authors are grateful to Principal, Sheth M. N. Science College, Hemchandracharya North Gujarat University, Patan, Gujarat, India for providing us the access to required resources for this research work.

DISCLOSURE STATEMENT

No potential conflict of interest was reported by the authors.

FUNDING

There was no fund for present research work.

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