Research Article

Solvent Free Synthesis, Spectral Studies and Antimicrobial Activities of Substituted (*E*)-1-Benzylidene-2-(4-Methylphenyl) Hydrazine's

Rajarajan M[1], Balaji S[1], Vijayakumar R[1], Senbagam R[1], Manikandan V[1], Vanangamudi G[1] and Thirunarayanan G[2]*

[1]*PG & Research Department of Chemistry, Government Arts College, C-Mutlur,Chidambaram-608 102, India*
[2]*Department of Chemistry, Annamalai University, India*

Abstract

A series of nine number of substituted (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazines has been synthesized by condensation of 4-methylphenylhydrazine with substituted benzaldehydes using Fly-ash–H_2SO_4 catalyst under solvent free condition. This method is experimentally simple, high yielding process with shorter reaction time. The synthesized substituted (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazines have been assigned by physical constants, UV, IR and NMR spectral data. This UV, IR and NMR spectral data have been utilized for spectral correlation analyses. From the results of spectral correlation analysis assessment of substituent effect have been studied. The antimicrobial activities of all the substituted (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazines have been studied using disc diffusion method.

INTRODUCTION

Hydrazone containing azomethine –NH—N=CH— protons constitute an important class of compounds for development of new drugs. The general method for the synthesis of hydrazones by the condensation reaction of hydrazine with carbonyl compounds such as aldehydes or ketones in ethanol [1-3]. The azomethine (-CH=N-) carbon important role for biological activity that has been demonstrated various pharmacological activities such as antimicrobial [4], antiplatelet [5], anticancer [6], antiviral [7], antitubercular [8], anticonvulsant [9], analgesic [10], anti-inflammatory [11], antitumoral [12], antimalarial [13], analgesic [14], ntiprotozoal [15], antiparasitic [16], cardioprotective [17], antidepressant [18] and anti-HIV [19] activities.

Green chemistry is used for designing chemical products and processes by reducing or eliminating the use of harmful chemicals and/or generating hazardous substances. Various green chemistry methods involving stirring, phase transfer catalyst, ionic liquid and many more techniques like microwave techniques include approaches for the creation of ''benign-by-design'' synthetic methods which are now accepted worldwide [20]. This processes designed by green synthesis help in the promotion of resource and efficient utilization of energy. They involve low level of waste and hence the processes are becoming economically and environmentally beneficial.

The catalyst promoted organic synthesis has become one of the hot areas in organic synthesis in the last few decades. At present, a wide range of methods for synthesizing Schiff bases in the presence of catalysts are available viz., $ZnCl_2$ [21], $TiCl_4$ [22], K-10 clay [23], CaO [24] P_2O_5-SiO_2 [25] and MnO_2 [26]. No report has been found in the literature regarding the synthesis of (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazines with Fly-ash-H_2SO_4 catalyst under grinding method and spectral as well as biological activities. Therefore the authors have taken efforts for the synthesis of (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazines from 4-methylphenylhydrazine with substituted benzaldehyde using Fly-ash-H_2SO_4 catalyst under solvent-free conditions. The intention of the present methodology is a green protocol offering several advantages such as, excellent yield of products, minimizing production of chemical wastes, shorter reaction profile, mild reaction conditions, simple operational procedure, easy preparation of catalyst and its recyclability up to five cycles without any appreciable loss in catalytic activity. The various spectral data of these (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazines have been utilized for studying the Hammett spectral correlation analyses. The antimicrobial activities of all the

SciMedCentral

substituted (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazines have been studied using disc diffusion method [27].

MATERIALS AND METHODS

All the chemicals and solvents used in this work were purchased from Sigma-Aldrich and E-Merck chemical companies. Melting points of all the (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazines were determined in open glass capillaries on Mettler FP51 melting point apparatus and are uncorrected. The UV spectra of all substituted (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazines were recorded with SHIMADZU-1650 SPECTROMETER spectrophotometer λ_{max} (nm) in spectral grade ethanol solvent. The IR spectra were obtained on a SHIMADZU-2010 Fourier transform spectrophotometer. The NMR spectra are recorded in Bruker AV400 NMR spectrometer operating at 400 MHz for ^{1}H NMR spectra and 100 MHz for ^{13}C spectra in CDCl$_3$ solvent, using TMS as internal standard.

Preparation of Fly-ash–H$_2$SO$_4$ catalyst

In a 50 mL borosil beaker, 3 g of Fly-ash and 3 mL of sulphuric acid have been taken and mixed thoroughly with glass rod at room temperature. This mixture has been heated on a hot air oven at 10°C for 1 h, cooled to room temperature, stored in a borosil bottle and tightly capped.

Synthesis of substituted (E)-1-benzylidene-2-(4-methylphenyl) hydrazines

An appropriate equimolar quantity of 4-methylphenylhydrazine (2 mmol) with *meta-* and *para-* substituted benzaldehydes (2 mmol) and Fly-ash-H$_2$SO$_4$ (0.5 g) was thoroughly ground with a pestle in an open mortar at room temperature for 5-10 minutes. Completion of the reaction was monitored by TLC. After the completion of reaction 10 mL of dichloromethane was added and the organic layer separated by filtration, yield solid product on evaporation. The crude product was obtained upon recrystallization from ethanol to give the pure substituted (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazines and their melting points have been observed. The insoluble catalyst has been recycled by washing with ethyl acetate (10 mL) followed by drying in an air oven at 100°C for 1h and reused for further reactions. The general reaction is as shown in (Scheme 1).

RESULTS AND DISCUSSION

In this work, we have synthesized a series of (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazine derivatives by condensation of 4-methylphenylhydrazine with substituted benzaldehydes and Fly-ash-H$_2$SO$_4$ was thoroughly ground with a pestle in an open mortar at room temperature for 5-10 minutes. After the completion of the reaction, the products have been isolated followed by washing the catalyst with ethyl acetate and filtered. To make this catalyst reusable for further reactions, it has been heated to 100°C. In this method is interesting to note that during the condensation there has been no appreciable change in the percentage of the yield of (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazine, on reusing the same catalyst. It is noteworthy to mention that the green route method requires simple work-up procedure involving simple filtration to isolate the products as they are insoluble in water.
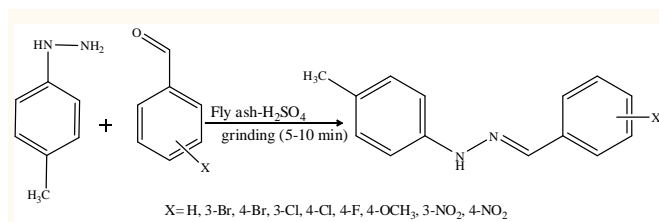
Scheme 1 Synthesis of substituted (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazines.

The synthesized (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazines have been evaluating the assessment of substituent effect on the group frequency such as UV λ_{max} (nm), IR ν(cm^{-1}) of CH=N, ^{1}H NMR chemical shift δCH=N(ppm) and ^{13}C NMR chemical shift δC=N (ppm) carbon have been correlated with Hammett substituent constants *F* and *R* Swain-Lupton's parameters using single and multi-linear regression analysis. The physical constants of synthesized (*E*)-1-benzylidene-2-(4-methylphenyl)hydrazineshave been given in (Table 1). The UV, IR and NMR spectral data of synthesized (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazines given in (Table 2).

UV Spectral study

The UV absorption λ_{max} (nm) values are correlated [29-33] with Hammett substituent constants and *F* and *R* Swain-Lupton's parameters using single and multi-linear regression analyses [34]. In correlation analysis the Hammett equation employed, as shown in equation (1)

$$\lambda = \rho\,\sigma + \lambda_0 \qquad (1)$$

Where λ_0 is the absorption maximum of the parent member of this series.

The single parameter correlations are given in (Table 3). The correlation analysis of UV absorption maxima λ_{max} (nm) values of all substituted (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazines with Hammett substituent constants and *F* and *R* parameters are found to have failure correlations (r < 0.900) with negative ρ values. This shows that the reverse substituent effects operate in all the systems. The resonance conjugative structure of the substituted (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazines is shown in (Figure 1)**.** The multi regression analysis of the UV absorption maximum λ_{max}C=N (nm) values of all the *(E)*-1-benzylidene-2-(4-methylphenyl) hydrazine compounds with inductive, resonance and *F* and *R* Swain-Lupton's [34] parameters produce satisfactory correlations as shown in equations (2) and (3).

$$\lambda_{max} \text{(nm)} = 347.61(\pm5.23) + 4.90(\pm1.68)\sigma_I + 4.61(\pm1.43)\sigma_R \quad (2)$$

(R = 0.925, n = 9, P > 90%)

$$\lambda_{max} \text{(nm)} = 349.29\ (\pm5.22) + 1.26\ (\pm0.24)F + 4.98\ (\pm1.44)R \quad (3)$$

(R = 0.919, n = 9, P > 90%)

IR Spectral study

The infrared νCH=N stretching frequencies (cm^{-1}) values of all the synthesized substituted *(E)*-1-benzylidene-2-(4-methylphenyl) hydrazines have been recorded and presented

SciMedCentral

Table 1: Physical constants of substituted (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazines.

Entry	X	m.p (°C)	Mol. formula	Mol. weight	Yield (%)
1	H	93-94 93-94 [28]	$C_{14}H_{14}N_2$	210	94
2	3-Br	134-135	$C_{14}H_{13}N_2Br$	289	92
3	4-Br	151-152	$C_{14}H_{13}N_2Br$	289	90
4	3-Cl	139-140	$C_{14}H_{13}N_2Cl$	244	95
5	4-F	119-120	$C_{14}H_{13}N_2F$	228	91
6	4-CH$_3$	129-130	$C_{15}H_{16}N_2$	224	90
7	4-OCH$_3$	162-163	$C_{15}H_{16}N_2O$	240	93
8	3-NO$_2$	137-138	$C_{14}H_{13}N_3O_2$	255	95
9	4-NO$_2$	127-128	$C_{14}H_{13}N_3O_2$	286	96

Table 2: UV, IR and NMR Spectral data of substituted (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazines.

Entry	X	UV λ$_{max}$ (nm)	IR(ν, cm^{-1}) C=N	^{1}H NMR (δ, ppm) CH=N	^{13}C NMR (δ, ppm) C=N
1	H	345.5	1620.21	7.905	148.72
2	3-Br	357.5	1573.91	7.852	147.93
3	4-Br	358.5	1573.91	7.878	146.10
4	3-Cl	354.5	1570.06	7.953	143.44
5	4-F	339.5	1570.06	8.111	144.53
6	4-CH$_3$	344.0	1604.76	7.871	145.60
7	4-OCH$_3$	345.8	1610.56	7.833	148.62
8	3-NO2	351.5	1583.56	7.868	147.96
9	4-NO2	343.0	1593.20	7.988	146.88

Table 3: The results of statistical analysis of UV λ$_{max}$ (nm), IR ν (cm^{-1}) of CH=N, ^{1}H NMR chemical shift δCH=N (ppm) and ^{13}C NMR chemical shift δC=N (ppm) data of substituted (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazines with Hammett constants σ, σ+, σ$_I$ & σ$_R$ and *F* and *R* parameters.

Frequency	Constant	r	I	ρ	s	n	Correlated derivatives
λ$_{max}$ nm	σ	0.862	350.67	-3.01	4.81	9	H, 3-Br, 4-Br, 3-Cl, 4-F, 4-CH$_3$, 4-OCH$_3$, 3-NO$_2$, 4-NO$_2$
	σ$^+$	0.802	349.97	-0.65	4.93	9	H, 3-Br, 4-Br, 3-Cl, 4-F, 4-CH$_3$, 4-OCH$_3$, 3-NO$_2$, 4-NO$_2$
	σ$_I$	0.816	351.46	-3.75	4.87	9	H, 3-Br, 4-Br, 3-Cl, 4-F, 4-CH$_3$, 4-OCH$_3$, 3-NO$_2$, 4-NO$_2$
	σ$_R$	0.827	349.02	-5.76	4.75	9	H, 3-Br, 4-Br, 3-Cl, 4-F, 4-CH$_3$, 4-OCH$_3$, 3-NO$_2$, 4-NO$_2$
	F	0.820	351.76	-4.26	4.83	9	H, 3-Br, 4-Br, 3-Cl, 4-F, 4-CH$_3$, 4-OCH$_3$, 3-NO$_2$, 4-NO$_2$
	R	0.826	348.98	-4.63	4.76	9	H, 3-Br, 4-Br, 3-Cl, 4-F, 4-CH$_3$, 4-OCH$_3$, 3-NO$_2$, 4-NO$_2$
ν(cm^{-1}) CH=N	σ	0.846	1594.58	-24.04	17.98	9	H, 3-Br, 4-Br, 3-Cl, 4-F, 4-CH$_3$, 4-OCH$_3$, 3-NO$_2$, 4-NO$_2$
	σ$^+$	0.752	1592.32	-19.55	17.36	9	H, 3-Br, 4-Br, 3-Cl, 4-F, 4-CH$_3$, 4-OCH$_3$, 3-NO$_2$, 4-NO$_2$
	σ$_I$	0.907	1608.95	-52.92	14.40	7	H, 3-Br, 4-Br, 3-Cl, 4-F, 3-NO$_2$, 4-NO$_2$
	σ$_R$	0.717	1590.96	15.49	20.05	9	H, 3-Br, 4-Br, 3-Cl, 4-F, 4-CH$_3$, 4-OCH$_3$, 3-NO$_2$, 4-NO$_2$
	F	0.900	1610.06	-52.00	13.96	7	H, 3-Br, 4-Br, 3-Cl, 4-F, 3-NO$_2$, 4-NO$_2$
	R	0.750	1591.12	11.65	20.11	9	H, 3-Br, 4-Br, 3-Cl, 4-F, 4-CH$_3$, 4-OCH$_3$, 3-NO$_2$, 4-NO$_2$
δ(ppm) CH= N	σ	0.901	7.90	0.03	0.09	7	H, 3-Br, 4-Br, 3-Cl, 4-CH$_3$, 4-OCH$_3$, 4-NO$_2$
	σ$^+$	0.900	7.91	0.02	0.09	7	H, 3-Br, 4-Br, 3-Cl, 4-CH$_3$, 4-OCH$_3$, 4-NO$_2$
	σ$_I$	0.739	7.86	0.13	0.08	9	H, 3-Br, 4-Br, 3-Cl, 4-F, 4-CH$_3$, 4-OCH$_3$, 3-NO$_2$, 4-NO$_2$
	σ$_R$	0.900	7.90	-0.10	0.09	8	H, 3-Br, 4-Br, 3-Cl, 4-CH$_3$, 4-OCH$_3$, 3-NO$_2$, 4-NO$_2$
	F	0.875	7.84	0.17	0.07	9	H, 3-Br, 4-Br, 3-Cl, 4-F, 4-CH$_3$, 4-OCH$_3$, 3-NO$_2$, 4-NO$_2$
	R	0.902	7.90	-0.07	0.09	8	H, 3-Br, 4-Br, 3-Cl, 4-CH$_3$, 4-OCH$_3$, 3-NO$_2$, 4-NO$_2$
δ(ppm) C=N	σ	0.903	148.33	-1.31	1.32	8	H, 3-Br, 4-Br, 3-Cl, 4-CH$_3$, 4-OCH$_3$, 3-NO$_2$, 4-NO$_2$
	σ$^+$	0.904	148.23	-1.22	1.26	8	H, 3-Br, 4-Br, 3-Cl, 4-CH$_3$, 4-OCH$_3$, 3-NO$_2$, 4-NO$_2$
	σ$_I$	0.711	148.24	-0.59	1.41	9	H, 3-Br, 4-Br, 3-Cl, 4-F, 4-CH$_3$, 4-OCH$_3$, 3-NO$_2$, 4-NO$_2$
	σ$_R$	0.820	147.65	-2.79	1.26	9	H, 3-Br, 4-Br, 3-Cl, 4-F, 4-CH$_3$, 4-OCH$_3$, 3-NO$_2$, 4-NO$_2$
	F	0.711	147.78	-0.57	1.41	9	H, 3-Br, 4-Br, 3-Cl, 4-F, 4-CH$_3$, 4-OCH$_3$, 3-NO$_2$, 4-NO$_2$
	R	0.810	147.55	-2.45	3.60	9	H, 3-Br, 4-Br, 3-Cl, 4-F, 4-CH$_3$, 4-OCH$_3$, 3-NO$_2$, 4-NO$_2$

Abbreviations: r: Correlation coefficient; I: Intercept; ρ: Slope; s: standard deviation; n: Number of correlated derivatives
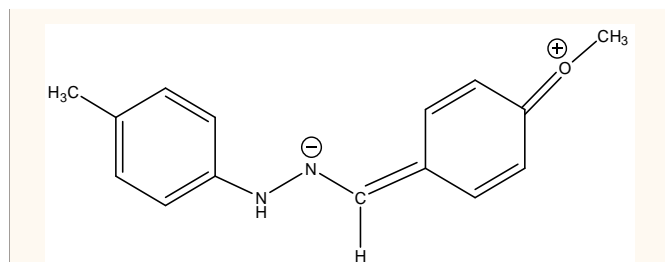
Figure 1 The resonance-conjugative structure.

in (Table 2). These infrared νC=N stretching frequencies (cm^{-1}) values are correlated [29-33] with Hammett substituent constants and Swain-Lupton's parameters using single and multi-linear regression analyses [34]. The structure parameter correlation involving group frequencies, the employed Hammett equation is shown in equation (4).

$$\nu = \rho\,\sigma + \nu_0 \qquad (4)$$

Where ν_0 is the frequency for the parent member series

The results of single parameter statistical analysis [29-33] of all the *(E)*-1-benzylidene-2-(4-methylphenyl) hydrazine compounds except those with 4-CH$_3$ and 4-OCH$_3$ substituent's have shown better correlation with Hammett substituent constants σ_I (r = 0.907) and Swain-Lupton *F* (r = 0.900) parameters. When the substituent's that have been given exception are included in regression, they reduce the correlations considerably. The infrared frequency νC=N (cm^{-1}) values of all the *(E)*-1-benzylidene-2-(4-methylphenyl) hydrazine compounds have shown failure correlations (r < 0.900) with the remaining Hammett constants σ σ^+, σ_R and *R* parameters. This is due to the reason stated earlier, with resonance conjugative structure as shown in (Figure 1)**.** All the correlations gave negative ρ values (except σ_R and *R*) parameters. This indicates that reverse substituent effect operates in *(E)*-1-benzylidene-2-(4-methylphenyl) hydrazines. The multi regression analysis of the infrared νC=N (cm^{-1}) values of all the *(E)*-1-benzylidene-2-(4-methylphenyl) hydrazines with inductive, resonance and Swain-Lupton's [34] parameters produce satisfactory correlations are given in equations (5) and (6).

$$\nu C=N(cm^{-1}) = 1613.34(\pm9.80) - 55.91(\pm4.00)\sigma_I + 24.66(\pm3.27)\sigma_R \qquad (5)$$

(*R* = 0.957, n = 9, P > 95%)

$$\nu C=N\ (cm^{-1}) = 1611.74(\pm10.01) - 51.64(\pm3.63)F + 9.61(\pm2.02)R \qquad (6)$$

(*R* = 0.972, n = 9, P > 95%)

NMR Spectral study

^{1}H NMR Spectral study: The ^{1}H NMR spectra of synthesized *(E)*-1-benzylidene-2-(4-methylphenyl) hydrazines have been recorded in deuterio chloroform solutions employing tetramethylsilane (TMS) as internal standard. The δCH=N (ppm) chemical shifts values of all the substituted *(E)*-1-benzylidene-2-(4-methylphenyl) hydrazines were assigned and are presented in (Table 2). These δCH=N (ppm) chemical shifts have been correlated [29-33] with reactivity parameters. Hammett

correlation involving group frequencies, the form of the Hammett equation employed is as shown in (7).

$$\delta = \rho\sigma + \delta_0 \qquad (7)$$

Where δ_0 is the chemical shift of the corresponding parent compound.

The results of statistical analysis are presented in (Table 3). From (Table 3), it is evident that the ^{1}H NMR chemical shift δCH=N (ppm) values of all the *(E)*-1-benzylidene-2-(4-methylphenyl)hydrazine compounds except those with 4-F and 3-NO$_2$ substituent's have shown better correlations with Hammett substituent constants namely σ (r = 0.901) and σ^+ (r = 0.900) parameters.

The entire *(E)*-1-benzylidene-2-(4-methylphenyl) hydrazine compounds except those with 4-F substituent has shown satisfactory correlations with Hammett substituent constants viz., σ_R (r = 0.900) and *R* (r = 0.902) parameters. When the substituent's that have been given exception are included in regression they reduce the correlations considerably. The ^{1}H NMR chemical shift δCH=N (ppm) values of all the *(E)*-1-benzylidene-2-(4-methylphenyl) hydrazine compounds have shown poor correlations (r < 0.900) with the remaining σ_I and *F* parameters. This is attributed to the weak inductive and field effects of the substituent's for predicting the reactivity on the δCH=N (ppm) chemical shift through resonance as per the conjugative structure given in (Figure 1). All the correlations with few Hammett substituent constants namely σ, σ^+, σ_R and *F* and *R* parameters have shown positive ρ values. It indicates that the normal substituent's effect operates with respect to^{1}H NMR chemical shift δCH=N (ppm) values of all the *(E)*-1-benzylidene-2-(4-methylphenyl) hydrazine compounds.

The multi regression analysis of the ^{1}H NMR chemical shift δCH=N (ppm) values of all the *(E)*-1-benzylidene-2-(4-methylphenyl) hydrazine compounds with inductive, resonance and Swain-Lupton's [34] parameters produce better correlations as shown in equations (8) and (9).

$$\delta CH=N(ppm) = 7.84(\pm0.60) - 0.14(\pm0.01)\,\sigma_I - 0.12(\pm0.01)\sigma_R \qquad (8)$$

(*R* = 0.949, n = 9, P > 90%)

$$\delta CH=N(ppm) = 7.83(\pm0.56) + 0.17(\pm0.01)\,F - 0.06(\pm0.01)\,R \qquad (9)$$

(*R* = 0.957, n = 9, P > 95%)

^{13}C NMR Spectral study: The chemical shift δC=N (ppm) values of *(E)*-1-benzylidene-2-(4-methylphenyl) hydrazines carbon, have been assigned and are presented in (Table 2). The assigned δC=N chemical shift (ppm) values have been made to correlate [29-33] with Hammett substituent constants, field and resonance parameters, with the help of single and multi-regression analyses [34] the results of statistical analysis are presented in (Table 3). From (Table 3), it is evident that the ^{13}C NMR chemical shift δC=N (ppm) values of all the *(E)*-1-benzylidene-2-(4-methylphenyl) hydrazine compounds except that with 4-F substituent has shown better correlations with Hammett substituent constants namely σ (r =0.903) and σ^+ (r =0.904) parameters. When the substituent's that have been given

exception included in regression they reduce the correlations considerably.

The ^{13}C NMR chemical shift δC=N (ppm) values of all the *(E)*-1-benzylidene-2-(4-methylphenyl) hydrazines have shown failure correlations (r < 0.900) with the remaining Hammett substituent constants σ_I, σ_R and *F* and *R* parameters. The failure correlation is attributed to the conjugated structure shown in (Figure 1). All the correlations have shown negative ρ values with all the Hammett substituent constants and *F* and *R* parameters. This indicates the operation of reverse substituent effect with respect to chemical shift δC=N (ppm) values of all the *(E)*-1-benzyliden-2-(4-methylphenyl) hydrazine compounds. The multi regression analysis of the chemical shift δC=N (ppm) values of all the *(E)*-1-benzylidene-2-(4-methylphenyl) hydrazine compounds with inductive, resonance and Swain-Lupton's [34] parameters produce satisfactory correlations as shown in equations (10) and (11).

$$\delta C=N(ppm) = 147.75(\pm0.93) - 0.25(\pm0.09)\sigma_I - 2.75(\pm0.12)\sigma_R \tag{10}$$

(*R* = 0.946, n = 9, P>90%)

$$\delta C=N(ppm) = 147.36(\pm0.90) + 0.48(\pm0.07)F - 2.43(\pm0.18)R \tag{11}$$

(*R* = 0.949, n = 9, P>90%)

Antimicrobial Activities

Antibacterial activity: The antibacterial activities of all the substituted (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazines have been evaluated against three gram positive pathogenic strains *Bacillus substilis*, *Micrococcus luteus* and *Staphylococcus aureus* and two gram negative strains *Escherichia coli* and *Pseudomonas aeruginosa*. The disc diffusion technique was followed using the Kirby–Bauer [27] method, at a concentration of 250 μg/mL with ciprofloxacin used as the standard drugs. The measured antibacterial activities of all substituted (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazines are presented in (Table 4). The antibacterial activity of all the synthesized (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazines are shown in

(Figure 2) (Plates 1-10) and the clustered column chart is shown in (Figure 3). There is considerable antibacterial activity was possessed by all substituent's on the five bacterial species in general.

The (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazines with 3-Cl, 4-CH₃, 4-OCH₃ and 3-NO₂ substituent's have shown good antibacterial activity against *B. subtilis*. The (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazines with parent (H), 3-Br, 4-Br, 4-F and 4-NO₂ substituent's have shown moderate antibacterial activity against *B. subtilis*. The 3-Cl, 4-CH₃, 4-OCH₃, 3-NO₂ and 4-NO₂ substituted (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazine have shown good antibacterial activity against *M. luteus*. The (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazines with 3-Cl, 4-F, 4-CH₃, 4-OCH₃, 3-NO₂ and 4-NO₂ substituent's have shown good antibacterial activity against *S. aureus*. The (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazines with parent (H), 3-Br and 4-Br substituent's have shown moderate antibacterial activity against *S. aureus.*

The parent (H), 4-Br, 3-Cl, 4-F, 4-CH₃, 4-OCH₃, 3-NO₂ and 4-NO₂ substituted (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazine compounds have shown good antibacterial activity against *E. coli* alone*.* The 3-Br substituted (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazine compound has shown improve the antibacterial activity against *E. coli*. The 3-Cl, 4-F, 4-CH₃ and 4-NO₂ substituted (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazine compounds have shown good antibacterial activity against *P. aeruginosa.* The (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazines with parent (H), 3-Br and 3-NO₂ substituent's have shown moderate antibacterial activity against *P. aeruginosa.* The remaining 4-Br and 4-OCH₃ substituted (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazine compounds have shown poor antibacterial activity against *P.aeruginosa.*

Antifungal activity

Measurement of antifungal sensitivities of all the (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazines have been performed using disc diffusion technique [27]. The antifungal activity while for the three strains *A. niger, M. species* and *T. viride*

Table 4: Antibacterial activity of substituted (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazines.

S. No.	X	Zone of inhibition (mm)				
		Gram positive Bacteria			Gram negative Bacteria	
		B. subtilis	*M. luteus*	*S. aureus*	*E. coli*	*P. aeruginosa*
1	H	7	0	8	10	6
2	3-Br	6	0	6	8	6
3	4-Br	6	8	9	16	0
4	3-Cl	12	11	10	15	14
5	4-F	9	9	10	12	10
6	4-CH₃	13	10	12	13	10
7	4-OCH₃	16	13	10	10	0
8	3-NO₂	14	12	10	16	9
9	4-NO₂	8	16	12	15	10
Standard	Ciprofloxacin	16	15	16	16	15
Control	DMSO	0	0	0	0	0

Plate-1 Plate-2

Plate-3 Plate-4

Plate-5 Plate-6

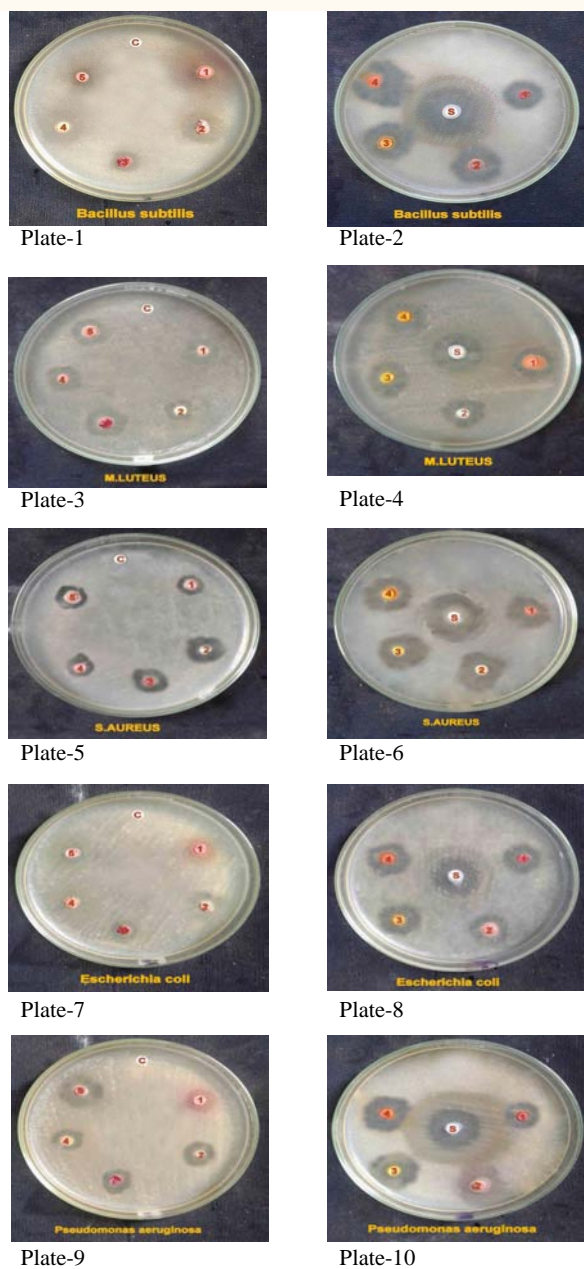Plate-7 Plate-8

Plate-9 Plate-10

Figure 2 Antibacterial activity of substituted (E)-1-benzylidene-2-(4-methylphenyl) hydrazines (petri dishes).

the disc diffusion method was adopted. The drugs dilution was 250μg/mL. Miconazole has been taken as the standard drug. The zone of inhibition values of all the (E)-1-benzylidene-2-(4-methylphenyl) hydrazine compound presented in (Table 5). The anti fungal activities of all the (E)-1-benzylidene-2-(4-methylphenyl) hydrazines compounds in the present study are shown in (Figure 4) (Plates1-6) and the clustered column chart, shown in (Figure 5). All the (E)-1-benzylidene-2-(4-methyl) hydrazine compounds have shown good to moderate activity against all the three fungal species evaluated in general.

The (E)-1-benzylidene-2-(4-methylphenyl)hydrazines having parent (H), 3-Br, 3-NO$_2$ and 4-NO$_2$substituents have shown good antifungal activity against *A. niger.*The4-Br, 3-Cl, 4-F, 4-CH$_3$

and 4-OCH$_3$ substituted (E)-1-benzylidene-2-(4-methylphenyl) hydrazine compounds have shown improved activity antifungal activity against *A. niger.* The (E)-1-benzylidene-2-(4-methylphenyl)hydrazines having parent (H), 3-Br, 4-CH$_3$, 4-OCH$_3$, 3-NO$_2$ and 4-NO$_2$ substituent's have shown good antifungal activity against *M. Species*alone. The (E)-1-benzylidene-2-(4-methylphenyl) hydrazines compounds with parent (H), 3-Br, 4-F, 4-CH$_3$, 4-OCH$_3$, 3-NO$_2$ and 4-NO$_2$ substituent's have shown good antifungal activity against *T. viride.* The (E)-1-benzylidene-2-(4-methylphenyl)
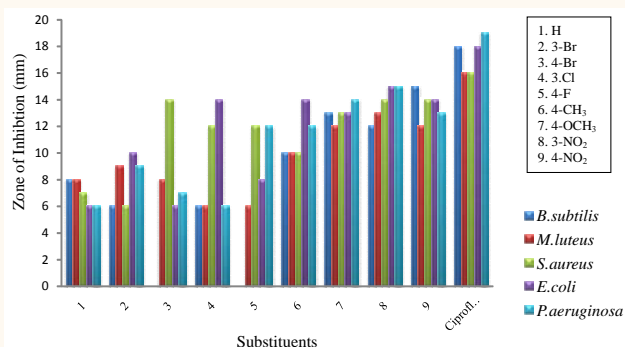

Figure 3 The antibacterial activities of substituted (E)-1-benzylidene-2-(4 methyl phenyl) hydrazines (clustered column chart).
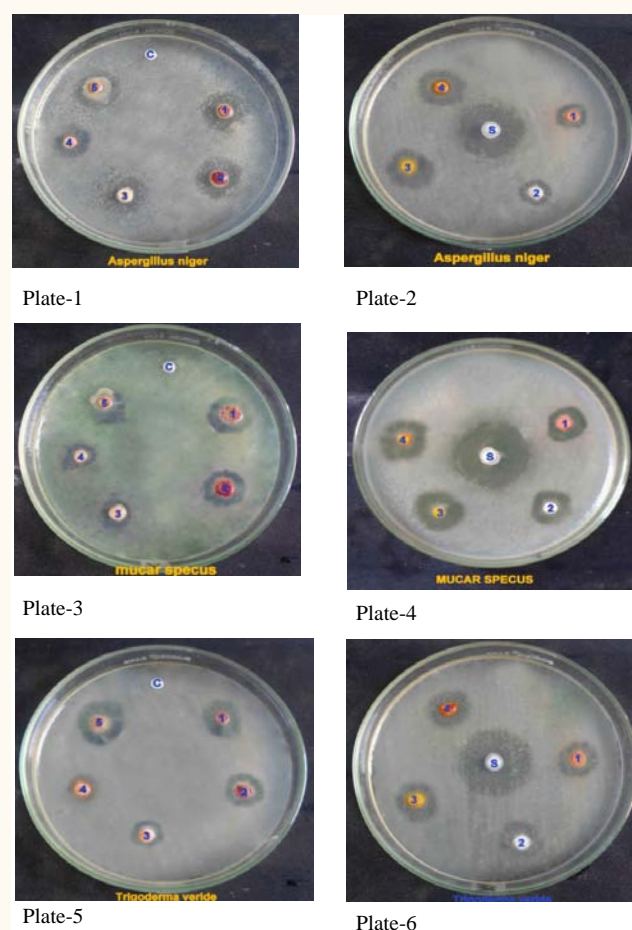

Plate-1 Plate-2

Plate-3 Plate-4

Plate-5 Plate-6

Figure 4 Antifungal activity of (E)-1-benzylidene-2-(4-methylphenyl) hydrazines compounds (petri dishes).

SciMedCentral

Table 5: Antifungal activity of substituted (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazines.

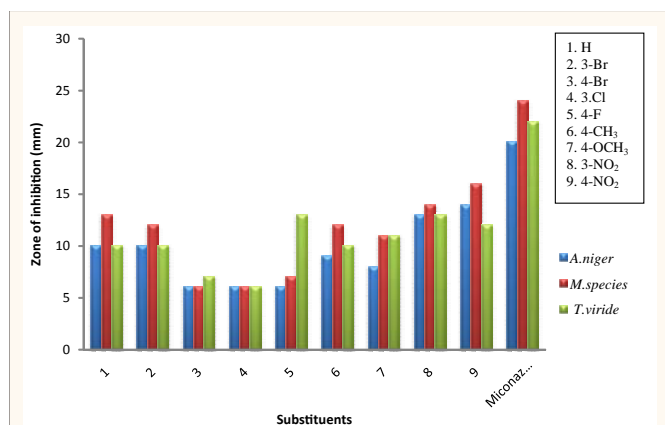| S. No.	X	Zone of inhibition(mm)		
		A. niger	*M. species*	*T. viride*
1	H	10	13	10
2	3-Br	10	12	10
3	4-Br	6	6	7
4	3-Cl	6	6	6
5	4-F	6	7	13
6	4-CH$_3$	9	12	10
7	4-OCH$_3$	8	11	11
8	3-NO$_2$	13	14	13
9	4-NO$_2$	14	16	12
Standard	Miconazole	20	24	22
Control	DMSO	0	0	0

Figure 5 Antifungal activity of substituted (*E*)-1-benzylidene-2-(4-methyl phenyl) hydrazines (clustered column chart).

hydrazines having 4-Br, 3-Cl and 4-F substituent's have shown improve antifungal activity against *M. Species*. The 4-Br and 3-Cl substituted (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazines compounds have shown improved antifungal activity against *T. viride* alone.

CONCLUSION

In the present research work, we have synthesized a series of substituted (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazine compounds from substituted benzaldehydes with 4-methylphenylhydrazine using Fly-ash-H$_2$SO$_4$ catalyst under solvent free condition. This method involves simple work-up procedure, shorter reaction time (5-10 minutes), eco-friendly, non-hazarders with excellent yield more than 90%. The synthesized substituted (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazine compounds conformed by their physical constant, UV, IR and NMR spectral data. The UV λ$_{max}$ (nm), IR ν (cm^{-1}) of CH=N, ^{1}H NMR chemical shift δ CH=N (ppm) and ^{13}C NMR chemical shift δC=N (ppm) data have been correlated with Hammett substituent constants and Swain-Lupton's constants using single and multi-regression analysis. From the results some of the single parameter correlations gave satisfactory correlation coefficients. The multi-regression analysis gave satisfactory correlation coefficients in all spectral data. The antimicrobial activity of all the substituted (*E*)-1-benzylidene-2-(4-methylphenyl) hydrazine compounds have shown good to moderate and poor activity against all the microorganism

REFERENCES

1. Berdinskii S. The chemistry of hydrazones. Chem Hetero Comp.1979; 15: 238-244.

2. Smith PAS. Benjamin Cummings. Reading Mass. 1983.

3. Polshettiwar V, Varma RS. Polystyrene Sulfonic Acid Catalyzed Greener Synthesis of Hydrazones in Aqueous Medium Using Microwaves. Tetrahedron Lett. 2007; 48: 5649-5652.

4. Rollas S, Gulerman N, Erdeniz H. Synthesis and antimicrobial activity of some new hydrazones of 4-fluorobenzoic acid hydrazide and 3-acetyl-2,5-disubstituted-1,3,4-oxadiazolines. Farmaco. 2002; 57: 171-174.

5. Silva GA, Costa LM, Brito FC, Miranda AL, Barreiro EJ, Fraga CA. New class of potent antinociceptive and antiplatelet 10H-phenothiazine-1-acylhydrazone derivatives. Bioorg Med Chem. 2004; 12: 3149-3158.

6. Savini L, Chiasserini L, Travagli V, Pellerano C, Novellino E, Cosentino S, et al. New alpha-(N)-hetero cyclic hydrazones: evaluation of anticancer, anti-HIV and antimicrobial activity. Eur J Med Chem. 2004; 39: 113-122.

7. Abdel-Aal MT, El-Sayed WA, El-Ashry el-SH. Synthesis and antiviral evaluation of some sugar aryl glycinoyl hydrazones and their oxadiazoline derivatives. Arch Pharm (Weinheim). 2006; 339: 656-663.

8. Imramovský A, Polanc S, Vinsová J, Kocevar M, Jampílek J, Recková Z, et al. A new modification of anti-tubercular active molecules. Bioorg Med Chem. 2007; 15: 2551-2559.

9. Dimmock JR, Vasishtha SC, Stables JP. Anticonvulsant properties of various acetylhydrazones, oxamoylhydrazones and semicarbazones derived from aromatic and unsaturated carbonyl compounds. Eur J Med Chem. 2000; 35: 241-248.

10. Lima PC, Lima LM, da Silva KC, Léda PH, de Miranda AL, Fraga CA, et al. Synthesis and analgesic activity of novel N-acylarylhydrazones and isosters, derived from natural safrole. Eur J Med Chem. 2000; 35: 187-203.

11. Salgin-Gökşen U, Gökhan-Kelekçi N, Göktaş O, Köysal Y, Kiliç E, Işik S, et al. 1-Acylthiosemicarbazides, 1,2,4-triazole-5(4H)-thiones, 1,3,4-thiadiazoles and hydrazones containing 5-methyl-2-benzoxazolinones: synthesis, analgesic-anti-inflammatory and antimicrobial activities. Bioorg Med Chem. 2007; 15: 5738-5751.

12. El-Hawash SA, Abdel Wahab AE, El-Demellawy MA. Cyanoacetic acid hydrazones of 3-(and 4-) acetylpyridine and some derived ring systems as potential antitumor and anti-HCV agents. Arch Pharm (Weinheim). 2006; 339: 14-23.

13. Belkheiri N, Bouguerne B, Bedos-Belval F, Duran H, Bernis C, Salvayre R, et al. Synthesis and antioxidant activity evaluation of a syringic hydrazones family. Eur J Med Chem. 2010; 45: 3019-3026.

14. Duarte CD, Barreiro EJ, Fraga CA. Privileged structures: a useful concept for the rational design of new lead drug candidates. Mini Rev Med Chem. 2007; 7: 1108-1119.

15. Caputto ME, Fabian LE, Benítez D, Merlino A, Ríos N, Cerecetto H, et al. Thiosemicarbazones derived from 1-indanones as new anti-Trypanosoma cruzi agents. Bioorg Med Chem. 2011; 19: 6818-6826.

16. Aslam MA, Mahmood SU, Shahid M, Saeed A, Iqbal J. Synthesis, biological assay in vitro and molecular docking studies of new Schiff base derivatives as potential urease inhibitors. Eur J Med Chem. 2011; 46: 5473-5479.

17. el-Sabbagh OI, Shabaan MA, Kadry HH, Al-Din ES. New octahydroquinazoline derivatives: synthesis and hypotensive activity. Eur J Med Chem. 2010; 45: 5390-5396.

18. Catto M, Aliano R, Carotti A, Cellamare S, Palluotto F, Purgatorio R, et al. Design, synthesis and biological evaluation of indane-2-arylhydrazinylmethylene-1, 3-diones and indol-2-aryldiazenylmethylene-3-ones as beta-amyloid aggregation inhibitors. Eur J Med Chem. 2010; 45: 1359-1366.

19. Kajal A, Bala S, Sharma N, Kamboj S, Saini V. Therapeutic potential of hydrazones as anti-inflammatory agents. Int J Med Chem. 2014; 2014: 761030.

20. V Koteswara Raoa, S Subba Reddya, B Satheesh Krishnaa, K Reddi Mohan Naidua, C Naga Rajua, SK Ghoshb, et al. Synthesis of Schiff's bases in aqueous medium: a green alternative approach with effective mass yield and high reaction rates. Green Chem Lett Rev. 2010; 3: 217-223.

21. Billman JH, Tai KM. Reduction of Schiff bases II: benzhydrylamines and structurally related compounds. J Org Chem. 1958; 23: 535-539.

22. White WA, Weingarten H. A versatile new enamine synthesis. J Org Chem. 1967; 32: 213–214.

23. Varma RS, Dahiya R, Kumar S. Clay catalyzed synthesis of imines and enamines under solvent-free conditions using microwave irradiation. Tetrahedron Lett.1997; 38: 2039–2042.

24. Gopalakrishnan M, Sureshkumar P, Kanagarajan V, Thanusu J. New environmentally-friendly solvent-free synthesis of imines using calcium oxide under microwave irradiation. Res Chem Intermed. 2007; 33: 541-548.

25. Hasaninejad A, Zare A, Sharghi H, Shekouhy M. P2O5/SiO2 an efficient, green and heterogeneous catalytic system for the solvent-free synthesis of N-sulfonyl imines. Arkicov. 2008; 1: 64-74.

26. Blackburn L, Taylor RJ. In situ oxidation-imine formation-reduction routes from alcohols to amines. Org Lett. 2001; 3: 1637-1639.

27. Bauer AW, Kirby WMM, Sherris JC, Truck M. Antibiotic susceptibility testing by a standardized single disk method. Am J Clinical Pathology. 1966; 45: 493-498.

28. Rong Hu J, Jia Zhang W, Gui Zheng D. A one-pot synthesis of bisarylhydrazones by Cu (I)-catalyzed aerobic oxidation. Tetrahedron. 2013; 69: 9865-9869.

29. R Senbagama, M Rajarajana, R Vijayakumara, V Manikandana, S Balajia, G Vanangamudia, et al. Synthesis, Assessment of substituent effect and Antimicrobial activities of some substituted (E)-N-benzylidene-4H-1,2,4-triazol-4-amines. World Scientific News. 2015; 2: 211-226.

30. R Senbagam, R Vijayakumar, V Manikandan, S Balaji, G Vanangamudi, G Thirunarayanan, et al. Synthesis, spectral studies and antimicrobial activities of some substituted (E)-1-benzylidene-2- (4-bromophenyl) hydrazines. World Scientific News. 2015; 9: 155-171.

31. R Senbagam, M Rajarajan, R Vijayakumar, V Manikandan, S Balaji, G Vanangamudi, et al. Synthesis, spectral correlations and antimicrobial activities of 2-pyrimidine Schiff's bases. Int Lett Chem Phy Astro. 2015; 53: 154-164.

32. R Senbagama, R Vijayakumara, M Rajarajana, S Balajia, V Manikandana, G Vanangamudia, et al. Synthesis, assessment of substituent effect and antimicrobial activities of (4E)-4-(benzylideneamino)-1,2-dihydro-2,3-dimethyl-1-phenylpyrazol-5-one compounds. Karbala Int J Modern Sci. 2016; 2: 56-62.

33. Rajarajan M. Synthesis, spectral correlations and antimicrobial activities of substituted 4-((E)-2-benzylidenehydrazinyl) benzonitriles. Indian J Chem. 2016; 55: 197-206.

34. Swain CG, Lupton EC. Field and resonance components in substituent effects. J Am Chem Soc.1968; 90: 4328-4337.