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SOLVENT-FREE SYNTHESIS OF AZOMETHINES, SPECTRAL CORRELATIONS AND ANTIMICROBIAL ACTIVITIES OF SOME *E*-BENZYLIDENE-4-CHLOROBENZENAMINES

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ABSTRACT. Some azomethines including substituted benzylidene-4-chlorobenzenamines (*E*-imines) have been synthesized by fly-ash: PTS catalyzed microwave assisted condensation of 4-chloroaniline and substituted benzaldehydes under solvent-free conditions. The yield of the imines has been found to be more than 85%. The purity of all imines has been checked using their physical constants and UV, IR and NMR spectral data. These spectral data have been correlated with Hammett substituent constants and F and R parameters using single and multi-linear regression analysis. From the results of statistical analysis, the effect of substituents on the above spectral data has been studied. The antimicrobial activities of all imines have been studied using standard methods.

KEY WORDS: Solvent-free synthesis, Fly-ash: PTS, Aryl imines, Spectral correlations, Antimicrobial activities

INTRODUCTION

Azomethines are the imine compounds, which contain –C=N– group. These compounds are also known as imines but most commonly, they are known as Schiff bases to honor Hugo Schiff, who synthesized these compounds first [1]. Schiff bases can be hemiaminal which followed by a dehydration to generate imines. Schiff bases are synthesized from aromatic amines and carbonyl compound by nucleophilic condensation forming important class of organic compounds, especially in the medicinal and pharmaceutical fields. Now-a-days the development and synthesis of novel Schiff's base derivatives as potential chemotherapeutics still attract the attention of organic and medicinal chemists [2, 3]. Many studies reported the biological activities of Schiff's bases, including their anticancer, antibacterial, antifungal, and herbicidal activities [4]. Spectroscopic data is very useful for studying the ground state equilibrium of organic molecules through linear relationships [5]. The infrared and NMR spectroscopic data determines the structure of unsaturated systems, such as *E*- or Z, *s-cis* and *s-trans* conformers of styrenes, enones, unsaturated acid chlorides, acyl chlorides and their esters [6, 7]. Recently, Suresh *et al.* [8] have synthesized some aryl *E*-imines and studied the UV, IR and NMR spectral liner correlation analysis through Hammett equation. In their investigations they observed satisfactory and good correlations for the above spectroscopic data with Hammett substituent constants and Swain Luptons' [9] parameters.

It is well known that microwave (MW) irradiation can accelerate a great number of chemical processes and in particular, the reaction time and energy input are supposed to be mostly reduced in the reactions that are run for a long time at high temperatures under conventional conditions [10]. The most successful applications of microwave irradiation are found to be

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*Corresponding author. E-mail: drmsgacplcmcdm@gmail.com related to the use of solvent-free systems, in which microwaves interact directly with reagents. Therefore, it can more efficiently accelerate chemical reactions. In classical organic synthesis of Schiff bases, the common problems are removal of solvents from the reaction mixture, solvent extraction especially in the case of aprotic dipolar solvents with high boiling point and product isolation through solvent-solvent extraction. The absence of solvent reduces the risk of hazardous explosions when the reaction takes place in a closed vessel or a microwave oven [11]. Local overheating, which can lead to product, substrate and reagent degradation is avoided in microwave-assisted synthesis [12]. The solvent-free organic synthesis mediated by microwave irradiation offers several advantages such as they involve shorter duration, operational simplicity, easy workup procedure, less hazardousness to humans and environment and better yields. Therefore the authors have taken efforts to synthesis imines from substituted benzaldehydes and 4-chloroaniline using fly-ash: PTS catalyst with microwave irradiation under solvent-free conditions. The spectral data of these imines have been utilized for studying the quantitative structure-activity relationships through Hammett correlations. The biological activities of these imine derivatives have been studied using standard method [13].

EXPERIMENTAL

General

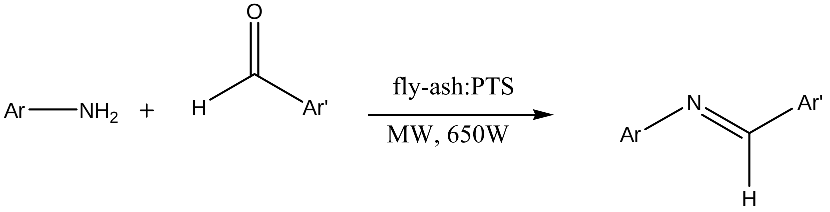
All the chemicals involved in the present investigation, have been procured from Sigma-Aldrich and E-Merck chemical companies. Melting points of all imines have been determined in open glass capillaries on SUNTEX melting point apparatus and are uncorrected. The UV spectra of all the imines, synthesized, have been recorded with ELICO-BL222 spectrophotometer (λ_{max}, nm) in spectral grade methanol solvent. Infrared spectra (KBr, 4000-400 cm^{-1}) have been recorded on SHIMADZU-2010 Fourier transform spectrophotometer. Bruker AV400 NMR spectrometer has been utilized for recording ^1H NMR, operating at 400 MHz spectra and 100 MHz for ^{13}C spectra in $CDCl_3$ solvent using TMS as internal standard. Electron impact (EI) (70 eV) and chemical ionization mode FAB+ mass spectra have been recorded with a JEOL JMS600H spectrometer.

Synthesis of catalyst

The fly-ash:PTS catalyst has been prepared following the procedure published in literature [14]. In a 50 mL Borosil beaker, 1 g of fly-ash and 0.8 mL (0.5 mol) of PTS have been taken and mixed thoroughly with glass rod. This mixture has been heated on a hot air oven at 85 °C for 1 h, cooled to room temperature, stored in a borosil bottle and tightly capped.

Synthesis of imines

An appropriate equimolar quantity of aryl amines (2 mmol), substituted benzaldehydes (2 mmol) and fly-ash:PTS (0.5 g) have been taken in borosil tube and tightly capped. The mixture was subjected to microwave irradiation for 6-12 min in a microwave oven at 650W (Scheme 1) (Samsung GW73BD Grill Model microwave Oven, 100-750 W, 230 V - 50 Hz, 2450 MHz) and then cooled to room temperature. After separating the organic layer with dichloromethane the solid product has been obtained on evaporation. The solid, on recrystallization with benzene-hexane mixture afforded glittering product. The insoluble catalyst has been recycled by washing with ethyl acetate (8 mL) followed by drying in an oven at 100 °C for 1 h and reused for further reactions.



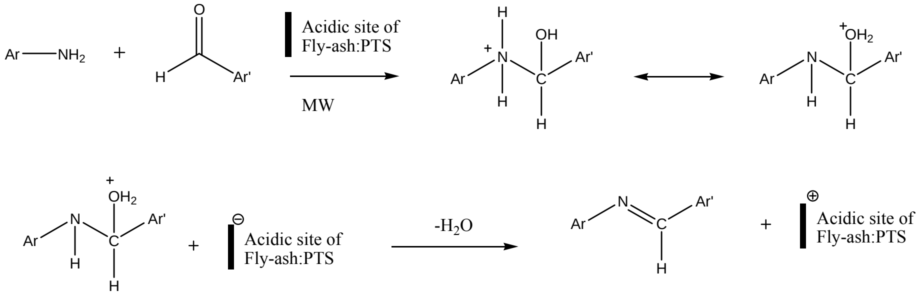
(1-36)

Entry 30: Ar= 4-ClPh
Ar'=Ph

Scheme 1. Synthesis of substituted benzylidene-4-chloroanilines in presence of fly-ash:PTS catalyst under microwave irradiation.

RESULTS AND DISCUSSION

Fly-ash has been converted into useful catalyst fly-ash:PTS by mixing fly-ash and PTS. The PTS and other chemical species present in the fly-ash have enhanced the catalytic activity [7, 8, 10, 14, 15]. During the course of the reactions these species are responsible for promoting condensation between the aryl amines and aryl aldehydic groups leading to the formation of imines. In these experiments the products were isolated and the catalyst was washed with ethyl acetate, heated to 100 ^{o}C then reused for further five run reactions. In this protocol the reaction gave better yields of the imines during the condensation without any environmental discharge. We have investigated the catalytic effect of fly-ash: PTS on the synthesis of benzylidene-4-chloroaniline (entry 30, Table 2) by varying the catalyst quantity from 0.1 g to 1 g. As the catalyst quantity was increased from 0.1 g to 1 g, the percentage of yield of product was increased from 85 to 92%. The effect of catalyst loading was shown in Figure 1. The proposed mechanism for the condensation of the amine and aldehydes is shown in Scheme 2. Further increase the catalyst amount there was no significant increasing of the percentage of product. The optimum quantity of catalyst loading was found to be 0.4 g. The reusability of catalyst on condensation of 4-chloroaniline and substituted benzaldehyde are studied. First two runs gave 92% product. The third, fourth and fifth runs of reactions gave the yields 91.5, 91.5 and 91% of imines. The reusability of the catalyst on the yields of the imines is presented in Table 1. There was no appreciable loss in its effect of catalytic activity was observed up to fifth run. The analytical and mass spectral data were presented in Table 2. The complete UV, IR and NMR spectral data of all imines were presented Table 3. The IR and NMR spectra of selective compounds are provided in supplementary data.



30: Ar=4-ClPh
Ar'=Ph

Scheme 2. The proposed mechanism for the synthesis of *E*- imines in presence of fly-ash:PTS catalyst under microwave irradiation.

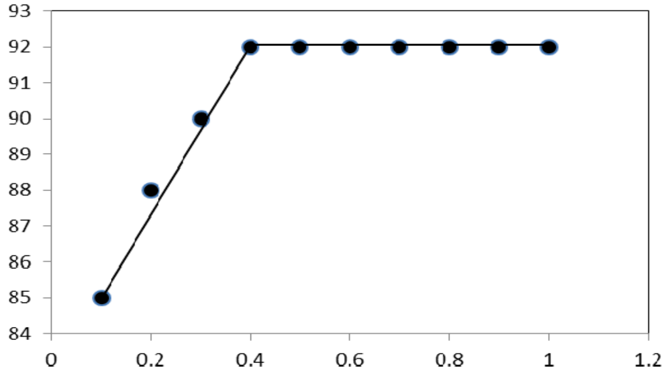


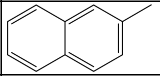
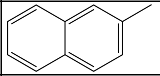
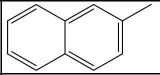
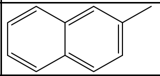
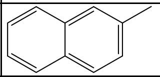
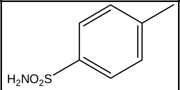
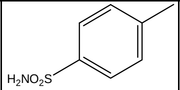
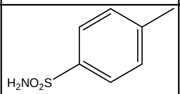
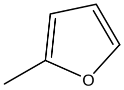
Figure 1. The effect of loading of catalyst on synthesis of substituted benzylidene-4-chloroanilines in presence of fly-ash: PTS catalyst under microwave irradiation (entry 30).

Table 1. Reusability of catalyst on condensation of 4-methoxy aniline (2 mmol) and benzaldehydes (2 mmol) (entry 30) under microwave irradiation.

Run	1	2	3	4	5
Yield	92	92	91.5	91.5	91

Table 2. Analytical and mass spectral data of aryl imines synthesized by aryl amines and substituted benzaldehydes reaction of the type Ar-NH_2 + Ar'-CHO → Ar-N=CH-Ar' under microwave irradiation.

Entry	Ar	Ar'	Product	M.W.	Yield (%)	M.p. (°C)	Mass (m/z)
1	C_6H_5	C_6H_5	$C_6H_5N{=}CHC_6H_5$	181	87	52-53 (50-52) [8]	---
2	C_6H_5	4-ClC_6H_4	$C_6H_5N{=}CHC_6H_4Cl(4)$	215	85	61 (60-62) [8]	---
3	C_6H_5	4-CH_3O-C_6H_4	$C_6H_5N{=}CHC_6H_4OCH_3(4)$	211	87	56 -57	---

						(54-56) [8]	
4	C_6H_5	4-$CH_3C_6H_4$	$C_6H_5N{=}CHC_6H_4CH_3$(4)	195	85	36-37 (35-37) [8]	---
5	C_6H_5	4-$NO_2C_6H_4$	$C_6H_5N{=}CHC_6H_4NO_2$(4)	226	86	88-90 (87-89) [8]	---
6		C_6H_5	$C_{10}H_7N{=}CHC_6H_5$	231	88	120-121 (118-120) [8]	---
7		4-ClC_6H_4	$C_{10}H_7N{=}CHC_6H_4Cl$(4)	265	85	94-95 (90-93) [8]	---
8		4-CH_3O-C_6H_4	$C_{10}H_7N{=}CHC_6H_4OCH_3$(4)	261	87	85-86 (84-86) [8]	---
9		4-$CH_3C_6H_4$	$C_{10}H_7N{=}CHC_6H_4CH_3$(4)	245	88	71 (68-70) [8]	---
10		4-$NO_2C_6H_4$	$C_{10}H_7N{=}CHC_6H_4NO_2$(4)	276	85	156-158 (153-155) [8]	---
11	2-Cl, 4-$NO_2C_6H_3$	4-Cl C_6H_4	2-Cl,4-$NO_2C_6H_3N{=}CHC_6H_4Cl$(4)	295	85	83-84 (78-82) [8]	---
12	2-Cl,4-$NO_2C_6H_3$	3-$NO_2C_6H_4$	2-Cl,4-$NO_2C_6H_3N{=}CHC_6H_4NO_2$(3)	306	85	77-78 (72-76) [8]	---
13		C_6H_5	$NO_2SC_6H_6$ $N{=}CHC_6H_5$	260	89	171-181 (170-180) [8]	---
14		3-NO_2-C_6H_4	$NO_2SC_6H_6$ $N{=}CH$ $C_6H_4NO_2$(3)	306	85	182-183 (180-182) [8]	---
15			$NO_2SC_6H_6N{=}CHC_4H_4O$	251	86	192-193 (190-192) [8]	---
16	4-$CH_3C_6H_4$	C_6H_5	4-$CH_3C_6H_4N{=}CHC_6H_5$	195	90	114-115 (112-115) [8]	---
17	4-$CH_3C_6H_4$	4-$CH_3OC_6H_4$	4-$CH_3C_6H_4N{=}CHC_6H_4OCH_3$(4)	226	92	95-96 (93-95) [8]	---
18	4-$CH_3C_6H_4$	4- $NO_2C_6H_4$	4-$CH_3C_6H_4N{=}CHC_6H_4NO_2$(4)	241	85	125-126 (121-125) [8]	---
19	3-$CH_3OC_6H_4$	C_6H_5	3-$CH_3OC_6H_4N{=}CHC_6H_5$	211	86	156-157 (154-157) [8]	---
20	4-$CH_3OC_6H_4$	4-OH C_6H_4	4-$CH_3OC_6H_4N{=}CHC_6H_4OH$(4)	228	88	221-22 (215-220) [8]	---
21	4-BrC_6H_4	C_6H_5	4-$BrC_6H_4N{=}CHC_6H_5$	260	85	62-63	---

						(61-62) [8]	
22	4-$NO_2C_6H_4$	C_6H_5	4-$NO_2C_6H_4N$=CHC_6H_5	226	88	140-141 (138-140) [8]	---
23	4-$NO_2C_6H_4$	4-$NO_2C_6H_4$	4-$NO_2C_6H_4N$=$CHC_6H_4NO_2$(4)	272	85	182-183 (178-182) [8]	---
24	4-IC_6H_4	4-OH, 3-OCH_3, 5-$NO_2C_6H_2$	4-IC_6H_4N=CHC_6H_2 4-OH,3-OCH_3,5-NO_2	398	85	135-36 (135) [8]	---
25	2-Cl-4-CH_3- C_6H_3	C_6H_5	2-Cl-4-CH_3-C_6H_3N=CHC_6H_5	228	88	120-121 119-120 [16]	---
26	2-Cl-4-CH_3- C_6H_3	2-Cl C_6H_4	2-Cl-4-CH_3-C_6H_3N=CHC_6H_4Cl(2)	261	85	65-66 64-65 [16]	---
27	2-Cl-4-CH_3- C_6H_3	2-FC_6H_4	2-Cl-4-CH_3-C_6H_3N=CHC_6H_4F(2)	247	85	61 60-61 [16]	---
28	2-Cl-4-CH_3- C_6H_3	4-$CH_3C_6H_4$	2-Cl-4-CH_3-C_6H_3N=$CHC_6H_4CH_3$(4)	243	87	61 60-61 [16]	
29	4-$OCH_3C_6H_4$	C_6H_5	(4)$H_3COC_6H_4N$=CHC_6H_5	177	90	50-51 [8]	---
30	4-ClC_6H_4	C_6H_5	4-ClC_6H_4N=CHC_6H_5	215	92	56	215[M^+], 217[M^{2+}], 180, 138, 111, 104, 77
31	4-ClC_6H_4	4-BrC_6H_4	4-ClC_6H_4N=CHC_6H_5-Br(4)	295	86	117 (119-120) [17]	295[M^+], 297[M^{2+}], 299[M^{4+}], 258, 182, 155, 138, 111
32	4-ClC_6H_4	2-ClC_6H_4	4-ClC_6H_4N=CHC_6H_5-Cl(2)	249	85	58-59	249[M^+], 251[M^{2+}], 253[M^{4+}], 214, 138, 111
33	4-ClC_6H_4	4-ClC_6H_4	4-ClC_6H_4N=CHC_6H_5-Cl(4)	249	87	106 (110-111)[17]	249[M^+], 251 [M^{2+}], 253[M^{4+}], 214, 138, 111
34	4-ClC_6H_4	4-FC_6H_4	4-ClC_6H_4N=CHC_6H_5-F(4)	233	86	66-67 64[18]	233[M^+], 235[M^{2+}], 198, 138, 122, 111, 95
35	4-ClC_6H_4	4-$OCH_3C_6H_4$	4-ClC_6H_4N=CHC_6H_5-OCH_3 (4)	245	92	92(90-94)[19]	245[M^+], 247[M^{2+}], 210, 138, 134, 111, 107
36	4-ClC_6H_4	4-$CH_3C_6H_4$	4-ClC_6H_4N=CHC_6H_5- CH_3 (4)	229	91	102-103	229[M^+], 231[M^{2+}], 194, 138, 122, 118, 111
37	4-ClC_6H_4	2-$NO_2C_6H_4$	4-ClC_6H_4N=CHC_6H_5-NO_2 (2)	261	87	98-99	261[M^+], 263[M^{2+}], 225, 149, 138, 122, 111
38	4-ClC_6H_4	3-$NO_2C_6H_4$	4-ClC_6H_4N=CHC_6H_5-NO_2 (3)	261	86	104-105	261[M^+], 263[M^{2+}], 225,

							149, 138, 122, 111
39	4-ClC_6H_4	4-$NO_2C_6H_4$	4-$ClC_6H_4N{=}CHC_6H_5$-NO_2 (4)	261	88	113-114 (110-113)[19]	261[M^+], 263[M^{2+}], 225, 149, 138, 122, 111

UV spectral study

In the present study the spectral linearity of synthesized imines (entries 30-39) has been studied by evaluating the substituent effects. The spectral data observed for the imines, UV (λ_{max}, nm), infrared νC=N, the proton chemical shifts δ (ppm), of C-H and carbon chemical shifts of C=N are correlated with various substituent constants. The measured absorption maxima (λ_{max}, nm) of synthesized imines are presented in Table 3. These values have been correlated with Hammett substituent constants and F and R parameters using single and multi-linear regression analysis [5-8, 14, 15] Hammett equation employed, for the correlation analysis, involving the absorption maxima is as shown below in equation (1).

$$\lambda = \rho\sigma + \lambda_o \tag{1}$$

where λ_o is the frequency for the parent member of the series.

The results of statistical analysis [5-8, 14, 15, 20] of these values with Hammett substituent constants are presented in Table 4. From Table 4, it is evident that Hammett σ and σ^+ constants and R parameters give satisfactory correlations with λ_{max}. The σ_I, σ_R and F values give poor correlations. All constants give negative ρ values. This is due to the fact that the polar, resonance, filed and inductive effects of the substituents are sufficiently weaker for predicting the reactivity on the absorption through conjugation. This is attributed to the resonance conjugative structure shown in Figure 2. The multi regression analysis of these UV spectral data of all imines with inductive, resonance and Swain-Lupton's [9] constants produce satisfactory correlations as shown in equations 2 and 3.

$$\lambda_{max}\,(\text{nm}) = 263.345(\pm 11.926) - 39.007(\pm 15.523)\,\sigma_I + 38.677\,(\pm 10.729)\,\sigma_R \tag{2}$$
$$(R = 0.970,\ n = 10,\ p > 95\%)$$

$$\lambda_{max}\,(\text{nm}) = 269.501(\pm 13.210) - 10.646\,(\pm 4.952)\,F - 40.724\,(\pm 24.589)\,R \tag{3}$$
$$(R = 0.955,\ n = 10,\ p > 95\%)$$

Table 3. The ultraviolet absorption maxima (λ_{max}, nm), infrared absorptions (ν, cm^{-1}) and NMR chemical shifts (δ, ppm) imines (entries 30-39).

Entry	X	λ_{max} (nm)	ν_{max} (cm^{-1})		^1H			^{13}C		
			CN	X	CH	Ar-H	X	C=N	Ar-C	X
30	H	262.5	1622.13	---	8.487	7.196-7.982 (*m*, 9H)	---	160.72	122.04-150.42	---
31	4-Br	273.0	1626.57	---	8.384	7.093- 7.799 (*m*, 8H)	---	159.27	119.683-150.562	---
32	2-Cl	266.2	1614.42	---	8.952	7.168- 8.320 (*m*, 8H)	---	158.56	122.330-150.355	---
33	4-Cl	270.5	1624.06	---	8.416	7.127- 8.226 (*m*, 8H)	---	159.12	122.205-150.113	---

34	4-F	269.0	1625.99	---	8.416	7.160- 7.934 (*m*, 8H)	---	159.10	122.179-150.289	---
35	4-CH_3	269.5	1614.42	---	8.428	7.160- 7.820 (*m*, 8H)	2.410 (*s*, 3H)	160.68	122.270-150.723	21.67 (CH_3)
36	4-OCH_3	317.5	1608.26	1236.8	8.379	7.002- 7.820 (*m*, 8H)	3.780 (*s*, 3H)	160.99	122.183-159.048	55.43 (OCH_3)
37	2-NO_2	237.5	1620.62	---	8.889	7.501- 8.372 (*m*, 8H)	---	160.84	122-85-155.536	---
38	3-NO_2	259.0	1602.72	---	8.556	7.210- 8.362 (*m*, 8H)	---	159.6	122.337-150.432	---
39	4-NO_2	289.5	1589.34	---	8.559	7.103-8.360 (*m*, 8H)	---	157.65	122.336-149.433	--

Table 4. Results of statistical analysis of UV λ_{max} (nm), νC=N (cm^{-1}) IR, NMR δ^1H (ppm) CH=N and $\delta^{13}C$ (ppm) C=N of substituted benzylidene-4-chlorobenzamines (entries 30-39) with Hammett substituent constants σ, σ^+, σ_I, σ_R, F and R parameters.

Frequency	Constants	r	I	ρ	s	n	Correlated derivatives
λ_{max} (nm)	σ	0.951	278.39	-27.67	18.91	7	H, 4-Br, 2-Cl, 4-Cl, 4-F, 4-CH_3, 3-NO_2
	σ^+	0.961	276.50	-25.23	17.33	7	H, 4-Br, 2-Cl, 4-Cl, 4-F, 4-CH_3, 3-NO_2
	σ_I	0.819	277.91	-15.76	21.59	10	H, 4-Br, 2-Cl, 4-Cl, 4-F, 4-CH_3, 4-OCH_3, 2-NO_2, 3-NO_2,4-NO_2
	σ_R	0.748	266.37	-44.29	19.19	10	H, 4-Br, 2-Cl, 4-Cl, 4-F, 4-CH_3, 4-OCH_3, 2-NO_2, 3-NO_2,4-NO_2
	F	0.718	277.39	-13.94	21.66	10	H, 4-Br, 2-Cl, 4-Cl, 4-F, 4-CH_3, 4-OCH_3, 2-NO_2, 3-NO_2,4-NO_2
	R	0.952	264.81	-41.56	18.60	7	H, 4-Br, 2-Cl, 4-Cl, 4-F, 4-CH_3, 3-NO_2,
νC=N	σ	0.838	1617.83	-11.83	11.67	10	H, 4-Br, 2-Cl, 4-Cl, 4-F, 4-CH_3, 4-OCH_3, 2-NO_2, 3-NO_2,4-NO_2
	σ^+	0.821	1615.99	-5.67	12.25	10	H, 4-Br, 2-Cl, 4-Cl, 4-F, 4-CH_3, 4-OCH_3, 2-NO_2, 3-NO_2,4-NO_2
	σ_I	0.723	1619.69	-11.76	12.21	10	H, 4-Br, 2-Cl, 4-Cl, 4-F, 4-CH_3, 4-OCH_3, 2-NO_2, 3-NO_2,4-NO_2
	σ_R	0.843	1612.30	-22.41	11.38	10	H, 4-Br, 2-Cl, 4-Cl, 4-F, 4-CH_3, 4-OCH_3, 2-NO_2, 3-NO_2,4-NO_2
	F	0.809	1618.59	-8.73	12.38	10	H, 4-Br, 2-Cl, 4-Cl, 4-F, 4-CH_3, 4-OCH_3, 2-NO_2, 3-NO_2,4-NO_2
	R	0.826	1612.05	-17.60	11.60	10	H, 4-Br, 2-Cl, 4-Cl, 4-F, 4-CH_3, 4-OCH_3, 2-NO_2, 3-NO_2,4-NO_2
δCH=N	σ	0.752	8.48	0.26	0.19	10	H, 4-Br, 2-Cl, 4-Cl, 4-F, 4-CH_3, 4-OCH_3, 2-NO_2, 3-NO_2,4-NO_2
	σ^+	0.758	8.50	0.24	0.18	10	H, 4-Br, 2-Cl, 4-Cl, 4-F, 4-CH_3, 4-OCH_3, 2-NO_2, 3-NO_2,4-NO_2
	σ_I	0.832	8.42	0.30	0.20	10	H, 4-Br, 2-Cl, 4-Cl, 4-F, 4-CH_3, 4-OCH_3, 2-NO_2, 3-NO_2,4-NO_2
	σ_R	0.832	8.58	0.31	0.21	10	H, 4-Br, 2-Cl, 4-Cl, 4-F, 4-CH_3, 4-OCH_3, 2-NO_2, 3-NO_2,4-NO_2
	F	0.722	8.45	0.21	0.21	10	H, 4-Br, 2-Cl, 4-Cl, 4-F, 4-CH_3, 4-OCH_3, 2-NO_2, 3-NO_2,4-NO_2
	R	0.741	8.60	0.32	0.20	10	H, 4-Br, 2-Cl, 4-Cl, 4-F, 4-CH_3, 4-OCH_3, 2-NO_2, 3-NO_2,4-NO_2
δC=N	σ	0.941	159.95	-1.18	1.09	8	H, 4-Br, 2-Cl, 4-Cl, 4-F, 4-CH_3, 4-OCH_3, 4-NO_2
	σ^+	0.951	159.88	-1.42	1.02	8	H, 4-Br, 2-Cl, 4-Cl, 4-F, 4-CH_3,

							4-OCH_3, 4-NO_2
	σ_I	0.957	160.68	-2.51	0.97	7	H, 4-Br, 2-Cl, 4-Cl, 4-F, 4-CH_3, 4-NO_2
	σ_R	0.704	159.62	-0.24	1.19	10	H, 4-Br, 2-Cl, 4-Cl, 4-F, 4-CH_3, 4-OCH_3, 2-NO_2, 3-NO_2,4-NO_2
	F	0.905	160.63	-2.30	1.00	7	H, 4-Br, 2-Cl, 4-Cl, 4-F, 4-CH_3, 4-NO_2
	R	0.811	159.58	-0.43	1.18	10	H, 4-Br, 2-Cl, 4-Cl, 4-F, 4-CH_3, 4-OCH_3, 2-NO_2, 3-NO_2,4-NO_2

r = correlation co-efficient; ρ = slope; I = intercept; s = standard deviation; n = number of substituents.

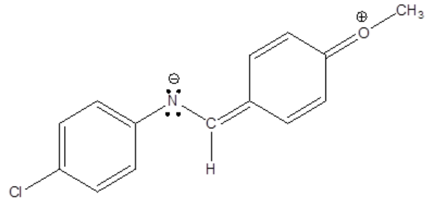


Figure 2. The resonance-conjugative structure.

IR spectral study

The infrared νC=N stretching frequencies (cm^{-1}) of the synthesized imines (entries 30-39) have been recorded and presented in Table 3. These data are correlated [5-8, 14, 15, 20] with Hammett substituent constants and Swain-Lupton's [9] parameters. In this correlation the structure parameter relation equation employed is as shown in equation (4).

$$\nu = \rho\sigma + \nu_o \tag{4}$$

where ν_o is the frequency for the parent member of the series.

The observed νC=N stretching frequencies (cm^{-1}) are correlated with various Hammett substituent constants and F and R parameters through single and multi-regression analyses including Swain-Lupton's [9] parameters. The results of statistical analysis of single parameter correlation are shown in Table 4. The correlation of νC=N (cm^{-1}) frequencies of imines with all the Hammett substituent constants and F and R parameters fail to give a satisfactory correlation, All correlations give negative ρ value. This is due to the absence of polar, field and inductive effects of the substituents and hence they are unable to predict the reactivity on C=N stretches. This is associated with the conjugative structure shown in Figure 2. In short all the single parameter correlations of νC=N (cm^{-1}) frequencies with Hammett substituent constants fail in correlation. So, the authors think that it is worthwhile to seek the multi regression analysis which may produce a satisfactory correlation with Resonance, Field and Swain-Lupton's [9] constants. This is shown in the following equations 5 and 6.

$$\nu C{=}N\ (cm\text{-}1) = 1618.593(\pm 9.264) - 2.943\ (\pm 1.054)\ \sigma_I - 7.640\ (\pm 3.875)\ \sigma_R \tag{5}$$
$$(R = 0.972,\ n = 10,\ p > 95\%)$$

$$\nu C{=}N(cm\text{-}1) = 1615.292\ (\pm 8.216) - 7.352\ (\pm 1.510)\ F - 17.013(\pm 1.284)\ R \tag{6}$$
$$(R = 0.945,\ n = 10,\ p > 95\%)$$

^{1}*H NMR spectral study*

The ^{1}H NMR spectra of the imine derivatives (entries 30-39) under investigation have been recorded in deuteriochloroform solution employing tetramethylsilane (TMS) as internal standard. The signals of the imine protons have been assigned and are presented in Table 3. In nuclear magnetic resonance spectra, the ^{1}H or the ^{13}C chemical shifts (δ, ppm) depends on the electronic environment of the nuclei concerned. These chemical shifts have been correlated with reactivity parameters. Thus the Hammett equation has been used in the form as shown in (7).

$$\log \delta = \log \delta_0 + \rho\sigma \tag{7}$$

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

The assigned proton chemical shifts (ppm) of imines have been correlated [5-8, 14, 15, 20] with various Hammett sigma constants, F and R parameters. The results of statistical analysis are presented in Table 3. These proton chemical shifts (ppm) fail in correlation with Hammett substituent constants and F and R parameters. All correlations give positive ρ values. This shows that the normal substituent effect operates in all systems. The failure in correlation is attributed to the conjugative structure shown in Figure 2. In view of the inability of the Hammett σ constants to produce individually satisfactory correlations with the imine proton chemical shifts, the authors think that, it is worthwhile to seek multiple correlations involving either σI and σR constants or Swain-Lupton's [9] F and R parameters. This is shown in the following equations (8) and (9) for protons:

$$\delta\text{CH(ppm)} = 8.470(\pm0.144) + 0.253\ (\pm0.028)\ \sigma_I + 0.244(\pm0.031)\ \sigma_R \tag{8}$$
$$(R = 0.945,\ n = 10,\ p > 95\%)$$

$$\delta\text{CH(ppm)} = 8.515\ (\pm0.140) + 0.187\ (\pm0.026)\ F + 0.309(\pm0.025)\ R \tag{9}$$
$$(R = 0.948,\ n = 10,\ p > 95\%)$$

^{13}*C spectral study*

The chemical shifts (ppm) of imine C=N carbon, have been assigned and are presented in Table 2. Attempts have been made to correlate the δC=N chemical shifts (ppm) with Hammett substituent constants, field and resonance parameters, with the help of single and multi-regression analyses to study the reactivity through the effect of substituents. The chemical shifts (ppm) observed for the δC=N has been correlated [5-8, 14, 15, 20] with Hammett constants and the results of statistical analysis are presented in Table 4. The δC=N chemical shifts (ppm) give satisfactory correlation with Hammett σ, σ^+ and $\sigma_{I,}$ constants and F parameter. The remaining Hammett σ_R, constant and R parameter are found to fail in correlation. This is due to the reason stated earlier with resonance conjugative structure shown in Figure 2. In view of inability of some of the σ constants to produce individually satisfactory correlation, the authors think that it is worthwhile to seek multiple correlation involving σ_I, σ_R, F and R parameters. This is given in the following correlation equations (10) and (11).

$$\delta\text{C=N (ppm)} = 160.776(\pm0.716) - 2.606(\pm1.388)\ \sigma_I + 0.439\ (\pm0.054)\ \sigma_R \tag{10}$$
$$(R = 0.958,\ n = 10,\ p > 95\%)$$

$$\delta\text{C=N (ppm)} = 160.587(\pm0.719) - 2.288(\pm1.059)\ F - 0.254(\pm0.033)\ R \tag{11}$$
$$(R = 0.954,\ n = 10,\ p > 95\%)$$

Antibacterial activity

Antibacterial sensitivity assay has been performed by using [13] disc diffusion technique. In each Petri plate about 0.5 mL of the test bacterial sample has been spread uniformly over the solidified Mueller Hinton agar using sterile glass spreader. Then the discs with 5 mm diameter made up of Whatmann No. 1 filter paper, impregnated with the solution of the compound have been placed on the medium using sterile forceps. The plates have been incubated for 24 hours at 37 °C by keeping the plates upside down to prevent the collection of water droplets over the medium. After 24 hours, the plates have been visually examined and the diameter values of the zone of inhibition were measured. Triplicate results have been recorded by repeating the same procedure.

The antibacterial screening effect of synthesized Schiff bases (entries 30-39) is shown in (Figure 3) (Plates 1-10). The antibacterial activities of all the synthesized imines have been studied against three gram positive pathogenic strains *Micrococcus luteus*, *Bacillus substilis*, *Staphylococcus aureus* and two gram negative strains *Escherichia coli* and *Pseudomonas species*. The disc diffusion technique was followed, at a concentration of 250 µg/mL with ampicillin taken as the standard drug. The zone of inhibition is compared using Table 5 and the corresponding clustered column chart is shown in (Figure 4). A good antibacterial activity has been possessed by all substituents on the microorganisms in general. The substituents H, 4-OCH_3 and 3-NO_2 have shown very good activity against *Micrococcus luteus.* The substituents 4-Br, 4-Cl, 4-CH_3 and 4-OCH_3 have improved antibacterial activity against *E. coli* species. The substituent 4-CH_3 has very good activity against *Pseudomonas* and *Bacillus subtilis.* The substituents H, 4-Cl, 4-OCH_3, 3-NO_2 have good antibacterial activity against *Staphylococcus aureus.*

Table 5. Antibacterial activity of substituted benzylidene-4-chloroanilines

Entry	X	Zone of inhibition (mm)				
		Gram positive bacteria			Gram negative bacteria	
		B. subtilis	*M. luteus*	*S. aureus*	*E. coli*	*P. aeruginosa*
30	H	7	12	10	7	8
31	4-Br	9	8	7	11	7
32	2-Cl	9	9	7	8	8
33	4-Cl	8	8	9	10	7
34	4-F	9	8	8	9	8
35	4-CH_3	10	8	7	11	10
36	4-OCH_3	7	12	9	10	8
37	2-NO_2	8	7	8	8	9
38	3-NO_2	9	11	9	9	7
39	4-NO_2	7	7	7	7	8
Standard	AMPICILLIN	13	17	15	15	13
Control	DMSO	-	-	-	-	-

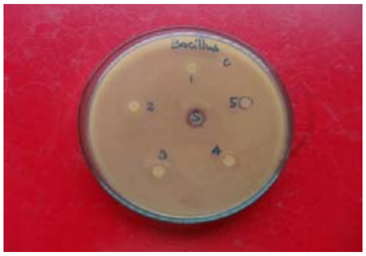


PLATE I

PLATE II

PLATE III

PLATE IV

PLATE V

PLATE VI

PLATE VII

PLATE VIII

PLATE IX PLATE X

Figure 3. Antibacterial activity of substituted benzylidene-4-chloroanilines-petri dishes (30-39).

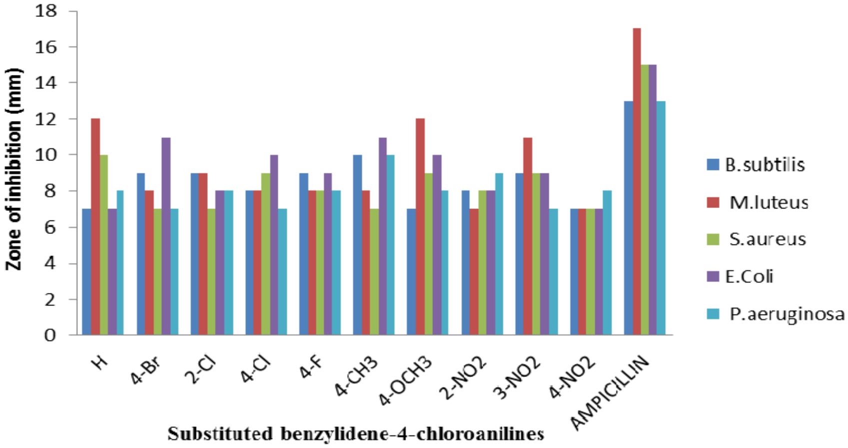


Figure 4. The antibacterial activities of substituted benzylidene-4-chloroanilines-clustered column chart (30-39).

Antifungal activities

Antifungal sensitivity assay has been performed using [13] disc diffusion technique. PDA medium was prepared and sterilized as above. It has been poured (ear bearing heating condition) in the Petri-plate which has been already filled with 1 mL of the fungal species. The plates have been rotated clockwise and counter clock-wise for uniform spreading of the species. The discs have been impregnated with the test solution. The test solution has been prepared by dissolving 15 mg of the Schiff bases in 1 mL of DMSO solvent. The medium have been allowed to solidify and kept for 24 hours. Then the plates have been visually examined and the diameter values of zone of inhibition have been measured. Triplicate results have been recorded by repeating the same procedure. The antifungal activities of substituted imines (entries 30-39) have been studied and are shown in (Figure 5) for Plates (1-4) and the zone of inhibition values of the effect is given in Table 6. The clustered column chart, shown in (Figure 6) reveals that all the compounds have moderate antifungal activity against both the fungal species namely *A. niger* and *Penicilium scup.*

Table 6. Antifungal activity of substituted benzylidene-4-chloroanilines

Entry	**X**	Zone of inhibition (mm)	
		A. niger	*P. scup*
30	H	8	7
31	4-Br	6	7
32	2-Cl	7	-
33	4-Cl	-	6
34	4-F	-	6
35	4-CH_3	8	6
36	4-OCH_3	7	7
37	2-NO_2	7	-
38	3-NO_2	7	-
39	4-NO_2	6	-
Standard	Miconazole	13	12
Control	DMSO	-	-

PLATE I PLATE II

PLATE III PLATE IV

Figure 5. Antifungal activity of substituted benzylidene-4-chloroanilines-petri dishes (entries 30-39).

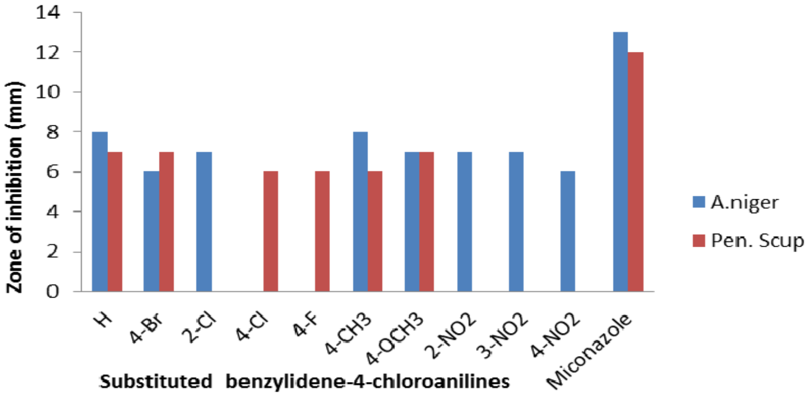


Figure 6. Antifungal activity of substituted benzylidene-4-chloroaniline-clustered column chart (entries 30-39).

CONCLUSIONS

A series of some 2*E*-imines including substituted benzylidene-4-chloroanilines have been synthesized by condensation of 4-chloroaniline and substituted benzaldehydes using microwave irradiation in the presence of fly-ash:PTS under solvent-free conditions. This reaction protocol offers a simple, eco-friendly; nonhazardous, easier work-up procedure and good yields. These imines are characterized by their physical constants, spectral data. The UV, IR, NMR spectral data of these imines has been correlated with Hammett substituent constants, F and R parameters. From the results of statistical analyses the effects of substituent on the spectral data have been studied. The antimicrobial activities of all synthesized imines have been studied using Bauer-Kirby method.

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REFERENCES

1. Schiff, H.J. *Lieb. Ann. Chem.* **1864**, 131, 118.
2. (a) Bharti, S.K.; Nath, G.; Tilak, R.; Singh, S.K. *Eur. J. Med. Chem*. **2010**, 45, 651. (b) Shabani, F.; Saghatforoush, L.A.; Ghammamy, S. *Bull. Chem. Soc. Ethiop*. **2010**, 24, 193.
3. Rosu, T.; Pahontu, E.; Maxim, C.; Georgescu, R.; Stanica, N.; Gulea, A. *Polyhedron* **2011**, 30, 154.
4. Gudipati, R.; Anreddy, R.N.; Manda, S. *Saudi Pharm. J.* **2011**, 19, 153.
5. Thirunarayanan, G.; Vanangamudi, G. *Spectrochim. Acta* **2011**, 81A, 390.
6. Thirunarayanan, G.; Ananthakrishna Nadar, P. *Indian J. Chem. Soc*. **2006**, 83, 1107.
7. Thirunarayanan, G.; Vanangamudi, G.; Sathiyendiran, V.; Ravi, K. *Indian J. Chem*. **2011**, 50B, 593.
8. Suresh, R.; Kamalakkannan, D.; Ranganathan, K.; Arulkumaran, R.; Sundararajan, R.; Sakthinathan, S.P.; Vijayakumar, S.; Sathiyamoorthi, K; Mala, V.; Vanangamudi, G.; Thirumurthy, K.; Mayavel, P.; Thirunarayanan, G. *Spectrochim. Acta* **2013**, 101A, 239.
9. Swain, C.G.; Lupton, E.C., Jr. *J. Am. Chem. Soc*. **1968**, 90, 4328.

10. Thirunarayanan, G.; Vanangamudi, G. *IUP. J. Chem*. **201**1, 1, 54.
11. Yang, H.J.; Sun, W.H.; Li, Z.L.; Ma, Z. *Chin. Chem. Lett*. **2002**, 13, 3.
12. Lidstorm, P.; Tierney, J.; Wathey, B.; Westman, J. *Tetrahedron* **2001,** 57, 9225.
13. Bauer, A.W.; Kirby, W.M.M.; Sherris, J.C.; Truck, M. *Am. J. Clin. Pathol*. **1966**, 45, 493.
14. Arulkumaran, R.; Vijayakumar, S.; Sakthinathan, S.P.; Kamalakkannan, D.; Ranganathan, K.; Suresh, R.; Sundararajan, R.; Vanangamudi, G.; Thirunarayanan, G. *J. Chil. Chem. Soc.* **2013**, 58, 1684.
15. Thirunarayanan, G.; Mayavel, P.; Thirumurthy, K. *Spectrochim. Acta* **2012**, 91A, 18.
16. Vananja, G.; Kanagambal, P.; Thirunarayanan, G. National Conference on Exploring Excellence in Chemistry, NCEEC-12, December 21, 2012, Department of Chemistry, Dhanalakshmi Srinivasan College of Arts and Science for Women, Perambalur-621 212, India; **2012**.
17. Neuvonen, H.; Neuvonen, K.; Koch, A.; Kleinpeter, E. *J. Mol. Struct*. **2007**, 815, 95.
18. Karki, S.S.; Butle, S.R.; Shaikh, R.M.; Zubaidha, P.K.; Pedgaonkar, G.S.; Shendarkar, G.S.; Chitra G.R. *Res. J. Pharm. Biol. Chem. Sci*. **2010**, 1, 707.
19. Patil, S.; Jadhav, S.D.; Shinde, S.K. *Org. Chem. Int.* **2012**, 2012, 1-5; Article ID 153159, http://dx.doi.org/10.1155/2012/153159.
20. Sakthinathan, S.P.; Suresh, R.; Mala, V.; Sathiyamoorthi, K.; Kamalakkannan, D.; Ranganathan, K.; Arulkumaran, R.; Vijayakumar, S.; Sundararajan, R.; Vanangamudi, G.; Thirunarayanan, G. *Int. Lett. Chem. Phys. Astro*. **2013**, 6, 77.