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Research Article

CONVENTIONAL AND MICROWAVE ASSISTED SYNTHESIS AND QSAR STUDIES OF COUMARINYLCHALCONES AS POTENT ANTIMICROBIAL AGENTS

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ABSTRACT

Synthesis of 3-aryl-1-(3′-coumarinyl) propen-1-ones was undertaken by the conventional and the microwave–assisted methods by reacting 3-acetylcoumarin and various substituted aromatic aldehydes. The microwave-assisted synthesis provided an eco-friendly and an efficient method of synthesis since it resulted in the shorter reaction time, higher yields and simple operations, when compared with the conventional heating method. The synthesized compounds were screened for the antimicrobial activity against gram positive and gram negative bacteria as well as pathogenic fungi. The quantitative structure activity relationships (QSAR) studies of these compounds were performed using the software *Strike* from Schrodinger, USA, by simple linear regression analysis. The best correlated QSAR model depicted that the weakly polar component of the total solvent accessible area (WPSA) was significant for the antibacterial activity of coumarinylchalcones against *E. coli*.

Keywords: Coumarinylchalcones, conventional synthesis, microwave-assisted synthesis, antimicrobial activity, QSAR.

INTRODUCTION

Infectious diseases caused by bacteria and fungi affect millions of people worldwide. A number of antimicrobial agents have been used to treat these infections but most of them cause adverse effects and also pose a problem of resistance. Hence, there is an urgent need for the new and effective antimicrobial agents.

Coumarins are the compounds that display a special role in nature and have raised considerable interest because of their interesting biological and pharmacological activities[1, 2]. Coumarin derivatives have been reported to possess anticoagulant[3], antibacterial[4], anti-inflammatory[5], anti-oxidant[6], anthelmintic[7], anti- HIV[8] and anticancer[9] activities. Chalcones are α, β-unsaturated ketones containing a reactive ketoethylenic group, –CO-CH=CH. The presence of α, β- unsaturated carbonyl system in their structure makes them biologically active. Depending on the substitution pattern on the aromatic rings attached to the ketoethylenic group, a wide range of pharmacological activities have been identified[10, 11]. Encouraged from these findings, it was decided to combine two potent moieties, coumarin and a chalcone, to synthesize

coumarinylchalcones and screen them against pathogenic microbes.

In the recent decades, the microwave heating has taken an incontestable place in the analytical and organic laboratories, for providing an efficient and non-polluting method of activation. The microwaves consist of electromagnetic energy and the energy transfer is produced by the dielectric loss[12].

CHEMISTRY

The general method for the synthesis of coumarinylchalcones followed the mechanism of Knoevenagel condensation[13] and is presented as **Scheme1**. 3-Aryl-1-(3′-coumarinyl) propen-1-ones (**2a-2k**) were synthesized by the reaction of 3-acetyl coumarin with various substituted aromatic aldehydes (**1a-1k**) in the presence of piperidine and glacial acetic acid, using ethanol as a solvent[14,15]. An appropriate reaction medium is of crucial importance in the successful microwave-promoted organic transformations. The microwave irradiation was carried out at 560 Watts, after optimizing the power from lower to higher in relation to the time of reaction. The reaction time for all the products was optimized by monitoring the completion of a reaction at frequent intervals using Thin Layer Chromatography (TLC). The formation of compounds **2a-2k** was evidenced by the appearance of a doublet in the range of δ = 6.86-7.69 ppm in the ^{1}HNMR spectrum, due to the presence of a CH=CH unit in coumarinylchalcones.

MATERIALS AND METHODS

The starting material, 3-acetylcoumarin, was obtained as the gift sample from Syn-Fine chemicals Pvt. Ltd., Hyderabad, India. The solvents were dried and purified when necessary according to the conventional procedures mentioned in the literature. The reactions were monitored by TLC on the pre-coated silica gel plates from E. Merck and Co. (Darmstadt, Germany). The melting points of the synthesized compounds were determined by the open capillaries using the Expo-Hi-tech melting point apparatus, and are uncorrected. The structures of the synthesized derivatives were characterized by their IR spectra, recorded on JASCO FTIR 5300 spectrometer, by the KBr disc method. The ^{1}H NMR spectra were recorded on AS - I -10 spectrometer using TMS as an internal reference. Chloroform and DMSO were used as solvents for recording ^{1}H NMR spectra.

General procedure for the synthesis of 3-aryl-1-(3′-coumarinyl) propen-1-ones (2a-k):

Conventional method: Coumarinylchalcones were synthesized by reacting 3-acetylcoumarin with various substituted aromatic aldehydes **(1a-1k)**. 3-Acetylcoumarin (1.88 g, 0.01 M) and the substituted aromatic aldehydes (0.02 M) were dissolved in 10 mL of ethanol by heating. Piperidine (0.4 mL) was added to this reaction mixture, followed by the addition of glacial acetic acid (0.3 mL). The reaction was carried out under reflux till its completion, as indicated by TLC. Ethanol was recovered by vacuum distillation after the completion of a reaction and the residue was triturated with 1 mL of methanol. The reaction mixture was filtered off and the crude product was recrystallized using an appropriate solvent.

Microwave (MW) irradiation method: The reaction mixture, as in the conventional method of synthesis, was subjected to the microwave irradiation at 560 watts and refluxed till the completion of a reaction, which was monitored by TLC. The reaction mixture was then processed as per the procedure followed in the conventional method. The melting points, R_f values and the spectral data for coumarinylchalcones are given below:

1-(3'-Coumarinyl)-3-phenyl-2-propen-1-one (2a)

M.P. 82-84 ˚C, **TLC** [Benzene: ethyl acetate (3: 0.5)], **R_f**: 0.42, **Molecular formula**: $C_{18}H_{12}O_3$, **FTIR** (KBr, cm^{-1}): 3030 (Ar, C-H str*), 2960 (alkane, C-H str), 1739 (C=O str), 1680 (C=O str, α, β-unsaturated, cyclic), 1612 (C=C str); **^{1}H NMR** ($CDCl_3$, δ ppm): 8.592 (s, 1H, C-H), 7.932 (d, 2H, -CH=CH-), 7.640 (m, 4H, Coumarin), 7.419-7.266 (m, 5H, Ar-H)

1-(3'-Coumarinyl)-3-(4"-methoxyphenyl)-2-propen-1-one (2b)

M.P. 140-142 ˚C, **TLC** [Benzene: ethyl acetate (3: 0.5)], **R_f**: 0.85, **Molecular formula**: $C_{19}H_{14}O_4$, **FTIR** (KBr, cm^{-1}): 2924 (Ar, C-H str), 1707 (C=O str), 1633 (C=O str, α, β-unsaturated, cyclic), 1604 (C=C str), 1454 (alkane, C-H str),

1340 (C-O str); **^{1}H NMR** ($CDCl_3$, δ ppm): 8.517 (d, 1H, Coumarin), 7.845 (d, 1H, =C-H), 7.688-7.634 (m, 4H, Ar-H), 7.417-7.263 (m, 3H, Coumarin), 6.950 (d, 1H, =C-H), 3.863 (s, 1H, Coumarin), 2.736 (s, 3H, O-CH_3)

1-(3'-Coumarinyl)-3-(4"-chlorophenyl)-2-propen-1-one (2c)

M.P. 188-190 ˚C, **TLC** [Benzene: ethyl acetate (3: 0.5)], **R_f**: 0.80, **Molecular formula**: $C_{18}H_{11}O_3Cl$, **FTIR** (KBr, cm^{-1}): 2920 (Ar, C-H str), 1718 (C=O str), 1666 (C=O str, α, β-unsaturated, cyclic), 1610 (C=C str), 758 (C- Cl str); **^{1}H NMR** ($CDCl_3$, δ ppm): 8.599 (s, 1H, Coumarin), 7.966 (d, 1H, =C-H), 7.831 (d, 1H, =C-H), 7.697-7.592 (m, 4H, Coumarin), 7.416-7.268 (m, 4H, Ar-H)

1-(3'-Coumarinyl)-3-(2"-chlorophenyl)-2-propen-1-one (2d)

M.P. 204-206 ˚C, **TLC** [Benzene], **R_f**: 0.75, **Molecular formula**: $C_{18}H_{11}O_3Cl$, **FTIR** (KBr, cm^{-1}): 2924 (Ar, C-H str), 1724 (C=O str), 1664 (C=O str, α, β-unsaturated, cyclic), 1610 (C=C str), 756 (C-Cl str); **^{1}H NMR** ($CDCl_3$, δ ppm): 8.615 (s, 1H, Coumarin), 8.262 (d, 1H, =C-H), 7.967 (d, 1H, =CH), 7.707-7.652 (m, 4H, Coumarin), 7.453-7.264 (m, 4H, Ar-H)

1-(3'-Coumarinyl)-3-(4"-hydroxyphenyl)-2-propen-1-one (2e)

M.P. 176-178 ˚C, **TLC** [Benzene: ethyl acetate (3: 0.5)], **R_f**: 0.44, **Molecular formula**: $C_{18}H_{12}O_4$, **FTIR** (KBr, cm^{-1}): 3261 (O-H str), 2920 (Ar, C-H str), 1689 (C=O str, α, β-unsaturated, cyclic), 1602 (C=C str), 1338 (O-H b†); **^{1}H NMR** ($CDCl_3$, δ ppm): 10.174 (s, 1H, O-H), 8.670 (s, 1H, Coumarin), 7.949 (d,1H, Coumarin), 7.772-7.608 (m, 4H, Ar-H), 7.508-7.432 (m, 3H, Coumarin), 6.859 (d, 2H, -CH=CH-)

1-(3'-Coumarinyl)-3-(2"-hydroxyphenyl)-2-propen-1-one (2f)

M.P. 78-79 ˚C, **TLC** [Benzene: ethyl acetate (3: 0.5)], **R_f**: 0.58, **Molecular formula**: $C_{18}H_{12}O_4$, **FTIR** (KBr, cm^{-1}): 3416 (O-H str), 2928 (Ar, C-H str), 1732 (C=O str), 1682 (C=O str, α, β-unsaturated, cyclic), 1608 (C=C str), 1365 (O-H b); **^{1}H NMR** ($CDCl_3$, δ ppm): 8.670 (s, 1H, O-H), 7.976 (s, 1H, Coumarin), 7.754 (d, 2H, -CH=CH-), 7.486-7.424 (m, 4H, Ar-H), 7.100-6.937 (m, 4H, Coumarin)

1-(3'-Coumarinyl)-3-(2"-furyl)-2-propen-1-one (2g)

M.P. 101-103 ˚C, **TLC** [Benzene], **R_f**: 0.45, **Molecular formula**: $C_{16}H_{10}O_4$, **FTIR** (KBr, cm^{-1}): 2930 (Ar, C-H str), 1726 (C=O str), 1608 (C=C str); **^{1}H NMR** ($CDCl_3$, δ ppm): 8.584 (d, 1H, =C-H), 7.832 (d, 1H, =C-H), 7.702-7.633 (m, 3H, Coumarin), 7.572 (s, 1H, Coumarin), 7.495-7.343 (m, 3H, furyl), 6.859 (d, 1H, Coumarin)

1-(3'-Coumarinyl)-3-(2", 4", 6"-trimethoxy phenyl)-2-propen-1-one (2h)

M.P. 158-160 ˚C, **TLC** [Benzene: ethyl acetate (3: 0.5)], **R_f**: 0.55, **Molecular formula**: $C_{21}H_{18}O_6$, **FTIR** (KBr, cm^{-1}): 2945 (Ar, C-H str), 1728 (C=O str), 1660 (C=O str, α, β-unsaturated, cyclic), 1610 (C=C str), 1356 (C-O str); **^{1}H NMR** ($CDCl_3$, δ ppm): 8.612 (s, 1H, Coumarin), 7.898 (d, 2H, Coumarin), 7.782-7.668 (d, 2H, Coumarin), 7.443-7.362 (d, 2H, -CH=CH-), 6.923 (s, 2H, Ar-H), 3.942 (s, 9H, 3xOCH_3)

1-(3'-Coumarinyl)-3-(3", 5"-dimethoxyphenyl) -2-propen-1-one (2i)

M.P. 147-149 ˚C, **TLC** [Benzene: ethyl acetate (3: 0.3)], **R_f**: 0.63, **Molecular formula**: $C_{20}H_{16}O_5$, **FTIR** (KBr, cm^{-1}): 2926 (Ar, C-H str), 1722 (C=O str), 1655 (C=O str, α, β-unsaturated, cyclic), 1606 (C=C str), 1344 (C-O str); **^{1}H NMR** ($CDCl_3$, δ ppm): 8.605 (s, 1H, Coumarin), 7.856 (d, 2H, Coumarin), 7.686 (t, 2H, Coumarin), 7.712-7.658 (d, 2H, -CH=CH-), 7.486 (s, 1H, ArH), 7.226 (s, 1H, Ar-H), 6.932 (s, 1H, Ar-H), 3.972 (d, 6H, 2xOCH_3)

1-(3'-Coumarinyl)-3-(4"-dimethylamino phenyl)-2-propen-1-one (2j)

M.P. 108-110 ˚C, **TLC** [Benzene: ethyl acetate (3: 0.3)], **R_f**: 0.60, **Molecular formula**: $C_{20}H_{17}O_3N$, **FTIR** (KBr, cm^{-1}): 3435 (N-H, str), 1344 (C-N str), 1734 (C=O str), 1645 (C=O str, α, β-unsaturated, cyclic), 1606 (C=C str), 1568 (N-H, b); **^{1}H NMR** ($CDCl_3$, δ ppm): 8.583 (s, 1H, Coumarin), 7.929 (d, 1H, Coumarin), 7.777-7.656 (m, 3H, Coumarin), 7.626 (d, 2H, Ar-H), 7.424 (d, 2H, -CH=CH-), 6.720 (d, 2H, Ar-H), 3.077 (s, 6H, 2xCH_3)

1-(3'-Coumarinyl)-3-(4"-fluorophenyl)-2-propen-1-one (2k)

M.P. 171-173 ˚C, **TLC** [Benzene: ethyl acetate (3: 0.3)], **R_f**: 0.70, **Molecular formula**: $C_{18}H_{11}O_3F$, **FTIR** (KBr, cm^{-1}): 1718 (C=O str), 1666 (C=O str, α, β-unsaturated, cyclic), 1612

(C=C str); **^{1}H NMR** ($CDCl_3$, δ ppm): 8.626 (s, 1H, Coumarin), 7.899 (d, 2H, Coumarin), 7.721-7.675 (t, 2H, Coumarin), 7.444-7.359 (d, 2H, -CH=CH-), 7.157-7.100 (d, 4H, Ar-H)
*str = stretching, † b = bending

Antimicrobial activity
All the synthesized coumarinylchalcones were screened against gram positive bacteria (*S. aureus* and *P. aeruginosa*), gram negative bacteria (*E. coli, K. pneumoniae*) and fungi (*C. albicans* and *A.* niger) at 100, 250, 500 and 1000 µg/ mL concentrations using an agar cup plate method[16]. The zones of inhibition (ZOI) of the compounds were compared with those of the standard antibacterial agent, sulphamethoxazole (100-1000 µg/mL) and the standard antifungal agent, miconazole (100-1000 µg/mL). Dimethylsulfoxide was used as the negative control.

Quantitative structure activity relationships (QSAR) studies
The quantitative structure activity relationships (QSAR) studies were carried out on all the eleven synthesized coumarinylchalcones in order to identify the physicochemical parameter responsible for their antimicrobial activity. The structures of the synthesized compounds were fed to the computer using the software *Ligprep* from Schrödinger, USA, which produced a single, low-energy, 3D structure with the correct chirality, for each successfully processed input structure. The software *QikProp* from Schrodinger predicts physically significant descriptors and pharmaceutically relevant properties of organic molecules. Simple regression analysis was performed using the software *Strike* from Schrodinger, to determine the correlation between the physicochemical descriptors and the Log ZOI values of coumarinylchalcones against *E. coli*. The best QSAR model was chosen based on the statistical parameters like the square of the correlation coefficient (r^2), the Fischer's value of significance (F) and the standard error of estimate (s). The best QSAR model was validated by the *internal* and the *external validation* methods. For this purpose, the dataset of 11 coumarinylchalcones was divided into a *training set* (6 compounds) and a *test set* (5 compounds), randomly. The QSAR models were developed using 6 coumarinylchalcones in the training set and correlating their Log ZOI values with various physicochemical descriptors using the program *Strike*[17]. The best QSAR model represented the relation between the weakly polar component of the total solvent accessible area (WPSA) and Log ZOI values for *E. coli* at 250 µg/mL and is represented by **equation 2**. For the *internal predictivity,* the antibacterial activity of six compounds in the *training set* was predicted by using **equation 2** and this predicted antibacterial activity was correlated with their experimentally determined antibacterial activity. The most vital validation in QSAR is the *external validation*, which consists of making predictions for the compounds, which are not used in the *training set*. These compounds form a *test set*. The best QSAR model (**equation 2**) was used to predict the antibacterial activity of the five compounds from the *test set*. The experimentally obtained antibacterial activity of these compounds was then correlated with their predicted antibacterial activity using the software *"Strike"*. The predictivity of the best QSAR model was judged based on the values of the square of the correlation coefficient (r^2), Fischer's value (F) and the standard error (s) of estimate.

RESULTS AND DISCUSSION

Synthesis
The comparison between the conventional and MW-assisted synthesis for the reaction time and % yield of coumarinylchalcones is shown in **Table 1.** The reaction time for the reactants differed based on the functional groups present on them and their electronic effects. The reaction time was reduced considerably from hours to minutes in the MW irradiation method. The MW irradiation also resulted in the increase in the yield of the products in the range of 4-10 %.

Antimicrobial activity
The results of the antibacterial and antifungal activities are tabulated in **Tables 2** and **3**, respectively. The antibacterial activity against all the bacterial strains was significant when compared with the standard, sulfamethoxazole, and is represented graphically in **Figures 1, 2, 3** and **4**. **Table 4** lists the compounds having antibacterial activity greater than that of sulfamethoxazole

and **Table 5** lists the compounds having antibacterial activity comparable to the antibacterial activity of sulfamethoxazole. All the compounds showed good antibacterial activity against *P. aeruginosa* at all the tested concentrations.

Quantitative structure activity relationships (QSAR) studies

The antibacterial activity was measured in terms of zone of inhibition (ZOI). Various QSAR models were developed by the simple linear regression analysis. The highest value of the square of the correlation coefficient (r^2 = 0.55), and the satisfactory values of F and s (11 and 0.06, respectively), were obtained when the antibacterial activity (Log ZOI) of all the eleven coumarinylchalcones, against *E. coli* at 250 µg/mL, was correlated with the weakly polar component of the total solvent accessible surface area (WPSA). This correlation is represented by **equation 1**. This correlation helped to identify the parameter, which contributed the maximum to the antibacterial activity of coumarinylchalcones.

Log (ZOI) = 1.1433 - 0.00218 WPSA .....Equation 1

n = 11, r^2 = 0.55, F = 11, s = 0.06

The negative sign associated with the parameter, WPSA, indicated that the lower the value of WPSA, the higher would be the antibacterial activity of coumarinylchalcones. Thus, the polar functional groups like hydroxy, alkoxy, amino, etc., on the aromatic rings, are required for antibacterial activity of coumarinylchalcones.

For the validation of the best QSAR model, eleven coumarinylchalcones were placed in two different sets, a *training set* consisting of six compounds and a *test set* consisting of five compounds. The best QSAR model was obtained for the six compounds in the training set when their Log ZOI values against *E. coli* at 250µg/mL were correlated with WPSA. **Equation 2** shows the best QSAR model obtained by the simple linear regression analysis for the coumarinylchalcones in the training set. It is expressed as:

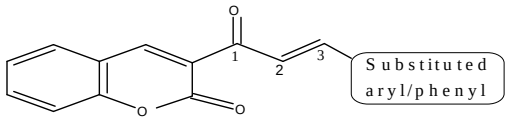
Log(ZOI) = 1.1418 - 0.003317 WPSA.....Equation 2

***n* = 6, r^2 = 0.849, *F* = 22.6, *s* = 1.001**

For the *internal validation* of the best QSAR model **(equation 2)**, the Log ZOI values of the compounds in the *training set* were predicted by substituting the values of the physicochemical descriptor, WPSA, for 6 coumarinylchalcones, in **equation 2**. This was done using the software *"Strike"*. The actual antibacterial activity of these compounds was already determined experimentally. The predicted antibacterial activity of the compounds in the training set was then correlated with their experimentally obtained antibacterial activity. The observed and the predicted values of Log ZOI for the compounds in the *training set*, against *E. coli* at 250 µg/mL, are summarized in **Table 6**. Good correlation between the observed and the predicted antibacterial activities was obtained as the square of the correlation coefficient (r^2) was 0.8867. This correlation is represented graphically in **Fig. 5**. Thus, **equation 2** provided good *internal* predictivity.

For the *external validation* of the best QSAR model **(equation 2)**, the compounds from the *test set* were used. The best QSAR model (**equation 2**) was used to predict the antibacterial activity of the *test set* compounds. The antibacterial activity of the *test set* compounds was also determined experimentally. The observed and the predicted antibacterial activities of the *test set* compounds are listed in **Table 6** and the correlation between them is represented graphically in **Fig. 6**. The higher values of the square of the correlation coefficient (r^2 = 0.8868) proved good external predictivity of the best QSAR model, represented by **equation 2**.

Table 1: Comparison of the conventional and microwave-assisted synthesis of coumarinylchalcones



Compd.	3-Aryl/phenyl substituent	Reaction time		% Yield	
		C (h)	M (min)	C	M
2a	-C_6H_5	5·5	20	50	60
2b	- C_6H_4-OMe (p)	4·5	47	58	64
2c	-C_6H_4-Cl (p)	20	145	56	60
2d	-C_6H_4-Cl (m)	5	40	60	65
2e	-C_6H_4-OH (p)	6	20	44	48
2f	-C_6H_4-OH (o)	6	50	42	48
2g	-C_4H_3O (o) or (o-Furyl)	5	40	70	78
2h	-C_6H_2-$(OMe)_3$ (2, 4, 6)	7	125	41	50
2i	-C_6H_3-$(OMe)_2$ (3, 5)	12·5	110	48	54
2j	-C_6H_4-$N(CH_3)_2$ (p)	7	50	41	45
2k	-C_6H_4-F (p)	6	32	42	49

C = Conventional heating, MW = Microwave irradiation at 560 W

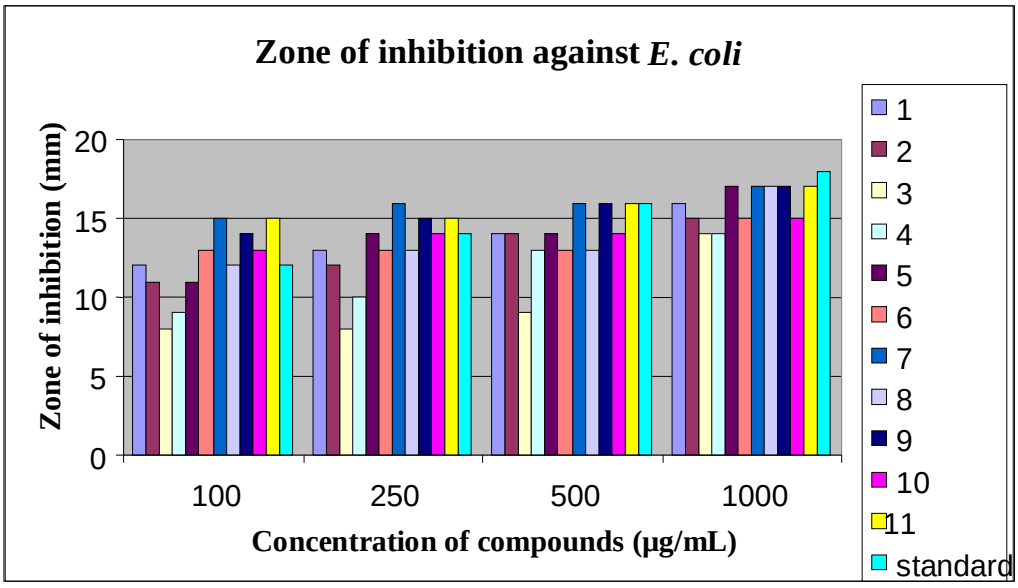


Fig. 1: Zone of inhibition of coumarinylchalcones and sulfamethoxazole at various concentrations against *E. coli*

Table 2: Antibacterial activity of coumarinylchalcones and sulfamethoxazole

Compd.	Zone of inhibition (ZOI)															
	E. coli				*S. aureus*				*P. aeruginosa*				*K. pneumoniae*			
	Conc. (ug/mL)				Conc. (ug/mL)				Conc. (ug/mL)				Conc. (ug/mL)			
	100	250	500	1000	100	250	500	1000	100	250	500	1000	100	250	500	1000
2a	**12**	13	14	16	**14**	**15**	15	**18**	**12**	**14**	**13**	**15**	**13**	**14**	**14**	**16**
2b	11	12	14	15	**16**	**16**	**17**	17	**10**	**12**	**15**	**21**	**10**	**12**	**15**	**16**
2c	8	8	9	14	10	11	11	13	**16**	**16**	**18**	**20**	**10**	10	**14**	**16**
2d	9	10	13	14	10	**14**	15	17	**12**	**12**	**13**	**15**	6	8	10	13
2e	11	**14**	14	17	10	**13**	15	17	**10**	**10**	**11**	**15**	9	11	**14**	**16**
2f	**13**	13	13	15	8	9	12	15	**11**	**12**	**12**	**13**	**11**	**12**	12	14
2g	**15**	**16**	16	17	10	11	13	15	**13**	**14**	**14**	**15**	**12**	**12**	13	14
2h	**12**	13	13	17	10	11	11	14	**10**	**12**	**15**	**16**	**15**	**14**	**16**	**17**
2i	**14**	**15**	**16**	17	**12**	**13**	13	14	**13**	**11**	**11**	**15**	**13**	**14**	**14**	**17**
2j	**13**	**14**	14	15	**13**	**14**	15	16	**6**	**15**	**15**	**16**	6	8	9	**16**
2k	**15**	**15**	**16**	17	7	**15**	15	17	**11**	**13**	**14**	**17**	8	10	11	14
Std*	12	14	16	18	11	13	16	18	4	10	10	12	10	12	14	16

*Std: Sulfamethoxazole

Table 3: Antifungal activity of coumarinylchalcones and miconazole

Compd.	Zone of inhibition (ZOI) in mm							
	C. albicans				*A. niger*			
	Conc. (µg/mL)				Conc. (µg/mL)			
	100	250	500	1000	100	250	500	1000
2a	9	12	15	18	6	8	10	11
2b	8	10	10	11	4	8	9	12
2c	6	8	8	14	6	8	9	10
2d	11	14	16	20	7	8	11	14
2e	8	11	14	16	6	9	10	13
2f	9	10	13	15	7	9	12	14
2g	10	12	14	16	6	8	10	13
2h	12	14	15	17	8	7	11	14
2i	12	13	15	16	5	8	10	12
2j	13	14	14	20	6	9	9	11
2k	15	16	16	20	6	9	10	15
Std*.	16	28	30	34	8	10	12	16

*Std: Miconazole

Table 4: Compounds showing antibacterial activity higher than that of sulfamethoxazole at various concentrations

Micro-organisms	Compounds and concentrations			
	100 µg/mL	250 µg/mL	500 µg/mL	1000 µg/mL
E. coli	2f, 2g, 2i, 2j, 2k	2g, 2i, 2k	-	-
S. aureus	2a, 2b, 2i, 2j	2a, 2b, 2d, 2j, 2k	2b	-
P. aeruginosa	All	All except 2e	All	All
K. pneumoniae	2a, 2f, 2g, 2h, 2i	2a, 2h, 2i	2b, 2h	2h, 2i

Table 5: Compounds showing antibacterial activity comparable to sulfamethoxazole, at various concentrations

Micro-organisms	Compounds and concentrations			
	100 µg/mL	250 µg/mL	500 µg/mL	1000 µg/mL
E. coli	2a, 2h	2e, 2j	2g, 2i, 2k	-
S. aureus	-	2e, 2i	-	2a
P. aeruginosa	-	2e	-	-
K. pneumoniae	2b, 2c	2b, 2f, 2g	2a, 2c, 2e, 2i	2a, 2b, 2c, 2e, 2j

Table 6: Observed and the predicted antibacterial activities (Log ZOI) of the training and test set compounds against *E. coli* at 250 µg/mL using the best QSAR model (Equation 2)

Compounds (Training set)	Log ZOI		Compounds (Test set)	Log ZOI	
	Observed	Predicted		Observed	Predicted
2b	1.0790	1.1168	**2a**	1.1130	1.1418
2c	0.9030	0.9369	**2e**	1.1400	1.1418
2d	1.0000	0.9609	**2g**	1.2000	0.9349
2f	1.113	1.1168	**2i**	1.1400	1.1418
2h	1.113	1.1168	**2k**	1.1700	0.9850
2j	1.1400	1.1168			

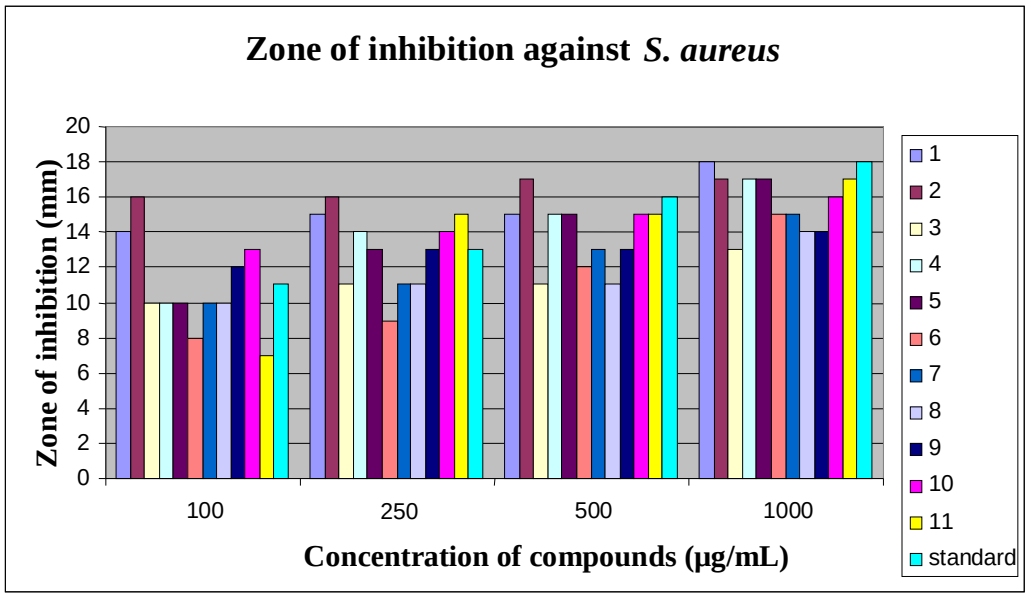


Fig. 2: Zone of inhibition of coumarinylchalcones and sulfamethoxazole at various concentrations against *S. aureus*

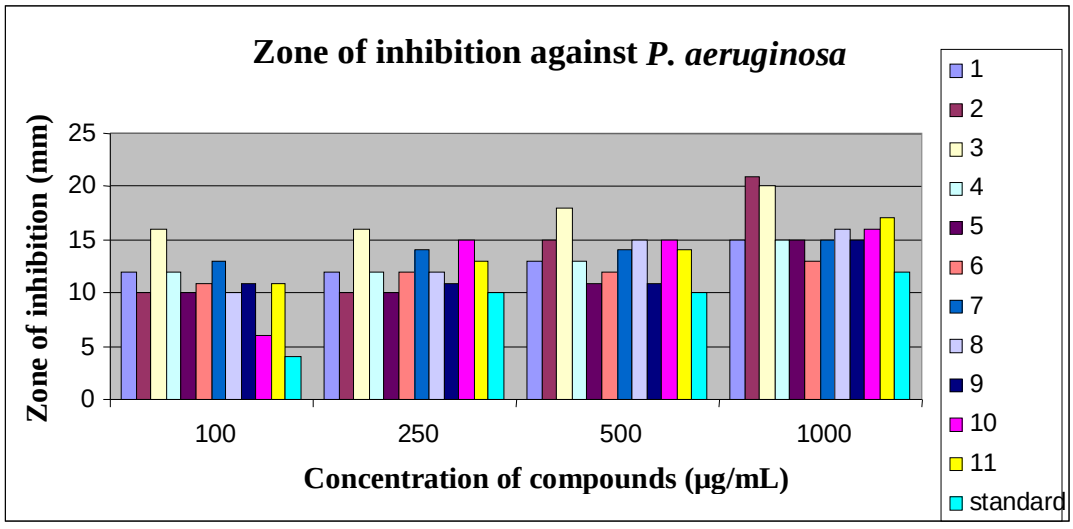


Fig. 3: Zone of inhibition of coumarinylchalcones and sulfamethoxazole at various concentrations against *P. aeruginosa*

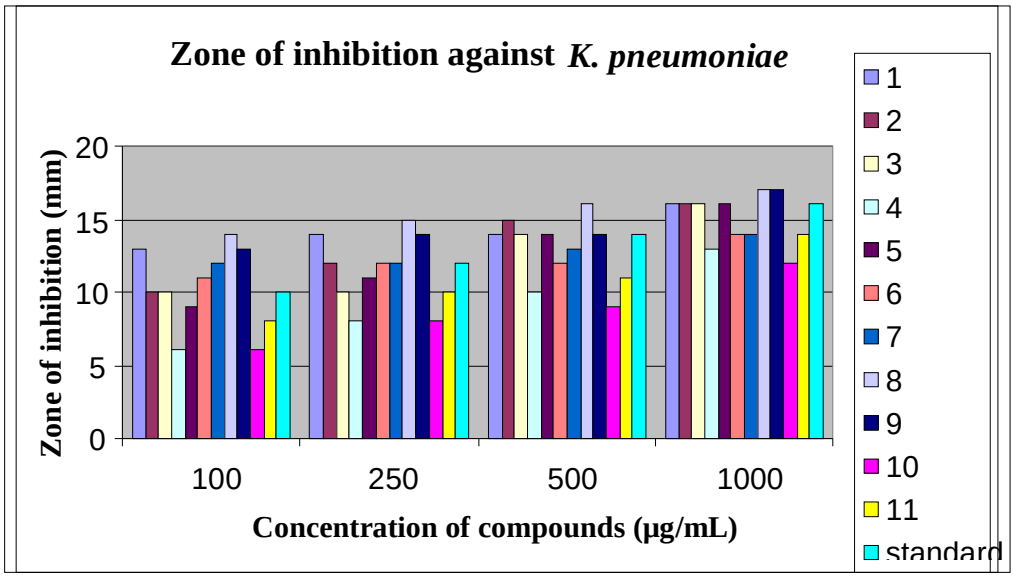


Fig. 4: Zone of inhibition of coumarinylchalcones and sulfamethoxazole at various concentrations against *K. pneumoniae*

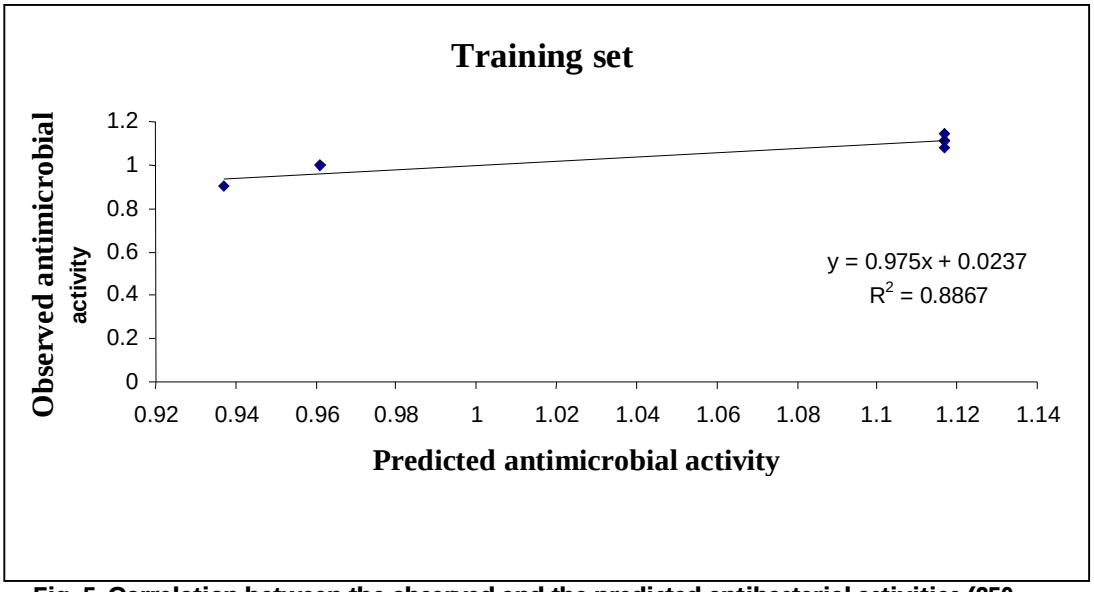


Fig. 5: Correlation between the observed and the predicted antibacterial activities (250 µg/mL) using the best QSAR model for the training set compounds against *E. coli*

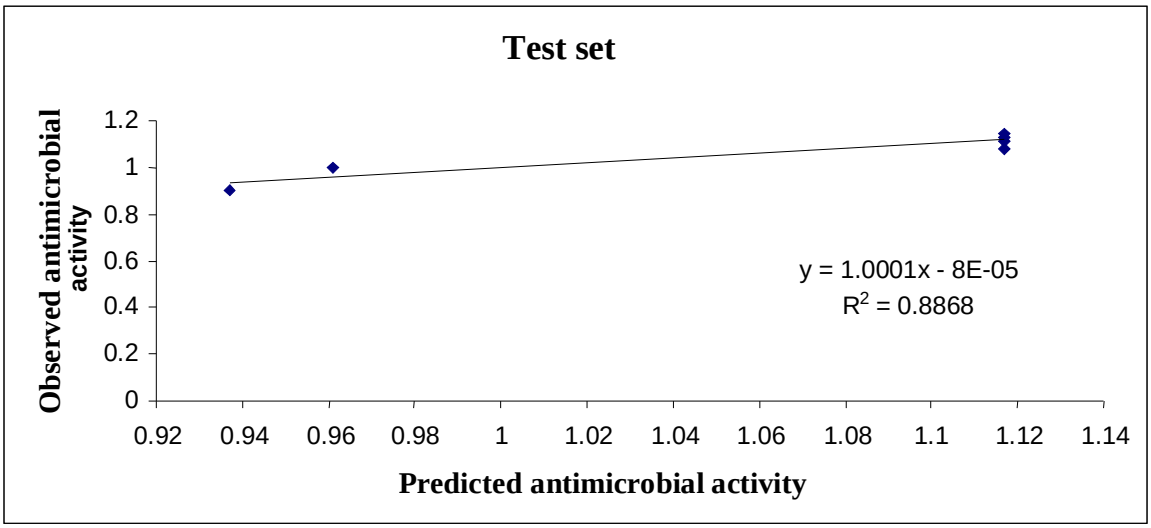
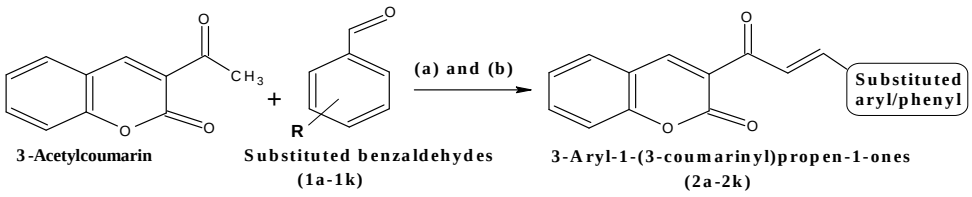


Fig. 6: Correlation between the observed and the predicted antibacterial activities (250 µg/mL) using the best QSAR model for the test set compounds against *E. coli*

Scheme 1: Synthesis of coumarinylchalcones 2a-2k



(a) = Piperidine/ethanol/glacial acetic acid; MW at 560 W
(b) = Piperidine/ethanol/glacial acetic acid; conventional reflux

CONCLUSIONS

An efficient and eco-friendly process for the synthesis of coumarinylchalcones was developed in the laboratory. The microwave-assisted reaction resulted in the higher yields and significant reduction in the reaction time. Many synthesized compounds depicted antimicrobial activity higher than that of sulfamethoxazole against gram positive as well as gram negative bacteria and many showed activity equivalent to sulfamethoxazole. All the compounds showed good antibacterial activity against *P. aeruginosa*. The parameter, WPSA, was significantly correlated to the antibacterial activity of coumarinylchalcones against *E. coli* at 250 µg/mL. A close correlation between the observed and the predicted antibacterial activities (Log ZOI values) for the compounds in the test and the training sets indicated the development of the best QSAR model. This model can be used for the design of newer antibacterial compounds from the series of coumarinylchalcones.

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