SYNTHESIS, CHARACTERIZATION AND ANTIMICROBIAL ACTIVITY OF SCHIFF BASE COMPOUNDS

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AbstractIn the present study five differentSchiff base compounds were synthesized and characterized by UV, IR, NMR and elemental analysis. The synthesized Schiff base compounds were tested for antimicrobial activity. The 4-Cl and 3-NO_2 substituted Schiff base compounds have shown good antimicrobial activity against three bacterial species, namely *B. subtilis, S. pyogenes*and *P. aeruginosa.* The 3-NO_2 substituted Schiff base compound has shown good antifungal activity against *Aspergillus flavus* and *Aspergillus niger.*

***Key words**:*Schiff bases, antimicrobial activity& antifungal activity.

1. Introduction

Schiff base derivatives have arouse considerable interest of chemists due to their versatile practical applications as well as their wide range of biochemical properties. Schiff base have been reported to possess a broad spectrum of biological activities namely antimicrobial, anticancer, anti-inflammatory, antifungal, antiproliferative, anticonvulsant, antioxidant, and antitubercular activities. Due to its wide range of biological activity Schiff base constitutes a relevant synthetic target in pharmaceutical industry.

Schiff bases are condensation products of primary amines with carbonyl compounds and they were first reported by Hugo Schiff (Schiff, 1864) in 1864.

The common structural feature of these compounds is the azomethine group with a general formula R-N=CH-R', where R and R' are substituted or unsubstituted alkyl, aryl, cycloalkyl or heterocyclic groups. These compounds are also known as imines or azomethines (Cimerman *et al.,* 1986; Keller, 1998; Patel et al., 1986; Layer, 1963).

Schiff bases containing aryl substituent's are substantially more stable than those with alkyl substituents. Schiff

bases of aliphatic aldehydes are relatively unstable and they are readily polymerizable. Schiff bases of aromatic aldehydes have effective conjugation and stability. The formation of a Schiff base from an aldehyde or ketone is a reversible reaction with an intermediate carbinolamine known as hemiaminol and generally takes place under acid or base catalysis, or upon heating (Cozzi, 2004).

Large amount of work has been done on this class of compounds due to its multi azetidinone (Bongini et al., 2000), thiazolidinone (Mulwad et al., 2002), formazone (Ainsworth et al., 1995), arylacetamide (Weber et al., 2005), metal complexes (Singh et al., 2007; Zhu et al., 2008; Yuan et al., 2009) and many other derivatives (Wang et al., 2008;Ceng et al., 2009).

Further, these Schiff bases are widely used as complexing agents (Cai et al., 2004) and perfumery reagents (Pilecki et al., 1984). Many reagents have been used as molecular sieves in ionic liquids (Chang et al., 2004), K-10 montmorillonite (Abid et al., 2007), MnO_2 (Blackburn et al., 2001), Tandamcatalysts (Barr et al., 1989), CaO (Gopalakrishnan et al., 2007), $MgSO_4$-PPTS(Chakraborti et al., 2004), $ZnCl_2$ (McBurney et al., 2012), P_2O_5-SiO_2 (Hasaninejad et al., 2008), phenyliodine(III)bis-(trifluoroacetate) (PIFA) (Varmaet al., 1999), etc., through microwave irradiation technique (Barretal., 1989).

Some Schiff bases have been represented as good corrosion inhibitors. Hegazy*et.al*., (2012) had examined the corrosion inhibition effect of 2-((pyridin-2-ylimino)methyl)phenol (S_1), 2-((hexadecylimino)methyl)phenol (S_2), 2-((4-hydroxyphenylimino) methyl)phenol and 1-(4-(2-hydroxybenzylideneamino) phenyl)ethanone for carbon steel. As a final point

Schiff base derivatives exhibitsinteresting antimicrobial& antifungal activity based on special substitution in diverse position. In this paper, we report synthesis, characterization, antimicrobial & antifungal activities of five unique Schiff bases.

Experimental

2.1. Materials and methods

All the chemicals and solvents were purchased from Sigma Aldrich Chemical Company, Bengaluru-100. The melting points were taken in open capillaries in electrical apparatus and are uncorrected.

2.2. Instrumentation

The UV spectra of all the(*E*)-N-benzylidene-4-methylbenzo[d]thiazol -2-amine under investigation were recorded on SHIMADZU-1650

spectrophotometer ($\lambda_{max,}$ nm) in spectral grade methanol, The IR spectra were recorded on SHIMADZU-FT-IR. The ^{1}H and ^{13}C NMR Spectra of all aryl imines under investigation were recorded using BRUKER, 400MHz,Elemental analyses of all compounds were performed in Thermofinnigan analyser at CAS in Marine biology, Annamalai University.

2.3. Synthesis of (*E*)-N-benzylidene-4-methylbenzo[d]thiazol-2-amines

Equimolar quantities of benzaldehyde (0.01 mol) and 4-methylbenzo[d]thiazol-2-amine (0.01 mol) were refluxed for 4hrs with 20 ml of absolute ethanol (Abd-Elzaher et al., 2016) and it is shown in **Scheme-1**. After the completion of the reaction, as monitored by TLC, the mixture was cooled at room temperature. The resulting precipitate was filtered and washed with cold water. The product appeared as pale yellow solid. Then it was recrystallized using ethanol to obtain pale yellow glittering solid melting at 161-162^{o}C

The same procedure has been followed to synthesized the remaining four more substituted heterocyclic Schiff base compounds namely, (*E*)-N-(4-chlorobenzylidene)-4-methylbenzo[d]thiazol-2-amine, (*E*)-4-methyl-N-(4-methylbenzylidene)benzo[d]thiazol-2-amine, (*E*)-N-(4-methoxybenzylidene)-4-methylbenzo[d]thiazol-2-amine and (*E*)-4-methyl-N-(3-nitrobenzylidene)benzo[d]thiazol-2-amine.

2.4 Antimicrobial activity

The synthesized Schiff base compounds during the present investigation were screened for their antibacterial activity on five common microorganisms such as, *Bacillus subtilis, Staphylococcus aureus, Streptococcus pyogenes, Escherichia coli and Pseudomonas aeruginosa*.Using Kirby-Bauer (Bauer et *al.,* 1966) disc diffusion technique (Collins et *al.*, 1989) antibacterial sensitivity assay was performed.

3. Results and discussion

In the present research work, Substituted (*E*)-N-benzylidene-4-methylbenzo[d]thiazol-2-amine from condensation of different aromatic aldehydes with4-methylbenzo[d]thiazol-2-amine were synthesized in presence of CH_3COOHcatalyst. The general reaction of substituted (*E*)-N-benzylidene-4-methylbenzo[d]thiazol-2-amine is as given in Scheme 1.The synthesized(*E*)-N-benzylidene-4-methylbenzo[d]thiazol-2-aminehave been characterized by UV, IR & NMR spectral data given in**Table (1-5).**

Table 1: Analytical and spectral data of (*E*)-N-benzylidene-4-methylbenzo[d]thiazol-2-amine(1)

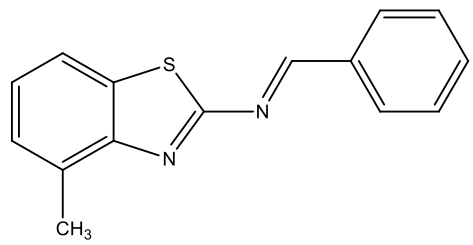
Molecular Formula $C_{15}H_{12}N_2S$	**Molecular Weight** 252.33	**Melting Point (°C)** 161-162	**% of yield** 90	**Elemental analysis (%):** C,71.40, H,4.79,N, 11.10,S,12.71
UV:345 nm, 222 nm, **IR(KBr, cm^{-1}):**3024.38 (Ar-CH), 2922.16 (Aliphatic-CH),1562.34 ($C=N_{thiazole}$), 1602.85 (CH=N), 754.17 (C-S-C)				
^{1}H NMR (400 MHz, $CDCl_3$, δ, ppm):7.068-8.185(m, 8H, Ar-H), 2.600(S, 3H, -CH_3), 8.111(S, 1H, -N=CH-)				
^{13}C NMR (100 MHz, $CDCl_3$, δ, ppm): 18.11(CH_3),169.44($C=N_{thiazole}$), 146.10 (C=N), 145.66-118.96 (aromaticcarbons)				

Table 2: Analytical and spectral data of (*E*)-N-(4-chlorobenzylidene)-4-methylbenzo[d]thiazol-2-amine (2)

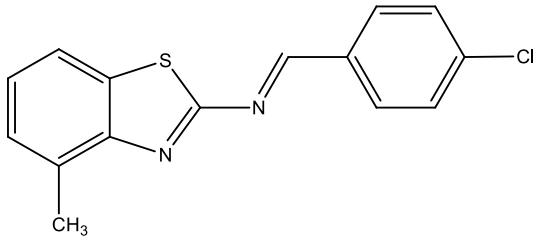
Molecular Formulae $C_{15}H_{11}N_2SCl$	**Molecular Weight** 286.78	**Melting Point (°C)** 130-131	**% of yield** 82	**Elemental analysis (%)** C, 62.82, H, 3.87, N,9.77, S, 11.18, C, l12.36
UV:322.5, 271 **IR(KBr, cm^{-1}):** 3064.89 (Ar-CH), 2922.16 (Aliphatic-CH),1550.77 ($C=N_{thiazole}$), 1612.49 (CH=N), 744.52 (C-S-C)				
^{1}H NMR (400 MHz, $CDCl_3$, δ, ppm): 7.056-7.645 (m, 8H, Ar-H), 2.586(S, 3H, -CH_3), 7.878(S, 1H, -N=CH-)				
^{13}C NMR (100 MHz, $CDCl_3$, δ, ppm): 18.03 (CH_3), 167.43 (C=Nthiazole), 150.55(C=N), 142.78-118.66 (aromaticcarbons)				

Table 3: Analytical and spectral data of (*E*)-4-methyl-N-(4-methylbenzylidene)benzo[d]thiazol-2-amine (3)

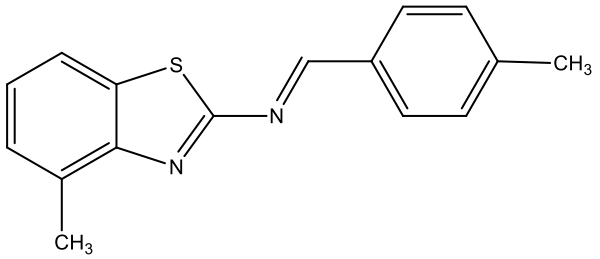
Molecular Formulae $C_{16}H_{14}N_2S$	**Molecular Weight** 266.36	**Melting Point (°C)** 107-108	**% of yield** 86	**Elemental analysis (%)** C, 72.15, H, 5.30, N,10.52, S, 12.04
UV:340.5,225 **IR(KBr, cm^{-1}):** 3001.24 (Ar-CH), 2922.16 (Aliphatic-CH),1566.20 (C=N $_{thiazole}$), 1606.70 (CH=N), 738.74 (C-S-C)				
^{1}H NMR (400 MHz, $CDCl_3$, δ, ppm): 7.050-7.595 (m, 8H, Ar-H), 2.388(S, 3H, -CH_3), 2.550(S, 3H, -CH_3) 7.868(S, 1H, -N=CH-)				
^{13}C NMR (100 MHz, $CDCl_3$, δ, ppm): 18.06 (CH_3), 21.48 (CH_3), 167.43 (C=Nthiazole), 148.67(C=N), 144.00-118.83 (aromaticcarbons)				

Table 4: Analytical and spectral data of (*E*)-N-(4-methoxybenzylidene)-4-methylbenzo[d]thiazol-2-amine (4)

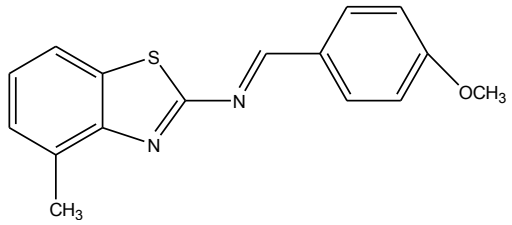
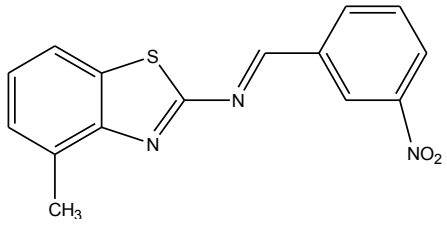
Molecular Formulae $C_{16}H_{14}N_2OS$	**Molecular Weight** 282.36	**Melting Point (°C)**	**% of yield** 78	**Elemental analysis (%)** C, 68.06, H, 5.00, N,9.92, O, 5.67, S, 12.04
UV:337.5,227 **IR(KBr, cm^{-1}):** 3061.03 (Ar-CH), 2924.09 (Aliphatic-CH),1508.33 (C=$N_{thiazole}$), 1600.92 (CH=N), 740.67 (C-S-C)				
^{1}H NMR (400 MHz, $CDCl_3$, δ, ppm): 6.915-7.626 (m, 8H, Ar-H), 2.538(S, 3H, -CH_3), 3.847(S, 3H, -OCH_3)7.833(S, 1H, -N=CH-)				
^{13}C NMR (100 MHz, $CDCl_3$, δ, ppm): 18.13(-CH_3), 55.38(-OCH_3), 168.14(C=$N_{thiazole}$), 161.04(C=N), 148.62-114.24 (aromaticcarbons)				

Table 5: Analytical and spectral data of(*E*)-4-methyl-N-(3-nitrobenzylidene)benzo[d]thiazol-2-amine.(5)

Molecular Formulae	**Molecular Weight**	**Melting Point (°C)**	**% of yield**	**Elemental analysis (%)**
$C_{15}H_{11}N_3O_2S$	297.33	189-190	80	C, 60.59, H, 3.73, N,14.13,O, 10.76, S, 10.78
UV: 339.5, 225.5 **IR(KBr, cm^{-1}):** 3088.03 (Ar-CH), 2976.16 (Aliphatic-CH),1533.41 (C=$N_{thiazole}$), 1598.99 (CH=N), 723.31 (C-S-C)				
^{1}H NMR (400 MHz, $CDCl_3$, δ, ppm): 7.066-8.217 (m, 8H, Ar-H), 2.524 (S, 3H, -CH_3), 8.477(S, 1H, -N=CH-)				
^{13}C NMR (100 MHz, $CDCl_3$, δ, ppm): 17.86(CH_3), 167.41(C=$N_{thiazole}$), 148.71(C=N), 141.94-119.06(aromatic carbons)				

3.1 Evaluation of antibacterial activity

The synthesized Schiff base derivatives were screened for the antibacterial activity against three Gram-positive bacteria viz*., Bacillus subtilis, Staphylococcus aureus* and *Streptococcus pyogenes* and two Gram-negative bacteria viz., *Escherichia coli* and *Pseudomonas aeruginosa*at various concentrations was given in **Table-6**by using the disk diffusion method. Ciprofloxacin was used as reference standard for comparing the results. The corresponding cclustered column chart is given in **Fig-6**.

The substituents place a vital role in imparting enhanced antibacterial activity to the compounds. The 4-Cl and 3-NO_2 substituted Schiff base compounds have shown good activity against *B. subtilis*. The remaining three parent (H), 4-CH_3 and 4-OCH_3 substituted Schiff base compounds have shown moderate antibacterial activity against *B. subtilis*. All the five Schiff base compounds have shown moderate antibacterial activity against *S. aureus.* The two **1** and **5** Schiff

base compounds with 4-Cl and 3-NO_2 substituents have shown good activity against *S. pyogenes.* The remaining three parent (H), 4-CH_3 and 4-OCH_3 substituted Schiff base compounds have shown moderate antibacterial activity against *S. pyogenes*. All the five Schiff base compounds have shown moderate antibacterial activity against *E. coli.* The 3-NO_2 substituted Schiff base compound has shown good activity against *P.aeruginosa*. The remaining four Schiff base compounds have shown moderate activity against *P. aeruginosa.*

Table- 6 Antibacterial activity of Schiff base derivatives

DISC DIFFUSION METHOD		**ANTIBACTERIAL ACTIVITY**				
		ZONE OF INHIBITION 10µg/ml				
		Gram +*ve* Bacteria			**Gram –*ve* Bacteria**	
S. No	**Comp. No**	***B. subtilis***	***S. aureus***	***S. Pyrogenes***	***E. coli***	***P.aeruginosa***
1	**1**	12	19	14	10	13
2	**2**	23	21	22	`18	21
3	**3**	17	12	17	16	12
4	**4**	21	19	20	16	15
5	**5**	28	20	25	21	27
Ciprofloxacin		31	30	29	31	32
Control		0	0	0	0	0

3.2 Antifungal activity of substituted Schiff base compounds

The minimum inhibitory concentration of antifungal activity of all the Schiff base compounds have been measured at various concentrations is given in **Table-7**. It is evident that most of the compounds have shown significant antifungal activity at the concentration of 10 mg/mL in general. The antifungal activity of all the five Schiff base compounds have been studied three fungal species namely *Aspergillus flavus, Aspergillus niger and Trigoderma veride* by using disk diffusion method.The results of this evaluation were compared with Amphotericin–B as reference standard. The zone of inhibition values are given in **Table-7.** The corresponding clustered

column chart is given in **Fig-7**.The 4-OCH_3 and 3-NO_2 substituted Schiff base compounds have shown good antifungal activity against *Aspergillus flavus.* The remaining three Schiff base compounds with parent (H), 4-Cl and 4-CH_3 substituents have shown moderate antifungal activity against *Aspergillus flavus.* The only one 3-NO_2 substituted Schiff base compound has shown good antifungal activity against *Aspergillus niger.* The parent (H), 4-Cl, 4-CH_3 and 4-OCH_3 substituted Schiff base compounds have shown moderate antifungal activity against *Aspergillus niger.* The only one 4-CH_3 substituted Schiff base compound has shown good antifungal activity against *Trigoderma veride.* The remaining four parent (H), 4-Cl and 4-OCH_3 and 3-NO_2 substituted Schiff base compounds have shown moderate antifungal activity against *Trichoderma veride.*

Table-7 Antifungal activity of Schiff base derivatives

DISC DILUTION METHOD		**ANTIFUNGAL ACTIVITY**		
		ZONE OF INHIBITION 10µg/ml		
S. No.	**Comp. No.**	***A. flavus***	***A. niger***	***T. veride***
1	**1**	14	14	13
2	**2**	14	13	11
3	**3**	15	16	15
4	**4**	16	15	14
5	**5**	19	18	14
Amphotericin-B		26	22	20
Control		**0**	**0**	**0**

4. Conclusion

In the present study five differentSchiff base compounds were synthesized and determined with different spectroscopy data like UV, IR, NMR and elemental analysis. The synthesized Schiff base compounds tested for antimicrobial

activity. The 4-Cl and 3-NO_2 substituted Schiff base compounds have shown good antimicrobial activity against three bacterial species, namely *B. subtilis, S. pyogenes* and *P. aeruginosa.* The 3-NO_2substituted Schiff base compound has shown good antifungal activity against *Aspergillus flavus* and *Aspergillus niger.*

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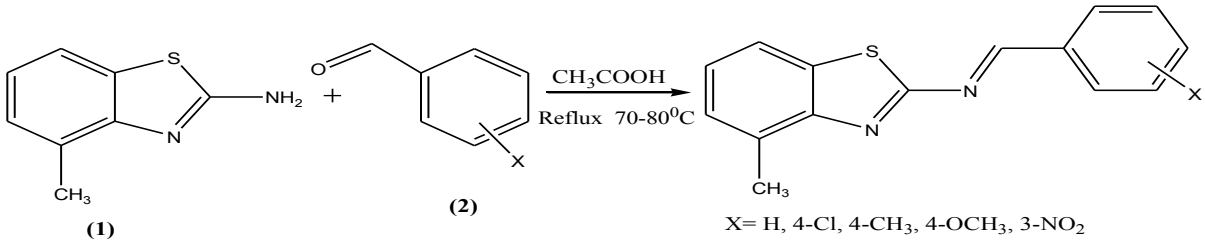
Figure and Figure Captions



Scheme1. Synthesis of (*E*)-N-benzylidene-4-methylbenzo[d]thiazol-2-amine compounds

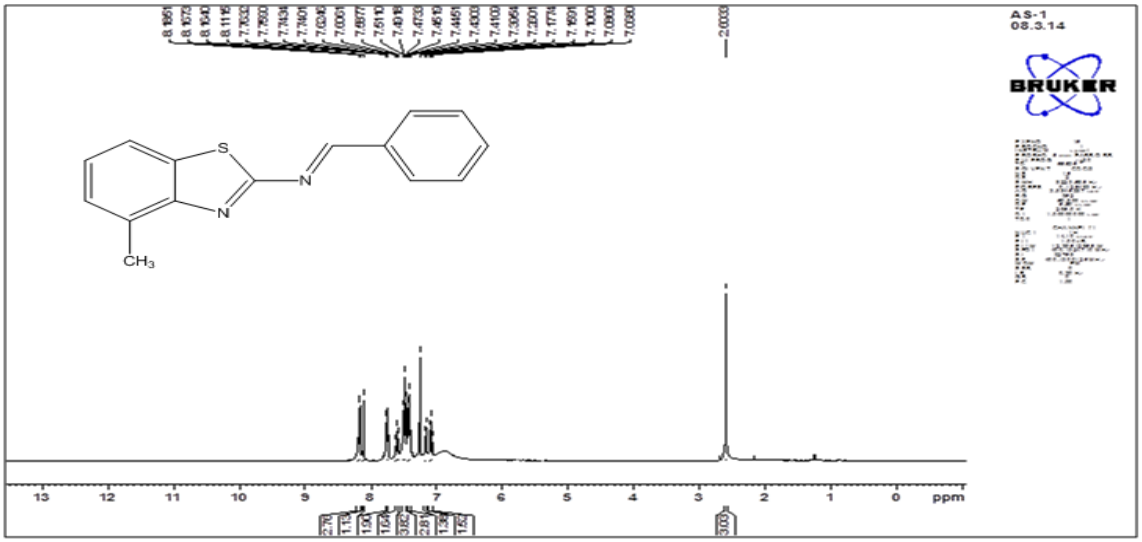


Fig-1:^{1}H NMR Spectrum of (*E*)-N-benzylidene-4-methylbenzo[d]thiazol-2-amine

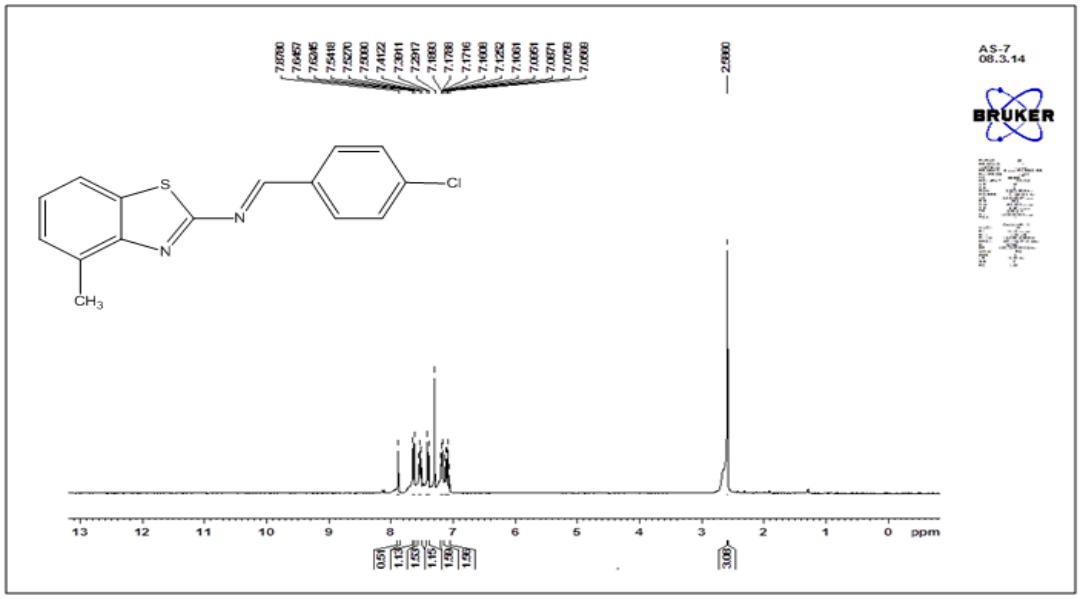


Fig-2: ^{1}H NMR Spectrum of (*E*)-N-(4-chlorobenzylidene)-4-methylbenzo[d]thiazol-2-amine

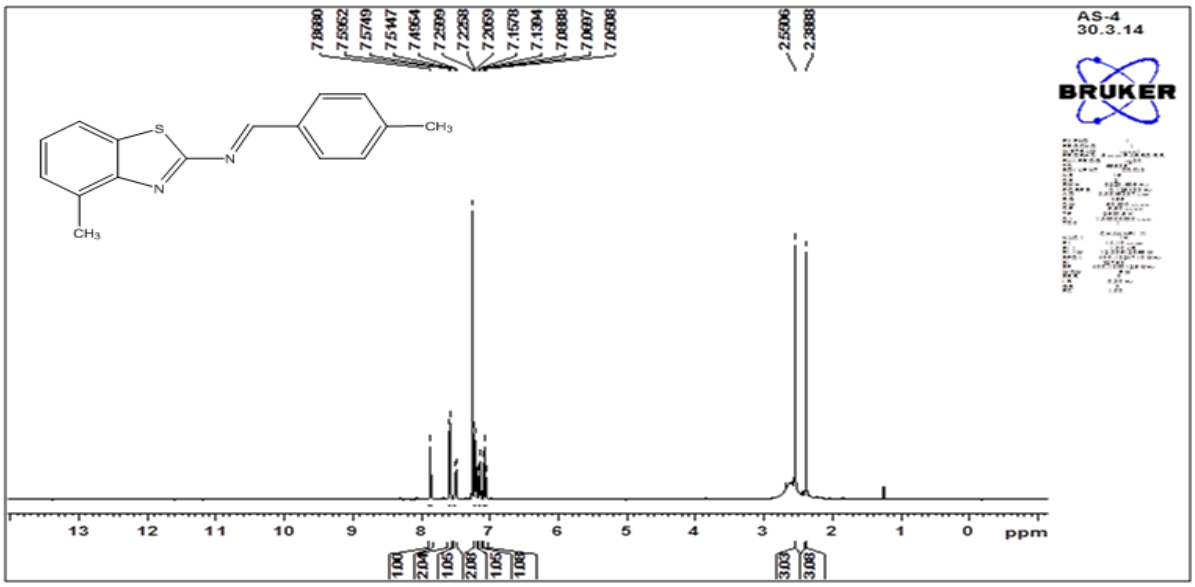


Fig-3: ^{1}H NMR Spectrum of (*E*)-4-methyl-N-(4-methylbenzylidene)benzo[d]thiazol-2-amine

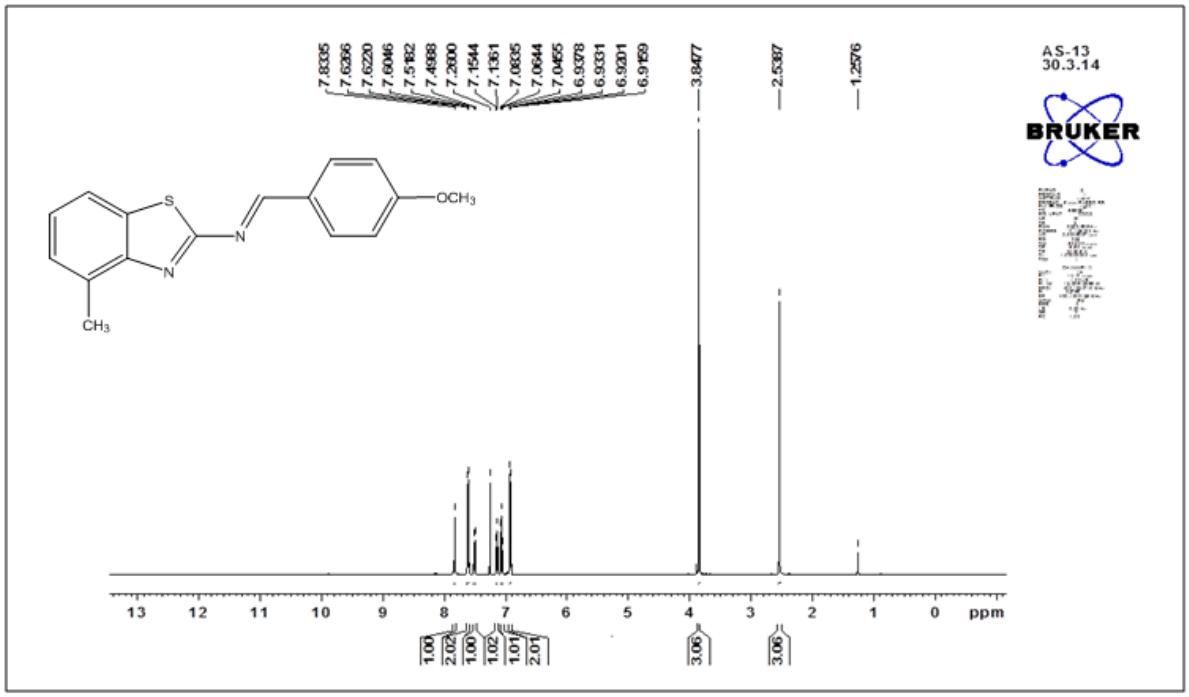


Fig-4: ^{1}H NMR Spectrum of (*E*)-N-(4-methoxybenzylidene)-4-methylbenzo[d]thiazol-2-amine

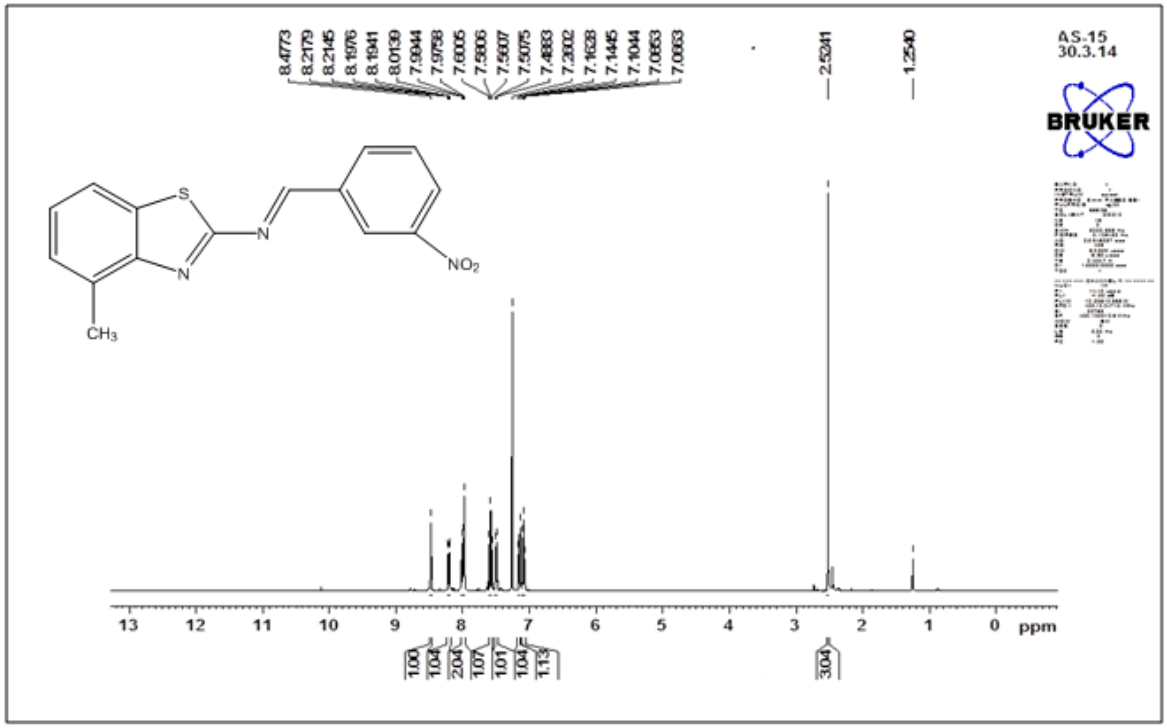


Fig.-5: ^{1}H NMR Spectrum of (*E*)-4-methyl-N-(3-nitrobenzylidene)benzo[d]thiazol-2-amine

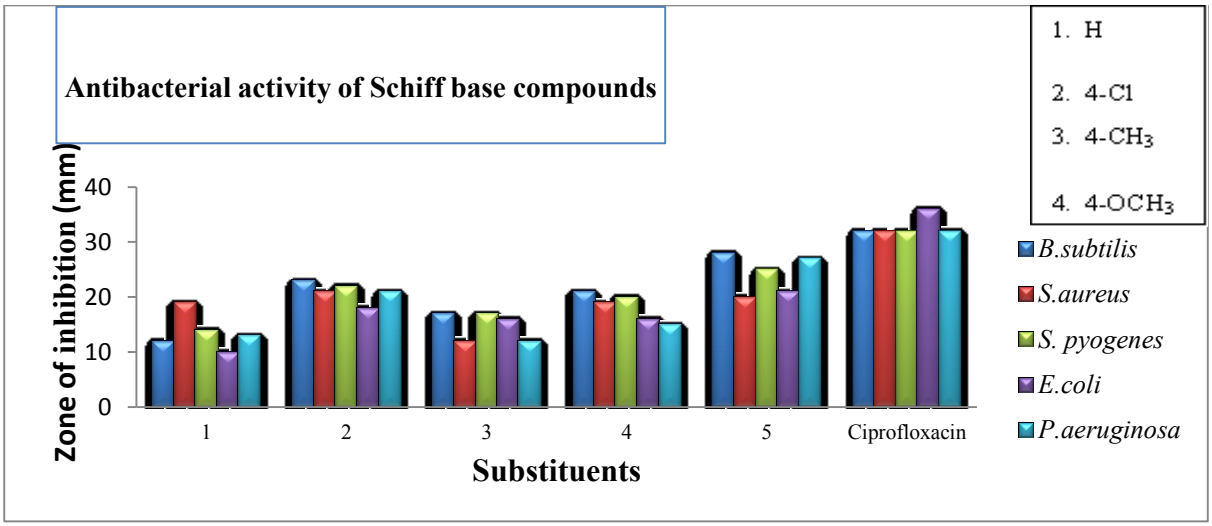


Fig-6: Antibacterial activity of substituted (*E*)-2-benzylidenehydrazinecarboximidamide compounds

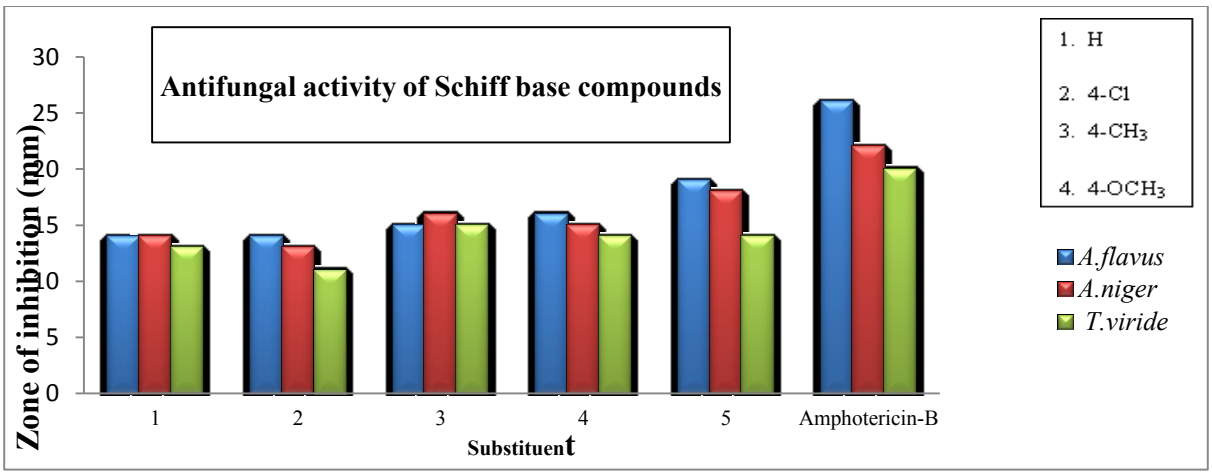


Fig-7: Antifungal activity of substituted (*E*)-2-benzylidenehydrazinecarboximidamide compounds

References

Abd-Elzaher M. M, Hegazy W., Gaafar A.,(2005),Synthesis, characterization and biological studies of ferrocenyl complexes containing thiophene moiety, *Appl. Organometal. Chem.***,** 19, 911 - 916.

Abd-Elzaher M. M.,(2004), Synthesis, characterization, and antimicrobial activity of cobalt (II), nickel (II), copper (II) and zinc (II) complexes with ferrocenyl Schiff bases containing a phenol moiety, *Appl. Organometal. Chem.*, 18, 149 - 155.

Abd-Elzaher M. M., (2004),Synthesis and spectroscopic characterization of some ferrocenyl Schiff bases containing pyridine moiety and their complexation with cobalt, nickel, copper and zinc, *J. Chin. Chem. Soc*, 51, 499 - 504.

Abd-Elzaher M. M., Labib A. A., Mousa H. A., Moustafa S. A., Ali M. M., El-Rashedy A. A., (2016), Synthesis, anticancer activity and molecular docking study of Schiff base complexes containing thiazole moiety,*Beni–Suef UniversityJ. Basic. App. Sci.*, 5, 85 - 96.

Abid M., Savolainen M., Landge S., Hu J., SuryaprakashG.K., Olah G. A., B. Torok, (2007), Synthesis of trifluoromethyl-imines by solid acid/superacid catalyzed microwave assisted approach,,*J. Fluo. Chem*., 128, 587 - 594.

Alang G., Kaur R., Kaur G., Singh A., Singla P., (2011), *Acta Pharm. Sci.,* 52, 213.

Barr D.A., Donegan G., Grigg R.,(1989), Tandem Michael addition-1,3-dipolar cycloaddition of imines of α-amino acid estersandaminoacetonitrile, *J. Chem. Soc., PerkinTrans,* 1, 1550 - 1551.

Bauer A. W., Kirby W. M. M., Sherris J. C., Truck M., (1966), Antibiotic susceptibility testing by a standardized single disk method, *Am. J. Clin. Pathol.,* 45, 493.

Blackburn L., Taylor R. J. K., (2001),In Situ Oxidation − Imine Formation − Reduction Routes from Alcohols to Amines*, Org. Lett*.,3, 1637 - 1639.

Bongini A., Panunzio M., Piersanti G., Bandini E., Martelli G., Spunta G., Venturini A., (2000), Stereochemical Aspects of a Two-Step Staudinger Reaction− Asymmetric Synthesis of Chiral Azetidine-2-ones, *Euro. J. Org. Chem.,*13, 2379.

Cai T., Peng Z., Deng Q., Long Y., Deng T., Yu K.,(2004), *Wuji, HuaxueXuebao.*, 20, 731.

Chakraborti A.K., Bhagat S., Rudrawar S.,(2004),Magnesium perchlorate as an efficient catalyst for the synthesis of imines and phenylhydrazones, *Tetrahedron Lett.,*45, 7641.

Chang W., Ahn B. J.,(2004), Microwave-Assisted Mesoporous Molecular Sieve-Catalyzed Knoevenagel Condensation and Imine Formation Reactions under Solvent-free Conditions, *J. Ind. Eng. Chem*., 10, 690.

Cimerman Z., Miljanic S., C*Croatica* N. G., (2000), Ultra Sensitivity Bio-Sensor based on Photonic Crystal Ring Resonator, *ChemicaActa.,*73(1), 81.

Cozzi P. G., (2004), Metal–Salen Schiff base complexes in catalysis: practical aspects, *Chem. Soc. Rev.,* 33, 410.

Gopalakrishnan M., Sureshkumar P., Kanagarajan V., (2007), *Thanusu J*., *Res. Chem. Int. Med.,*33, 541

Hasaninejad A., Zare A., Sharghi H., Shekouhy M., (2008), *Arkivoc*., 11, 64.

McBurney R. T., Fernando P. C., Walton J. C., (2012), A helicopter view of microwave application to chemical processes: reactions, separations, and equipment concepts, *RSC Adv.,* 12, 1264.

Pilecki M., Marczewski Z., Gora J., (1984), Schiff bases, their application in perfumery*, Kosmetyki,* 28, 223.

Schiff H., (1864), *Ann. Chem.,*131, 118.

Weber G., Messerschmidt J., (2005),Signal enhancement in adsorptive stripping voltammetry of Pt by forced convection during the measurement step, *Ana. Chimi. Acta.,* 545, 166.

Varma R. S., Kumar D., (1999), Solid state oxidation of 1, 4-dihydropyridines to pyridines using phenyliodine (III) bis (trifluoroacetate) or elemental sulfur, *J. Chem. Soc., Perkin Trans*., 1, 1755.