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RESEARCH ARTICLE

Synthesis¸ Characterization and Biological Activity of Schiff Bases derived from 3-(4-Substituted)-1-phenyl-1*H*-pyrazole-4-carbaldehyde and *o*-Aminophenol

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Abstract: A series of new Schiff bases have been synthesized by reaction of 3-(4-substituted)-1-phenyl-1*H*-pyrazole-4-carbaldehyde and *o*-aminophenol in ethanol. The newly prepared Schiff bases were characterized by elemental analysis and spectroscopic (IR, ^{1}H NMR, ^{13}C NMR and Mass) data. They have been screened for their *in vitro* biological activities against bacteria and fungi.

Keywords: Schiff base, Pyrazole aldehyde, Antimicrobial activity

Introduction

Schiff bases are condensation products of primary amines with carbonyl compounds. They were first reported by Schiff[1] in 1864. The common structural feature of these compounds is azomethine group with a general formula RHC=N-R1, where R and R1 are alkyl, aryl, cyclo alkyl or heterocyclic groups which may be variously substituted. These compounds are also known as imines or azomethine. Several studies[2-5] showed that the presence of a lone pair of electrons in a sp^2 hybridized orbital of nitrogen atom of the azomethine group is of considerable chemical and biological importance. Due to straight forward preparation, synthetic flexibility and the special property of C=N group, Schiff bases are generally excellent chelating agents[6-12], especially when a functional group like –OH is present close to the azomethine group. Versatility of Schiff base ligands and biological, analytical and industrial applications of their complexes make further investigations in this area highly desirable. Nowadays, the research field dealing with Schiff base coordination chemistry has expanded enormously. The importance of Schiff base complexes for bioinorganic chemistry, biomedical applications, supramolecular chemistry, catalysis and material science, separation and encapsulation processes and formation of compounds with unusual properties and structures has been well recognized and reviewed[13-14]. Schiff bases resulted from aromatic aldehydes *ortho-substituted* with a hydroxyl group have initially arouse the researchers interest because of their ability to act as bidentate ligands for transitional metal ions[15-19]. Schiff bases have been reported in their biological properties, such as, antibacterial,

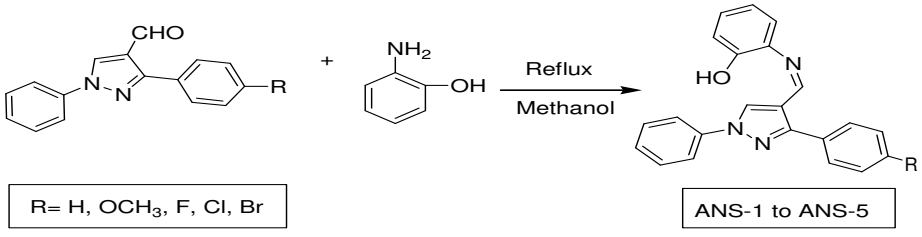
antifungal activities[20-21]. Schiff bases are active against a wide range of organisms for example; *Candida Albicans, Escherichia coli Staphylococcus aureus, Bacillus polymxa, Trychophyton gypseum, Mycobacteria, Erysiphe graminis* and *Plasmopora.* They serve as models for biologically important species.

Experimental

All chemical reagents and solvents used were of laboratory grade or higher and used directly. Pyrazole aldehyde was synthesized by very well-known Vilsmeier-haack[22] reaction and purified by usual separation methods. Elemental analyses were carried out on EURO EA-3000 RS-232. IR spectra were recorded on 8400 FTIR Simadzu spectrometer. ^{1}H NMR spectra were recorded on a Bruker Advance II 400 spectrometer at room temperature using TMS as internal standard. UV-Visible spectra were recorded on Shimadzu, Pharmaspec UV-1700, UV visible spectrometer. Mass spectra were recorded on GC-MS QP-2010 spectrometer.

Preparation of 2-((3-(4-substitutedphenyl)-1-phenyl-1H-pyrazol-4yl)methylene amino) phenol

The series of new Schiff base were synthesized by using a synthetic approach (Scheme 1) in which solution of *o*-aminophenol (0.01 M) added drop wise in to the solution of pyrazol aldehyde (0.01 M) in 30 mL methanol. The resulting reaction mixture was reflux for 8 h. The progress of reaction was monitored by TLC (solvent system, benzene: acetone- 8:2). After completion of the reaction, the reaction mixture was poured into crushed ice. The separated solid product was filtered and washed with cold saturated sodium bisulphate solution and recrystallized with ethanol.



Scheme 1

Table 1. Physical characterization of Schiff base ligands

Comp. code	R	Yield%	M. P. ºC	R_f
ANS-1	-H	86	146	0.31
ANS-2	-OCH_3	82	148	0.34
ANS-3	-F	87	144	0.39
ANS-4	-Br	89	156	0.41
ANS-5	-Cl	88	148	0.42

*2-((1,3-Diphenyl-1H-pyrazol-4-yl)methylene amino)phenol (**ANS-1**)*

Colour: Off white; Anal. Calcd. For $C_{22}H_{17}N_3O$ (339.14 g/mol): C, 77.86%; H, 5.05%; N, 12.38%; O, 4.71%;. Found: C, 77.76%; H, 5.15%; N, 12.32%; O, 4.68%;. MS (*m/z*): 339 (M); IR (KBr, cm^{-1}): *ν*(OH) 3425; *ν*(-HC=N) 1614; *ν*(-C=C), *ν*(-C=N), *ν*(-N-N) of pyrazol at 1456, 1492, 1109 respectively, *ν*(Ar–C-H) 3142; *ν*(Ar–C=C) 1498; ^{1}H NMR (DMSO-d_6): δppm): δppm 9.28, (s, 1H, OH); 8.71, (s, 1H, -HC=N); 7.96, (s, 1H, Ar-H); 7.94, (s,1H, Ar-H);

7.8,(s, 1H, Ar-H), 7.78,(s, Ar-H); 7.44-7.54,(m, 4H, Ar-H); 7.43 (d, 1H, Ar-H); 7.33-7.37, (t, 1H, Ar-H); 7.17-7.19, (d, 1H, Ar-H); 7.0-7.03,(t, 1H, Ar-H); 6.89-6.92, (d,1H, Ar-H); 6.79-6.82, (d, 1H, Ar-H); ^{13}C NMR (DMSO-d_6): δppm 184, (-HC=N); 152 (phenolic, C); 114.38, 115.41, 116.85, 117.74, 118.60, 119.39, 120, 122.10, 126.82, 127.38, 128.56, 129.34, 131, 132.26, 132, 134.22, 137.07, 139, 144.10, 150, 151.33 (Ph); UV-Vis:(DMF) (λ_{max}/ nm): 294, 280.

*2-((3-(4-Methoxyphenyl) -1-phenyl-1H-pyrazol-4-yl)) methylene aminophenol (**ANS-2**)*

Colour: Brown; Anal. Calcd. For $C_{23}H_{19}N_3O_2$ (369.15 g/mol): C, 74.78%; H, 5.18%; N, 11.37%; O, 8.66%; Found: C, 74.14%; H, 5.29%; N, 11.30%; O,8.44%; MS (*m/z*):369.15 (M); IR (KBr, cm^{-1}): *v*(-OH) 3379; *v*(-HC=N) 1616; *v*(N-N) 1026; *v*(Ar–C-H) 2956-3138; *v*(Ar –C=C) 1541; ^{1}H NMR (DMSO-d_6): δppm 9.21, (s, 1H, OH); 8.68, (s, 1H, -HC=N); 8.5, (s, 1H, Ar-H); 7.90-7.92, (d, 2H, Ar-H); 7.69-7.71(d, 2H, Ar-H); 7.47-7.51,(t, 2H, Ar-H); 7.30-7.36 (m, 1H, Ar-H); 7.15-7.17, (d, 1H, Ar-H); 6.97-7.06, (m, 3H, Ar-H); 6.8,(s,1H, Ar-H); 6.67-6.81(t,1H, Ar-H); ^{13}C NMR (DMSO-d_6): δppm 184,(-HC=N); 55.02,(O-CH_3); 113.7, 114.58, 115, 117.87, 118.99, 119.43, 120.06, 123.06, 124.49, 126.71, 127.06, 129.28, 134.44, 129.35, 129.67, 137.29, 139.05, 144.07, 151.30, 152.37, 159.62, (Ph); UV-Vis:(DMF) (λ_{max}/ nm): 338, 316.

*2-((3-(4-Fluorophenyl)-1-phenyl-1H-pyrazol-4-yl) methylene aminophenol (**ANS-3**)*

Colour: Greenish; Anal. Calcd. For $C_{22}H_{16}FN_3O$ (357.13 g/mol): C, 73.94%; H, 4.51%; F, 5.32%; N, 11.76%; O, 4.48%. Found: C, 73.91; H, 4.49; F, 5.29; N, 11.66; O, 41; MS (*m/z*): 357 (M); IR (KBr, cm^{-1}): ν(-OH) 3356; *v*(-HC=N) 1627; *v*(N-N) 1053; *v*(Ar–C-H) 3122-2895; *v*(Ar–C=C) 1445; *v*(C-F)1197; ^{1}H NMR (DMSO-d_6): δppm 9.27-30,(d,1H, Ar-OH *J* = 15 Hz); 8.68-8.71 (d, 1H, HC=N); 8.0-8.2, (d, 2H, Ar-H); 7.85-7.88, (d, 2H, Ar-H); 7.42-7.55, (t, 2H, Ar-H); 7.1-7.4,(m, 3H, Ar-H), 7.0, (s, 1H, Ar-H); 6.82-6.89, (d, 1H, Ar-H), 6.78-6.82, (d, 1H, Ar-H); ^{13}C NMR (DMSO-d_6): δppm 184,(-HC=N); 115.37, 115.38, 115.58.9,118.09, 119.32, 120.24, 126.91, 127.11, 128.51, 129.41,129.71,130.42, 130.50, 137.23, 150.10, 151.25, 151.40,(Ph); UV-Vis:(DMF) (λ_{max}/ nm): 286, 318.

*2-((3-(4-Bromophenyl)-1-phenyl-1H-pyrazol-4-l) methylene aminophenol (**ANS-4**)*

Colour: Yellowish; Anal. Calcd. For $C_{22}H_{16}BrN_3O$ (417.05 g/mol): C, 63.17%; H, 3.86%; Br, 19.10%; N, 10.05%; O, 3.82%. Found: C, 63.17%; H, 3.46%; Br, 19.08%; N, 10.15%; O, 3.62%. MS (*m/z*): 417 (M); IR (KBr, cm^{-1}): *v*(-OH) 3333; *v*(-HC=N) 1595; *v*(N-N) 1114; *v*(Ar–C-H) 2848; *v*(Ar–C=C) 1491; *v*(C-Br) 610; ^{1}H NMR (DMSO-d_6): δppm 9.28 (s,-OH Ar); 8.68, (s, 1H, HC=N); 8.66 (s, 1H, Ar-H); 7.88-7.94, (m, 2H, Ar-H); 7.75-7.77, (d, 2H, Ar-H); 7.62-7.79 8.51 (d, 2H, H-Ar); 7.49-7.53,(t, 1H, Ar-H); 7.33-7.39 (s, 1H, Ar-H); 7.19-7.21(s,1H,Ar-H), 7.02-7.06, (t,1H, Ar-H), 6.87-6.89, (d,1H, Ar-H), 6.77-6.81 (t,1H, Ar-H); ^{13}C NMR (DMSO-d_6): δppm 114.35; 114.50, 115.48, 118.04 118.65, 119.08, 119.08,119.32, 120.36, 122.09, 126.95, 127.17, 127.51, 129.39, 129.88, 130.28, 130.39, 131.27, 131.51, 135.37, 137.12, 138.92, 149.82, 151.13, 151.30, (Ph); 183.98 (HC=N); UV-Vis:(DMF) (λ_{max}/ nm): 286, 370.

*2-((3-(4-Chlorophenyl)-1-phenyl-1H-pyrazol-4- yl) methylene aminophenol (**ANS-5**)*

Colour; Yellow, Anal.Calcd. For $C_{22}H_{16}ClN_3O$ (373.1 g/mol): C, 70.68%; H, 4.31%; Cl, 9.48%; N, 11.24%; O, 4.28%. Found: C, 70.65%; H,4.33%; Cl, 9.41%; N, 11.14%; O, 4.21%. MS (*m/z*): 373 (M); IR(KBr, cm^{-1}): *v*(-OH) 3385, *v*(-HC=N) 1622, *v*(N-N) 1039; *v*(Ar–C-H) 3045; *v*(Ar–C=C) 1217; *v*(C-Cl) 754; ^{1}H NMR (DMSO-d_6): δppm 9.28 (s,1H, -

OH Ar); 8.67, (s, 1H, HC=N); 7.92- 7.95(d, 2H, Ar); 7.78-7.80, (d, 2H, Ar-H); 7.43-7.51, (q, 4H, Ar-H); 7.31-7.37, (d, 1H, Ar-H); 7.17-7.19, (d,1H, Ar-H); 7.02-7.06,(d, 1H, Ar-H); 6.77-6.81, (d, 1H, Ar-H); ^{13}C NMR (DMSO-d_6): δppm 114.45; 115.51, 116.53, 118.19, 119.33, 119.32, 120.38, 126.98, 127.53, 128.35, 129.91, 130.15, 130.89, 133.54, 135.45, 136.25, 137.22, 138.93, 150.30, 151.09, 151.26 (Ph); 184.04 (HC=N); UV-Vis:(DMF) (λ_{max}/ nm): 260, 312.

Results and Discussion

The mass spectra of the all synthesized ligands [L] showed corresponding molecular ion peak [M]. The IR spectrum of ligands showed the characteristic ν(HC=N) bands in the region 1616-1630cm^{-1} and ν(−OH) bands at 3300−3425 cm^{-1} region. The UV-Visible spectra of Schiff bases were recorded in DMF at room temperature. The electronic spectrum of Schiff base shows only two bands which was assigned to n–π* transition of the C=N chromospheres and π-π* transition. The ^{1}H NMR Spectra of the Schiff bases in DMSO exhibit signals at 8.71, 8.68, 8.68-8.71, 8.68, 8.67 for compounds **ANS-1** to **ANS-5** attributed to CH=N- protons. The multi signals within the range 6.89-8.1 ppm were assigned to the aromatic protons of **ANS-1** to **ANS-5**. The signals at 9.91, 9.21, 9.27, 9.28 and 9.28 attributed to phenolic protones of compounds **ANS-1** to **ANS-5** respectively. The ^{13}C NMR spectra provide further support for the structural characterization of the Schiff bases. ^{13}C NMR spectral data of Schiff bases **ANS-1** to **ANS-5** have been given. The number of signals found corresponds with the presence of magnetically nonequivalent carbon atoms, which were assigned by comparison with literature values. The ^{13}C NMR spectral data of the Schiff bases were in accord with the proposed structures.

Antimicrobial evaluation

All the synthesized compounds were tested for their antibacterial and antifungal activity (MIC) *in vitro* by broth dilution method[20] with two Gram-positive bacteria *Staphylococcus aureus* MTCC-96, *Streptococcus epidermidis* MTCC 442, one Gram-negative bacteria *Escherichia coli* MTCC 443 and four fungal strains *Candida albicans* MTCC 227, *Aspergillus niger* MTCC 282, *S. cerevisiae* MTCC 170, *E. floccosum* MTCC 7880 taking gentamycin, ampicillin, chloramphenicol, ciprofloxacin, norfloxacin, nystatin and greseofulvin as standard drugs. The standard strains were procured from the microbial Type Culture Collection (MTCC) and microcare laboratory, surat, gujarat, India.

The minimal inhibitory concentration (MIC) values for all the newly synthesized compounds, defined as the lowest concentration of the compound preventing the visible growth, were determined by using microdilution broth method according to NCCLS standards. Serial dilutions of the test compounds and reference drugs were prepared in Muellere-Hinton agar. Drugs (10 mg) were dissolved in dimethylsulfoxide (DMSO, 1 mL). Further progressive dilutions with melted Mueller-Hinton agar were performed to obtain the required concentrations. In primary screening 1000 μgmL^{-1}, 500 μgmL^{-1} and 250 μgmL^{-1} concentrations of the synthesized drugs were taken. The active synthesized drugs found in this primary screening were further tested in a second set of dilution at 200 μg mL^{-1}, 100 μg mL^{-1}, 50 μgmL^{-1}, 25 μgmL^{-1}, 12.5 μgmL^{-1} and 6.25 μgmL^{-1} concentration against all microorganisms. The tubes were inoculated with 108 cfu mL^{-1} (colony forming unit/mL) and incubated at 37 ºC for 24 h. The MIC was the lowest concentration of the tested compound that yields no visible growth (turbidity) on the plate. To ensure that the solvent had no effect on the bacterial growth, a control was performed with the test medium supplemented with DMSO at the same dilutions as used in the experiments and it was observed that DMSO had no effect on the microorganisms in the concentrations studied.

The results obtained from antimicrobial susceptibility testing are depicted in Table 2 and Table 3. Compounds 1, 4, 5, in Table 1 showed significant antibacterial activity against *S. Aureus*, while compound 3 were found to be more active against *E. Coli.* The results obtained by the antifungal activity it was found that the compounds bases were moderately active against all tested fungi.

Table 2. Antibacterial activity of synthesized Schiff base

Minimum inhibition concentration, µg/mL				
Comp. No.	Code No.	*E, Coli* MTCC 443	*S. Pidermidis* MTCC 442	*S. Aureus* MTCC 96
1	**ANS-1**	200	100	62.5
2	**ANS-2**	500	250	150
3	**ANS-3**	65.2	100	62.5
4	**ANS-4**	125	500	250
5	**ANS-5**	100	100	62.5

Table 3. Antifungal activity of synthesized Schiff base

Minimum fungicidal concentration, µg/mL					
Comp No.	Code No.	*C.Albicans* MTCC 227	*A.Niger* MTCC 282	*S.Cerevisiae* MTC 170	*E. Floccosum* MTCC 7880
1	**ANS-1**	500	200	500	>1000
2	**ANS-2**	150	200	150	500
3	**ANS-3**	250	500	250	500
4	**ANS-4**	250	500	250	>1000
5	**ANS-5**	500	200	500	>1000

Conclusion

The synthesized compounds 2-((3-(4-substituted)-1-phenyl-1*H*-pyrazol-4-yl)methyleneamino) phenol were characterized by spectral data and subjected them for biological assay. Some compounds exhibited significant activity against all the tested microorganisms. Compounds were very good antioxidants due to the presence of hydroxyl group in them.

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References

1. Cimerman Z, Miljanic S and Galic N, *Croatica Chemica Acta*, 2000, **73(1)**, 81- 95.
2. Singh P, Goel R L and Singh B P, *J Indian Chem Soc*., 1975, **52**, 958-959.
3. Perry B F, Beezer A E, Miles R J, Smith B W, Miller J and Nascimento M G, *Microbois*., 1988, **45**, 181.
4. Elmali A, Kabak M and Elerman Y, *J Mol Struct*., 2000, **477**, 151.
5. Patel P R, Thaker B T and Zele S, *Indian J Chem*., 1999, **38A**, 563-566.
6. Patai S, Ed., "The Chemistry of the Carbon-Nitrogen Double Bond", J Wiley & Sons, London, 1970.
7. Jungreis E and Thabet S, "Analytical Applications of Schiff Bases", Marcell Dekker, New York, 1969.

8. Metzler C M, Cahill A and Metzler D E, *J Am Chem Soc*., 1980, **102**, 6075.
9. Dudek G O and Dudek E P, *Chem Commun*., 1965, 464.
10. Cimerman Z and Stefanac Z, *Polyhedron*, 1985, **4,** 1755.
11. Galic N, Cimerman Z and Tomisic V, *Anal Chim Acta*, 1997, **343**, 135-143.
12. Pfeiffer P, Breith E, Llibbe E and Tsumaki T, *Justus Liebigs Ann Chem.*, 1933, **503**, 84.
13. Hunter L and Marriott J A, *J Chem Soc*., 2000, 1937.
14. Sacconi L, Ciampolini M, Maggio F and Cavasini F P, *J Am Chem Soc*., 1962, **84**, 3246.
15. Holm R H and Swaminathan K, *Inorg Chem*., 1962, **1**, 599.
16. Perryand G C and Thornton D A, *J Inorg Nue Chem*., 1972, **34**, 3357.
17. Williams D R, *Chem Rev*., 1972, **72**, 203.
18. Campos A, Anacona J R and Campos-Vallette M M, *Mian Group Metal Chem*., 1999, **22**, 283.
19. National Committee for Clinical and Laboratory Standards, Method for Dilution Antimicrobial Susceptibility Tests for Bacteria that Grow Aerobically Approved Standard, Fourth Ed., NCCLS, Villanova, Italy, 1997 Document M 100-S7. S100-S157.
20. Isenberg D H, Essential Procedure for Clinical Microbiology, American Society for Microbiology, Washington, 1998; (b) Zgoda J R and Porter J R, *Pharm Biol.,* 2001, **39**, 221-225.
21. Chambhare R V, Khadse B G, Bobde A S and Bahekar R H, *Eur J Med Chem*., 2003, **38(1)**, 89-100.
22. Ambika Shrivastava and Singh R M, *Indian J Chem.,* 2005, **44B,** 1868-1875.