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Synthesis and biological activity a new Substituted Schiff bases.

KEYWORDS: schiff bases; antibacterial; antifungal.

Vishakha P. Bodade
Department of Chemistry, G.S.College of science,arts and commerce, ,Khamgaon,Buldhana, Maharashtra-India

Dr. Y.K.Meshram
Department of Chemistry, G.S.College of science,arts and commerce, Khamgaon,Buldhana, Maharashtra-India

ABSTRACT *A new series of biologically active variously substituted Schiff bases with general formula, R1N=CHR2. were synthesized by the reaction of differently substituted benzaldehyde with second, aryl amines in dimethyl formamide by microwave synthesis.*

Such compounds were characterized by different physico-chemical techniques like, melting point, elemental analysis, multinuclear NMR (1H). The free ligands and their metal complexes have been screened for their in vitro biological activities against bacteria, fungi and yeast.

1 Introduction
A Schiff base is a nitrogen analog of an aldehyde or ketone in which the C=O group is replaced by RC=N group. It is usually formed by condensation of an aldehyde or ketone with a primary amine.

Because of the relative easiness of preparation, synthetic flexibility, and the special property of C=N group, Schiff bases are generally excellent chelating agents,[1-7] especially when a functional group like –OH or –SH is present close to the azomethine group so as to form a five or six membered ring with the metal ion. 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 [8]. Schiff bases have been reported in their biological properties, such as, antibacterial, antifungal activities

[9-12]. Their metal complexes have been widely studied because they have anticancer and herbicidal applications [13,14]. They serve as models for biologically important species.

This paper presents a series of new Schiff bases with a potential biological activity resulted from the acid catalyzed condensation of aryl aldehydes with aromatic and hetero aromatic amines. These compounds could also act as valuable ligands.

2. METHOD AND MATERIAL:
2.1 Typical experimental procedure:-
3,4,5trihydroxy-benzohydrazide:
3, 4,5-trihyydroxy benzohydrazide was synthesized by refluxing propyl gallete (0.01mol) and hydrazine hydrate in presence of 25 cm^3ethanol for about 6 hrs. While refluxing 2-3 drops of conc. H_2SO_4 was added .After refluxing the crude compound was filtered and washed with distilled water .It was then recrystallized from ethanol. This compound is further used for the synthesis of substituted Schiff bases.

1a. 3,4,5trihydroxy benzoamido-4-chliroimine.
0.01 mol.3,4,5 trihydroxy benzohydrazide was mixed with equimolar amount of p-chloro benzaldehyde. In this reaction mixture Dimethyl sulphoxide was added as solvent .It was then irradiated to microwave. After completion of reaction ,the crude product was washed with distilled water ,and recrystallized from ethanol.

IR:(v max) cm-1**:** 3045(OH), 1622.18(C=N), 1588.8,1484 (C=C Aromatic),819.6(p sub.Cl)

Yield :69%,time required for completion of reaction:51 sec.

2b. 3,4,5trihydroxy benzoamido-4bromoimine.
0.01 mol.3,4,5 trihydroxy benzohydrazide was mixed with equimolar amount of p-bromo benzaldehyde. In this reaction mixture Dimethyl sulphoxide was added as solvent .It was then irradiated to microwave. After completion of reaction ,the crude product was washed with distilled water ,and recrystallized from ethanol.

IR:(v max) cm-1**:** 3422(OH), 1623.8(C=N), 1583, 1481 (C=C Aromatic),699.9(Br)

Yield :80%,time required for completion of reaction:1min.02 sec

3c. 3,4,5trihydroxy benzoamido-4-fluoroimine.
0.01 mol.3,4,5 trihydroxy benzohydrazide was mixed with equimolar amount of p-fluoro benzaldehyde. In this reaction mixture Dimethyl sulphoxide was added as solvent .It was then irradiated to microwave. After completion of reaction ,the crude product was washed with distilled water ,and recrystallized from ethanol.

IR:(v max) cm-1**:** 3400(OH), 1630(C=N), 1506 (C=C Aromatic),1153.7(-fl)

Yield :77%,time required for completion of reaction:45 sec

4d. 3,4,5trihydroxy benzoamido-4-iodoimine.
0.01 mol.3,4,5 trihydroxy benzohydrazide was mixed with equimolar amount of p-iodo benzaldehyde. In this reaction mixture Dimethyl sulphoxide was added as solvent .It was then irradiated to microwave. After completion of reaction ,the crude product was washed with distilled wa-

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ter ,and recrystallized from ethanol.

IR:(v max) cm-1: 3447(OH), 1617(C=N), 1576,1549 (C=C Aromatic), 620.5(-I)

Yield :75%,time required for completion of reaction:50 sec

3.Biological activity
3.1. ANTIBACTERIAL ACTIVITY
Procedure:
The antibacterial activity was measured by agar cup method..

The bacterial cultures selected were, two gram negative cultures viz. *Escherichia coli; Salmonella typhi and* two Gram positive cultures viz. *Staphylococcus aureus, Bacillus subtilis*. This seeded preparation was then poured in sterile Petri plate under aseptic condition and allowed it to solidify.

Cups of 10mm diameter were bowered in the agar plate with sterile cork borrer. 100. ~I ·of compound solution prepared in Dimethyl Sulphoxide (1%) was added in the cup under aseptic condition with the help of micropipette. 100 ul of DMSO was also placed in one of the cup as blank (negative control). A standard antibiotic disk impregnated with 10 units of Penicillin was also placed on the seeded nutrient agar surface as standard reference antibiotic (positive control). Plates were incubated at 37°C for 24 hours. After incubation the average zone of inhibition was recorded in mm. [15-19]

3.2 ANTIFUNGAL ACTIVITY
Procedure:
Antifungal activity was performed by Poison plate method. The "medium used was Potato Dextrose Agar (Himedia). The medium was prepared and sterilized at 10 Psi in autoclave for 15 minutes. Then the compound to be tested is added to the sterile medium in aseptic condition so as to get final concentration as 1%. A plate with DMSO was prepared as blank (negative control) similarly a plate with 1% Gresiofulvin was prepared as standard reference plate (positive control).

Aspergillus niger, Penicillium chrysogenum, Fusarium moneliforme, Aspergillus flavus were selected as test fungal cultures. They were allowed to grow on slant for 48 hours so as to get profuse sporulation. The fungal suspension was spot inoculated on the plates prepared using. Compound with the help of nicrome wire loop. The plates were incubated at room temperature for 48 hours. After incubation. plates were observed for the growth of inoculated fungi.

Method - Agar Cup method.
4.. Result and discussion:
4.1 Antibacterial activity
The results of the antibacterial screening of the Schiff bases at a concentration of 20mg/ml against all bacteria have been found. The inhibition zones were measured in mm and results are shown in following table . The results of antimicrobial screening, indicate that substituted Schiff bases show significant activity against Staphylococcus aureus, Escherichia coli, Bacillus subtilis and salmonella typhi. When we increase concentration, area of inhibited growth also increased.

The antibacterial results are shown in table no.1

Table No.1

Sr.No.	Compound	Escherishi coli	Salmonella coli	Staphylococcus typhi	Bacillus aureus subtilis
1	1a	23mm	16mm	17mm	13mm
2	2b	40mm	32mm	30mm	36mm
3	3c	-ve	-ve	12mm	-ve
4	4d	14mm	-ve	13mm	13mm
5	DMSO	-ve	-ve	-ve	-ve
6	Penicillin	24mm	19mm	34mm	18mm

Legends- - ve No Antibacterial Activity
Zone of inhibition -- mm

4.2 Antifungal activity
From the results obtained by the antifungal activity it is found that the compounds e,f and g active against all tested fungi .

The greater activity of these compounds is probably due to the presence of hydroxyl group, fluoro, and chloro group.. Compound b show good activity against all tested fungi as compared to standard drug.. The antifungal activity results are shown in Table no.2

Table No.2

Sr.No.	Compound	Aspergillus niger	Penicillium chrysogenum	Fusarium moneliforme	Aspergillus flavus
1	1a	+ve	+ve	+ve	+ve
2	2b	+ve	+ve	+ve	+ve
3	3c	-ve	-ve	-ve	-ve
4	4d	-ve	-ve	-ve	-ve
6	-ve control	-ve	-ve	-ve	-ve

4 Conclusion
Schiff bases of substituted aldehydes and secondary amines were synthesized by standard and microwave irradiation technique. These compounds exhibited significant activity against all the tested microorganisms.

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