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Study of Synthetic Based and Plant Based Compounds Potentially Active against Microbes in Life Cells by Using Chemical Techniques

Agarwal Charu

Ex. Lecturer, Deptt. of Chemistry, P.C. Bagla P.G. College, Hathras

Abstract: ***Various pharmacological properties of chemical compounds are need of time. Chemical examination of a plant Cyperus Scariosus commonly called "Nagarmotha" belonging to family cyperaceae shows anti-inflammatory, anti-convulsant properties. Various compounds obtained were examined and identified viz. sesquieterpenoids etc. In contradiction to that synthetic compounds of metal Schiff bases of substituted coumarin and aromatic aldeyhyde with Zn(II) was also prove active against various bacteria and fungi in life cells. All examination indulges steps like preparation, characterization, identification, biocidal studies etc. In view of that for enrichment of human welfare aforesaid compounds were proceeded with the aim of retrieving knowledge about their pharmacological studies.***

Keywords: Schiff bases, Anti Microbial properties

1. Introduction

Plant derived substances have recently become of great interest owing to their versatile application. Medicinal plants are the richest bio-source of drugs of traditional systems, medicinal nutra-ceuticals, food supplements and chemical entities for synthetic drugs. A number of interesting outcomes have been found with the use of a mixture of natural products to treat diseases, most notably the synergistic effects. Plant belonging to the family cyperacea viz., *Cyperus scariosus* commonly called '*Nagarmotha*' examined, separated, eluted and obtained compounds were terpenoids, *sesquieterpenes* and many more. They were found active against different microbes. The need of more efficient drugs in methodology studied. In contradiction more potential of synthetic metal Schiff bases complexes synthesized, characterized and examined against microbes. Exhibited good anti microbial performances than plant derived compound with low MIC ≥ 250 µg/ml. Bacteriocidal effects and broad activity spectrum qualifies these compound as suitable candidates to the next step of drugs fabrication. Nevertheless, further studies on the mechanism of growth inhibition and toxicity are needed in order to evaluate their potential. Annually millions of people die due to infections caused by micro organism resistant to current anti-biotics (W.H.O. 2012). In the search for new anti-microbial effective in the treatment of infections caused by multi resistant bacteria. Due consideration ought to be given for the synthesis of drugs with new activation target as well as potentialisation of the activity of compounds with known anti-microbial activity.

2. Material and Methods

a) ***Cyperus scariosus*:** Extraction of mechanically purified plant material (600gms) with light petroleum extract for 36 hours at its boiling point under refluxed conditions. 11 gms dark brownish solid extract obtained gave positive test for sterols, terpenes. Further spotted on T.L.C. with 2% silver nitrate and developed with carbon tetrachloride- ethyl aceto-acetate (95.5 v/v). Dried and sprayed with 10% methanolic H_2SO_4 and kept in electric oven for 5 Min. Four spots with Rf values 1.0, 0.62, 0.30 and 0.20 respectively gets visible. Further extract was separated by column chromatography and components obtained was flavonoids, terpenoids, mono and sesquiterpene, steroids, saponins, ketones, 1,8–cineole, alkaloids. Among them *Sesquiterpenes* were found to be more active. Evidently longiverbenone isolated and exhibited moderate to good anti bacterial activity against the organism. It gives anti bacterial activity against all the test bacteria viz. 11 potential human pathogenic bacteria by disc diffusion method using Mueller-Hinto (Agar and Broth) medium. All the results were compared with the standard anti-bacterial, anti-biotic ampicillin (20 µg/disc). The largest zone of inhibitors lie 30, 22, 15 and 8 mm in diameter against *vibrio cohlearae* at the concentration 160, 80, 40, 20 µg/disc respectively. (Ref. Table.1)

b) **Zinc (II) SAHMC complex:** 0.01 mole $ZnSO_4$ in CH_3OH to 0.01 mole of methanolic solution of salicylaldehyde and 0.01 mole of methanolic solution of 8-amino-7-Hydroxy-4-methyl coumarin were refluxed for about 3 hours then concentrated, cooled, filtered, washed with ether and recrystallised from methanol. Zinc complex formed. M.F- Zn C_{34} H_{24} O_{28} N_2; molecular weight- 653.3; M.P- 294 °C.

Table 1: Anti-bacterial activity of *longiverbenone* isolated from rhizome of *Cyperus scariosus*

Test organism	Diameter of zone of inhibition (mm)					
	Longiverbenone (µg/disc)					Ampicillin (20 µg/disc)
	160	80	40	20	10	
Bacillus subtilis	25	14	7	0	0	19
Bacillus cereus	26	17	12	7	0	18
Bacillus megaterium	25	14	7	0	0	16
Staphylococcus aureus	0	0	0	0	0	22
Escherichia coli	15	10	0	0	0	10
Vibrio cholerae	30	22	15	8	0	15
Shigella dysenteriae	24	13	8	0	0	22
Shigella sonnei	10	6	0	0	0	20

Salmonella typhi	15	8	0	0	0	20
Salmonella paratyphi	12	10	6	0	0	17
Pseudomonas aeruginosa	0	0	0	0	0	0

3. Anti- Microbial Activity

Substances that are used against micro-organism may be described as "cidal" if they kill cells quickly or described as "static" if their effect is mainly to inhibit the growth of cells. Most anti- microbial agents are obviously good ligands. It appears, therefore, that the activity of these ligands is increased in the form of metal complex on being coordinated to a suitable metal ion. This is either due to more lipo soluble nature of the metal complex than the organic ligand alone. It is believed that the metal ion helps to transport it across the cell membrane. They penetrate the cells and destroy the oxidation and reduction system of the cells.

4. Experimental

1 mg/ml of metal complex (Zn (II))In Propylene Glycol were sterilized by passing into the G-5 sintered crucible. Minimum inhibitory concentration obtained by serial dilution method "2 fold" obtained for metal complexes. Graded solution of the metal complexes in nutrient medium are inoculated with organism "two fold serial dilution" method used. 1 ml of the seeded broth was taken in 10 sterilized tubes (3x 100 mm size). First tube was kept emptied, separately 2 ml of the seeded broth was prepared, 100 mg /ml and 150 mg /ml of metal complexes in tubes **A** and **B** respectively (by dissolving 0.2 ml and 0.3 ml of the stock solution in 1.8 and 1.7 broth respectively). Tube- **A** contents placed in first empty tube, 1 ml of tube B was added to second tube. Similarly, 1 ml contents of the first tube was withdrawn and added to third tube and mixed well. 1 ml content of second tube was pipette out and added into fourth tube. This gradient dilution process continued for 10 tubes. Tubes were labeled with 100 mg/ml, 75 mg/ml, 50 mg/ml, 25 mg/ml, 12.5 mg/ml, 6.25 mg/ml, 3.125 mg/ml, 1.56 mg/ml, 0.78 mg/ml and 0.39 mg/ml respectively. For control of culture 1 ml of each of the seeded broth and broth was placed in two separate tubes. All above sets of tubes were incubated in BOD incubator at the desired temperature and time for the respective indicated micro organism. The tube having the highest dilution showing no visible turbidity was chosen. The amount of the test compound in this tube was MIC. (Refer to Table.2)

Table 2: Minimum Inhibitory Concentration (MIC) in Molar Concentration (x 10^{-4}) of SAHMC (salicyl aldehyde–8- amino- 7- hydroxy- 4– methyl coumarin) and Its Metal Complexes

Sr.No.	Compound	Bacteria		Fungi	
		S.aureus	E.coli	A.niger	C.albicans
1.	SAHMC	>3.5	>3.5	>3.5	>3.5
2.	$[Zn(SAHMC)_2]$	0.17	0.15	0.16	0.15

5. Discussion

On comparing with plant eluted compounds and synthetic metal Schiff complexes, both were found active against various microbes but their efficiency varies. The minimum inhibitory concentration of natural compound was slightly more than synthetically derived compounds. Extracts of cyperus scariosus proved to be as anti-inflammatory, anti-diabetic, analgesic, hepato-protective, astringent, oils are used in perfume industry. Many chemical constituents viz. flavonoids, terpenoids, mono and sesquiterpenes, steroids, saponine etc were obtained but main active component appear to be sesquiterpene. Among them longiverbe none showed anti-microbial activity and MIC were obtained. These were injected in guinea pigs, rats but the death ratio was found to be more with synthetic chemical compound but less when cyperus scariosus extract were injected. Metal Schiff bases which are discussed found to be more fatal but the required minimum inhibitory concentration is less for the Zn(II) SAHMC complexes. Here discussion is being restricted to obtain the minimum inhibitory concentration of metal complexes.

6. Conclusion

The above collected information regarding the uses of pharmacological activities of Cyperus scariosus were matched with available literature. And metal Schiff bases were obtained, characterized and bio-cidal screening had been done. Bio-cidal screening of both compounds compared in the best classical approach had been performed. In the search of new drugs for management of various diseases it was found, compounds of plant origin is popular remedy among the various ethnic groups. MIC of both were compared and presented above. The further research are exploring may provides therapeutic potential of this plant and therapeutic of metal complexes. Our results could stimulate further pharmacological studies seeking new antimicrobial agents from the plant resources. The studies revealed that Schiff bases containing amino group were found more active against selected bacteria and fungi. It is also observed that Zn(II) metal complexes have exhibited higher antimicrobial activity than the respective schiff bases as cleared from data in table 2.

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