Synthesis of the novel type of phenyl acridine and pyridine derivatives as an antifungal agent

by

I. N. D. Warnakulasuriya

Dissertation submitted in partial fulfillment of the requirements for the degree of MASTER OF SCIENCE in ANALYTICAL CHEMISTRY, Faculty of Science, University of Colombo, Sri Lanka.

January 2012

ABSTRACT

The continuously increase in multi drug resistant to pathogenic fungi and shortage of new antimicrobial agents are a serious problem in treatment with infectious diseases. Therefore it is essential to synthesis novel compounds and evaluates the antimicrobial properties of these compounds. Acidinium and pyridinium salts are used as an effective antimicrobial agent and various antimicrobial applications in industrially and domestically, from a long time.

This research mainly focus on synthesis of acidinium based salt and study of the *in vitro* interaction of aqueous solution of this compound with fungi. In this experiment *Aspergillus niger ,Rhizopus sp., Penicillium sp., Fusarium oxysporum, Mucor sp., Colletotrichum sp.* were used as models to screen for antifungal activity.

The secondary aim of this research, previously synthesized pyridinium derivative, 4-phenyl-1-(2-phenyl-allyl)-pyridinium bromide was used to investigate its interaction with fungi and were evaluated *in vitro* by disk diffusion method against *Aspergillus niger ,Rhizopus sp., Penicillium sp., Fusarium oxysporum, Mucor sp., Colletotrichum sp.*

In the conventional synthesis scheme, N,N-diphenylamine and benzoic acid were reacted in the presence of zinc chloride in 1:1:1 mole ratio under high temperature. This synthesis was carried out according to the previously published methods. A mixture of N,N-diphenylamine benzoic acid, and zinc chloride was irradiated in a domestic microwave oven(900 W) for 1.5 minutes. The crude product was dissolved in dichloromethane and washed with 10% aqueous sodium hydroxide and water. The organic layer was dried with anhydrous Na_2SO_4 and evaporated to yield a brown colourr crystal like solid product. Resulting acidinium was later recrystallized using ethanol/diethylether solvent system. Then the isolated product was (subjected to) carried out the protonation reaction. The dark colored solution was evaporated to obtain a yellow colored viscous liquid. Then recrystallization was carried out by adding diethyl ether until a precipitate was formed.

Then the investigation was undertaken to assess antimicrobial properties of acridinium compound and pyridinium based salts in order to establish the possibility of these compounds as antimicrobial agents. It was initially evaluated the susceptibility of these compounds against above mentioned fungi using disk diffusion method. Only acridinium compound showed an antimicrobial activity. According to the antimicrobial test results strong inhibitory effect was observed on *Aspergillus niger* (larger inhibition zone was formed) and *Rhizopus sp*

showed very small inhibition. (very small inhibition zone) towards the 4-phenyl-1-(2-phenyl-allyl)-pyridinium bromide while all the other tested fungi revealed resistance against the compound.

Aspergillus niger was selected to determine the MIC (Minimum Inhibitory Concentration) by the disk diffusion method and it was found as ≤ 1800 μg/ml.

Mode of interaction and the exact mechanism of binding of this compound to the fungi are still not understood.

Finally comparison of antibacterial activity of 4-phenyl-1-(2-phenyl-allyl)-pyridinium bromide with known antibiotic Clotrimazole, which are given to cure *Aspergillus sp* infections was carried out using disk diffusion method.