Executive Summary of the final report of the work done on the project sanctioned under Minor Research Project by UNIVERSITY GRANT COMMISSION

THE PRINCIPAL INVESTIGATOR: - Dr. Savanta V.Raut, Department of Microbiology, Bhavan's college, Munashi Nagar Andheri[w], Mumbai -400058

TENURE OF THE PROJECT :- Two year 5th Oct.2010 to March 2012 TOTAL GRANT ALLOCATED: - Rs. 100,000/-

TITLE OF THE PROJECT:-"Studies on Antimicrobial principles of extract of *Sonneretia alba*. OBJECTIVES OF THE PROJECT :-

- 1. Solvent Extraction of antimicrobial compounds from leaves and fruits of Sonneratia alba.
- 2. Study of the antimicrobial effect of extracts of leaves and fruits on various human pathogens and phytopathogenic bacteria and fungi.
- **3.** Determining the Minimum Inhibitory Concentration of the extract against human and phytopathogenic organisms to elucidate their potential applications in drug therapy.
- 4. Activity guided fractionation with various organic solvents differing in their polarity.
- 5. Isolation of the bioactive compound by Qualitative analysis, HPTLC, Bioautography.
- **6.** Purification and Identification of the bioactive compound by using sophisticated analytical tools like Preparative HPTLC, UV-Spectrophotometer, GC-MS, LC-MS, FTIR and CHNSO analyzer.

SUMMARY OF THE FINDINGS; -

A] Fruit Extract :-The antimicrobial activity of the fruit extract, hot extract using Soxhlet's apparatus showed higher activity (10-25mm inhibition zone of tested cultures) as compared to cold extraction method /(15-24mm inhibition zone of tested cultures), Bioactivity guided fractionation shows Methanol and Acetone fractions gave the maximum activity than other solvent fractions, with methanol and acetone fractions showing 20-25mm and 21-24mm range inhibition zones of sensitive test cultures respectively. Bioautographic studies of acetone and methanol fractions revealed the separation of three the major bioactive compounds in HPTLC shows antimicrobial activity against the sensitive strains tested. The above three compounds as characterized by UV absorption, showed the maximum absorption in UV light at 310nm while the maximum absorption in the visible range was at 340 nm and CHNSO analysis indicated that it contained no sulphur group while carbon is 24.057%, hydrogen 7.979%, nitrogen 0.484% while oxygen is 10.152%. Thus it can be said that the active compound of interest may not contain sulphur group. Finally GCMS,LCMS and FTIR analysis shows that the antimicrobial component present in the Fruit methanol fraction may be 2-[3,4-Dichlorophenyl]-6-methoxy cinchoninic acid or 7,8-Epoxylanostan-11-ol,3-acetoxy .(15)

B] Bark Extract :-Methanol extract of Bark showed the highest antimicrobial activity, followed by Acetone and Ethyl acetate extracts, while in cold extraction Alcohol gave the highest activity followed by Hydroalcohol. In this case, *Staphyloccocus aureus* MTCC 1144 (23mm inhibition zone) and *Salmonella typhi* and *Shigella flexneri* MTCC1457 (22mm inhibition zone), showed maximum activity . MIC of crude hot extract was found to be 2.5 mg/ml, while cold methanol extract was less than 1.5 mg/ml and in MBC extract it was less than 2.5 mg/ml. Experimentation with Bioactivity guided fractionation revealed maximum activity in Methanol and Acetone fractions as compared to other fractions. Inhibition zones ranging from 13-22 mm and 11-17mm were observed in methanol and acetone fractions respectively. In bioautography and

HPTLC experiments with methanol and acetone fractions the presence of one separated bioactive compound was detected. The semi purified bioactive compound was subjected to preparative HPTLC was carried out and the separated band isolated through elution. AST of the same showed inhibitory action of the test strain. Analysis of Acetone fraction by UV Visible absorption, CHNSO, GCMS, FTIR and NMR shows probable compound may be N-βchloropropionyltryptamine [3-N-ethyl (1-Cloropropionamide)Benzoate] with probability 60.2% and in methanol fraction probable compound Myo-inositol 4C methyl with probability 36.7% as detected in GCMS analysis and its dell value matches with standard dell value. C] Leaves Extract :- The antimicrobial activity of leaves extract showed higher antimicrobial activity of test cultures except in case of Bacillus subtilis, and Klebsiella pneumoniae which showed a lower activity. MIC range was found to be 1.56 - 3.12 mg/ml, 6.25 - 12.5 mg/ml and 12.5 – 25 mg/ml with extracts of methanol, hydro alcohol and alcohol respectively. However in case of Ethyl acetate, Acetone and Alcohol extracts MIC was less than 1.5 mg/ml.Activity guided fractionation of extract revealed maximum activity with Methanol and Acetone fractions as compared to other fractions. The range of inhibitory zones of tested cultures was found to be 22-26mm and 23-28mm per 50µL methanol and acetone extracts respectively. Target directed bioautography showed one bioactive spot separation in acetone fraction. The phytochemical analysis showed the presence of tannins, saponins, flavonoids and alkaloids in acetone and methanol fractions. The compounds in the leaf acetone fraction could not be conclusively identified, necessitating further purification in future studies.

NO. OF PUBLICATIONS OUT OF THE PROJECT: - 03 publications

 Title - Studies on antimicrobial activity of leaves extract of *Sonneratia alba*" in Current Research in Microbiology and Biotechnology. Vol. 1, No. 5 (2013): 203-213 ISSN: 2320-2246.
Title –" Studies on antimicrobial activity of leaves extract of *Sonneratia alba*" in journal Trends in Biosciences.[ISSN-0974-8] [NAAS Rating -2.7] Vol.No.6[6] ,Dec.2013 PP.831-837
Title "Studies on antimicrobial compounds of extract of fruit of *Sonneratia alba*" in *Journal* Advances in Life Sciences vol.3 No.2.Nov-2014.pp 85-96 ISSN-2278-3849 [peer reviewed

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