ABSTRACT
Natural product chemistry has long served as a consistent resource to pharmaceutical needs. Although to contrary, the need for scientific guidelines and criterions as a qualitative and qualitative tool to measure its accuracy seems to be deficient. This explains the fact that even though more than 50% of people around the globe use herbal formulations as curatives but very few gain acknowledgment. In this study, the natural products are attempted to be used as aquaculture drugs considering the apparently related biology of the aquatic ecosystem. The allelopathic interferences of aquatic macrophytes collected from the floodplain wetlands of West Bengal are used as a natural warfare against fish diseases. The antimicrobial product screening is inclusive of fish toxicity markers with antioxidant activity, brine shrimp lethality test, duckweed toxicity test and most importantly anti-algal assay against harmful algal blooms.

Three aquatic macrophytes have been considered for the study, *Vallisneria spiralis* L., *Ipomoea aquatica* Forssk. and *Cyperus rotundus* L. The use of banana (*Musa paradisiaca*) leaves is an auxiliary addition which was necessary realising its potential role in maintain and restoring the pH of the water body.

Two bioactive fractions have been isolated from *V. spiralis* leaf extract, Ethanol 80% (VsE80) and semi-purified methanol fraction (VsF7). The VsF7 has shown a strong antioxidant activity with IC50 0.258 mg/ml with antimicrobial activity against *Aeromonas* species. The GCMS data shows the fraction to be rich in long chain alkenes and esters of phthalic acid. The VsE80 was more proficient in anti-algal assay supressing the Chlorophyll-pigmentation. It also showed antifungal activity against *Malassezia globosa*. The m/z of the crystal obtained had molecular mass of 359 and the FT-IR spectra revealed significant functional groups at fingerprint regions with sulphoxides (S=O) at 1360cm⁻¹ being one such.
ABSTRACT

Bio-minerals are one of the plant secondary metabolites that are imperative for their biological communication. Raphides or calcium oxalate (\(\text{CaC}_2\text{O}_4\cdot2\text{H}_2\text{O}\)) crystals from the aqueous fraction of Ipomoea aquatica leaves \((F2_{a1})\) are abundantly synthesized bio-minerals in plants which help channelize the excess calcium absorbed. 1.6% raphide content have been isolated and purified by chromatographic process. The structural characterization of the crystal by the magnetic resonance and infra-red spectra further confirmed the presence of raphides. Direct bio-autography followed by agar well diffusion assay revealed the in-vitro antimicrobial property of the crystals against fish pathogen Citrobacter freundii with MIC< 50 ppm. The crystal exhibited strong free radical scavenging activity with AAI 1.38 and IC50 at 0.144 mg/ml. The atomic force microscopy studies predicted a hydrous inorganic salt displaying average peaks between 4-5 nm with homogeneity of dispersion.

The Cyperus rotundus contained three bioactive fraction of diverse polarity, CrP isolated from Petroleum ether fraction, CrCh isolated from chloroform fraction and CrM isolated from the methanol fraction. The CrP fraction showed strong bactericidal activity against fish pathogens Pseudomonas putida, Bacillus safensis and Citrobacter freundii. It showed an ABTS radical scavenging activity at IC50 70.8mg/ml. The fraction constituted of methyl esters with commercially known antimicrobials like methyl Linoleate, Methyl Stearate and Eicosanoic Acid methyl ester. The CrM fraction was the most feasible fraction with least toxicity and with strong antimicrobial property against Aeromonas. popoffi, A. hydrophila and Edwardsiella tarda. The fraction mainly consisted of Mycorradicin, Annosquamosin A and Cinnamylcinnamate. They are known standards of antimicrobial and antioxidant protocols. The fraction has also shown chlorophyll-a inhibition from 60% to 24%.
As for *Musa paradisiaca*, the ethyl acetate fraction was found to be most bioactive and holds the bacteriostatic property against *Edwarsiella tarda* and *Streptococcus aureus*. The brine shrimp lethality assay states the LC50 for 6hrs at 0.845µg/ml and that of 1 hr at 15.005µg/ml which concludes the fraction to be toxic in nature.

The in-vivo application of F2a1a and CrM was tested against Aeromonas infected *Pangasius hypophthalmus*. Two experimental conditions were studies with plant sample formulated feed and banana leaf extract in the medium. The prophylaxis set was found to show significant results than curatives. The infected fish fed with formulated feed had almost the same histopathology like the uninfected fish samples with only exception of cytoplasmic vacuoles. The intact triad and the proportionate hepatocytes of the liver were seen. The kidney tissue samples were marked with prominent and integrated Bowman’s capsule.

Comprehensively, impact study of environmental chemistry on aquaculture point of view has also been made for isolation and molecular identification of bioactive allelo-compounds.