SUMMARY
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The generation of wastewaters through human activity or by industries is a major environmental threat. This can be suitably addressed by developing biological options, which are environment-friendly and cost effective. Investigations were undertaken to characterise the sewage wastewater in terms of physicochemical parameters and microalgal diversity, followed by screening of microalgae for wastewater treatment and biomass production. Consortia (native and non-native strains) were developed and evaluated for nutrient removal, wastewater treatment and biomass production potential. The sewage grown microalgal biomass was evaluated for its efficacy as a biofertilizer in wheat.

Observations revealed a wide variation in the physicochemical characteristics and microalgal diversity of sewage wastewater during the study period (at monthly interval over a period of one year). Diversity analyses revealed the presence of algal members belonging to all major divisions (viz. Cyanophyta, Chlorophyta, Bascillariophyta, Xanthophyta and Euglenophyta), with a predominance of Cyanophycean members. The indices of microalgal diversity showed a positive correlation with nutrients and a negative correlation with COD and heavy metal concentrations, implying the significant role of these factors in influencing the algal population. The highest microalgal species richness was recorded in the month of June, correlated with high NO$_3$-N and low COD levels. Low values of Shannon-Weiner and Simpson’s diversity indices revealed that sewage wastewater was heavily polluted. *Phormidium* sp. being a robust organism, was present throughout the year as it can tolerate high COD and heavy metal concentrations. Sewage wastewater in this study also exhibited high NO$_3$-N, NH$_4$-N and COD levels. Spectrometric analyses of sewage wastewater revealed the presence of heavy metals (Cr (III), Co (II), Pb (II), As and Cd), with Cr (III) ranging from 3 - 4 mg L$^{-1}$ in all the samples collected over the year. A positive correlation was found between COD and total heavy metal concentration ($r = 0.77$).

In preliminary studies, all the tested strains of microalgae from germplasm *viz.* *Calothrix* sp., *Lyngbya* sp., *Ulothrix* sp. and *Chlorella* sp. were unable to flourish in
full strength sewage wastewater, therefore, sewage wastewater diluted with tap water (1:1 ratio; TW+SW) was used to study the wastewater treatment potential and biomass production. Among the inoculated strains, *Calothrix* showed the highest dry cell weight (916.67 mg L\(^{-1}\)), chlorophyll and carotenoid content in tap water + sewage water (1:1) treatment. The total dissolved solids and electrical conductivity of tap water + sewage water after incubation with *Calothrix* sp. decreased by 28.5 and 28.0 %, accompanied by higher increase in dissolved oxygen compared to other microalgae on the 20\(^{th}\) day. Microalgal monocultures revealed significant removal of NO\(_3\)-N ranging from 57-78% and PO\(_4\)-P (44-91%) from tap water + sewage water (1:1) with higher NO\(_3\)-N removal of 78.9% in *Chlorella* sp. inoculated wastewater. The study revealed a differential response in the uptake of nutrients, water quality improvement and biomass production. Therefore, three types of microalgal consortia were developed viz. MC1, comprising microalgae from germplasm (species of *Calothrix*, *Lyngbya* and *Ulothrix* and *Chlorella*); MC2 (native filamentous strains isolated from sewage wastewater - species of cyanobacteria - *Phormidium*, *Anabaena*, *Westiellopsis*, *Fischerella*, and green alga - *Spirogyra*); and MC3 (comprising native unicellular strains of sewage -species of green algae - *Chlorella*, *Scenedesmus*, *Chlorococcum*, and cyanobacterium- *Chroococcus*). These consortia were evaluated for their potential for nutrient removal, water quality improvement and biomass production using primary treated sewage water. The highest NO\(_3\)-N (90%) and PO\(_4\)-P (97.8%) removal were observed with MC2 inoculated sewage water. Highest decrease in TDS from 1120 to 806 mg L\(^{-1}\) and the highest increase in DO was shown in MC2 inoculated sewage water on 6\(^{th}\) day. The biomass production was also highest in MC2 (1.07 g L\(^{-1}\)) followed by MC1 and MC3 (0.90 and 0.94 g L\(^{-1}\) respectively) on 6\(^{th}\) day. The consortium of filamentous strains from native environment proved promising, not only in nutrient removal efficiency, but also led to enhanced biomass.

Further, an investigation was aimed to evaluate the activity of different enzymes related to N and P utilization, in microalgal consortia grown in sewage wastewater and analyze the quality of biomass generated. All the consortia recorded higher biomass production in sewage wastewater as compared to the Bold’s Basal Medium (BBM). The consortium of native filamentous strains (MC2) exhibited significantly higher glutamine synthetase (GS) and nitrate reductase activity (NR) of 17.6 and 3.7 \(\mu\)moles min\(^{-1}\) mg protein\(^{-1}\) respectively in sewage wastewater, compared
to the consortia of native unicellular strains (MC3) and selected microalgae from germplasm (MC1). The highest acid phosphatase (AcPA) and alkaline phosphatase (APA) activity were recorded with the consortium MC3. MC2 exhibited significantly the highest nitrogen (4.7%) and potassium (5.7%) in the biomass generated. The present study revealed that the activity of nutrient mobilizing enzymes play an important role in wastewater nutrient removal as their activity was concomitant with the rates of nutrient removal by different consortia in the sewage wastewater. Due to presence of heavy metals in the sewage wastewater used, investigations were undertaken for studying the metal removal potential of various formulated microalgal consortia was evaluated in single (Cr (III)) and multiple metal components [Cr (III), As (V), Pb (II) and Co (II)] containing wastewater substrates under outdoor conditions. The highest Cr removal (64%) was observed with the consortium of native filamentous strains (MC2) in both types of sewage wastewater, with single and multiple metal components, although it was higher in the former on 6th day. However, with the increase in incubation period to 12th day, it was higher in MC3 (the consortium of native unicellular strains) inoculated sewage wastewater. In sewage wastewater with multiple metal components, 48.7 – 68.7%, 34.8 – 53.8% and 69.3 – 91.6% of As, Pb and Co removal was recorded after treatment with the different microalgal consortia, respectively. This study highlighted the possible involvement of both bioaccumulation and bioadsorption mechanisms in effective metal removal and the promise of such consortia in bioremediation of sewage wastewater.

Investigations were undertaken to evaluate the utility of sewage wastewater as an alternative growth medium for microalgae and potential of microalgal consortia developed in terms of mobilization of wastewater nutrients (N, P, heavy metals etc.) and production of nutrient rich biomass. The potential of two formulations prepared using sewage grown microalgal biomass (filamentous/unicellular microalgal consortia with vermiculite: compost as a carrier), was further assessed as a biofertilizer [N, P, K and micronutrients (Zn, Fe, Cu and Mn)] in wheat crop. Analyses of sewage grown microalgal dry biomass revealed high N, P, and K along Cr (III) above permissible limits given by WHO. However, after preparing formulations with carrier, Cr (III) level decreased considerably, much below the permissible limits. A total of five treatments were taken viz. T1 (Recommended dose of fertilizer N: P: K in 120: 60: 60), T2 (75% N + full dose PK), T3 (75% N + full dose PK + carrier), T4 (75% N +
full dose PK + MC3) and T5 (75% N + full dose PK + MC2). A significant enhancement was observed in the microalgal consortia inoculated treatments in terms of soil nutrient characteristics at mid crop stage, as compared to uninoculated treatments. The highest values of available N, P and K content in soil and acetylene reduction activity were recorded in treatment T5. Significantly higher available Zn, Fe, Cu and Mn content was recorded in soil samples from treatments belonging to microalgal consortia inoculation, as compared to uninoculated treatments, at both mid and harvest stage of wheat crop. A significant enhancement of 35.1 - 51% in organic carbon content was recorded in microalgal consortia inoculated treatments over control, with T5 recording the highest values. Significantly higher dehydrogenase activity was recorded in T4. MBC was also enhanced by 31.8 – 67.0% in both the inoculated treatments over control, with the highest values in T4. Both the microalgal formulations significantly increased the N, P and K content of roots, shoots and grains. Microalgal consortia inoculated treatments also revealed 37.3 - 48.0% increase in grain yield with significantly higher micronutrient (Zn, Fe, Cu and Mn) concentration in grains, as compared to control. At harvest stage, the treatments inoculated with microalgal formulations (T4 and T5) also recorded 7.4 – 33% increase in plant dry weight and upto 10% enhanced spike weight. The values of 1000 grain weight showed an enhancement of 5.6 – 8.4%, as compared to T1. A positive correlation was observed between soil nutrient availability at mid crop stage and plant biometrical parameters at harvest stage. A strong positive correlation was also recorded between the availability of micronutrients in soil at mid crop stage and grain yield.

The present study highlighted the promise of wastewater as a medium for the growth of microalgae and as a rich bioresource for identifying robust microalgae. Useful microalgal consortia were developed which proved useful in bioremediation and as a nutrient supplement or a low cost biofertilizer for wheat crop.