Microalgal-based approach for wastewater treatment for thriving environment sustainability- A Review

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Abstract:

Over a period of time, pollution as well as changes in the atmosphere particularly due to industrialization has gained much attention. For the protection of the ecosystem, government authorities imposed strict guidelines on the discharge of wastewater in the streams. Industrial wastewater contains huge amounts of organic and inorganic waste compounds which acts as a massive source of pollution. Microalgal-based treatment of wastewater is an emerging biotechnological process with Advantages such as easy availability, handling as well their prolonged viability. However, culturing microalgae at an industrial scale has many challenges such as economic restrictions, etc. Microalgae treatment showed promising results for detoxifying the components of wastewater. Microalgae can easily grow and develop itself on wastewater and convert it into biomass which can be further used as biofertilizer, animal feed, etc. This review primarily focuses on recent microalgal studies for wastewater treatment, their mechanisms, potentials, benefits as well as challenges associated with the processes. . This study also discusses the recent co-cultivation techniques of culturing microalgae with other microorganisms to enhance the efficiency of the treatment process.

KEYWORDS- Microalgae, biofertilizer, wastewater , ecosystem, Animal feed

Introduction :

Due to fast industrialization and population growth, developing nations like India produce a staggering volume of wastewater each day.[1]By 2050, there will be a 20-30% increase in the amount of water used by agriculture, industry, and towns worldwide [2]. A deeper awareness of the magnitude of the issues ahead has been developed due to the rising volumes of produced wastewater and the need to provide fresh water to a rising population [3].

Due to its capacity to absorb large amounts of carbon dioxide during the process of producing oxygen and glucose through photosynthesis, microalgae-based treatment systems are among the most effective methods of dealing with environmental issues like worldwide warming, the growth of the ozone hole, and climate change [4]. The most intriguing and popular alternative biomass in use today in wastewater treatment applications is microalgae [5]. The many functional groups present in algae's cell walls allow pollutants to attach to the cell surface through a procedure termed biosorption [6]. This quick and reversible procedure can be used with either living or dead biomass because it is not dependent on the metabolism of the microalgae [7]. Assessment of algal growth potential is determined by the connection between the maximum biomass yield and the nutrients physiologically utilized for microalgal growth. Depending on the nutrients contained in the water, nutrients in a body of water may be eaten partially or entirely [8].

The level of industrialization, non-renewable water sources quality, the economic position in the region, as well as institution building, are just a few of the variables that influence the relative degree of quality in various Middle Eastern nations [9]. Concern over the long-term viability of conventional treatment systems for wastewater in terms of their economic viability and ecological consequences has grown in light of the need to achieve more rigorous effluent regulations to improve water quality. Energy utilization and emissions of greenhouse gases from treating wastewater are two elements that have emerged as crucial features of a treating wastewater system's overall effectiveness [10]. In order to safely dispose of or reuse the water, conventional wastewater treatment relies on consecutive aerobic and anaerobic processes that turn wastewater pollutants into benign chemicals. These conventional techniques remove enough carbon, nitrogen, and phosphorus, but at the cost of considerable energy use and nutritional loss. Urban wastewater up to 500 Mm3 per year is processed by Aqualia, the third-largest European wastewater treatment company. Data from Aqualia shows that conventional treatment of wastewater includes an average amount of energy consumed of 0.5 kWh/m3, costing 0.2 €/m3 - 50% of the cost subsequent to the energy consumption [11]. By utilizing nonarable land, microalgae-based biofuels, presently regarded as the third generation of biofuels, can significantly help to meet primary energy demands. Production of microalgae biofuel from brackish and wastewater does not interfere with the growth of food crops [12,13]. The potential uses of microalgae are highlighted by the combination of biofuel generation, biohydrogen synthesis, and wastewater treatment. With downstream processes accounting for 60% of the cost of producing biodiesel, they are becoming more and more important in the production of microalgal-based biofuels [14]. The growth and removal of microalgae biomass can be aided by increasing microalgae as biofilms rather than planktonic cells in solution, and developments in photobiology can lower the price of increasing microalgal species, especially as biofilms. Since it's possible that researchers in these fields are not aware of the possibilities for collaboration in the use of microalgae for wastewater treatment, higher communication is intended to encourage research. Although the creation of microalgal biofilms, utilizing internal luminance in microalgae photobioreactors, and microalgae's use in wastewater treatment are not novel concepts, what is new is a greater understanding of reuse contaminants from wastewater, rapid invention using many different renewable energy sources, new photobiological findings, decreasing LED costs, an increase in the use of robotic systems to drive the economy, and an increase in interest in development [15].

Several types of microalgae can efficiently remove pathogens, heavy metals, pesticides, phosphorus, nitrogen, and nitrogen-fixing nutrients from wastewater. They can also effectively remove pesticides. Utilizing them in individual cells or storing them there is the primary method of removing contaminants [16]. For the treatment of wastewater, numerous species of microalgae including *Phormidium, Chlamydomonas, Chlorella, Scenedesmus, Botryococcus,* and *Arthrospira* have been successfully developed, according to a number of studies [17]. Unicellular, non-motile, spherical green microalgae with a diameter of 2 to 10 m make up the genus Chlorella. Because of its high photosynthesis rate and high nutritional value, chlorella is nowadays the microalgae that are studied and grown the most extensively worldwide [18]. *Scenedesmus* green microalgae colonies are frequently found in groups of four or eight cells, arranged next to one another or inside the relatively similar mother wall. Scenedesmus is typically discovered in freshwater lakes and rivers, where it frequently displaces various types of microalgae [19]. Despite its rarity, a strain of the slow-growing alga *Botryococcus braunii* could be used to treat sewage and value biomass. The majority of the colonies that make it up this greenish freshwater microalga circulate on the water's surface in enormous groups. The ability of this algae to create substantial lipid and long-chain hydrocarbon amounts that can be used to produce biofuel is widely known.[20]

Composition of wastewater:

Many distinct sources, with a wide range in volume and strength, pollute waterways. The lifestyles and technology used in the producing civilization are reflected in the wastewater's composition.[21] It is a complex interaction of synthetic molecules, as well as naturally occurring both organic and inorganic elements. 75% of the organic matter in sewage is composed of lipids, proteins, amino acids, volatile acids, and carbohydrates. Large amounts of heavy metals, ammonium salts, calcium, potassium, magnesium, sulfur, phosphate, and bicarbonate are among the inorganic components. [22,23] as shown in Fig.1

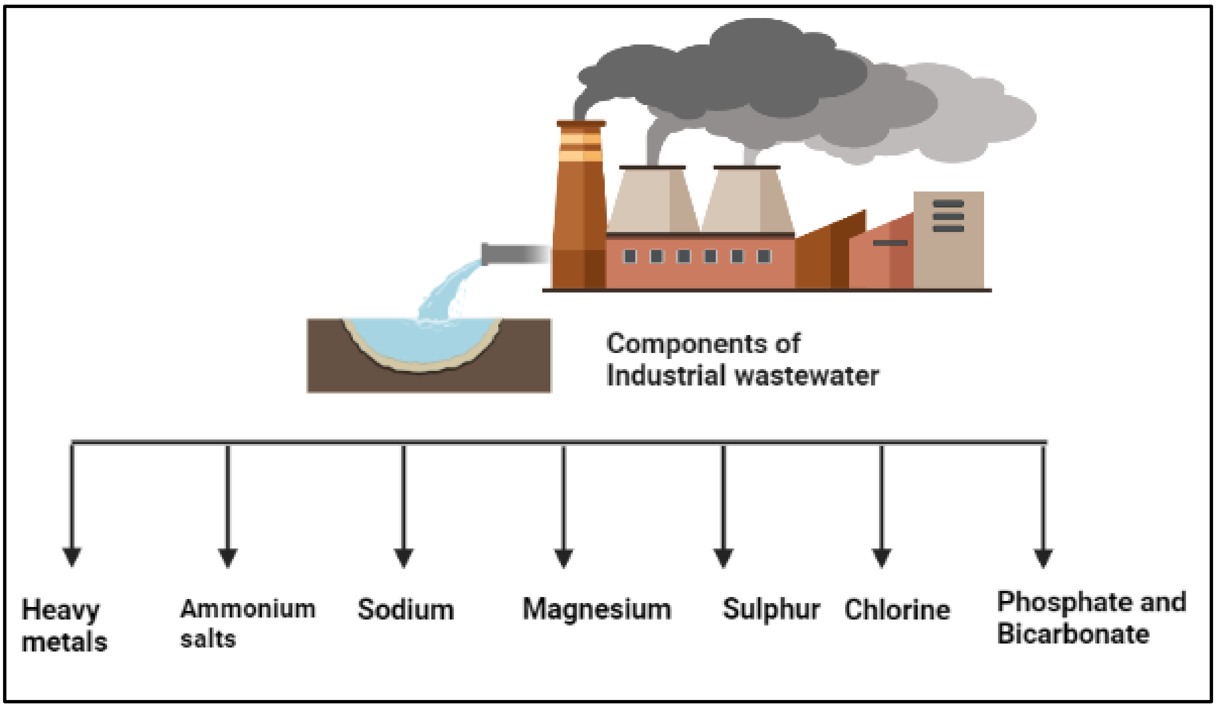

Figure1.- Components of the wastewater

Microalgae studies:

The numerous studies carried out to determine wastewater treatment utilising microalgae are discussed below.

According to Dalrymple et al(2013) .'s research, There are significant benefits to trying to integrate microalgal production technology with nutrition waste streams. While vital services like water purification can be significantly provided by these systems, the electricity produced by algae will play a crucial part in ensuring energy security. It also demonstrates that 94% ammonia, 89% TN, and 81% TP were eliminated by the end of the 14-day batch growth with the assistance of algae.[24]

Lau et al.(1996) investigated the capacity of Chlorella vulgaris to eliminate nutrients in published papers. They found that the data indicated that inorganic N and inorganic P were removed with an efficiency of 86% and 70%, respectively.In a previous study, Colak and Kaya (1988) observed that the treatment of industrial wastewater by algae eliminated phosphorus (85.7%) and nitrogen (50.2%), while household wastewater treated by algae eliminated phosphorus (97.8%) [25].

The successful use of algal cells to extract nutrients from water that is high in nitrogenous and phosphorus compounds has been demonstrated in numerous studies [26]. *Scenedesmus sp.* is fairly typical across all types of freshwaters, which Mohamed (1994) noted is important because it acts as a primary producer and helps to clean up eutrophic waters. According to the author, the presence or absence of specific Scenedesmus species can be utilized to assess water quality [27,28]. Harvesting or physically recovering the algal cells is also necessary to prevent the recirculation of nutrients in water sources and to collect the created biomass, and it is one of the most significant technological and financial challenges to be addressed [29].

Sekaran et al. (2013) investigated an integrated Bacillus species immobilized cell reactor and *Synechocystis species* algal reactor for the treatment of tannery effluent with a CAACO reactor. COD, BOD5, TOC, VFA, and sulfur dioxide were completely removed by the clarifier, chemo autotrophs immobilized cell reactor, and additional treatment of the effluent, respectively. In a *Synechocystis species* batch reactor for microalgae injection, the remaining organics in the treated tannery wastewater were further processed. The algal pond was capable of discharging treated wastewater that complied with the BOD5, 207 mg/l, COD, 16779 mg/l, and TOC, 7816 mg/l limits established by pollution control authorities. In the current experiment, the cumulative percentage removal of BOD5, COD, TOC, VFA, and sulfide was 98%, 95%, 93%, 86%, and 100%, respectively.[30]

One study evaluated the effectiveness of the sewage treatment facility using algae in Mysore, India. According to the study, the removal of 62 percent of total COD, 50 percent of filterable COD, 82 percent of total BOD, and 70 percent of filterable BOD resulted in moderate levels of treatment. The effectiveness of nitrogen removal was lower. However, a major decline in the suspended particles going to follow a greater euglenoid growth tends to suggest that algal consumption is going to remove particulate carbon [31]

The deletion of metal ions, phosphorus, nitrogen, and chemical oxygen demand from wastewater treatment by the development of green algae Chlorella species in a study was assessed by wang et al. (2010). The wastewater was sampled at four divided stages of the treatment process flow of a nearby municipal water treatment plant. The study found that for four distinct types of wastewaters, the typical specific growth rate during the exponential growth phase was 0.412, 0.429, 0.343, and 0.948 day1. It also found that the removal rates for NH4-N, phosphorus and COD were 74–82%, 83–90%, and 50–83.0%, respectively. Additionally, it was discovered that metal ions were effectively eliminated.[32]

The transformation of pollutants from wastewater, including heavy metals, nutrients, and other contaminants has been the subject of numerous studies and appears to be a sensible choice. Microalgae are thought to be a source of future energy generation and are successful at removing nutrients and CO2 from wastewater Microalgae can use the inorganic and organic nitrogen found in ammonia, nitrite, and nitrates, unlike traditional wastewater treatment methods. In order to eliminate inorganic N and P from wastewater treated by algae, there is no need to switch between different operational settings, which simplifies and saves energy during the treatment process. In fact, this technique is utilized to get rid of PPCPs and emerging pollutants such as heavy metals. To remove organic poisons, a variety of freshwater or marine microalgae can be used, especially in tropical climates. Another finding removed 30%–70% of PPCPs from home wastewater using green microalgae. As a result, the method of wastewater treatment has a number of encouraging qualities, such as the potential to photosynthesize and use less energy. There are few reviews of the studies that have been done to evaluate the efficiency of using microalgae to remediate wastewater. The study compiles the outcomes of a thorough literature search with the goal of critically analysing the utilization of microalgae in wastewater treatment, with an emphasis on current developments and potential roadblocks. An extensive explanation of the physiological and molecular mechanisms employed by microalgae to decrease nutrients (N, P, and C), heavy metals, and some other toxins in wastewater are provided after a brief description of the fundamental properties of wastewater. This study discusses a wide range of algal efficiency criteria from various wastewater sources, as well as the drawbacks of microalgae-based wastewaters [33].

Cyanidium caldarium, another name for *Galdieria sulphuraria,* is one of the most intriguing microalgae with extremophilic growth capabilities. According to a 1995 study by Gross and *Schnarrenberger,* this species of red algae (Rhodophyta) can grow both heterotrophically and mixotrophically on distinct sugars and alcohols.[34] *Galdieria sulphuraria* can acidify its environment through energetic proton efflux, which lowers the expense of pH regulation and the risk of complications. It can flourish in conditions that are both neutral and highly acidic, down to pH 1.8. [35]

Galdieria sulphuraria not only grows in an acidic environment but also grows thermophilically up to 56°C [36,37]. The phycobiliprotein phycocyanin, which is abundant in *Galdieria sulphuraria* and is increasingly considered a natural colorant the food business, cosmetics sector, and as a fluorescent identification in molecular biology, increases its economic significance [38,39]. Microalgal approach is shown in Figure 2.

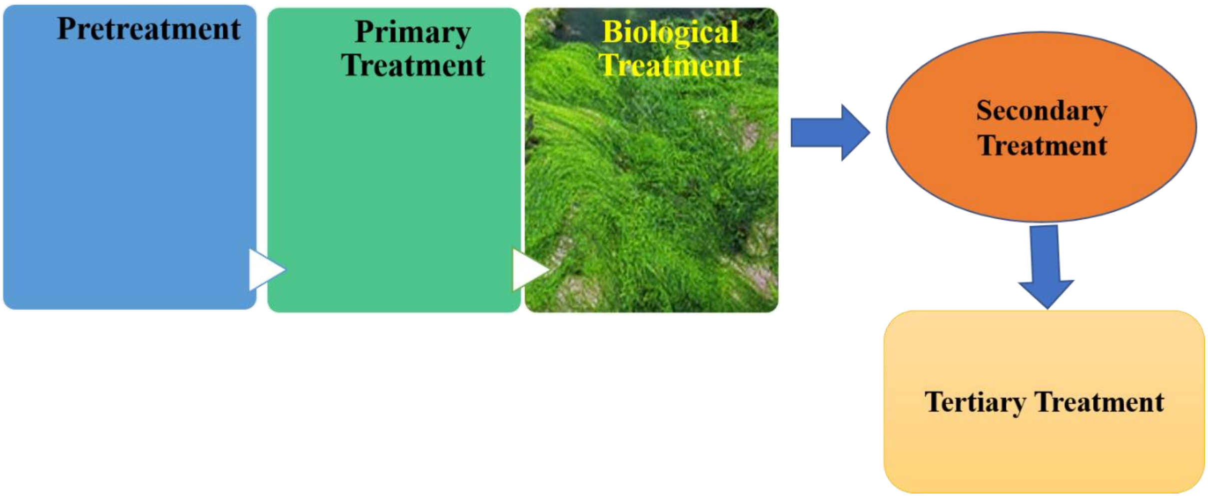

Figure-2 Microalgal approach of wastewater treatment

Due to its diverse metabolic profile and proficiency in creating products with value, *Galdieria. sulphuraria* is a very intriguing potential treatment of high chemical oxygen demand-loaded, , high-temperature wastewater. Phycocyanin [40]. Sloth et al. have shown that *Galdieria sulphuraria can* grow heterotrophically on protein hydrolysate uneaten from bakeries and restaurants [41]. According to Lammers and colleagues, *Galdieria sulphuraria* was able to flourish in primary-settled wastewater in a first field study while drastically reducing the amount of organic carbon (46-72%), ammonium (NH4-N) (63-89%), and phosphate (PO4) (71-95%). Chlorophyta contains the greatest number of promising acidophilic microalgal strains [42].

Chlamydomonas acidophilus has been discovered to grow in acidic waterways with pH between 1.7 and 3.1. It has been shown that *Chlamydomonas acidophilus* can expand mixotrophically at pH 2.5 without the need for CO2 by using various carbon sources, especially glucose, glycerol, and starch [43]. Its ability to eliminate NH4 has also been demonstrated [44].

Chlamydomonas acidophilus biomass derived from waste sources has added value due to its capacity to absorb large levels of antioxidants, along with the pigment lutein [45].

Chlorella protothecoides var. acidic-ola was discovered in acidic mine water and demonstrated strong heterotrophic expansion on glycolic acid, which is a portion of the wastewater pack of the fruit and vegetable manufacturing industry sectors.[46]. *Chlorella sorokiniana*, a well-examined thermophilic green microalga, has shown higher photoautotrophic rates of development up to 43°C [47]. It is a necessary prerequisite for several WWT processes that *C. sorokiniana* cultures cultivated heterotrophically, as demonstrated by kim et al. 2013, have effective Phosphorus and Nitrogen removal rates.[48] According to a subsequent investigation, heterotrophic *C. sorokiniana* cultures exhibit superior removal behavior in comparison to photo- and mixotrophic cultures. It is the ideal source for animal feed or the creation of biofuel because the sustainable sources bof iomass from *C. sorokiniana* can accumulate significant amounts of beneficial bioproducts. The co-immobilization of *C. sorokiniana* with the bacterium *Azospirillum brasilense,* which promotes microalgal development, considerably increased the P-removal effectiveness of *C. sorokiniana*.[49,50]. Psychrophilic species, like *Koliella antarctica*, which may have temperature optimums below 10°C, represent an intriguing potential biological process for having to clean wastewater from fruit processing facilities. Furthermore, it has been discovered that *Koliella antarctica* produces substantial amounts of DHA, EPA, astaxanthin, and lutein.[51]

Microalgal mechanism to detoxify wastewater:

Utilizing microalgae to clean up the environment uses a variety of mechanisms, including biosorption, bioaccumulation, and biodegradation.

- **Biosorption:**

The sorbent in every passive biosorption process is a biological substance that can attach to and focus water pollution completely. In essence, biosorption involves the mass transfer method of removing a substance from the liquid form and start binding it to the surface of the solid first. Several different mechanisms, including precipitation, surface complexation, electrostatic interaction, ion exchange, and absorption, encourage the physiological, chemical, and metabolic functions that constitute biosorption.[52] Target sorbate that is solubilized in water is needed for the mechanisms that underlie this phenomenon, along with a biosorbent (solid phase sorbent). Living or dead microorganisms, as well as their parts, maybe the biomaterial involved. The motivation behind the admiration of sorbate species is the equally huge potential of the biosorbent towards the desired sorbate, and the overall output is responsible for the number of sorbate molecules [53,54]. Until an equilibrium has been reached in relation to the substance that has been adsorbed by the biosorbent and its residual proportion in the liquid, the process is repeated. Additionally, a sorbate's distribution between the phases of solid and liquid depends on the biosorbent's level of affinity for that particular sorbate [55].

When radionucleotides and high-level metals (HMs) released from a nuclear power plant were largely focused on microalgae in the early 1970s, biosorption had first been noticed in a number of these organisms. Microalgae's cell wall, which is directly responsible for biosorption, plays a crucial role in the procedure and helps determine the mechanism through which this occurrence happens. Additionally, the pores and surface charge of microalgal surfaces aid in biosorption. The carboxyl, hydroxyl, and sulfate chemical groups found in the cell walls of microalgae serve as efficient ion exchangers and binding affinity to facilitate the complexity of metallic ions as well as the adsorption of organically contaminated water [56]. However, they are mostly present in the plasma membrane and cytosol and have the capacity to bind to metal cations in their multiple operational groups. Other molecules such as lipids, proteins, and nucleic acids can also be collected on the surface of the cell [57]. The intercellular spaces found on the cell wall, combined with the fibril matrix that generally gives microalgae's cell walls their high mechanical strength, and the amorphous fraction that gives them their flexibility, may all help to improve the biosorption process [58].

The removal efficiency of metal cations covered in an aqueous medium may well be decreased as a result to active site repulsion among microalgae surfaces, so having more biomass is not always better means having better biosorption properties. Additionally, as there are more biosorbent particles present, there are more active sites for biosorption [59]. A two-step mechanism is used by microalgae to bio sorb HMs ions. Following a procedure independent of metabolism that variety of experiences and changeable the adsorbate's connection to live sites located on the microalgae's skin, a slow stage that actually refers to favorablee intracellular diffusion and probably includes the occurrence of microalgal metabolism. In addition to cellulose, alginates, lipids, and proteins, microalgal cell walls may also constitute polymeric substances such as exopolysaccharides with uronic groups and peptides. These compounds offer a variety of functional groups, which include hydroxyl, phosphate (PO43), sulfonate, carboxyl, and thiol. Because they have numerous deprotonated carboxyl and sulfate groups, monomeric alcohols that promote biosorption, and a large number of cationic and anionic HM species, microalgae are able to capture both types of HMs [60].

- **Bioaccumulation**

As compared to biosorption, which is an inactive metabolic process, bioaccumulation utilizes a wide range of substances in the cell lumen. Compared to biosorption, bioaccumulation implies more energy and moves more slowly. Bioaccumulation is a technique for using living cells to detoxify wastes; these cells possibly take substances and either accumulate them or metabolized. Sulfates, phosphates, HMs, nitrate, and pesticides are examples of organic and inorganic pollutants that can enter cells, and this procedure is thought to be crucial for removing them.[61] It is challenging to determine the pollutants which are absorbed and bioaccumulated because of two modes are constantly evolving, despite the fact that the mechanisms involved in bioaccumulation or biosorption are very various procedures. Various pollutants, as well as nutrients and other small elements, are accumulated by microalgae [62]. Because they can adapt to their environment, Low concentrations of pollutants are not harmful to microalgae. Furthermore, microalgae are highly resistant to a range of toxic elements from the domestic, agricultural, and industrial sectors, which improves their capacity for bioremediation [63].

The quantity and concentration of a micropollutant adsorbent in a medium, or bioconcentration factor, is used to quantify bioaccumulation [64]. The step that comes before bioaccumulation is bioabsorption. But not all adsorbed molecules can result in bioaccumulation [65]. Spirogyra, for illustration, can absorb 850,000 times extra radio-phosphorus than the nearby water [66]. Physicochemical factors, including pH, temperature, contact duration, and the number of pollutants present, have a significant impact on bioaccumulation regardless of the mechanism [67].

- **Biodegradation**

The biodegradation process, wherein the complex compounds break down into safe and straightforward chemical building blocks, is one of the most efficient ways to remove pollutants from effluents. In contrast to bioavailability and biosorption, which depend on microorganisms trying to act as biological filtration to gather contaminants and remove them from the water's surrounding surface, Bioremediation includes the break-down of target pollutants either by fully mineralizing the parent molecules to H_2O and CO_2 or through biotransformation, which includes a variety of enzymatic reactions to yield new metabolic intermediates [68].

The two main types of degradable are (i) co-metabolism, where pollutants serve as an electron donor and a source of carbon for non-living aspects, and (ii) metabolic degradation, where pollutants serve as both a carbon source and an electron donor/acceptor for microalgae [69]. When microalgae are involved in the biodegradation process, the extracellular breakdown is chosen as the next step, followed by subcellular decomposition of the break-down intermediates. Both processes may occur simultaneously [70]. Organic pollutant degradation by microalgae involves three distinct stages of intricate enzymatic reactions. Cytochrome P450 enzymes, such as hydroxylase, decarboxylase, carboxylase, and monooxygenase, are needed for stage I. The enzyme’s objective is to increase or decrease the number of hydroxyl groups in the pollutant through oxidation-reduction reactions [71]. As the cell enters stage II, the addition of enzymes such as glutathione-S-transferases and glucosyltransferases can facilitate glutathione's conjugation with a variety of compounds that have electrophilic centres, trying to prevent oxidative cell damage [72]. Dehydrogenases, glutamyl-tRNA reductases, carboxylases, mono(di)oxygenases, laccases, transferases, hydrolases, pyrophosphatases, and dehydratases are a few of the enzymes used in stage III of detoxification to bio transform molecules into nontoxic intermediates [73].

Microalgae's Potential in Wastewater Treatment

- **Cost-Effectiveness**

It costs less to control microalgae production in wastewater than to treat it conventionally. Brewery wastewater contains organic loads that are conducive to microalgae growth making it a very alluring option for inexpensive and environmentally responsible wastewater treatment. Numerous types of microalgae can take nutrients from sewage and absorb them. This process has a lower initial cost when compared to conventional wastewater treatment methods [74],[75] as shown in Figure 3.

- **Low Energy consumption**

Microalgae produce oxygen as a result of wastewater treatment, which aerobic organisms use to further obliterate naturally occurring organic loads. When compared with the amount of energy stored required for aeration throughout conventional wastewater treatment processes, this lowers the energy cost. For the activated sludge procedure to remove 1 kg of biological oxygen demand, 1 kWh of electricity is required. As a result of the power generation in this process, 1 kg of fossil co 2 is produced. To start removing 1 kg of BOD from a sample of wastewater and generate 1 kWh of electricity through the development of methane by the algal biomass, microalgae need no energy input [76].

- **Sludge Formation Reductions**

Sludge reduction or elimination is the main objective of every wastewater treatment facility. Traditional wastewater treatment is characterised by the utilisation of extremely high levels of chemicals. Significant chemical use may lead to the advancement of sludge. Due to this, hazardous solid waste must be generated and discarded of in the surroundings. Chemical additives are not necessary for microalgae wastewater treatment, and sludge builds up as algal biomass [77].

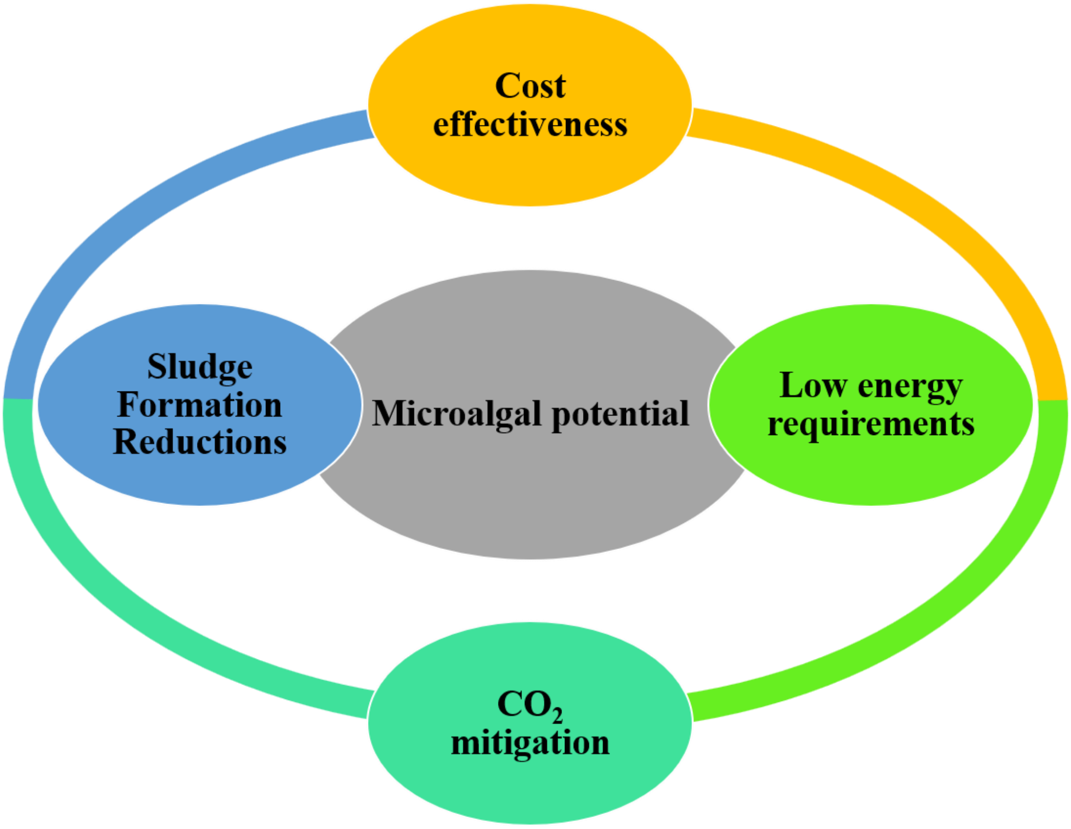

Figure 3 -Microalgal potential for wastewater

- **Emissions of Greenhouse Gases**

Around the world, there is a lot of concern about global warming. Wang et al. state that there are two strategic approaches to CO_2 mitigation [78], chemically and biologically. Separation, transport, and sequestration are all aspects of chemical approaches. These methods are costly and energy-intensive, so there is a requirement for longer-term, more affordable solutions to the threat [79].

There are estimated 2,000,000 types of microscopic algae that can be utilized for CO_2 sequestration, and they represent one of the developing varieties of biological to CO_2 mitigation. The potential for CO_2 fixation by photoautotrophic algal cultures to lower atmospheric CO_2 levels. 183 tonnes of CO_2 are fixed by microalgae to yield 100 tonnes of biomass. Microalgae have a superior capacity for absorbing solar energy, which allows them to grow more quickly than other terrestrial plants. Li et al. found that traditional forestry, agro-industrial, and aquatic plants cannot match the rates of development and CO_2 fixation of microalgae [80],[81].

Furthermore, microalgae involve carbon dioxide to grow and can be grown with any source of CO_2. Although using air eliminates the necessity for transportation, it may not contain enough CO_2 (0.04 w%) to support the growth of microalgae. pure CO_2 usage may also be extremely expensive. An adequate supply of CO_2 must be provided to ensure ideal algae growth. Industrial activity's CO_2 flue gas) can be applied to deal with these issues. Flue gas generally includes significant Carbon dioxide but the amount varies based on the origin and the methodology {82],[83]. For instance, CO_2 concentrations are relatively lower in exhaust gas from thermal power plants than in flue gas from natural gas-fired power plants. However, by placing photobioreactors close to the industries that produce flue gas, one can prepare to cope with the difficulties associated with utilizing CO_2 from industries, including in transporting exhaust gas to the treatment site.[84]. High-rate algal pond (HRAP) systems use sunlight, bacteria, and photosynthesis to start reducing 100 to 200 tonnes of carbon dioxide per ML of sewage treatment when opposite to electromechanical treatments in a standard oxidation pond.

Furthermore, nitrogen assimilation by algae could save 100-200 tons of Carbon dioxide per ML. As a result, biological CO_2 reduction can be achieved through microalgae-based wastewater treatment or its integration into some other wastewater treatment facilities. This method is cost-effective, economical, and environmentally friendly [85],[86].

Benefits of Wastewater Treatment Using Microalgae

- **Organic and inorganic fertilisers**

Through increasing and decreasing pH and soil acidity, excessive inorganic fertilizer application affects soil fertility. By increasing and decreasing soil pH and acidity, respectively, extreme use of inorganic fertilizers changes soil fertility. Microalgae are useful in the agro-industrial. The wastewater's derived algal biomass could be used to make fertilizers for plants. These fertilizers work to enhance the soil's mineral makeup and water-retentive capacity. Hasyim et al. claim that irrigation with raw wastewater samples as a fertilizer application source causes poor soil quality, low production, and poor plant growth [87].

- **Animal feed**

Because of their nutritional content and ease of digestion, microalgae are utilized as live feed in the aquaculture industry. Microalgae are made up of 39-71% protein and 10-57% carbohydrates, primarily polysaccharides starches, and cellulose [88]. Direct or indirect feeding of bivalve, oyster, and shrimp larvae is possible with harvested biomass. Additionally, zooplankton, which is fed to fish, can be improved by microalgae [89]. Phang et al. found that Spirulina biomass components could be used as better animal feed when sago wastewater was treated using an HRAP. The yellow hue of broiler skin and egg yolk can be enhanced by using microalgae biomass in poultry feed as an incomplete replacement for traditional proteins and carotenoids. According to several studies, animal feed should contain microalgae biomass made from treated wastewater. But it hasn't gotten much attention because of public opinion and requirements for acceptable food standards on animal feeds [90].

- **Biofuel Manufacturing**

  Biofuels are defined as substances made from biomass that have an intense temperature of combustion value, such as biohydrogen, biodiesel, ethanol, or bio methanol [91],[92]. The world's need for energy is growing. fossil fuels are responsible for almost 80% of all energy used worldwide [93]. However, the use and extraction of fossil energy tend to increase the number of greenhouse gases released into the atmosphere, which causes global warming [94]. Recent research has represented alternative sources of energy because of the drawbacks of using fossil fuels. Biofuels are thought to be a promising alternate power source and have drawn a lot of attention. Considered to be the most promising source of biofuel is microalgae. This is a result of their quick development and high photosynthesis process effectiveness [95]. Concerns about the lack of arable land, which could cause a global food shortage, were raised by first-generation biofuels. Recently, oil crops like palm, soya, and rapeseed have become more popular, but their planting requires a lot of arable lands. Second-generation biofuels are made from *Jatropha curcas,* but because of its slow growth rate and the high demand for arable land, it is not economically viable. As opposed to other bio – fuels sources (conventional crops), such as soya and oil palm, microalgae biofuel is asserted to produce 10–100 times the most fuel per unit surface area [96].

Co-cultivation of microalgae for the treatment of wastewater

Recently, microalgae have been widely integrated with activated sludge, bacteria, fungi, and nanoparticles to benefit from a co-culture system during bioremediation. The following co-culture systems will be discussed as shown in Figure 4.

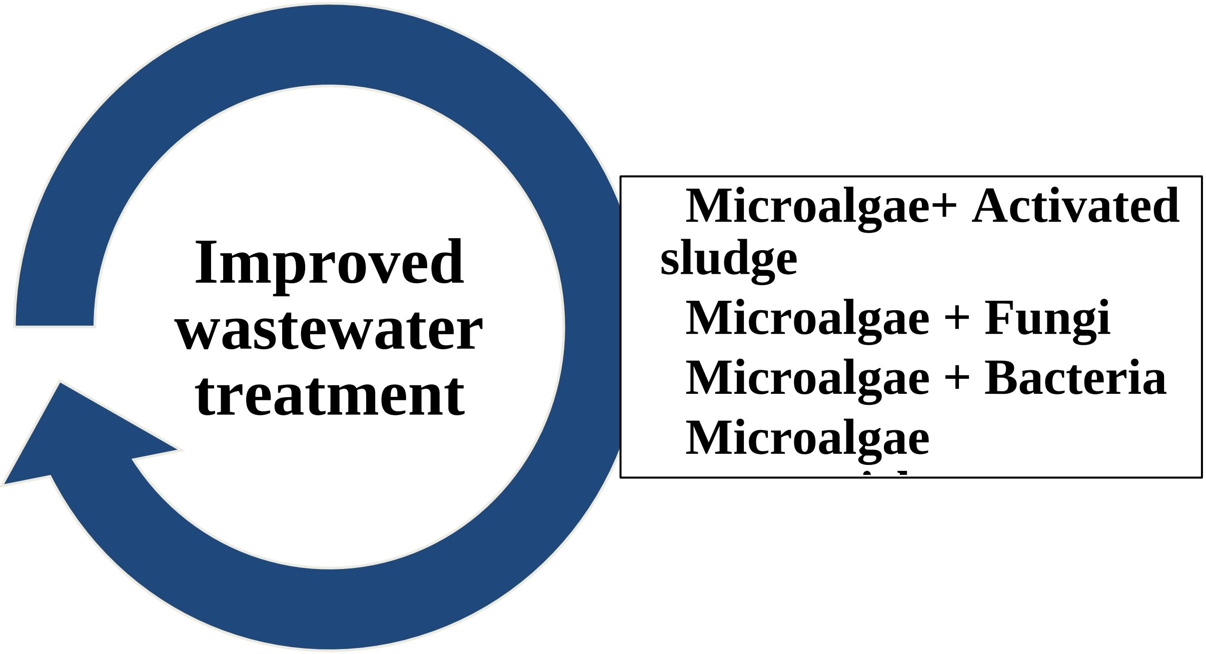

Figure 4 -Enhancement of microalgal potential for wastewater treatment

1. Microalgae-Bacteria:

When microalgae fix carbon through photosynthesis, they can begin to release organic oxygen molecules, which heterotrophic bacteria then break down into CO_2, which is required as a reagent in the photosynthetic process as shown in figure 5. The production and removal of biomass from wastewater can be made more effective by taking advantage of this synergistic relationship [97]. To increase the uptake of nutrients, microalgae were applied to a sequencing batch bioreactor. The findings demonstrated that TN removal could be increased by microalgal-bacterial symbiosis from 38.5 percentage points to 65.8% and TP removal from 31.9% to 89.3% [98]. Co-cultivating Chlorella with wastewater-borne bacteria can speed up the eradication rate of COD, TN, and TP from vinegar processing wastewater by 22.1%, 20.0%, and 18.1%, respectively, when compared to a microalgae-only system [99]. Co-cultivating the microalgae *C. sorokiniana* or bacteria *Pusillimonas* from anaerobic digester effluent can lower TN, NH4+, as well as COD by 34-67%, 65-97%, and 14-60%, respectively [100].

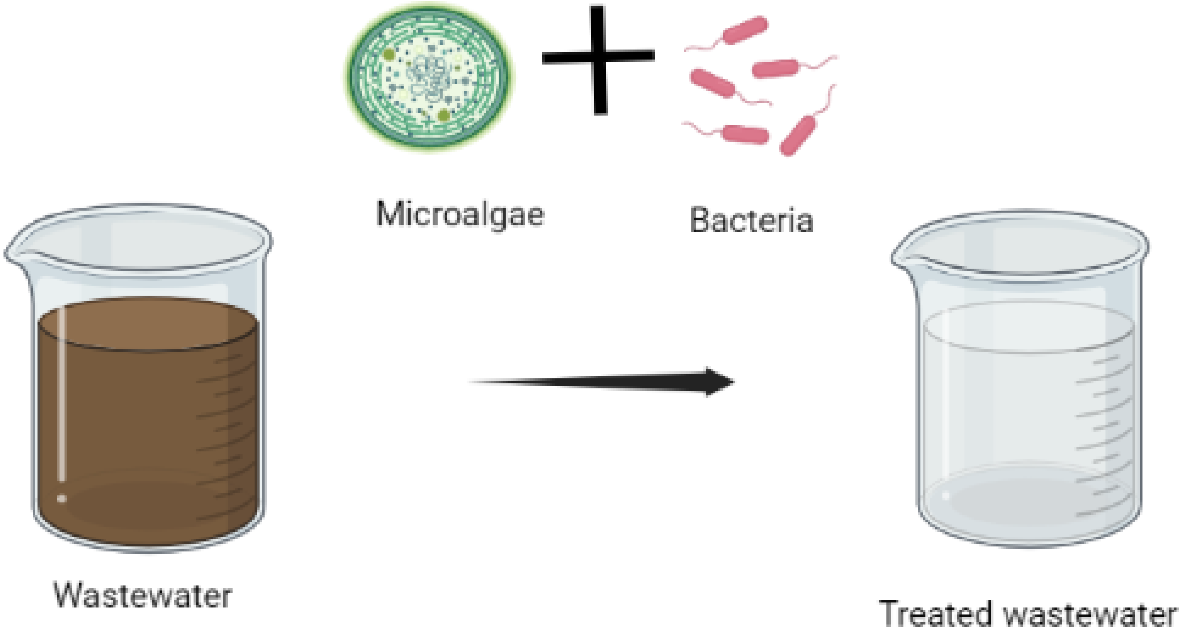

Figure 5: Microalgal-bacteria coculture for improved wastewater treatment

2.Microalgae activated sludge:

The advantages of the microalgae-activated sludge association in wastewater treatment over conventional mono-systems stem from the enhancement of COD removal by the microalgae and the assimilation of nutrients by the microalgae [101]. The ratio of microalgae to sludge depends on the composition and properties of wastewater. At a low sludge in municipal wastewater, microalgae growth doubled compared to pure microalgal culture, and more nutrient removal was completed [102]. In a study, Mujtaba et al. was using a co-culture of suspension-activated sludge or immobilized *Chlorella vulgaris* in a single reactor and discovered that as the inoculum ratio of activated sludge reduced, the removal of nutrients increased, eventually reaching the highest valuation of 0.5% with Nitrogen and Phosphorus removal services of 99.8% and 100%, respectively, within 24 hours [103]. However, at inoculum ratios between 90 and 95 percent COD removal, the variations in carbon matter degradation were insignificant. An activated sludge ratio of 3:1 was selected in a photobioreactor mixed culture to achieve effective organic and nutrient removal [104].

3.Microalgal fungi:

Numerous studies have also looked into the symbiotic relationship between fungi and microalgae and how it affects WW bioremediation. Microalgae generating O2 for biomass synthesis refers to the synergistic interaction between fungi and yeast. Then, as a result of using oxygen for cellular respiration, yeast releases CO_2 essential for microalgal photosynthesis [105]. Walls et al. co-culture algal cells with glucose-supplemented yeast to produce high levels of biomass and remove nutrients quickly [106]. In three days, the heterotrophic cultivation excluded 91% and 94% orthophosphate, 93 percentage points and 97 percent of the total number of nitrates, and 93% and 95% of total NH4+-N using 10 and 20 g L-1 glucose, respectively. It also produced 1.85 and 2.74 g L-1 of biomass concentration [107].

A hybrid culture of *Chlorella vulgaris* and *Aspergillus species* in molasses WW can begin by removing 70.7% COD, 67.1% TN, 94.7% NH4+-N, and 88.4% TP, as opposed to 26%, 44.4%, 79%, and 33%, respectively, when compared to mono-microalgae and mono-fungi.[108] Co-culturing Chlorella species and Penicillium species can yield 46.13% COD, 13% TN, 6% NH4+-N, and 88.4% TP from the by-product of the hydrolysis of wet biomass. In order to remove nutrients from WW, the co-cultivation system performed best than a mono-system of microalgae [109].

4.Microalgae nanoparticles:

Due to their many advantages of having a high specific surface area, pore volume, and bioactivity, qualified nanofibers are chosen over the other conventional elements for the immobilization as well as accumulation of microorganisms, which include microalgae, to create hybrid materials that integrate biology with higher effectiveness of removing pollutants, simplification of use and recyclability [110]. Vasistha et al. combined microalgae and ZnO nanoparticles, and they evaluated the outcomes on the basis of leads to a reduction in nutrients from primary and secondary treated sewage wastewater. Co-cultured bio nanofiber removed with success capacities of 97.5%, 87.20 %, and 82.21% % for TOC, TN, and TP from both primary and secondary treated sewage wastewater, and 95.3%, 85.2%, 81.5%, respectively.[111] Luo et al. were using nano-TiO2 in a microalgae-TiO2 combining system to enhance the usage of microalgae to produce humic acid pig farm biogas slurry, ultimately trying to remove about 50% of the humic acid [112].

Treatment issues with microalgae and future directions for research:

- Extensive outdoor microalgae biomass cultivation typically produces between 15 and 30 g/m2/day of biomass on average. On the basis of the 4-6% nitrogen of microalgal cellular tissue, the rate of microalgal nitrogen uptake could range between 0.6 and 1.8 g N/m2/day. As a result, the nitrogen removal rate for a pond 20 cm deep would be 3.0-9 mg N/L/day [113],[114]. The rate of nitrogen removal from a commonly used activated sludge procedure, however, is about 30-78 mg N/L/day, which is several times greater than the nitrogen removal rate from microalgae [115].
- Lack of an external source of CO2 would inhibit the rate of microalgae biomass production, nutrient recycling, and wastewater treatment effectiveness. The entire procedure would be enhanced by having a CO2 point source (like flue gas) close to the cultivation site [116].

- Even though a particular contaminant may be removed by a microalga very effectively, exposing it suddenly to wastewater that contains that contaminant in very high levels, may be dangerous to its culture and lessen the efficacy of the testing process. Therefore, a regional procedure must be created to aid the strain's adaptation to the desired compound [117].
- There are times when wastewater contains extremely high levels of ammonia, which can be poisonous to some strains of microalgae. Some wastewaters may contain nitrogen as nitro, amino, or other groups in aromatic compounds. For effective microalgal bioremediation, wastewater's pH value should be adjusted if it is either extremely low or extremely high. However, the cost and required energy for every unit of separating biomass could be extremely high, trying to make the bioremediation using microalgae procedure as a whole economically unviable. Wastewater may have low concentrations of nitrogen and/or phosphorus, resulting in a lower biomass density [118].
- • It is very uncommon to observe a single microorganism entirely degrade a fragrant contaminant of xenobiotic pollutants. The use of microalgae in combination with other suitable microorganisms could increase the effectiveness of removal of such undesirable contaminants from wastewater [119].
- The amount of water lost through evaporation from an open cultivation system can range from 0.1-2 cm per day, depending on the climate. The efficiency of bioremediation would ultimately be impacted by the concentrations of pollutants in the culture that could arise due to water loss from evaporation. As required, the previous batch's treated wastewater could be used to make up for the water lost through evaporation [120].

- If coagulants are used to divide the microalgae biomass, the expense of bioremediation will increase. The step of harvesting biomass could be eliminated by attached cell growth, but creating the immobilization matrix could be very expensive. the ideal situations for growth should be used to promote auto-flocculation of the biomass [121].

Conclusions

This article has shown the potentiality of microalgal treatment methods. Thus, this technique is cost-effective, reliable as well as eco-friendly. The biggest outcome is biomass which is an immense source of biofuel in the future which is discussed in the above article. Co-cultivation of microalgae with other species also showed promising results and enhancement in the wastewater treatment process. However, this review also discusses the challenges of the process which need attention. Close investigation and integration of the process are required to achieve the desired results. Future findings should work on more diverse microalgal strains which can easily cope with other species, and adapt to process conditions to protect environment.

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