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Tertiary Treatment of Wastewater with Chlorella Vulgaris- A Literature Review

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Abstract: *Tertiary treatment of effluent is necessary process if this effluent is reused for various beneficial domestic purposes. The different tertiary treatment processes have many drawbacks like excessive generation of chemical and biological sludge and need of organic substrate for nitrification and de-nitrification processes. Open oxidation ponds are also used widely but due to enormous land requirement and it dependence on temperature variation and the availability of light this method is not very economical. As with the advancement in illumination technology removal of nutrients in algal photo bioreactor has become a feasible option. Domestic waste water is appropriate medium for algal growth. The most important factors for successful effluent with microalgae are light, pH, mixing and Hydraulic retention time*
Keywords: *light, microalgae, mixing, sludge*

I. Introduction

Even after secondary treatment, domestic sewage contains high concentrations of nutrients, i.e., nitrogen and phosphorus. Release of water containing high levels of nutrients to natural water bodies is undesirable due to the eutrophication problem. Tertiary treatment of domestic sewage is required for the removal of nutrients, thus rendering the treated effluent suitable for discharge into natural environment. Further, tertiary treatment of sewage is also essential if the treated effluent is to be reused and/or recycled for various beneficial uses.

Conventional tertiary treatment processes for nutrient removal, i.e., nitrification-denitrification for nitrogen removal and chemical precipitation for phosphorus removal, have certain drawbacks including the requirement of organic substrate for denitrification and excessive production of both biological and chemical sludge. Tertiary treatment of domestic sewage for nutrients removal is also possible in open oxidation ponds. However, the main drawbacks of such ponds are the excessive land requirement, and dependence of the system performance on temperature fluctuations and the availability of sunlight.

Due to recent advances in illumination technology, light can now be provided efficiently and cheaply using CFL and LED lamps. This has potentially made the use of artificially illuminated algal-bacterial photo bioreactors a feasible option. Thus, it now conceivable that secondary treated domestic sewage may be further treated in algal-bacterial photo bioreactors for simultaneous and comprehensive organic carbon and nutrients removal. Nutrients will be removed in such reactors primarily by assimilation by algal cells, while residual organic carbon shall be removed by bacterial oxidation. A symbiotic relationship between algae and bacteria is envisaged in such reactors, where oxygen produced through algal photosynthesis will be used by bacteria for respiration, while the inorganic carbon produced through bacterial carbon oxidation will be used by algae for photosynthesis.

However, before such reactors are technically feasible, the extent of nutrient uptake through algal assimilation in photo bioreactors must be studied in detail. The questions which require answering include the extent of nutrients removal possible in photo bioreactors at various hydraulic retention times (HRT), the impact of illumination intensity and illumination strategy on system performance, the optimal algal biomass concentration to be maintained in the reactor and the impacts of biomass recycling, effects of nutrient limitations on algal growth, optimal nutrient ratios, etc. on reactor performance. The experiments described in this dissertation have been designed to answer some of the above questions so that technical feasibility of algal photo bioreactors can be assessed scientifically.

II. Literature Review

Wastewater and sewage management has been studied extensively over a long time. While earlier this was a necessity from the view of public health, in modern times the subject has been broadened as a tool to reduce pollution as the effects of man-made pollution have become clearer. Figure 2.1 below (Asano et al. 1985) shows a typical wastewater treatment process.

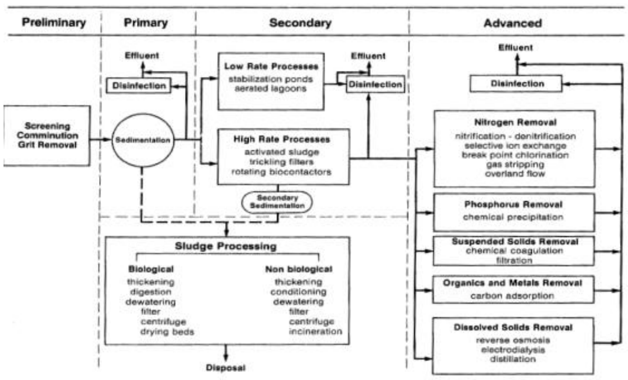

Fig 2.1 Typical wastewater treatment processes in different stages (Asano et al., 1985)

As seen from the figure, the primary treatment involves separation of the suspended matter through physical methods such as sedimentation. The secondary treatment uses techniques such as aeration and chemical methods to oxidize the organic matter present in the wastewater. The effluent coming out of the secondary stage carries large amounts of inorganic nitrogen and phosphorus as well as some heavy metals which are ultimately discharged to large water bodies (Nöie et. al., 1992).

When the effluent from secondary treatment is discharged directly to the water bodies, it increases the level of nutrients present in water to very high levels as compared to their natural state. It has been shown that effluents from urban, agricultural or industrial sourceshave a total nitrogen concentration up to three orders of magnitude higher (Nöie et al. 1992) than natural levels. The same holds true for total phosphorus. This increased level of nutrients give rise to high growth of phyto-planktons and algae in the water bodies, which is also referred to as eutrophication. US National Academy of Sciences (1969) defined eutrophication as, "..... eutrophication refers to natural or artificial addition of nutrients to bodies of water and to the effects of the added nutrients. When the effects are undesirable, eutrophication may be considered a form of pollution........".

The problem with eutrophication lies in the imbalance it creates in the marine and freshwater ecosystems. This imbalance starts with the increased growth of certain species like phyto-planktons and algae in the ecosystem. Ultimate decomposition of algae results in additional oxygen demand, which is stressful for other fauna and flora living in these ecosystems, (McGriff and McKinney, 1972)

In freshwater ecosystems nitrogen pollution has been shown to cause the acidification of water bodies and development of primary products which ultimately cause eutrophication and toxic conditions which impair the ability of aquatic animals to survive, grow and reproduce. Drinking of such polluted water has been linked to many health problems in humans (Camargo and Alonso, 2006). Nitrogen pollution in marine ecosystems has been shown to cause increase in plant production, discoloration, algae blooms and ecosystem-scale oxygen depletion, which can cause the loss of habitats (Paerl and Piehler, 2008).

2.1 Conventional Nutrient Removal Processes

2.1.1 Nitrogen removal process

The methods available for nitrogen removal can be classified into two categories, i.e., bio-chemical treatment processes and physical/chemical treatment processes. Some physical/chemical processes for nitrogen removal are,

(i) Breakpoint Chlorination: Addition of chlorine to the wastewater which oxidises ammonia and nitrogen in the wastewater to nitrogen gas.
(ii) Selective ion exchange: Using zeolite ion-exchanger beds which have selectivity towards ammonia.
(iii) Air-Stripping ammonia: Increasing pH of wastewater to 11.5 which converts the ammonium ion into ammonia gas form which is then removed by air contact.

Bio-chemical treatment processes for nitrogen removal consist of nitrification and denitrification of the wastewater. Nitrogen in fresh sewage is largely in the form of ammonium ion or urea (from which ammonia is derived) and some nitrogen is in the form of more complex organic materials such as proteins and amino acids (Sawyer and McCarty, 1967). In the first step, i.e., the nitrification process, aerobic oxidation of urea/ammonia to nitrate is done by bacteria Nitrosomonas and Nitrobacter. In the second step, i.e., denitrification, heterotrophic and autotrophic bacteria like Pseudomonas convert nitrate ions to nitrogen gas (Reeves 1972). Denitrification process can only take place in the anoxic conditions.

2.1.2Phosphorus Removal Process

Phosphorus is present in sewage in the form of orthophosphate, complex polyphosphate, and organic phosphate. All of the polyphosphates gradually hydrolyze in aqueous solution and revert to orthophosphate form.Chemical removal is a common technology used for phosphorous removal from the wastewater. Addition of lime and coagulants like alum and ferric chloride in water result in the precipitation of metal hydroxide precipitates, which incorporate the dissolved orthophosphate ions into the precipitate matrix. The additional sludge production during phosphorus removal can be a major concern (Tchobanoglous et al., 2003).

2.2 Microalgae for Tertiary Treatment

Microalgae has long been studied and used for nutrient removal. There have been many studies suggesting the use of algal cultivation as tertiary treatment process, i.e., removal of nutrients from water (Przytocka-Jusiak et al., 1984; Rodrigues & Oliveira, 1987; Noüe&Eidhin, 1988).

Algal growth results uptake of nitrogen and phosphorus from wastewater for assimilation into the algal biomass (Oswald, 1988b; 1988c; Richmond, 1986). Due to the high pH prevalent in algal systems, nutrients also removed in such systems by ammonia volatilisation and phosphorous precipitation. Microalgae systems thus provide a means for nutrient removal from waterand result in the production of algal biomass (Nöie et al., 1991). In addition to nutrient removal, microalgae have also been shown to have the capacity to remove heavy metals (Hammouda et al., 1995), and some toxic organic compounds (Redalje et al., 1989) from water.

2.3 Factors limiting algal growth

2.3.1 Carbon

Most algae are photo-autotrophs, i.e., they use light energy to convert inorganic carbon into complex organic compounds by the photosynthesis process. Hence algal growth depends on the presence of alkalinity in water. Under some circumstances, some algal species also demonstrate photo-heterotrophy i.e. the capability to use light energy for assimilating organic carbon. Algal species which show heterotrophy can use organic sources such as organic acids, sugars, acetate or glycerol. Some algal species show shift between heterotrophy and autotrophy under different conditions. For example, Chlorella and Scenedesmus are capable of growing using sunlight during the day and using some organic compounds during the night (Becker, 1994).

2.3.2 Nutrients: Nitrogen and Phosphorous

Nitrogen is an important nutrient for algal growth. Algae can assimilate nitrogen as nitrates, ammonia or urea. Some algal species have been shown to utilize even gaseous nitrogen through fixation (Fidalgo et al., 1998). Ammonium ion has been shown to be the preferred form of nitrogen for uptake (Haines and Wheeler, 1978). The suppression of nitrate uptake by ammonia availability has also been studied (Syrett and Morris, 1963). This can be explained by the difference in the uptake mechanisms between both forms. Ammonium can be assimilated directly but nitrate is enzymatically reduced to ammonium before assimilation. The reduction of nitrate to ammonia involves two independent enzymatic steps. In the first step, the nitrate is reduced to nitrite catalysed by NADH2-nitrate reductase, and secondly, the reduction of nitrite to ammonia catalysed by the ferredoxin-nitrite reductase (Losada et al., 1970, Serra et al., 1978). This nitrate uptake is regulated by the levels of both nitrate and ammonia in the system (Herrero et al., 1981). Very high nitrate reductase activity may also result in high accumulation of nitrite inside the cells, which causes reduced nitrate uptake and inhibition of growth (Jeanfils et al., 1993).

Phosphorous is another important nutrient which is essential for algal growth. It plays an important role in the photosynthesis process in the form of ATP and NADPH which are responsible for generating metabolic energy. Uptake of phosphorous is dependent on the algal physiology and can change depending on whether the environment is phosphorous -starved or has excess phosphorous. It has been observed that algal cells with lower initial phosphorous concentrations show higher maximum uptake rate (Vymazal, 1995). Luxury phosphorus uptake is another phenomenon which has been well studied in microalgae cultures. Microalgae can assimilate phosphorous in much higher quantities then their immediate growth requirements. The excess phosphorus is stored for later use (Miyachi et al., 1964).

In an environment where nutrients are present in excess, the ratio of carbon, nitrogen and phosphorous uptake by algae has been reported as 106: 16:1 (Stumm and Morgan, 1981). As noted earlier, the actual uptakes of these elements can vary as the algal cell exhibit luxury uptake. Figure 2.2shows how the cellular nitrogen and phosphorus concentration in algae varies with the N: P ratio in the bulk media.

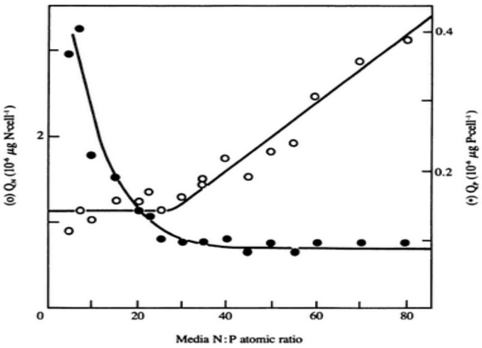

Fig 2.2 Dependence of cellular N and P concentration on the media N: P ratio (Source: Rhee ,1978)

2.3.3 Temperature

Since temperature affects algae metabolism, algal growth is sensitive to temperature. Algal growth increases with temperature and reaches an optimum value after which growth declines on further temperature increase. It has been shown that for an algal culture grown at the optimal temperature, the photo-inhibition, i.e., decline in the rate of algal growth due to the excessive presence of light, will occur at higher light intensities (Borowitzka et al., 1998).

2.3.4 Light

Being photo-autotrophs, algae use light energy to assimilate inorganic carbon. Hence,light is an important determinantfor algal growth. Increase in light intensity increases the algal growth rate up to a point. At higher intensities, photo-inhibition occurs and algal growth rates decrease with any further increase in light intensity (Tschiersch and Ohmann, 1993; Vonshak, Chnawongse et al., 1996). Figure 2.3 shows the growth rates of *Chlorella Pyrenoidosa* at limiting, saturating and inhibiting light intensities.

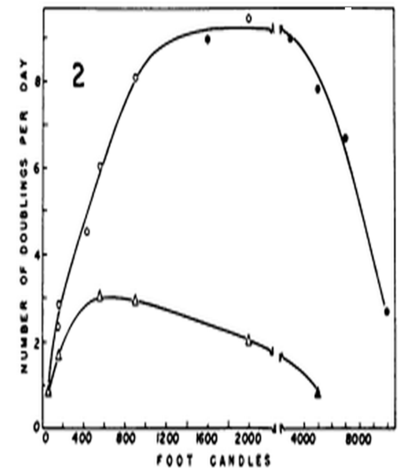

Fig 2.3 Growth rates of *Chlorella pyrenoidosa* at different light intensities and at 39^{o}C and 25^{o}C (Sorokin and Krauss, 1958)

In dense algal cultures, algae are unable to get enough light due to mutual shading, resulting in growth inhibition (Borowitzka, 1998). This phenomenon can be reduced by increasing turbulence in the culture thus providing a more uniform distribution of light of all algae. Another way to avoid internal shading is by changing the depth of the culture. Oswald (1988) has given an empirical relationship between the concentration of green algae, C_c (mg/L) and the light penetration depth d_P (cm), $d_p = \frac{C_c}{6000}$. This shows that using shallow systems can increase the growth and consequently the nutrient removal efficiency of algae.

Further, artificial lighting has been shown to be more effective in nitrate removal than the natural sunlight (which has a diurnal cycle) for algal cultures. It has also been proposed that optimized photo-bioreactors with diurnal cycles can be used in conjunction with natural sunlight to save energy and improve the nutrient removal (Lee and Lee, 2001).

2.3.5 pH

Other factors remaining the same, an optimum value of pH will give maximum growth for a particular algae species.Acidic (3.0-6.2) and alkaline (8.3-9.0) pH values have been shown to reduce the growth of *Chlorella Vulgaris*, whose optimum growth was recorded in the pH range 7.5 – 8.0 (Rachlin and Grosso, 1991). The speciation of inorganic carbon in the solution changes with pH. Since the algae exhibit different rates of uptake for different inorganic carbon species, growth is affected by pH (Azov, 1982).

2.3.6 Mixing

Mixing is an important parameter affecting algal growth and the advantages of good mixing in an algal culture are numerous. The most important benefits of good mixing are the prevention of settling of algal biomass (Stengel, 1970) and reduced photo-inhibition (Soeder and Stengel 1974, Richmond and Vonshak, 1978). Mixing in an algal system is achieved either by bubbling air (in small systems) or by using paddles (in larger systems)(Persoone et al., 1980). Other methods of mixing like the use of jet pumps are which induce horizontal movement of the water have also been tried (Oswald et al. 1977, Stengel 1970, Shelef et al. 1977).

2.3.7 Inhibitory substances

The algal growth in wastewater can be inhibited by the presence of various inhibitory substances. Although NH_4^+ is an excellent nutrient source, free NH_3 has been shown to be toxic to the photosynthesis (Azov and Goldman, 1981), thus pH control (thereby controlling the amount of free ammonia) is critical for avoiding NH_3 toxicity in algal cultures. Surfactants used in washing agents, which ultimately end up in the wastewater streams, also act as inhibitors to the algal growth (Yamane et al., 1984).

2.3.8 Hydraulic Retention Time (HRT)

HRT refers to the average time for which is detained in a reactor. This is a crucial operating factor because it affects the nutrient removal rates and algal growth patterns. The selection of HRT should prevent the two extremes. It should be more than the minimum generation time of the algae and it should be less than a value where there is nutrient limitation and self-shading. An optimum HRT value of 2 to 7 days is common in natural algal treatment systems (Benemann, J.R., 1989).

2.4 Types of Algal Reactors

2.4.1 Open Pond Systems

Open ponds are commonly used for large-scale algae systems because of their simple construction and low operation and maintenance costs. These systems can be in the form of shallow unstirred, circularly stirred or raceway ponds and tanks (Zittelli et al., 2003, Kunjapur and Eldridge, 2010). Limitations of open pond systems as highlighted by Ugwu, (2008) are high evaporation, low mass transfer, and poor utilization of light. Contamination by protozoa, ciliates and bacteria etc. is another problem associated with open pond systems.

HRAP (High Rate Algal Ponds) and facultative ponds are two commonly used open pond systems. Facultative ponds are generally deeper than one meter and the growth of algae on in the surface layers. HRAP on the other hand are race-way ponds with depth of 0.2–1 m and paddlewheel mixing (Craggs, 2005). As per Oswald, (1988b), properly designed and operated HRAPs can remove more than 90% of the BOD and up to 80% of the nitrogen and phosphorous from wastewater.

2.4.2 Closed systems

Closed systems can have a variety of designs such as tubular, flat-plated, rectangular, continuously stirred reactor etc. These systems are called photobioreactors and have better light penetrating characteristics. Such systems can be operated at lower HRTs than open ponds (Borowitzka, 1998). However, photobioreactors are more difficult to operate due to overheating, fouling issues etc. and require higher maintenance costs. Thus photobioreactors are generally used for production of high value algae-derived products such as pharmaceuticals (Patil et al. 2008).

III. Conclusions

In the present study, a comprehensive review of the research works on the effect of various factor on algal growth has been studied. There is a long-standing interest in algal treatment of wastewater.Although, alternative treatment methods to remove nutrients from the wastewater streams are available and well developed, algal treatment provides a unique solution where the algal biomass produced can be harvested and applied as fertilizers, used for biodiesel production, or used in pharmaceutical industries. The properties of algae cultures have been studied extensively.

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