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Review article

The impact of wastewater characteristics, algal species selection and immobilisation on simultaneous nitrogen and phosphorus removal

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ABSTRACT

Nutrient removal from wastewater reduces the environmental impact of its discharge and provides opportunity for water reclamation. Algae can accomplish simultaneous nitrogen and phosphorus removal while also adding value to the wastewater treatment process through resource recovery. The application of algae to wastewater treatment has been limited by a low rate of nutrient removal and difficulty in recovering the algal biomass. Immobilising the algal cells can aid in overcoming both these issues and so improve the feasibility of algal wastewater treatment. Trends for nutrient removal by algal systems over different wastewater characteristics and physical conditions are reviewed. The impact that the selection of algal species and immobilisation has on simultaneous nutrient removal as well as the interdependence of nitrogen and phosphorus are established. Understanding these behaviours will allow the performance of algal wastewater treatment systems to be predicted, assist in their optimisation, and help to identify directions for future research.

1. Introduction

1.1. Algae for wastewater treatment

Most sources of wastewater contain nitrogen (N) and phosphorus (P). If not removed, a build-up of these nutrients can cause eutrophication and toxicity in aquatic environments. Consequently there are limits on their discharge to prevent environmental damage, example allowable median concentrations in wastewater to inland water bodies are total nitrogen (TN) < 10 ppm and total phosphorus (TP) < 0.5 ppm [1]. Conventional biological, chemical and physical treatment methods that separate N and P from wastewater have several downsides not shared with the use of algae. Critically, they become disproportionally more expensive the lower the nutrient concentration, making removal to very low concentrations uneconomical. Separate unit operations or enhanced designs are needed to enable both N and P removal. The concentrated form of the nutrients cannot be readily reused and the potential formation of harmful by-products creates a further need for safe disposal of any waste sludge generated [2].

Rather than viewing the nutrients of N and P as a waste that must be disposed of, wastewater treatment with algae facilitates their recovery in a cost effective and environmentally friendly manner. For example, utilising algae as bioavailable fertiliser removes the need to synthesise fertiliser through energy intensive and resource depleting pathways. This is especially important for added food security as there is a current reliance of mining P from finite reserves. As well as treating wastewater by removing N and P, potentially aiding in providing alternative water supplies, algae consume carbon dioxide (CO_2) and can be used to produce renewable energy, fuels and chemicals [3], thus reducing fossil fuel consumption and greenhouse gas emissions. Algae have the added benefit of removing additional contaminants from wastewater such as heavy metals [4] and persistent organic pollutants [5]. Some of these micropollutants are not, or not sufficiently, removed by conventional treatment processes. The removal of micropollutants is important as they can build up in the ecosystem and have negative health and environmental impacts [6]. These environmental and economic benefits of using algae make it a promising tool to meet the growing demand for wastewater treatment, water and nutrient reclamation, and energy production in a sustainable manner.

Wastewater characteristics vary between locations and fluctuate with time for a single source, with the concentration of nutrients likely to vary independently of each other for a particular wastewater stream [7]. This is an obstacle in simultaneously removing N and P. The complex interaction of wastewater characteristics and environmental conditions creates discrepancies in reported removal behaviours. Careful analysis is required to take into account their compounding interaction and understand their influence on nutrient removal. This is reviewed, with a focus on the interaction between N and P, to help

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identify and direct strategies that will enable algal treatment of wastewater to be completed in an efficacious and cost effective manner.

Applying algal systems to wastewater treatment has been limited by a low degree of nutrient removal as well as problems with harvesting the algae [8]. Newly developed reactors and immobilisation systems show promise to overcome these issues [3]. However, more understanding is needed on the impact that selection of reactor type and algal species has on remediation performance. This review isolates compounding factors pertaining to wastewater characteristics and environmental conditions from the impact of species selection and immobilisation. This approach can assist in improving treatment performance by allowing the design of the algal system to be tailored to the wastewater.

1.2. Wastewater characteristics

Algae are an option to treat different wastewaters, including those from industrial, agricultural and municipal sources [9]; this review focuses on the treatment of municipal wastewater. The major nutrients required for algal growth include nitrogen, phosphorus and carbon, with their concentration and form being important [10]. In municipal wastewater N mostly occurs as ammonium (NH_4^+) and nitrate (NO_3^-), but it can also occur as nitrite (NO_2^-) and organic nitrogen. The main form of P is orthophosphate (PO_4^{3-}) along with organic phosphorus [6]. The concentration and form of C containing compounds also varies. Algae are autotrophic, and predominately grow photosynthetically on inorganic carbon (IC) [11]. Minor nutrients are also required for growth and include sulphur, calcium, potassium and several other metals. For municipal wastewater the minor nutrients are normally in sufficient concentrations to not substantially influence treatment performance [2].

1.3. Treatment systems

Algal treatment systems can be categorised as suspended or immobilised. The former has cells growing freely in the medium. The low cost and simplicity of suspended systems are their main advantage, however they generally have low cell loadings leading to long hydraulic retention time (HRT) requirements [12]. Harvesting of suspended cells can be difficult causing inefficient resource recovery and risk of contaminating the discharged wastewater [8]. Due to the larger size of freshwater macroalgal cells, macroalgae are seen as an alternative to avoid the limitations in harvesting suspended microalgal systems [13].

Passive techniques of immobilisation, known as attached growth systems or biofilms, involve the algal cells accumulating on and attaching to a surface of a substrate through the excretion of extracellular polymeric substances [10, 14–16]. Active techniques of immobilisation involve physically separating the cells from the bulk liquid phase and include covalent coupling, adsorption, liquid–liquid emulsion, membrane separation, entrapment and encapsulation [4, 14, 17, 18]. In general, immobilisation allows for higher cell loading than suspended systems, giving a higher rate of nutrient removal. The resultant lower HRT and reduced reactor volume along with the ease of harvesting are the main advantages for wastewater treatment [19, 20]. These benefits need to be balanced against the additional costs of the immobilisation process and materials, and it should be noted that there have been few full scale applications of immobilised algae wastewater treatment systems.

1.4. Species selection

For the purpose of species selection many of the papers reviewed here are on pure monocultures to reduce interferences. In algal wastewater treatment systems the algae will exist as a consortium, and both bacteria and algae will participate in nutrient removal. Hence in real systems the influence of bacteria must also be considered [21, 22]. In addition, algae and bacteria form a synergistic relationship which can improve growth and nutrient removal, as the bacteria consume O_2 and produce CO_2, while the algae consume CO_2 and produce O_2 [12].

Immobilisation can prevent contamination of the inoculated algae, which enables more control over the selection of species within the culture [14, 23]. This allows the most appropriate algal species to treat the wastewater to be selected and offers a pathway to decouple the role of algae from that of bacteria. Compared with algal monocultures, co-cultures of selected algal and bacterial species, or of multiple algal species, can improve the rate and robustness of remediation [12]. Such co-cultures can be controlled by immobilisation, either by keeping each species in separate matrices or mixed within the same matrix [14, 24]. A carefully managed a co-culture of bacteria and algae may also be a way to improve nutrient removal during dark periods [25] or maintain the pH of the effluent [26].

Comparing monocultures, some algal species are equally capable of removing N and P and of adapting to different wastewaters [27–30]. In other cases the proportion of N to P removed differs significantly between algal species treating a single wastewater [31–33], indicating appropriate species selection can promote nutrient removal. There are species dependent responses to environmental conditions [34, 35] which mean in an open system seasonal variations can cause fluctuations in the species present and remediation rates [36]. One approach to selecting species that are suitable for the treatment conditions is to isolate them from the treatment plant or a local aquatic system [37, 38]. Starvation of algal cells before exposure to wastewater is a concept that may increase the rate of nutrient removal [39, 40]. Hernandez et al. [41] observed a higher P removal after starving alginate-immobilised *Chlorella sorokiniana,* but not with *C. vulgaris*, indicating caution needs to be applied as starvation leading to a positive effect is species-dependent.

2. Nutrients

2.1. Nitrogen

2.1.1. N removal mechanisms

Nitrogen is essential to many of the functional components of algae, including structural proteins and enzymes, nucleic acids, chlorophyll and energy transport molecules [11, 42]. This leads to a high requirement for N, however most algal species do not exhibit non-structural N storage [42]. The algal cell can adapt to assimilate N as needed based on the form and concentration available [43]. In addition to direct assimilation, NH_4^+ can be removed through volatilisation as ammonia (NH_3) under basic conditions, where an elevated pH can be a consequence of algal growth. NO_3^- is more stable and hence not removed by volatilisation [11]. Based on finding no clear relationship between influent and effluent NH_4^+ concentration from the compilation of a number of studies, Whitton et al. [19] suggested that the mechanism and rate of N removal depends on the treatment system and conditions.

2.1.2. N removal behaviour

As N is critical to many of the cellular components in algae it follows that the wastewater N concentration impacts N uptake. With alginate-entrapped *C. vulgaris*, when increasing the NO_3^- concentration Jeanfils et al. [44] found a proportional increase in the rate of NO_3^- removal. Increasing either NO_3^- or NH_4^+ also led to an increased rate of N removal for *C. vulgaris* and *Pseudokirchneriella subcapitata* [45]. Removal of NH_4^+ is especially important as it is the more toxic form of N in wastewater. Whitton [46] found that alginate immobilised *Scenedesmus obliquus* could adapt to 4.2 ppm NH_4-N and remove it to a residual of 0.06 ppm in 2 h bead contact time. *C. vulgaris* in attached growth exhibited greater N removal with increasing NH_4^+ concentration [47], as did suspended *Chlamydomonas acidophila* to a lesser extent [48], possibly due to inhibitory concentrations of NH_4^+ being reached. An increase in N available in the wastewater results in more N uptake, with