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Recovery of nutrients from swine wastewater using ultrafiltration: Applications for microalgae cultivation in photobioreactors

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ABSTRACT

The large-scale production of microalgae poses a number of challenges, including a costly fertilizer demand. While wastewater provides a concentrated source of nutrients, the presence of biological contaminants and the expense of heat treatment are challenging for large-scale production. The goal of this study was to use ultrafiltration to purify wastewater for use in the cultivation of microalgae. Swine waste was filtered, and the resulting permeate was utilized in a *Porphyridium cruentum* culture. Fluxes remained relatively constant during operation, and the complete rejection of bacteria was observed. The permeate contained high concentrations of total nitrogen (695.6 mg L^{-1}) and total phosphorus (69.1 mg L^{-1}). Higher biomass productivity and lipid contents were observed in the microalgae cultivated in the waste medium compared to that of a control medium. This suggests that, by using ultrafiltration as an alternative to heat treatment, agricultural wastewaters could be utilized as a nutrient source for microalgae.

1. Introduction

Algal biomass has been identified as a promising feedstock that could be used in a number of industrial applications, including biofuel production, aquaculture, and pharmaceutical production (Sandefur et al., 2014; Jones and Mayfield, 2012; Lam and Lee, 2012; Wang et al., 2013). Through the use of sunlight and CO_2, microalgae species are capable of producing biomass that is rich in lipids and carbohydrates, which can be extracted from the plant material and used in the production of commodities such as biofuels (Cai et al., 2013). Algae have been shown to have higher rates of productivity and higher lipid contents than traditional bioenergy crops, do not require high-quality land for cultivation, and would not compete with current agricultural products for space (Sandefur et al., 2011; Wiley et al., 2013). Examples of these high-value algae species include *Nannochloropsis oculata* and *Chlorella vulgaris*, which have been used in the production of biodiesel (Converti et al., 2009), in addition to *Porphyridium cruentum*, which has been identified as a potential source of omega-3 fatty acids (Ryckebosch et al., 2014).

Omega–3s are long-chain polyunsaturated fatty acids (PUFAs) and are a critical element of the human diet. There are several key types of dietary PUFAs, including docosahexaenoic acid (DHA), eicosapentaenoic acid (EPA), and arachidonic acid (AA). These fatty acids play an important role in neurological development, eye function, and cardiovascular health. The importance of omega–3 s in the neurological development of fetuses and small children has also been documented in the literature (Ruxton, 2004). However, traditional fish-based sources of omega-3 fatty acids have a number of challenges associated with their use in human and animal supplements, including the presence of carcinogenic compounds and heavy metal contaminants. As a result of these challenges, many pharmaceutical and nutraceutical companies are turning to microalgae as a source of omega-3 fatty acids. While DHA-rich microalgae products have emerged in recent years, microalgae-based sources of EPA are lacking (Yaakob et al., 2014). Research has shown that *Porphyridium cruentum*, a fast-growing red marine algae, has relatively high EPA and AA contents compared to other algae species (Yaakob et al., 2014; Ryckebosch et al., 2014).

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While the high productivity and lipid content of microalgae make it a promising feedstock for a number of industrial applications, there are several challenges associated with their cultivation in large-scale production systems. The production of microalgae can require large amounts of nitrogen and phosphorus fertilizer (Sialve et al., 2009). In order to make the production of microalgae economical, it is important that low cost sources of nitrogen and phosphorus be available to producers. Greenwell et al. (2013) reported a nitrogen fertilizer cost of $1.40 per kilogram. Considering the relatively high nitrogen content of microalgae (4–8%), the nitrogen inputs required for microalgae cultivation can be significant. In addition, the synthesis of nitrogen fertilizer produces around 2 kg of CO_2 kg^{-1}. The additional carbon generation, if added to the overall lifecycle, undermines the favorable carbon balance that is obtained by using microalgae biomass as a feedstock for biofuel production (Greenwell et al., 2013).

As a solution to this problem, agricultural wastes have been identified as an alternative to inorganic fertilizer in microalgae cultivation (Cai et al., 2013). Wastewater tends to have large amounts of nitrogen and phosphorus, and has been used as a nutrient source for microalgae cultivation in a number of studies (Fenton and hUallachain, 2012; Zhu et al., 2013; Honda et al., 2012). In addition, the problem of nutrient pollution from livestock wastes is a growing challenge for livestock producers. The development of a microalgae-based biological treatment system to replace current nutrient management plans could help to improve the sustainability of livestock production systems (Zhu et al., 2013).

There have been a number of studies utilizing waste streams in the cultivation of microalgae. Honda et al. (2012) used a simulated treated sewage in the cultivation of the microalgae *Chlorella vulgaris*, *Botryococcus braunii* and *Spirulina platensisin* in a flat plate reactor. The reported productivities were comparable to similar studies of microalgae cultivation systems that used traditional nutrient sources. Similarly, Zhu et al. (2013) successfully cultivated the microalgae *Chlorella zofingiensis* in a tubular photobioreactor using swine wastewater as a nutrient source.

While the high nutrient concentrations found in animal wastes make them desirable for use in the cultivation of microalgae, some problems still exist. Principally, the potential for bioreactor contamination from bacteria is increased when using waste streams as a nutrient source (FAO, 2012). In their review of contamination pathways for biological pollutants in microalgae cultivation, Wang et al. (2013) noted that the production of microalgal biomass has been historically constrained by biological contamination events that impede production at the industrial scale, even when waste streams are not used. Laboratory-scale studies of microalgae cultivation using waste often involve the preparation of mock waste instead of real wastewater samples (Wang and Lan, 2001; Voltolina et al., 2005; Feng et al., 2011). Studies that utilize authentic wastewater effluent samples have employed a number of treatment methods, including heat treatment and exposure to UV light (Zhu et al., 2013; Cho et al., 2011). For example, in order to avoid bioreactor contamination, Zhu et al. (2013) sterilized the swine waste in an autoclave prior to use. However, in a large-scale system, autoclaving the large volumes of wastewater prior to use is not economically feasible, and alternative methods for treatment must be developed (Wang et al., 2013).

Membrane separations technology constitutes an alternative method for the removal of biological contaminants from the nutrient-rich wastewater prior to use in microalgae cultivation. A membrane consists of a thin film that separates two phases, and can be used in liquid–liquid separations. Microfiltration and ultrafiltration involve the use of porous membranes to remove micro- or macro-particles from a solution (Teo, 2000). Wang (1999) used ultrafiltration in the purification of drinking water, and showed complete rejection of *E. coli* using a hollow fiber membrane system. Similarly, in their study of nutrient recovery from dairy sludge, Gerardo et al. (2013) used cross-flow microfiltration to obtain a bacteria-free solution while retaining the concentrations of nitrogen and phosphorus, although the use of the recovered nutrients in microalgae cultivation was not explicitly tested (Gerardo et al., 2013).

The objective of this study was to (1) evaluate the potential use of ultrafiltration technology in the removal of inorganic solids and biological contaminants (principally bacteria) from agricultural wastewater effluent, (2) determine if the treated wastewater was a viable source of nutrients for the production of the high-value microalgae *Porphyridium cruentum,* and (3) determine how resilient *P. cruentum* is to contamination from other algae strains under optimized growth conditions.

2. Materials and methods

2.1. Wastewater feed source

In order to test for the rejection of live cells from an agricultural waste stream, feed samples for the filtration process were obtained from a swine farm located in Savoy, AR (36°6′20″N 94°19′58″W). The feed sample source was an anaerobic digestion lagoon that was used to hold waste flushed from a 150 head grow-finish swine operation, which was used to house weaned pigs until they reach market weight. The facility was made up of a drop curtain style barn that housed pens with fully slatted floors for manure removal via flushing into an adjacent lagoon. For each ultrafiltration run, grab samples of wastewater were taken from the lagoon using a telescopic dipping sampler. The sample was refrigerated after collection for up to 24 h prior to each ultrafiltration run. Aliquot samples of the wastewater were taken and analyzed to determine the bacteria and total solid concentrations of the feed prior to ultrafiltration.

2.2. Ultrafiltration system operation

The ultrafiltration system used in this study was composed of a feed tank, pump, and 2–1 inch hollow fiber membrane cartridges (50,000 MWCO; Koch Romicon PM50). The pump capacity was 3 gpm at 25 psi. A valve and pressure gauge was located before and after the hollow fiber cartridges in order to control the transmembrane pressure (see Fig. 1). Valves 3 and 4 controlled the flow of permeate from the cartridges. The temperature of the fluid in the feed tank was monitored and maintained at 26 °C using a chiller unit.

To begin the ultrafiltration process, valves V-1 and V-2 were fully closed and opened, respectively (Fig. 1). The wastewater feed mixture was then placed in the reservoir and the pump was turned on. V-1 was then slowly opened until 10 psi was read from G-1. V-2 was then closed until a pressure of 15 psi was read from G-2 on the retentate side. V-1 was then adjusted until G-1 read 20 psi, resulting in an average transmembrane pressure of 17.5 psi. Permeate was recirculated through the system for two hours. After one hour, samples were taken from each membrane cartridge via the sampling port by opening valves 3a and 3b. This step was repeated at the two hour mark.

In addition to the sampling regime, the changes in permeate flux were measured during the ultrafiltration runs. In order to determine if the permeate flux would decrease with each new ultrafiltration cycle, the flux was measured every 10 min during three individual two hour runs. The permeate flux was also measured during a long term run, over the course of 8 h. The flux was determined by measuring the amount of time required to generate 100 mL of permeate, and was normalized against the total