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The Feasibility Study of Using Microalgae for Polishing Consumer-products Industrial Effluent Containing High Total Dissolved Solids

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Abstract

A consumer-products industrial effluent contained high phosphate and sulfate from the manufacturing and wastewater treatment processes, resulting in high total dissolved solids (TDS) concentration. It is believed that *Chlorella* sp. and *Scenedesmus* sp. can grow in high-sulfate wastewater. The objective was to evaluate the feasibility of cultivating both algae in the industrial effluent with high TDS to polish contaminants. The culture mediums with different pH values of 3, 5, 7, and 9 and different sodium sulfate concentrations of 400, 600, 800, and 1000 mg/L were used to determine the optimum pH values and to evaluate the effects of high TDS on both algae. For both algae, the optimum pH value was 7.0, and high TDS affected minimally the specific growth rates but decreased considerably the biomass productivities due to high ionic strength. The phosphorus removal efficiency of *Chlorella* sp. was greater than *Scenedesmus* sp. because of simultaneous phosphorus assimilation and calcium phosphate precipitation. The results revealed that the lower the TDS concentration in the effluent, the higher the biomass productivities of both algae were obtained. It appears that both algae are not applicable for polishing this effluent unless nitrogen and inorganic carbon are supplemented and the TDS is reduced.

Keywords : *Chlorella* sp.; Consumer-products effluent; *Scenedesmus* sp.; Sodium sulfate; Total dissolved solids

Introduction

Consumer-products like detergents, soaps, and other products typically contain surfactants including anionic, cationic, non-ionic surfactants; however, both linear alkylbenzene sulfonates (LAS) salt and acid are typically the largest group of anionic surfactant [1, 2]. The LAS are commonly manufactured by the sulfonation of both linear alkylbenzens (LAB) and sulfur trioxide to produce the LAS salt [2]. As a result of the washing process, the wastewater from this step contains a high concentration of sulfonic acid with high pH [3]. The aerobic biological wastewater treatment system is widely used for the removal of surfactants from the wastewater [4]. Coagulation-flocculation as a pretreatment step has been demonstrated to be a successful wastewater treatment system [5]. If aluminum sulfate, ferrous sulfate, or ferric sulfate is used as a coagulant, sulfate ion usually remains in the effluent. Moreover, the pH adjustment with chemicals such as sulfuric acid and sodium hydroxide is usually required for the coagulation-flocculation process for this industry, contributing both sodium and sulfate ions into the wastewater. Furthermore, LAS can be biodegraded with microbes in the biological wastewater treatment system with the oxidative pathway to form a sulfate ion [6, 7]; thus, sulfate ion can be prevalent anion in the effluent. Among other pollutants, phosphate is usually found in this industrial wastewater because phosphate is used as a builder in detergents for controlling and providing alkalinity to increase the overall detergency. Therefore, both sulfate and phosphate increase TDS in the effluent. Several physical and chemical treatment alternatives are available for TDS removal including softening, reverse osmosis (RO), chemical precipitation, adsorption, and electrophoresis, electrochemical, membrane separation, or electrodialysis [8]. However, each physical and chemical method has its own limitations and concerns such as cost-effective, chemical waste, sustainability, or environmentally friendly.

It is generally known that phytoremediation has been increasingly used in recent years with a primary objective to use plants or associated microorganisms to reduce the concentrations or toxic effects of contaminants in the environmental systems [9]. Several microalgae including *Chlorella vulgaris* [10-12], *Scenedesmus obligous* [13-14] have been reported to eliminate nitrogen and phosphorus effectively in wastewaters. *Oocystis* sp. obtained the sulfate uptake rate of 32% from the power plant wastewater containing sulfate concentration of about 5200 mg/L in a batch culture system with continuous carbon dioxide (CO_2) supply from the atmosphere [15]. Both *Chlorella* sp. and *Scenedesmus* sp. were the first and second top ranks of biomass productivity, respectively. Another study revealed that *Scenedesmus* sp. and *Chlorella* sp. obtained the sulfate removal efficiencies of 36% and 34% from the effluents of a municipal wastewater treatment plant, respectively [16]. It appears that both *Chlorella* sp. and *Scenedesmus* sp. could potentially grow in the rich-sulfate wastewater with their abilities to remove nitrogen and phosphorus from the wastewater.

The objective of this study was to evaluate the feasibility to cultivate *Chlorella* sp. and *Scenedesmus* sp. in the consumer-products industrial effluent containing high TDS resulting from both phosphate and sulfate ions. The factors involving the growth conditions of *Chlorella* sp. and *Scenedesmus* sp. including pH and sulfate concentration were studied in closed batch culture systems containing either BG-11 culture medium or industrial effluent.

Materials and Methods

The experiments were conducted in the Environmental Engineering Laboratory at the Department of Chemical Engineering, Burapha University of Thailand using 1-L glass bottles and 3-L small reactors at room temperature of about 29 °C. The growths of both *Chlorella* sp. and *Scenedesmus* sp. were evaluated at the initial pH values of 3, 5, 7, and 9 with the addition of sodium sulfate (Na_2SO_4) concentrations of 400, 600, 800, and 1000 mg/L (2.8, 4.2, 5.6, and 7.0 mM), respectively. Both BG-11 culture medium and effluent from industry were used to grow the algae.

Algal Inoculums and Culture Medium Preparations

The stock cultures of *Chlorella* sp. and *Scenedesmus* sp. were firstly prepared by culturing them in BG-11 nutrient medium [17] with initial pH of 7.0, which was prepared by using chemicals including $NaNO_3$ 1500 mg/L, $K_2HPO_4{\cdot}3H_2O$ 40 mg/L, $MgSO_4{\cdot}7H_2O$ 75 mg/L, $CaCl_2{\cdot}2H_2O$ 36 mg/L, Citric acid 6 mg/L, Ammonium ferric citrate 6 mg/L, Ethylene diaminetetraacetic acid disodium magnesium salt (EDTA-Na_2Mg) 1.0 mg/L, and Na_2CO_3 20 mg/L. One milliliter (mL) of trace metal stock solution was added to each liter of medium. A liter of trace nutrients stock solution consisted of H_3BO_3 2.86 g/L, $MnCl_2.4H_2O$ 1.81 g/L, $ZnSO_4.7H_2O$ 0.220 g/L, $Na_2MoO_4{\cdot}2H_2O$ 0.390 g/L, $CuSO_4{\cdot}5H_2O$ 0.079 g/L, $Co(NO_3)_2.6H_2O$ 0.049 g/L. The deionized water was used to prepare the stock solution. The BG-11 medium was sterilized for 15 minutes in the autoclave at the temperature of 121 °C and the pressure of 15 psi.

Pure cultures of both *Chlorella* sp. and *Scenedesmus* sp. were procured from The Institute of Marine Science, Burapha University. Both algae were transferred from agar into 50-mL of liquid BG-11 medium in the 250-mL Erlenmeyer flasks. All flasks were then placed on the shelves in the laboratory as illustrated in **Figure 1** under fluorescent light (Philips 36W, Cool Daylight, Color Code 865 [CCT of 6500K]) with the total light intensity of 4000 lux, at the temperature of about 29 °C with a 12:12 light-dark cycle. The liquid cultures were incubated and shaken for one minute three times each day for a week. For algal enrichments, *Chlorella* sp. and *Scenedesmus* sp. with the amounts of 50 mL were transferred to 750-mL of BG-11 liquid medium in 1-L bottles. A small fine bubble air stone diffuser connecting to an air supply pump was used to aerate the algae in each bottle. The exponential growth period of 4-12 days was found from the growth curve analysis for both algae; therefore, algae during this period with the concentrations of about 1000 mg/L (dry weight) were used for further experiments.

The specific growth rate (μ) was determined with the first-order approximation as listed in Eq. (1). The natural logarithm of algal concentration versus cultivation time was plotted in Microsoft Excel and the linear regression analysis was used to obtain the slope, indicating the specific growth rate. The fitness was identified by the coefficient of determination or R-squared (R^2) value.

$$\mu = (\ln C_t - \ln C_0) / (t - t_0) \qquad (1)$$

The algal biomass productivity (P) was also determined according to Eq. (2) from the slope of linear plots between the algal concentration and cultivation time.

$$P = (C_t - C_0) / (t - t_0) \qquad (2)$$

Both C_0 and C_t are the initial and final algal concentrations expressed as mg/L, t_0 and t are the initial and final time with a unit of day, μ is the specific growth rate with a unit of day^{-1}, and P is the algal biomass productivity with a unit of mg/L/day.

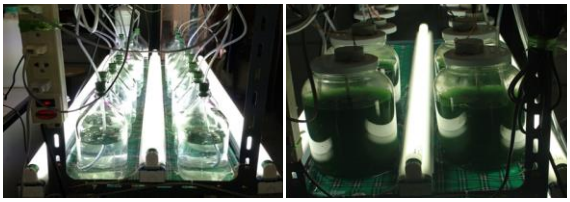

Figure 1 Microalgae in 1-L glass bottles and small 3-L bioreactors at the room temperature of about 29 °C

Optimal pH for Algal Growth in Culture Medium at Different TDS Concentrations from Sodium Sulfate Additions

The optimal pH for the algal growth in the BG-11 culture medium was determined by adjusting the BG-11 medium with 1 N of NaOH and 1 N of H_2SO_4 to the pH values of 3, 5, 7, and 9. At each pH value, the medium was spiked with the Na_2SO_4 stock solution of 100,000 mg/L, which was prepared with anhydrous chemical Na_2SO_4 ACS grade, to obtain a culture medium with different Na_2SO_4 concentrations of about 400, 600, 800, and 1000 mg/L. The medium contained sulfate at concentrations of about 270, 406, 541, and 676 mg SO_4^{2-}/L. Both *Chlorella* sp. and *Scenedesmus* sp. taken from stock cultures with the volumes of 80 mL were added into 1-L glass bottles containing 720-mL culture medium with different pH values and Na_2SO_4 concentrations to obtain the total solution volume of 800 mL. A set of bottles containing culture medium without the addition of algae was used as a control sample set. The samples were taken at a volume of 30 mL every 72 hours for a period of 288 hours (12 days) to measure the dry weight of algae, pH, TDS, and both anionic and cationic ions.

The Growth of Microalgae in the Consumer-products Industrial Effluent

Effluent from an industrial wastewater treatment plant manufacturing consumer-products in Thailand was collected and delivered to the laboratory. The industrial effluent volume of 2700 mL, after acclimatizing with the room temperature and adjusting the pH to 7.0 from the previous experiments, was subsequently added into a 3-L small reactor with 300 mL of each alga to obtain the initial algal concentration of 200 mg/L and the total volume of 3000 mL. In addition, the neutral pH of effluent is typically obtained from the biological wastewater treatment plant. The samples were collected every 3 days and were analyzed for parameters.

Additional experiments were conducted to evaluate the effects of TDS on the algal biomass productivities of *Chlorella* sp. and *Scenedesmus* sp. in the industrial effluent. The microalgae inocula at the concentration of about 400 mg/L were added into the 1-L glass bottles containing the industrial effluent volumes of 500, 600, 700, and 800 mL with the deionized water at the volumes of 300, 200, 100, and 0 mL to obtain the total volume of 800 mL. Same experimental conditions as described in the previous section were implemented.

Measurement and Analyses

All samples collected from the experiments were analyzed for several parameters including pH, algae concentration (dry weight), TDS, salinity, ammonium nitrogen (NH_4^+-N), nitrate nitrogen (NO_3^--N), phosphate (PO_4^{3-}), sulfate (SO_4^{2-}), chloride (Cl^-) sodium (Na^+), magnesium (Mg^{2+}), calcium (Ca^{2+}), and potassium (K^+). All samples were filtered with the 0.45-μm membrane filter to remove all particulates for measurements of anionic and cationic ions. The analyses of these parameters followed the Standard Methods for the Examination of Water and Wastewater [18]. Both anionic and cationic ions were injected into the Dionex 2010I Ionic Chromatograph (IC) installed with both anion column (IONPAC AS4A-SC) and cation column (IONPAC CS12A-4X) (Dionex Corp., Sunnyvale, CA). Both anionic and cationic mobile phases of 9.0 mM of Na_2CO_3 and 22 mN of H_2SO_4, were used in the IC equipment, respectively. A pH meter (CyberScan pH510, Eutech Instruments), a salinity meter (Ecoscan SALT6+, Eutech Instruments), and a TDS meter (EcoScan TDS6+, Eutech Instruments) were used to determine pH, salinity, and TDS concentrations, respectively. The dry weight of algal concentration was determined with the TSS method [18].

Results and Discussion

The experiments were conducted to evaluate the feasibility of using microalgae for polishing the industrial effluent of the biological wastewater treatment system treating the consumer-product industry. Various conditions including pH and sodium sulfate concentrations for the growths of both *Chlorella* sp. and *Scenedesmus* sp. were evaluated. The optimum conditions obtained from the BG-11 culture medium were applied to the industrial effluent.

Optimal pH for Algal Growth and Biomass Productivity in BG-11 Culture Mediums

Both *Chlorella* sp. and *Scenedesmus* sp. concentrations, cultured in the BG-11 medium with different pH values of 3, 5, 7, and 9 for a period of 12 days, are illustrated in **Figures 2(a)** and **2(b)**, respectively. The first-order specific growth rates of *Chlorella* sp. and *Scenedesmus* sp., which were determined from the slopes of linear equation retrieving from the graphical plots between the natural logarithm of algal concentration and cultivation time, at different pH values of BG-11 medium are shown in **Figures 2(c)** and **2(d)**, respectively. At the pH value of 3, *Chlorella* sp. ceased gradually at the decreasing specific growth rate of 0.02(0.14) day^{-1}. It is noted that the number in parenthesis is the R^2 value. The low R^2 value of 0.14 approaching zero indicates that *Chlorella* sp. concentration did not change with time during a cultivation period of 12 days. *Chlorella* sp. concentration increased minimally at the pH value of 5 with the overall growth rate of 0.04(0.48) day^{-1}, resulting from the slight decrease of algal growth during the first 6 days due to the pH adaptation and subsequently increased during the last 3 days of cultivation. The growth of this alga resulted in an increase of medium pH to the final pH value of about 8.2. At the initial pH of 9, *Chlorella* sp. concentration increased progressively during the first 9 days and ceased during the last 3 days, resulting in the net growth rate of 0.13 day^{-1} (R^2 = 0.84). During the last 3 days of culture, the death of *Chlorella* sp. was found due to the limitation of phosphorus. It was found that phosphate in the BG-11 medium was only 0.8 mg P/L on day 6. It is generally known that phosphate can be precipitated with calcium as hydroxyapatite [$Ca_5(PO_4)_3OH$] (HAP), depending on several factors including high pH, temperature, and both calcium and phosphate concentrations [21]. Both phosphate and calcium concentrations in the medium were minimum on day 6. It appears clearly from **Figure 2(a)** that the initial pH value of 7.0 was the optimum pH for the growth of *Chlorella* sp. in this study, producing the algal concentration of 840 mg/L at day 12 with the growth rate and the algal biomass productivity as listed in **Table 1**. Khalil et al. [19] reported that *Chlorella* sp. grew in a pH range of 4-10, but alkaline pH provided the maximum concentration for this alga. The final pH of the medium increased to 8.9 on day 12 at the initial pH of 7. As illustrated in **Figure 2(b)**, *Scenedesmus* sp. decreased gradually during the cultivation time of the first 9 days due to unsuitable pH for growth at the initial pH of 3. After 9 days of pH acclimation, the alga grew minimally; however, the overall decreasing specific growth rate of 0.06 day^{-1} (R^2 = 0.49) was obtained as indicated by the slope in **Figure 2(d)**. *Scenedesmus* sp. grew exponentially at the pH values of 5, 7, and 9 with the growth rates of 0.11(0.87), 0.14(0.92), and 0.07(0.70) day^{-1}, respectively. At the initial pH of 5, *Scenedesmus* sp. grew exponentially, reaching the maximum concentration of 390 mg/L and then ceased due to the final pH of about 10. The growth rate of *Scenedesmus* sp. was highest at the initial pH of 7 obtaining a maximum algal concentration of 745 mg/L with the algal biomass productivity of 47.2 mg/L/day as listed in **Table 1**. The results indicate that the neutral pH range was optimal for *Scenedesmus* sp. It was reported that the optimum pH for the growth rates of three *Scenedesmus* strains was about a neutral pH of 7 [20]. It can be concluded that the optimum pH value for the growths of both *Chlorella* sp. and *Scenedesmus* sp. was 7.0; however, *Chlorella* sp. grew slightly faster than *Scenedesmus* sp., providing higher algal biomass productivity under the same cultivation time. The specific growth rates and biomass productivities of both algae were used as control experiments for further comparisons.

Nitrogen and phosphorus are essential to algal production primarily to synthesize protein. It was found that nitrate and phosphate in the BG-11 medium were utilized as nutrients for the growths of both algae as shown in **Figure 3**. Excess nitrate was available in the medium; therefore, the zero-order kinetics of nitrate utilization rates could be evaluated. As listed in **Table 1**, the nitrate utilization rates of *Chlorella* sp. and *Scenedesmus* sp. were 5.72 and 2.51 mg N/L/day at the initial pH of 7.0, resulting in the nitrate removal efficiencies of 21.3 and 10.5%, respectively. The phosphate uptake of *Chlorella* sp. was continuously accomplished during the first 9 days resulting in the phosphate removal efficiency of 43.5%. However, *Scenedesmus* sp. assimilated phosphate rapidly during the first 6 days at the uptake rate of 0.69 mg P/L/day and then decreased minimally after that, resulting in the overall removal efficiency of 68.7%. It is not likely that phosphates were removed with chemical precipitation in this experiment because calcium concentrations in the BG-11 medium did not decrease as illustrated in **Figure 3**. It appears that *Chlorella* sp. was capable of removing nitrogen and *Scenedesmus* sp. was effective alga for eliminating phosphorus. TDS remained

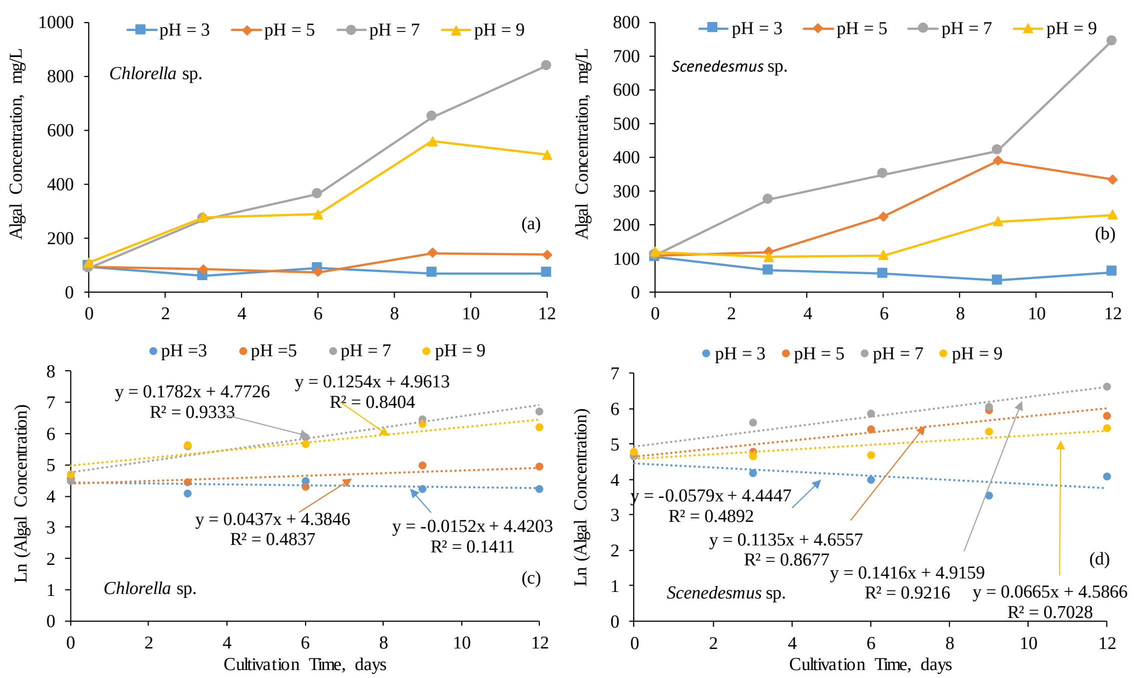

Figure 2 Algal concentrations and natural logarithm of algal concentrations versus time in the BG-11 medium with different pH values at different cultivation time

Table 1 Specific growth rates and algal biomass productivities of microalgae, nitrate and phosphate removal efficiencies and uptake rates at the optimum pH of 7 with different sodium sulfate concentrations

Na_2SO_4	Specific Growth Rate (day^{-1})		Nitrate Utilization Rate (mg N/L/day)		Phosphate Uptake Rate (mg P/L/day)	
	Chlorella	*Scenedesmus*	*Chlorella*	*Scenedesmus*	*Chlorella*	*Scenedesmus*
0	0.18(0.93)	0.14(0.92)	5.72	2.51	0.13	0.69
400	0.16(0.93)	0.15(0.98)	2.79	3.14	0.12	0.21
600	0.17(1.00)	0.13(0.91)	2.94	3.14	0.22	0.37
800	0.16(0.99)	0.13(0.98)	3.70	5.51	0.37	0.27
1000	0.16(0.98)	0.14(0.95)	3.15	5.59	0.26	0.20
Na_2SO_4	Biomass Productivities (mg/L/day)		Nitrate Removal Efficiency (%)		Phosphate Removal Efficiency (%)	
	Chlorella	*Scenedesmus*	*Chlorella*	*Scenedesmus*	*Chlorella*	*Scenedesmus*
0	62.7(0.98)	47.2(0.91)	21.3	10.5	43.5	68.7
400	48.5(0.81)	45.7(0.92)	10.6	11.9	36.9	53.4
600	53.7(0.89)	41.3(0.97)	11.8	11.9	64.7	77.5
800	50.3(0.91)	40.0(0.95)	12.6	20.5	99.2	79.1
1000	50.5(0.87)	43.3(0.97)	10.4	20.9	100.0	71.3

Number in parenthesis indicates R^2; Na_2SO_4 = 0 mg/L indicates that no additional Na_2SO_4 was added.

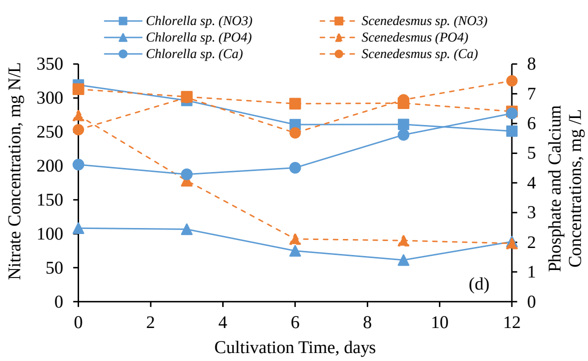

Figure 3 Nitrate, phosphate and calcium concentrations in BG-11 medium containing *Chlorella* sp. and *Scenedesmus* sp. during 12 days of cultivation time at the pH of 7 without Na_2SO_4 supplementation

approximately constant during the cultivation period of 12 days for both microalgae. The findings in this study are not consistent with the results of González et al. [22]. They reported that *Scenedesmus dimorphus* removed ammonia from secondary effluent of agro-industrial wastewater in a cylindrical bioreactor better than *Chlorella vulgaris* while both algae consumed phosphorus at the same removal efficiencies. This consistency may be due to different culture mediums (BG-11 and effluent), nitrogen sources (NH_3 and NO_3^-), and algal species (*Scenedesmus* sp. and *Scenedesmus dimorphus*).

As expected, the average final pH values in the BG-11 culture mediums containing *Chlorella* sp. and *Scenedesmus* sp. were increased to 9.6±0.4 and 9.4±0.2 at the initial pH of 7, respectively. The blank bottles revealed that the pH of the medium remained during a period of 12 days. It can be explained that the rise of pH in the BG-11 medium resulted from bicarbonate utilization as a carbon source for the growth of algae. Carbon dioxide (CO_2) dissolution from the atmosphere was limited because of the closed batch culture system. In addition, it is noted that the solubility of CO_2 from the atmosphere is very limited according to Henry's constant, especially at high temperature and high TDS solution [23]. Na_2CO_3 was a carbonate source in the BG-11 medium. According to the carbonate system, bicarbonate is known to convert from carbonate species (CO_3^{2-}) at neutral pH. As a result of CO_2 utilization from bicarbonate (HCO_3^-), which is facilitated through the CO_2 concentrating mechanism (CCM) [24] during photosynthesis, proton (H^+) is used to convert HCO_3^- to CO_2 according to the equilibrium ($H^+ + HCO_3^- \leftrightarrow CO_2 + H_2O$), resulting in OH^- inside the cell. The proton uptake from the bulk solution occurred to neutralize OH^- inside the cell [25]; therefore, the pH of BG-11 medium increased due to the reduction of H^+. It is expected that the total carbonate species decreased as a result of bicarbonate utilization for the growth of algae in the closed system and the ratio of CO_3^{2-} to HCO_3^- increased as the pH increased above 10; therefore, available HCO_3^- was limited for growth of algae. It is noted that carbonate, as a dominant species at high pH above 10, cannot be metabolized by microalgae [24].

Effects of Sulfate Concentrations on the Growth of Algae in BG-11 Culture Medium

The experiments were conducted to evaluate the effects of sodium sulfate concentrations on the growths of *Chlorella* sp. and *Scenedesmus* sp. at the selected optimum pH of 7.0 determined from previous experiments. The TDS concentrations in the BG-11 medium increased to 1450, 1560, 1780, and 1980 mg/L due to the Na_2SO_4 additions of 400, 600, 800, and 1000 mg/L, respectively. Due to the chemical constituents of the medium in section 2.1, the BG-11 culture medium contained about 1090 mg/L TDS and the sulfate concentration of about 30 mg/L (0.3 mM). The graphical plots between the natural logarithm of algal concentration and cultivation time and between algal concentration and cultivation time revealed the specific growth rates and algal biomass productivities of both *Chlorella* sp. and *Scenedesmus* sp., which are listed in **Table 1**. It was found that *Chlorella* sp. concentrations were 840, 725, 765, 720, and 710 mg/L, while *Scenedesmus* sp. concentrations were 745, 695, 645, 630, and 660 mg/L at the Na_2SO_4 concentrations of 30, 400, 600, 800, and 1000 mg/L, respectively. Furthermore, the specific growth rates of *Scenedesmus* sp. in **Table 1** indicate that sulfate concentrations did not influence the growth of this alga in the BG-11 medium, but the specific growth rates of *Chlorella* sp. decreased minimally as a result of the sodium sulfate addition. Studies on *Chlamydomonas moewusii* reported that sulfate concentrations higher than 3 mM increased ionic strength in the culture medium, indirectly inhibiting the growth of the alga in freshwater [26]. In this experiment, the sodium sulfate concentration of 400 mg/L (2.8 mM) resulted in the ionic strength in BG-11 medium of about 16 mM [26]; therefore, much higher ionic strength was expected at the Na_2SO_4 concentrations of 600, 800, and 1000 mg/L, respectively. However, the results of the present work indicated that high sulfate concentrations during this range influenced minimally the specific growth rates of both algae at the initial pH of 7.0, implying that both algae could possibly tolerate the medium with high ionic strength.

However, the experimental results listed in **Table 1** revealed that the algal biomass productivities of both algae were lower than the control experiments in the BG-11 medium containing only 30 mg SO_4^{2-}/L. **Figure 4(a)** exhibits that *Chlorella* sp. increased linearly as the cultivation time increased because inorganic carbon and nutrients were not limited, implying that a zero-order production rate was obtained. As the Na_2SO_4 concentration increased in BG-11 medium to 400 mg/L, it appears that *Chlorella* sp. required a lag period to acclimatize with additional sodium sulfate at least 6 days prior to grow rapidly. The pH of the medium remained approximately constant until day 6, supporting that the photosynthesis of *Chlorella* sp. was inhibited. As Na_2SO_4 increased in the medium at the concentrations of 600, 800, and 1000 mg/L, the lag periods decreased with the Na_2SO_4 concentrations because this alga responded to the inhibitory effect of ionic strength by the enhancement of photosynthetic CO_2 assimilation [27] and by the allocation of carbon and energy resources for synthesizing of organic osmolytes to tolerate the osmotic stress [26]. Furthermore, the nitrate utilization rates of *Chlorella* sp. as listed in Table 1 decreased considerably as compared with the control experiment at 400 mg/L and increased gradually with sodium sulfate concentrations, indicating the inhibitory effects induced by ionic strength on this alga. On the other hands, *Scenedesmus* sp. grew in the same way with the control experiment as illustrated by **Figure 4(b)**. However, the nitrate utilization rates immediately increased with the increase of sodium sulfate concentrations in the medium, demonstrating its efficiency of this alga for removing nitrogen under the high ionic strength of sodium sulfate. As listed in **Table 1**, phosphate removal efficiencies of both algae presented an initial decrease followed by a gradual increase for both algae as sodium sulfate concentrations increased in the medium. At the sodium sulfate concentrations of 800 and 1000 mg/L, it was found that phosphates in the culture medium containing *Chlorella* sp. were completely depleted after 9 days, explaining that phosphates were simultaneously removed by assimilation and the chemical precipitation of calcium phosphate at higher pH induced by the growth of algae [21]. The pH of the culture medium reached about 11.0 at the sodium sulfate additions of 800 and 1000 mg/L promoting the chemical precipitation, resulting in the calcium removal efficiencies of 74.1 and 52.6%, respectively. On the other hands, phosphates were assimilated for the growth of *Scenedesmus* sp. without the chemical precipitation. Calcium concentrations were remained constant in the culture medium during this study.

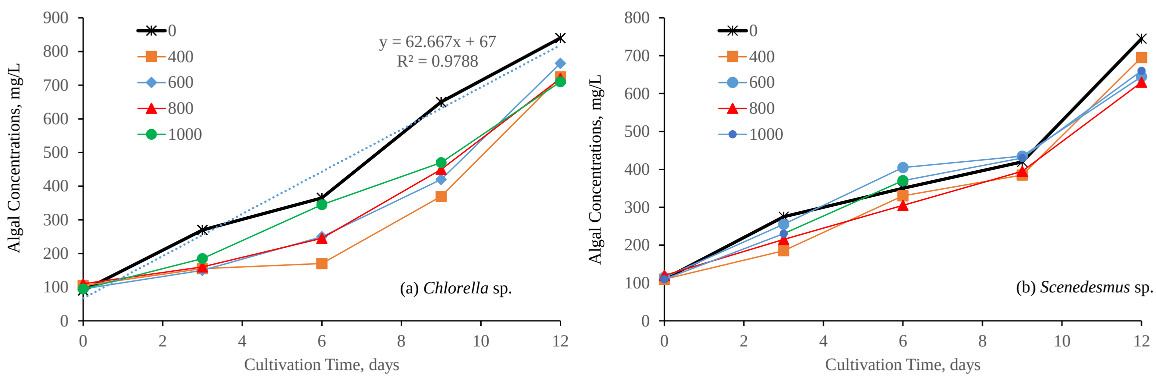

Figure 4 (a) *Chlorella* sp. and (b) *Scenedesmus* sp. concentrations in the BG-11 culture medium at different sodium sulfate concentrations and the optimum pH of 7.0

Feasible Use of Microalgae to Polish Consumer-products Effluents Containing High Sulfate and TDS Concentrations

The experimental results from the previous section revealed that both microalgae could possibly grow in the medium and utilize the nutrients including nitrogen and phosphorus. Both microalgae were grown in the effluents of the consumer-products industry. The characteristics of consumer-products industrial effluent are listed in **Table 2**. It appears that the consumer-products industrial effluent contained high sodium sulfate, phosphate, salinity, and TDS with low nitrogen; therefore, nitrogen deficiency was expected for cultivating both microalgae. However, the stock culture of microalgae in the BG-11 medium contained sufficient nitrate nitrogen; therefore, the seeding microalgae were not washed out with distilled water to clean up the nutrients. The final nitrate concentration of effluent was about 70 mg N/L.

Table 2 Effluent characteristics of the consumer-products industry

Parameters	Concentrations
pH	8.3
Total Dissolved Solids (TDS)	5700 mg/L
Total Suspended Solids (TSS)	180 mg/L
Salinity	11 ppt
Ammonium Nitrogen	ND
Nitrate Nitrogen	0.1 mg N/L
Sodium Sulfate	2042 mg/L
Calcium	18 mg Ca/L
Phosphate	608 mg P/L
Chloride	15.3 mg Cl/L
Sodium	2774 mg Na/L
Potassium	41.5 mg K/L
Magnesium	3.5 mg Mg/L

The dry weight of algal biomass and both ammonium and nitrate concentrations presented in **Figure 5** indicate that *Chlorella* sp. grew immediately in the industrial effluent containing high TDS during the first 3 days and then remained approximately constant until the end of the experiment. *Chlorella* sp. consumed nitrate nitrogen as a nitrogen source while ammonium nitrogen increased gradually after day 3 as a result of alga lysis. On the other hands, *Scenedesmus* sp began growing with an initial increase until day 6 and then declined continuously, indicating that the growth of this alga was declined. Nitrate nitrogen was remained constant after day 6. Both *Chlorella* sp. and *Scenedesmus* sp. reached the maximum concentrations of 306 and 393 mg/L because the inorganic carbon was limited as indicated by the final pH values of about 10.3 and 11.0, respectively. The specific growth rates of both *Chlorella* sp. and *Scenedesmus* sp. were 0.17(1.00) and 0.18(0.93) day^{-1}, respectively. In addition, the algal biomass productivities of *Chlorella* sp. and *Scenedesmus* sp. were 40.0(1.00) and 43.3(0.98) mg/L/day, respectively. Much lower algal biomass productivities of both *Chlorella* sp. and *Scenedesmus* sp. than the BG-11 culture medium was obtained. According to the algal growth in batch culture, the decline of both *Chlorella* sp. and *Scenedesmus* sp. concentrations after days 3 and 6 indicates that both algae were under the stationary and death phases, respectively. Both phosphate and sulfate were found excessive in the effluents; however, very little amounts of phosphate and sulfate were utilized due to the small biomass productivities of both algae. It was found that a tiny amount of calcium phosphate was precipitated due to the final pH of about 11.0 because only a small concentration of calcium existed in the effluent. It is hypothesized that high sulfate of about 14.4 mM, phosphate, and other ions resulting in high TDS concentration limited the growths of both algae due to high ionic strength [26]. Furthermore, the inorganic carbon was also depleted at the final pH of about 11.0. It is hypothesized that both algae could remove both sulfate and phosphate successfully as reported by another study [16] if the CO_2 is continuously supplied and nitrogen source is supplemented. Growth inhibition due to NaCl should be minimal because low NaCl concentration was found in the effluent. It can be concluded that *Scenedesmus* sp. could potentially tolerate better high TDS of the effluent than *Chlorella* sp. so higher the biomass productivity of *Scenedesmus* sp. than *Chlorella* sp. was obtained.

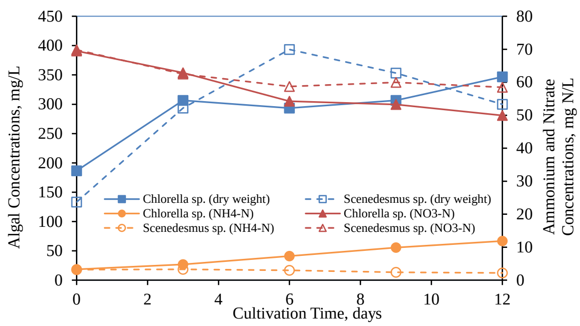

Figure 5 Algae, ammonium, and nitrate concentrations in the industrial effluents at the pH value of 7, initial algal concentration of about 200 mg/L, and TDS concentration of 5770 mg/L

To verify the effects of TDS concentrations on the growth of both algae, the effluents were diluted with different volumes of deionized (DI) water in 1-L glass bottles and added with a fixed amount of algae at about 11% (v/v) and pH of 7.0. **Figure 6** presents the algal biomass productivities of *Chlorella* sp. and *Scenedesmus* sp. cultivated in the industrial effluent diluted with DI water at the dilution rates of 0, 12.5, 25.0, and 37.5%. The TDS concentrations were 5770, 5030, 4420, and 3880 mg/L for reactors containing *Chlorella* sp. and were 5640, 5060, 4480, and 3820 mg/L for reactors containing *Scenedesmus* sp. at the dilution rates of 0, 12.5, 25.0, and 37.5%, respectively. It was found that *Chlorella* sp. increased in the effluent at the biomass productivity of 27.7 mg/L/day and the specific growth rate of 0.05(0.77) day^{-1} during the first 9 days and declined after that, indicating the death phase of this alga. However, the algal biomass productivities increased respectively to 29.7, 35.0, and 41.0 mg/L/day and the specific growth rates increased to 0.05(0.91), 0.07(0.90), and 0.08(0.88) day^{-1} when the effluent was diluted at the rates of 12.5, 25.0, and 37.5%, respectively. Furthermore, the biomass productivities of *Scenedesmus* sp. were 45.3, 53.7, 54.7, and 62.0 mg/L/day and the specific growth rates were 0.06(0.90), 0.08(0.86), 0.08(0.90), and 0.09(0.90) day^{-1} at the dilution rates of 0, 12.5, 25.0, and 37.5%, respectively. The results indicate that the biomass productivities and specific growth rates of both algae increased as the TDS concentration of effluent decreased, supporting the conclusion that the TDS of effluents inhibited the growths of both algae.

Conclusions

In this study, both *Chlorella* sp. and *Scenedesmus* sp. were cultured in the BG-11 medium to determine the optimum growth conditions under high sodium sulfate and TDS conditions. The optimum pH values for both algae were 7.0. High sulfate concentration inhibited slightly the growth of *Chlorella* sp., but did not affect the growth of *Scenedesmus* sp. However, the ionic strength of sodium sulfate reduced the biomass productivities of both algae. The photosynthesis of *Chlorella* sp. increased the pH of the medium to about 11 resulting in chemical precipitation of calcium phosphate along with phosphorus assimilation; therefore, the phosphorus was limited. The best growth conditions were applied to the consumer-products industry. It was found that the growths of both algae were limited due to the high ionic strength of effluent as a result of high TDS. Due to the low algal biomass productivities of both algae, nutrients were minimally eliminated from the effluent. It is suggested from the experimental results that both microalgae are not suggested to polish the effluent of the consumer-products industry unless the supplementations of nitrogen and inorganic carbon and the reduction of TDS concentration are implemented.

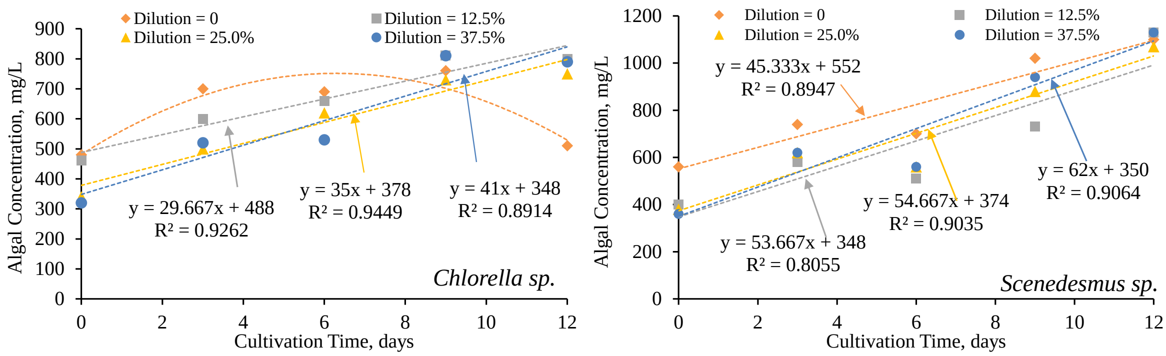

Figure 6 Algae productivities in the industrial effluents at the pH value of 7 with different dilution rates of 0, 12.5, 25.0, and 37.5%

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