Title:

Perspective assessment of algae-based biofuel production using recycled nutrient sources: The case of Japan

Authors:

Tunyen Wang*[a], Helmut Yabar [a] and Yoshiro Higano [a]

Affiliations:

[a] Graduate School of Life and Environmental Sciences, University of Tsukuba, JAPAN

* Corresponding author:

Tunyen Wang

Telephone & Fax number: +81-29-853-7221／E-mail: ianw005@gmail.com

Address: 1-1-1 Tennodai, Tsukuba, Ibaraki 305-8577, Japan

Abstract

In this study, an upper limit in the solar energy conversion efficiency which can be translated to a maximum potential algal yield of a large-scale culture is calculated based on the algal productivity model in which light and nutrient are made the growth rate limiting factors, and taking the design characteristics of the cultivation system into account. Our results indicate that for the production of low-cost biodiesel within the limits of the wastewater quality standards, that the culturing of high lipid content algae within a raceway pond would provide an appropriate solution for manufacturing biodiesel from algae. However, due to inefficient sunlight utilization and due to the large amount of fertilizer required in raceway ponds, a greater effluent recycle rate would have to be implemented to reduce the amount of fertilizer discharge to meet the wastewater quality standards and to maximize the attainable productivity of algal biomass.

Keywords:
algae; biodiesel; anaerobic digestion; nutrient recycling; fertilizer

1 Introduction

A sustainable development cannot be achieved without the support of robust energy supplies that are steady, cost-effective and low-hazard. Biofuels that can substitute fossil fuels are a hot topic today in many countries around the world. Algae as feedstock for biofuels have been highly regarded for several decades, due to its high biomass productivity and no competition with food supplies (Chisti, 2008; Mata et al., 2010; Sheehan et al., 1998). Despite these advantages, production of algae has larger potential environmental impacts in energy use, greenhouse gas emissions and water use, compared to conventional crops from a life cycle perspective (Clarens et al., 2010; Lardon et al., 2009).

Lardon et al. (2009) and Clarens (2010) et al. have also determined the impacts associated with fertilizer demand for algal cultivation according to the algae composition and life cycle inventory database, respectively, despite there are not many analyses focused on this topic. Their results showed fertilizer demand is a critical challenge for algae-derived biofuel due to the fact that mass algal cultivation requires significant amounts of nitrogen fertilizer, and production of chemical fertilizer is a principal burden driver of greenhouse gas emissions and eutrophication. Utilization of the wastewater as nutrient sources and recycling the spent biomass contained nutrients were suggested as a way to reduce the fertilizer consumptions and nutrient discharges. However, the actual environmental impact of fertilizer discharge may be underestimated since the algal culture must be enriched with nutrients to be viable for commercial purposes. For instance, the ammonium concentrations contained in enrichment media that are suitable for mass production of microalgae in large-scale extensive systems are generally above 100 mg per liter (Lavens & Sorgeloos, 1996), which can increase the fertilizer discharge beyond expected levels.

To mitigate the environmental impact of the consumption of great quantities of energy, anaerobic digestion used for energy recovery from microalgae residues after biofuel production was theoretically estimated in the studies of Chisti (2008), Sialve et al. (2009) and Heaven et al. (2011). The technology for anaerobic digestion using microalgae residues was further experimentally investigated and has been proved to be feasible by Ehimen et al. (2009; 2011). Harun et al (2011) and Davis et al. (2011) conducted techno-economic analyses on the integration of biofuel production systems with biogas production. Both of their studies completely assessed the processes from algae growth to oil refining and also compared two common types of algal cultivation systems, i.e. the raceway ponds and photobioreactors (PBRs). Their results underscored that the utilization of biogas can not only contribute to cost reduction of algal-derived biofuel, but can also indirectly lower carbon emissions resulting from the consumption of energy supplied by a power grid. However, these calculations were respectively based on using a given annual yield of algal-derived biofuel.

In this study, we presents a preliminary attempt to further evaluate the simultaneous biofuel production systems with biogas using algae, by establishing a detailed modeling method that can more effectively reflect the influence of vital factors of nutrient and light on algal growth.

2 Methods

In this section, an approach to calculation of the production of biofuel and biogas from algae in closed cultivation systems is presented. Basing on the thermodynamic model, a theoretical attainable algal productivity, as well as the yields of biofuel and biogas can be calculated gradually with the given available solar irradiation and nutrient loading. Next, to assess the environmental impact from nutrient discharge, three scenarios of nutrient supply designed for reducing chemical fertilizers use are considered. Also, the cases assume that technology improvement is applied in algal cultivation are taken into consideration, in order to discuss the possible limitation on mass production of algae for biofuels in the future. The scheme of a serial of models/calculations is shown as Fig. 1.

2.1 Theoretical maximum yield of algae

Considering the major environmental factors of light intensity (I), nutrients (S) and temperature (T), the photochemical conversion of radiant energy into algal biomass growth (p) over a period of time, following the basic concepts of Shelef et al. (1968) and Goldman (1979), can be described as:

$$p = (E_S / E_a) * I * S * T - D_r \quad (1)$$

where E_S is the total solar irradiation in terms of joules per area per time, E_a is the heat of combustion per unit mass of algae, and D_r is the overall decay in terms of grams of dry weight (DW) per area per time.

Goldman (1979) calculated the theoretical maximum yield of microalgae by considering physical laws. According to his study, a theoretically production rate of 60 g m^{-2} d^{-1} was possible. However, it should be considered that only 30-40 g m^{-2} d^{-1} was practical and that the production rate of microalgae was assessed in open ponds. In contrast, Weyer et al. (2010) and Zemke et al. (2010) developed similar theoretical models for estimating the thermodynamically possible production rates of microalgae, focused on projecting annual biodiesel productivity by converting available solar energy to algal lipids. They also considered the factor in a variety of cultivation systems. Those models employed basic physical laws, making the calculations of the theoretical upper limit of algae production more robust and variable in different growth locations by adjusting the given conditions of solar irradiation. However, the cultivation of algae using wastewater was not considered during their studies. Thus, attempts have been made to incorporate this issue into the models used in this paper, through subtle variation.

Ignoring decay and the factor of temperature, the equation of algal mass productivity in terms of mass of algae per area of land per time is rearranged as follows:

$$p = \left(E_{\mathrm{S}}/E_{\mathrm{a}}\right)\left(\tau \varepsilon_{\mathrm{a}} C_{\mathrm{PAR}}\right)\varepsilon_{\mathrm{s}} \quad (2)$$

where τ is the sunlight transmission efficiency and ε_{a} is the solar energy capture efficiency of the microalgae, used to express two phases of the transmission of sunlight to the surface of the algal mass and the absorption of light by the algae cell. C_{PAR} is the fraction of the solar spectrum corresponding to photosynthetically active radiation (PAR). Herein, ε_{S}, representing the factor of limited nutrient supply, is given by as:

$$\varepsilon_{\mathrm{s}} = S_{\mathrm{n}}/\left(K_{\mathrm{S}} + S_{\mathrm{n}}\right) \quad (3)$$

where S_{n} is set as a particular limiting nutrient by assuming all other nutrients are supplied in excess, and K_{S} is the half saturation constant.

2.2 Model and scenario design

2.2.1 Scenario design for nutrients supply

Chlorella vulgaris and *Botryococcus braunii*, two of the most researched microalgae as biodiesel feedstock, were selected as representative species for the analysis. Although the composition of the microorganism typically changes with the culture conditions, such as nitrogen concentration or salt stress (Brennan & Owende, 2010; Kumar et al., 2010; Lakaniemi et al., 2011; Mata et al., 2010; Mulbry et al., 2005), in our study the protein, lipid and carbohydrate fraction of the microalgae, based on the reports of Thomas et al. (1984) and Illman et al. (2000), were constant for the sake of computational convenience and also due to the reason of lack of data.

With regard to the condition under depletion of important nutrients such as nitrogen or phosphorous, nutrient starvation is widely applied in microalgal triacylglyceride production as a lipid induction technique. However, lipid accumulation in the algal cell attained through nitrogen-deficiency, as one of the conclusions that quoted in the literature review conducted by Rodolfi et al. (2009), does not increase oil productivity due to the higher lipid accumulation usually accompanies more decreases in growth rate. Thus, the regardless of nutrient starvation as a practicable process does not make a limitation on the assessment of the theoretical maximum oil production in this study. However, it should be noted that since the variation in chemical composition of algal cell with nutrient loading is ignored, a bias may exist in present model for projections of biofuel and biogas production despite it should be in a rational range while variation in algal biomass concentration with nutrient loading is considered in our analysis.

Lastly, to understand how the proposed closed cycle system will reduce the environmental load resulting from excessive nutrients, three nutrients supply scenarios were used for the comparison (see Fig. 2.). It was assumed that nutrients were available from the following four sources: (i) secondary treated sewage (STS) from sewage treatment plants (S_{STS}); (ii) extra inputs (S_{input}); (iii) the digestate from an anaerobic fermenter (S_{DIG}); and (iv) the effluent from culture medium after isolating the algal cells (S_{out}). In Scenario 1, the nitrogen requirement was supplied by STS only ($S_{\mathrm{in}} = S_{\mathrm{STS}}$). In Scenario 2, the nitrogen requirement was supplied with STS, extra inputs and digestate ($S_{\mathrm{in}} = S_{\mathrm{STS}} + S_{\mathrm{input}} + S_{\mathrm{DIG}}$).whereas in scenario 3 it was supplied by extra inputs and effluent ($S_{\mathrm{in}} = S_{\mathrm{input}} + S_{\mathrm{DIG}}$).

2.2.2 Modeling for the closed algal cultivation system

Based on equation (2), the total annual algal biomass yield (Y_{a}) using a closed cycle system is predicted as follows:

$$Y_{\mathrm{a}} = \sum_{t=365}^{T} y_{\mathrm{a}}(t) = \sum_{t=365}^{T} p(t)A = \sum_{t=365}^{T} \left[\varepsilon_{\mathrm{s}}(t) \left(\tau \varepsilon_{\mathrm{a}} C_{\mathrm{PAR}} E_{\mathrm{S}} \right)(t) / E_{\mathrm{a}} \right] A \quad (4)$$

$$\varepsilon_{\mathrm{s}}(t) = S_{\mathrm{n}}(t) / \left(K_{\mathrm{S}} + S_{\mathrm{n}}(t) \right) \quad (5)$$

where Y_{a} is expressed in terms of kg DW of algae a year, y_{a} is the daily yield of algal biomass expressed in terms of kg DW of algae, and A is the algae cultivation area expressed in m^2.

The total concentration of nutrient in the culture medium (i.e. the concentration of T-N) was calculated separately for the three scenarios. The rate of nutrient change in the medium is given as:

$$\begin{gathered} \text{Rate of change} = -\,\text{Consumption} + \text{Input} - \text{Output} \\ \Delta\left(SV\right)/\Delta t = -\nu x V + F_{\mathrm{in}} S_{\mathrm{in}} - F_{\mathrm{out}} S_{\mathrm{out}} \end{gathered} \quad (6)$$

where V is a unit volume of culture medium, v is the specific rate of substrate consumption x is the biomass concentration in terms of biomass per volume and F is the medium flow rate. In scenarios 1 and 2, where continuous culture was assumed to occur, once the culture was in a steady state, the rate of change in nutrient concentration would be zero. That is to say, to maintain a steady state, the amount of nutrient inputs has to equate to the amounts of the nutrients that are consumed by algal biomass and that flowed away. Thus,

$$S_{\mathrm{n}}(t) = S_{\mathrm{STS}}(t) + S_{\mathrm{input}}(t) + S_{\mathrm{DIG}}(t) + \Delta S_{\mathrm{n}}(t-1) \quad (7)$$

However, S_{input} and S_{DIG} are zero in scenario 1 and

$$S_{\mathrm{DIG}}(t) = y_{\mathrm{NH3\text{-}N}}(t) d^{-1} V^{-1} \quad (8)$$

where $y_{\mathrm{NH3\text{-}N}}$ is the theoretical NH_3-N recovery from anaerobically digested residuals expressed in mg NH_3-N of total solids (TS), and d is the ratio of the flow rate to the culture volume and is called the *dilution rate*. Compared with a continuous culture, a closed cultivation system in which nutrient-contented effluent is recycled for biomass culture can be regarded as a batch culture. Thus,

$$\Delta S / \Delta t = -v x \quad (9)$$

According to Tam and Wong (1996), the algal biomass concentration (mg L^{-1}) and the average daily nitrogen removal efficiency (mg NH_3-N L^{-1}) of *C. vulgaris* under different NH_3-N concentrations can be given as follows (see Fig. 3.). The average daily nitrogen removal efficiency of microalgae (R_{N})

within the range of 1 to 1000 mg NH_3-N L^{-1} is given by:

$$R_{\mathrm{N}} = 0.5131 S_{\mathrm{n}}^{0.3779} \quad (10)$$

The algal biomass concentration within the range of 10 to 50 mg NH_3-N L^{-1} is given by (see Fig. 4 (a)):

$$x = -0.3201 S_{\mathrm{n}}^{2} + 29.12 S_{\mathrm{n}} + 49.051 \quad (11)$$

The algal biomass concentration within the range of 51 to 1000 mg NH_3-N L^{-1} is given by (see Fig. 4 (b)):

$$x = -0.0002 S_{\mathrm{n}}^{2} + 0.0848 S_{\mathrm{n}} + 701.62 \quad (12)$$

Assuming that the variation of nutrient concentration due to algal assimilation is totally converted into biomass, then:

$$\nu = R_{\mathrm{N}} x^{-1} \quad (13)$$

For a continuous cultivation using recycled effluents, as in scenario 3, nutrient replenishment was required after nutrients were assimilated by microorganisms to sustain or enhance the biomass production capacity. Therefore, by taking effluent recycling rate (q) into account,

$$S_{\mathrm{n}}(t) = S_{\mathrm{in}}(t) + q S_{\mathrm{out}}(t-1) \quad (14)$$

Herein,

$$S_{\mathrm{out}}(t) = S_{\mathrm{n}}(t) - \Delta S(t) = S_{\mathrm{n}}(t) - \nu x(t) \quad (15)$$

$$S_{\mathrm{in}}(t) = -\Delta S(t-1) + (1-q) S_{\mathrm{out}}(t-1) = S_{\mathrm{input}}(t) + S_{\mathrm{DIG}}(t) \quad (16)$$

$$S_{\mathrm{DIG}} = y_{\mathrm{NH3\text{-}N}} V^{-1} \quad (17)$$

When the composition of the organic matter and the daily yield of algal biomass (y_a) are known, it is possible to calculate a theoretical specific methane yield associated with a theoretical ammonia release by using the formula adapted from Symons and Buswell (1933) (Angelidaki & Sanders, 2004; Heaven et al., 2011; Sialve et al., 2009). Table 1 shows the theoretical methane yields and ammonia concentrations for three types of organic compounds.

2.3 Cultivation system geometry and production costs

Two typical types of algal cultivation systems were discussed in this study; raceway ponds and horizontal tubular photobioreactor. The volume and area required, as well as energy consumption in the processes from biomass cultivation to dewatering, are scaled up or down for the basic case of one hectare scale, based on the design of Benemann and Oswald (1996) and Tapie and Bernard (1988) (see Table 2. and Table 3., respectively.).

In addition, the presuppositions for the economic analysis are: (i) a continuous supply of free CO_2 is available from flue gas, and a small amount of organic carbon is assumed to be supplied from recycled medium or digester effluent; and (ii) the nutrients required by the algae, notably nitrogen and phosphorous, have been assumed to be provided by animal manure. Carbon and fertilizer costs have therefore been excluded from the operating costs.

2.4 Cases of improving cultivation technology

To address the difficulty of developing future algal cultivation systems, four cases were used to compare the calculation results: (i) Base Case:

current technology of raceway ponds and PBRs; (ii) Case 1: improving the technology of raceway ponds; (iii) Case 2: improving the technology of PBRs; and (iv) Case 3: highly improving the technology of PBRs. The Base Case was based on the literature data. Cases 1 to 3 were based on the assumption that cultivation technology has improved (see Table 4.).

To illustrate the simulation model of algal biomass cultivation using closed recycle system, Tsukuba, a city located 65 km north of Tokyo, Japan, was selected as a demonstration example because of the moderate climate conditions and the average daily solar radiation in this region, which approximated to the national average observed in Japan over the last 30 years. The average daily solar radiation for January to December is between 8.35 and 17.56 MJ m^{-2} d^{-1}. In addition, owing to the proposed scenarios in which the domestic wastewater was assumed to be an option of nutrient sources, nearby Lake Kasumigaura, the second-largest lake in Japan, was identified as potential site for providing the nutrients for algal cultivation. The average daily T-N concentration among STS from the sewage treatment plant of Kasumigaura was 6 mg L^{-1}.

.

3 Results and Discussion

3.1 Algae-to-oil system as tertiary advantage wastewater treatment

As previously mentioned, it is practicable to grow lipid-rich algae with STS from wastewater treatment plants (Doušková et al., 2010; Órpez et al., 2009; Sawayama et al., 1992). In this study, Scenario 1 was designed to assess the feasibility of the idea of a comprehensive approach to energy and water management using algal biotechnology.

In our view, from an economic perspective, raceway ponds are more suitable than PBRs for the cultivation of algae and simultaneously provide the tertiary advantage of wastewater treatment. Nevertheless, the calculated net production costs of biodiesel from *C. vulgaris* and *B. braunii* presented the very costly results of ¥2,595 L^{-1} and ¥1,213 L^{-1}, respectively (Case 1). This resulted from a poor harvest of algal biomass caused by the nutritional deficiency in STS. The small population of organism also led to limitations in their nitrogen removal performance from wastewater. The maximum average daily nitrogen removal rates (or uptake rates) of *C. vulgaris* and *B. braunii* were calculated for scenario 1 to be 0.25 and 0.221 mg N L^{-1} d^{-1}, respectively. Both results were found to be lower than the experimental results of 1.05 and 0.852 mg N L^{-1} d^{-1} for *C. vulgaris* and *B. braunii*, respectively, as obtained from the studies of Kima and Lingarajua et al. (2010) and Sawayama et al. (1992). By converting with the annual yields of biomass, the total annual nitrogen uptake by algae was 126 kg N ha^{-1} yr^{-1} for *C. vulgaris* and 111 kg N ha^{-1} yr^{-1} for *B. braunii*.

In summary, the results indicated that algal cultivation in which only the STS used has a significant effect on the purification of wastewater would be ideal. However, if the purpose were for maximizing the bio-energy generation and minimizing its production cost, then the function as tertiary advantage wastewater treatment would be eliminated because large-scale cultivation always requires additional nutrient inputs that are accompanied by nutrient discharges from effluents.

3.2 Maximum biodiesel productivity and minimum production cost

Consideration of Cases 2 and 3 allowed for the realization that ideal algal productivity would be difficult to achieve under the limitations imposed by current algal cultivation technology. A maximum biodiesel productivity of 119,676 L ha^{-1} yr^{-1} was obtained in Case 3 involving the cultivation of *B. braunii* with PBRs at a net production cost of ¥236 L^{-1}. A minimum net biodiesel production cost was obtained in Case 1 involving the cultivation of *B. braunii* with raceway ponds, which reduced production costs to ¥193 L^{-1} at an annual biodiesel productivity of 53,854 L ha^{-1} yr^{-1}. The above results provide a clear indication that a lipid-rich algal species would be required to be cultured for biodiesel production. Depending on these calculated values of biodiesel production costs, the cost with raceway ponds cultivation was 22% less than with PBRs. However, owing to the limitation in technology of raceway ponds, biodiesel yields in raceway ponds cultivation were almost half of that obtained with PBRs cultivation.

From an energy balance perspective, the results indicated that raceway pond cultivation should be used for producing biodiesel from algae, in spite of its lower productivity. However, in a country that has small land area such as Japan, a massive cultivation using raceway pond technology may not only create conflicts over land resources but may also raise the production costs. Thus, in such countries, a new PBRs technology requiring less energy usage and with a high efficiency of land use would be the key to driving further cost reductions.

3.3 Minimizing fertilizer use and discharge in biodiesel production

Given that large-scale cultivation for algae-based biodiesel production not only requires a large amount of water and fertilizer usage but also causes plenty of fertilizer discharges at the same time, the nutrient-rich effluents have to be properly treated.

According to the latest revision to national wastewater treatment standards promulgated by the Ministry of the Environment of Japan, a permit average limit for nitrogen of 150 mg L^{-1} a day was set for agriculture wastewater. Considering the reuse of the culture medium as the only way to meet the effluent quality standards in this study, the amount of nitrogen used and discharged was calculated for scenarios 2 and 3, and were assessed according to the following assumptions: (i) the sources of nutrients based on priority order of utilization in scenario 2 are STS, digestate from anaerobic digestion and extra input; (ii) the sources of nutrients based on priority order of utilization in scenario 3 are reclaimed effluent water, digestate from anaerobic digestion and extra input; and (iii) if only a given nutrient level was reached, then any excessive amounts of nutrients fall into disuse and fertilizer discharge would occur.

Depending on the results of the calculation, the demand for additional nitrogen input and the total nitrogen discharge both revealed a trend towards decline occurring at a similar rate, with an increase in the ratio of the use of reclaimed medium. A greater demand for additional nitrogen input and total nitrogen discharge was observed in biodiesel production using *B.*

braunii in comparison to *C. vulgaris*, resulting from the smaller available quantity of digestate encountered using *B. braunii* in contrast to *C. vulgaris*. This feature of growing these two different strains of algae for oil production, however, resulted in a reverse effect in the cases where nutrient levels were limited (S_n is less than 300 mg N L^{-1}) and effluent recycle rates were high (greater than 0.7). As shown in Fig. 5., the total nitrogen discharge figures from cultivation using *C. vulgaris* were greater numbers than those encountered when using *B. braunii* because its demands for nutrient supply were almost satisfied with reclaimed medium, which resulted in an increase in the amounts of disused excessive nitrogen from digestate.

Under an effluent limitation for wastewater quality of 150 mg N L^{-1}, the production of lipids extracted from algal biomass and used for manufacturing biodiesel showed a 17% decline compared with projected maximum biodiesel production in every case. Associated with the increases on culture medium recycle ratio, the production of biodiesel has been improved because most of the nitrogen contained in effluent has been reused, and therefore, it was permitted to grow algae in the medium with higher nitrogen concentration levels, making the cultivation system more effective.

The variance of volumes experienced between the raceway ponds and PBRs creates a fundamental difference in the requirements and discharge of fertilizer. Based on the parameters adapted in this study for cultivation system designs, when the nitrogen concentration level was set to 1000 mg N and there was no effluent to be recycled, the results indicated that 489 to 493 ton N and 248 to 255 ton N per hectare per year were required as additional nitrogen inputs when using raceway ponds and PBRs, respectively. In contrast, the nitrogen discharges were forecasted to be approximately 500 and 260 ton N per hectare per year for cultivation using raceway ponds and PBRs, respectively.

As the effluent recycle rate was increased to 0.5, as in Case 1 involving the culturing of *B. braunii* in a raceway pond, the demand for additional nitrogen input was reduced from 491 to 248 ton N ha^{-1} yr^{-1}, accompanied by a decrease in the discharge of nitrogen from 499 to 251 ton N ha^{-1} yr^{-1} and in biodiesel production costs from ¥230 L^{-1} to ¥208 L^{-1}. With the same recycle rate of q = 0.5 for Case 3 involving the culturing of *B. braunii* in a PBR, additional nitrogen input was reduced from 252 to 123 ton N ha^{-1} yr^{-1}. The nitrogen discharge was also reduced from 259 to 130 ton N ha^{-1} yr^{-1}, accompanied by a reduction in biodiesel production costs from ¥273 L^{-1} to ¥251 L^{-1} (see Fig. 6.)

In summary, the results indicate that for the production of low-cost biodiesel within the limits of the wastewater quality standards, that the culturing of high lipid content algae within a raceway pond would provide an appropriate solution for manufacturing biodiesel from algae. However, due to inefficient sunlight utilization and due to the large amount of fertilizer required in raceway ponds, a greater effluent recycle rate would have to be implemented to reduce the amount of fertilizer discharge to meet the wastewater quality standards and to maximize the attainable productivity of algal biomass. The reuse of the culture medium is an important consideration for the large-scale production of algae. Several investigations, including reports by Burlew (1953), Leone (1963) and Kim et al. (2011), have demonstrated that the reuse of medium could be accomplished without special treatment and that there was no inhibition observed during cultivation periods of 16 to 72 days. These results suggest that the reuse or recycling of the culture medium is not only a practical procedure for algal cultivation, but also an important measure to conserve water and nutrients in any application of an algal system in a closed ecological system.

3.4 Opportunities for algal biodiesel production in Japan

Currently, microalgae are regarded as a suitable alternative feedstock for biodiesel production. To achieve their full processing capabilities, recent research efforts have concentrated on genetic engineering to identify an algal species optimized for high productivity and energy value. In addition, many different designs of algal culture systems such as PBRs have been developed with the aim of using the most effective and economical technology to produce large amounts of oil within large-scale algal cultivation units. However, in the current technology of algal cultivation, the raceway pond is still a low-cost method for producing biodiesel from algae despite its poor biomass productivity and high contamination risk. From our results for the scenario aimed at minimizing production cost, it is clear that the raceway pond cultivation technique required 545,120 ha of culture lands for a biodiesel supply of 29,357,000 kl, based on the diesel consumption in the transportation sector in 2008 Japan. Since the cost of land use can multiply the price of algae-derived biodiesel, the problem with using a large area of land may become a challenge for promoting the algae-derived biodiesel policy within an island country like Japan.

Application of a high productivity of biomass cultivation technique such as PBRs represents a better use of land. The projected per unit area of biodiesel output of PBRs was 1.3 times that of raceway ponds for the Base Case and was up to 2.8 times greater for Case 3. This means that the demand for land can be reduced at the same rate to produce an equivalent amount of biodiesel from algae using PBRs; for example, to meet the total consumption of diesel in the transportation sector in Japan, 195,000 and 419,000 ha would be required for algal cultivation in Case 3 and Base Case, respectively.

One of the main problems with PBRs is the requirement for a large energy input. Combining biomass gasification plants with the algal cultivation system as an integrated approach to algal biodiesel production and CO_2 utilization is possible to make up for the shortage of power required by PBRs, amounting to a projected reduction in the biodiesel production cost of up to 22% for the Base Case, or 20% from ¥236 to ¥189 L^{-1} for Case 3. Given the current pump price for petroleum diesel fuel that reported at ¥121 L^{-1} in July 2012, the currently projected costs of algae-based biodiesel are high. However, in the near future, this hybrid biomass energy generation system simultaneously providing power and fuels when they are consumed on site may provide a practicable measure for reducing our dependence on fossil fuels. Long-term basic and applied R&D is required to develop algal technology, as one of the many options that will be required in the future to reduce Japanese imports of oil and other foreign energy sources.

4 Conclusions

A potential problem for fertilizer discharge from mass cultivation of microalgae was studied. The writers have developed a theoretical approach to the modelling of the attainable biodiesel production with simultaneous biogas yield in algae and showed that nutrient recycling should be considered as a necessary process to reduce fertilizer discharges. Our results indicated the increase in cultivation system's illuminated surface can contribute to the cost reduction, however, it is still not cost-effective to compete with fossil fuels. Thus, future research should not only focus on improving cultivation technologies, but also explore the mechanism of algal lipid accumulation.

References

Angelidaki, I., Sanders, W. 2004. Assessment of the anaerobic biodegradability of macropollutants. *Reviews in Environmental Science and Biotechnology*, **3**(2), 117-129.

Benemann, J.R., Oswald, W.J. 1996. Systems and Economic Analysis of Microalgae Ponds for Conversion of CO2 to Biomass. Final Report to the Pittsburgh Energy Tecfmology Center. *U.S. Dept. of Energy*.

Brennan, L., Owende, P. 2010. Biofuels from microalgae—A review of technologies for production, processing, and extractions of biofuels and co-products. *Renewable and Sustainable Energy Reviews*, **14**(2), 557-577.

Burlew, J.S. 1953. Algal culture: from laboratory to pilot plant. Washington.

Chisti, Y. 2008. Biodiesel from microalgae beats bioethanol. *Trends in Biotechnology*, **26**(3), 126-131.

Clarens, A.F., Resurreccion, E.P., White, M.A., Colosi, L.M. 2010. Environmental Life Cycle Comparison of Algae to Other Bioenergy Feedstocks. *Environmental Science & Technology*, **44**(5), 1813-1819.

Davis, R., Aden, A., Pienkos, P.T. 2011. Techno-economic analysis of autotrophic microalgae for fuel production. *Applied Energy*, **88**(10), 3524-3531.

Doušková, I., Kaštánek, F., Maléterová, Y., Kaštánek, P., Doucha, J., Zachleder, V. 2010. Utilization of distillery stillage for energy generation and concurrent production of valuable microalgal biomass in the sequence: Biogas-cogeneration-microalgae-products. *Energy Conversion and Management*, **51**(3), 606-611.

Ehimen, E.A., Connaughton, S., Sun, Z., Carrington, G.C. 2009. Energy recovery from lipid extracted, transesterified and glycerol codigested microalgae biomass. *GCB Bioenergy*, **1**(6), 371-381.

Ehimen, E.A., Sun, Z.F., Carrington, C.G., Birch, E.J., Eaton-Rye, J.J. 2011. Anaerobic digestion of microalgae residues resulting from the biodiesel production process. *Applied Energy*, **88**(10), 3454-3463.

Goldman, J.C. 1979. Outdoor algal mass cultures--II. Photosynthetic yield limitations. *Water Research*, **13**(2), 119-136.

Harun, R., Davidson, M., Doyle, M., Gopiraj, R., Danquah, M., Forde, G. 2011. Technoeconomic analysis of an integrated microalgae photobioreactor, biodiesel and biogas production facility. *Biomass and Bioenergy*, **35**(1), 741-747.

Heaven, S., Milledge, J., Zhang, Y. 2011. Comments on 'Anaerobic digestion of microalgae as a necessary step to make microalgal biodiesel sustainable'. *Biotechnology Advances*, **29**(1), 164-167.

Illman, A.M., Scragg, A.H., Shales, S.W. 2000. Increase in Chlorella strains calorific values when grown in low nitrogen medium. *Enzyme and Microbial Technology*, **27**(8), 631-635.

Kim, D.-G., La, H.-J., Ahn, C.-Y., Park, Y.-H., Oh, H.-M. 2011. Harvest of Scenedesmus sp. with bioflocculant and reuse of culture medium for subsequent high-density cultures. *Bioresource Technology*, **102**(3), 3163-3168.

Kima, J., Lingarajua, B., Rheaumeb, R., Leea, J.-Y., Siddiqui, K. 2010. Removal of Ammonia from Wastewater Effluent by Chlorella Vulgaris. *Tsinghua Science & Technology*, **15**(4), 391-396.

Kumar, M.S., Miao, Z.H., Wyatt, S.K. 2010. Influence of nutrient loads, feeding frequency and inoculum source on growth of Chlorella vulgaris in digested piggery effluent culture medium. *Bioresource Technology*, **101**(15), 6012-6018.

Lakaniemi, A.-M., Hulatt, C., Thomas, D., Tuovinen, O., Puhakka, J. 2011. Biogenic hydrogen and methane production from Chlorella vulgaris and Dunaliella tertiolecta biomass. *Biotechnology for Biofuels*, **4**(1), 34.

Lardon, L., Hélias, A., Sialve, B., Steyer, J.-P., Bernard, O. 2009. Life-Cycle Assessment of Biodiesel Production from Microalgae. *Environmental Science & Technology*, **43**(17), 6475-6481.

Lavens, P., Sorgeloos, P. 1996. Manual on the production and use of live food for aquaculture. 04299345.

Leone, D.E. 1963. Growth of Chlorella pyrenoidosa in Recycled Medium. *Applied microbiology*, **11**, 427-9.

Mata, T.M., Martins, A.A., Caetano, N.S. 2010. Microalgae for biodiesel production and other applications: A review. *Renewable and Sustainable Energy Reviews*, **14**(1), 217-232.

Mulbry, W., Westhead, E.K., Pizarro, C., Sikora, L. 2005. Recycling of manure nutrients: use of algal biomass from dairy manure treatment as a slow release fertilizer. *Bioresource Technology*, **96**(4), 451-458.

Órpez, R., Martínez, M.E., Hodaifa, G., El Yousfi, F., Jbari, N., Sánchez, S. 2009. Growth of the microalga Botryococcus braunii in secondarily treated sewage. *Desalination*, **246**(1-3), 625-630.

Rodolfi, L., Chini Zittelli, G., Bassi, N., Padovani, G., Biondi, N., Bonini, G., Tredici, M.R. 2009. Microalgae for oil: Strain selection, induction of lipid synthesis and outdoor mass cultivation in a low-cost photobioreactor. *Biotechnology and Bioengineering*, **102**(1), 100-112.

Sawayama, S., Minowa, T., Dote, Y., Yokoyama, S. 1992. Growth of the hydrocarbon-rich microalga Botryococcus braunii in secondarily treated sewage. *Applied Microbiology and Biotechnology*, **38**(1), 135-138.

Sheehan, J., Dunahay, T., Benemann, J., Roessler, P. 1998. A Look Back at the U.S. Department of Energy's Aquatic Species Program: Biodiesel from Algae; Close-Out Report. National Renewable Energy Lab., Golden, CO. (US). NREL/TP-580-24190; TRN: US200420%%272.

Shelef, G., Oswald, W.J., Golueke, C.G. 1968. Kinetics of algal systems in waste treatment. Light intensity and nitrogen concentration as growth-limiting factors. *SERL Report*, **68**(4).

Sialve, B., Bernet, N., Bernard, O. 2009. Anaerobic digestion of microalgae as a necessary step to make microalgal biodiesel sustainable. *Biotechnology Advances*, **27**(4), 409-416.

Symons, G.E., Buswell, A.M. 1933. The Methane Fermentation of Carbohydrates1,2. *Journal of the American Chemical Society*, **55**(5), 2028-2036.

Tam, N.F.Y., Wong, Y.S. 1996. Effect of ammonia concentrations on growth of Chlorella vulgaris and nitrogen removal from media. *Bioresource Technology*, **57**(1), 45-50.

Tapie, P., Bernard, A. 1988. Microalgae production: Technical and economic evaluations. *Biotechnology and Bioengineering*, **32**(7), 873-885.

Thomas, W.H., Tornabene, T.G., Weissman, J. 1984. Screening for lipid yielding microalgae: activities for 1983. Final subcontract report. SERI/STR-231-2207; Other: ON: DE84004500.

Weyer, K., Bush, D., Darzins, A., Willson, B. 2010. Theoretical Maximum Algal Oil Production. *BioEnergy Research*, **3**(2), 204-213.

Zemke, P.E., Wood, B.D., Dye, D.J. 2010. Considerations for the maximum production rates of triacylglycerol from microalgae. *Biomass and Bioenergy*, **34**(1), 145-151.

Table captions

Nomenclature

Table 1. Theoretical methane yields for three types of organic compounds and ammonia concentrations for digested algal biomasses from the empirical formula of microalgae using the Buswell equation.

Table 2. Geometry parameters of raceway ponds and PBRs.

Table 3. Capital and operating cost parameters for algae production with raceway ponds and PBRs.

Table 4. Parameters for improving cultivation technology cases.

Table 5. Maximum production of biomass, biogas and biodiesel (Cases 1, 2 and 3).

Figure captions

Fig. 1. The scheme of a serial of calculations.

Fig. 2. Scenarios for different nutrient supplies.

Fig. 3. Average daily NH3-N removal efficiency of C. vulgaris.

Fig. 4. Biomass concentration of C. vulgaris under different NH3-N concentration (a) 0-50 mg NH3-N L-1; (b) 50-1000 mg NH3-N L-1.

Fig. 5. Nitrogen discharged by per kilogram algal biomass production in medium of Sn= 150 mg N L-1.

Fig. 6. Variation of biodiesel productivity and the net production cost of biodiesel for different ratios of effluent reuse under a nitrogen discharge limit of 150 mg N L-1.

Nomenclature

A	algae cultivation area (m^2)	S_{STS}	T-N concentration of the STS from sewage treatment plants (mg L^{-1})
C_{PAR}	fraction of sunlight that is photosynthetically active radiation	T	temperature factor
d	(daily) dilution rate (%)	V	total cultivation volume (L)
D_r	overall decay	x	algal biomass concentration
E_a	heat of combustion per unit mass of algae (MJ kg^{-1})	Y_a	total annual algal biomass yield (kg DW)
E_S	(daily) total solar irradiation (MJ m^{-2})	y_a	(daily) algal biomass yield (kg DW)
F_{in}	medium inflow rate	$y_{NH3\text{-}N}$	(daily) NH_3-N recovery from anaerobically digested residuals (mg NH_3-N TS)
F_{out}	medium outflow rate		
I	light intensity factor	*Greek letters*	
K_s	half-saturation constant (mg L^{-1})	ε_a	efficiency of conversion of incident sunlight to biomass in algae
p	algae productivity per time per area	ε_s	limiting nutrient supply factor
q	effluent recycling rate	ν	specific rate of substrate consumption
R_N	average daily nitrogen removal efficiency of algae	τ	efficiency of light transmission to algae
S	nutrients factor		
S_{DIG}	T-N concentration of digestate from anaerobic fermenter (mg L^{-1})	*Abbreviations*	
S_{in}	concentration of nutrient in the inflowing medium (mg L^{-1})	DW	dry weight
S_{input}	T-N concentration of extra nutrient inputs (mg L^{-1})	PBR	photobioreactor
S_n	specific limiting nutrient (mg L^{-1})	STS	secondarily treated sewage
S_{out}	concentration of nutrient in the effluent (mg L^{-1})	TS	total solids

Table 1. Theoretical methane yields for three types of organic compounds and ammonia concentrations for digested algal biomasses from the empirical formula of microalgae using the Buswell equation.

	Empirical formula (moles)				% in	% in	Methane yield	NH_3-N
	C	H	O	N	*C. vulgaris*	*B. braunii*	(l CH_4 g^{-1} TS)	(mg NH_3-N g^{-1} TS)
Protein	1.9	3.8	1.0	0.5	29	22	0.446	140.8
Carbohydrate	6.0	10.0	5.0	0.0	51	14.1	0.415	0.0
Lipid	57.0	104.0	6.0	0.0	18	44.5	1.014	0.0

Table 2. Geometry parameters of raceway ponds and PBRs.

Parameter	Raceway ponds	PBRs	Units
Illuminated surface	6,899	7,200	(m^2)
Space required	10,000	10,000	(m^2)
Total Volume	1,379,840	720,000	(L)
Land use efficiency[a]	0.690	0.720	
Electricity input	85.0	1888.9	(kWh d^{-1})

[a] Land use efficiency = Illuminated surface / Space required

Table 3. Capital and operating cost parameters for algae production with raceway ponds and PBRs.

Raceway pond		(¥ ha^{-1})	PBR	
Capital cost			Capital cost	
	Grading, earth works, etc.	773,000		Passive PBR
	CO_2 sumps	716,000		Circulation and agitation
	Mixing and carbonation system	732,000		Carbonation
	Harvesting	1,017,000		Harvesting
	Water storage reservoir and distribution	374,000		Construction and off-sites
	CO_2 delivery and distribution system	1,407,000		Anaerobic digestion system
	Nutrient supply system	61,000		
	Anaerobic digestion system	325,000		
	Other capital cost factors	5,097,000		
	Total capital cost	10,502,000		Total capital cost
Operating cost			Operating cost	
	Power (mixing, harvest,misc.)	150,000		Power (circulation, carbonation.,centrifugati
	CO_2(flue gas) blower power	161,000		Maintenance (5% of total capital)
	Maintenance (5% of total Capital)	457,000		Labor
	Labor	8,799,000		
	Total operating cost	9,567,000		Total operating cost
Annual fixed charge (10.3% of capital cost)		1,085,000	Annual fixed charge (10.3% of capital cost)	
Annual operating cost		9,567,000	Annual operating cost	
Total annual cost		10,652,000	Total annual cost	

* The conversion rate of 1 U.S. dollar = 77.9 Japanese yen, released on October 1, 2012. (http://www.federalreserve.gov/releases/h10/current/default.htm)

Table 4. Parameters for improving cultivation technology cases.

	Raceway ponds		PBRs	
	Land use efficiency	Environmental conditions coefficient	Land use efficiency	Environmental conditions coefficient
Base case	0.69	0.8	0.72	0.98
Case 1	0.98 (+42%)	0.9 (+12.5%)	0.72 (+0%)	0.98 (+0%)
Case 2	0.98 (+42%)	0.9 (+12.5%)	1.5 (+108%)	0.98 (+0%)
Case 3	0.98 (+42%)	0.9 (+12.5%)	2.0 (+178%)	0.98 (+0%)

Table 5. Maximum production of biomass, biogas and biodiesel (Cases 1, 2 and 3).

Theoretical maximum algal productivity[a] (S_n =1000 mg N L^{-1}, Scenario 3)	*C. vulgaris*	*Botryococcus braunii*			Units
	(P=0.29, C=0.51, L=0.18)[b]	(P=0.220, C=0.141, L=0.445)	(P=0.220, C=0.141, L=0.445)	(P=0.220, C=0.141, L=0.445)	
	Case 1	Case 1	Case 2	Case 3	
	Raceway ponds		PBRs		
Algal biomass yields	160,127.0	142,013.9	236,689.9	315,586.5	(kg DW)
Daily areal productivity	43.9	38.9	64.8	86.5	(g m^{-2})
Daily volumetric productivity	0.3	0.3	0.9	1.2	(g L^{-1})
Biodiesel production	24,562.1	53,854.2	89,756.9	119,675.9	(L)
Methane production using the residual algal biomass	54,601.7	22,244.4	37,073.9	49,431.9	(m^3 CH_4)
NH_3-N recovery from anaerobic digestion	6,540.4	4,400.4	7,334.1	9,778.7	(kg NH_3-N)
Energy recovery from combustion of CH_4	140,978.5	57,433.7	95,722.8	127,630.4	(kWh)
Energy consumption	31,025.0	31,025.0	689,453.9	689,453.9	(kWh)
Net energy gain from combustion of CH_4	109,953.5	26,408.7	-593,731.1	-561,823.5	(kWh)
Algal biomass production cost	67	75	112	84	(¥ kg^{-1})
Net biodiesel production cost[c]	222	193	310	236	(¥ L^{-1})
Nitrogen fixed quantity	1,906.2	1,690.6	2,817.7	3,756.9	(kg N)
Demand for additional nitrogen inputs	246,233.5	248,265.7	125,474.8	123,499.7	(kg N)
Nitrogen discharge	250,867.7	250,975.5	129,991.2	129,521.6	(kg N)

[a] 1 ha-scaled cultivation for one year.
[b] P, C and L is protein, carbohydrate and lipid, respectively.
[c] Transesterification cost is ¥25 L^{-1}, and selling price for excess electricity generated on-site is ¥40 per kWh.

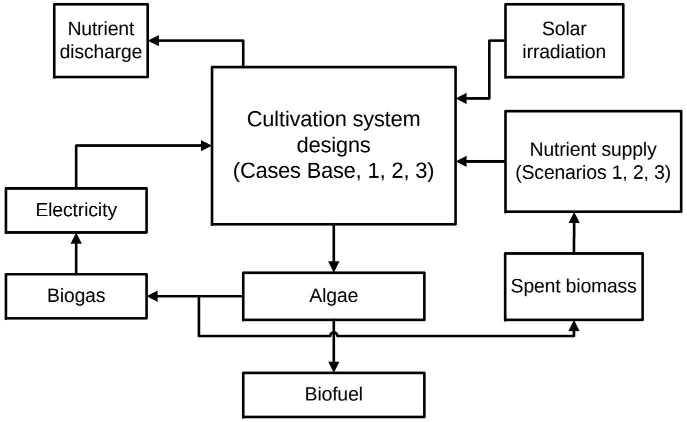

Fig. 1. The scheme of a serial of calculations.

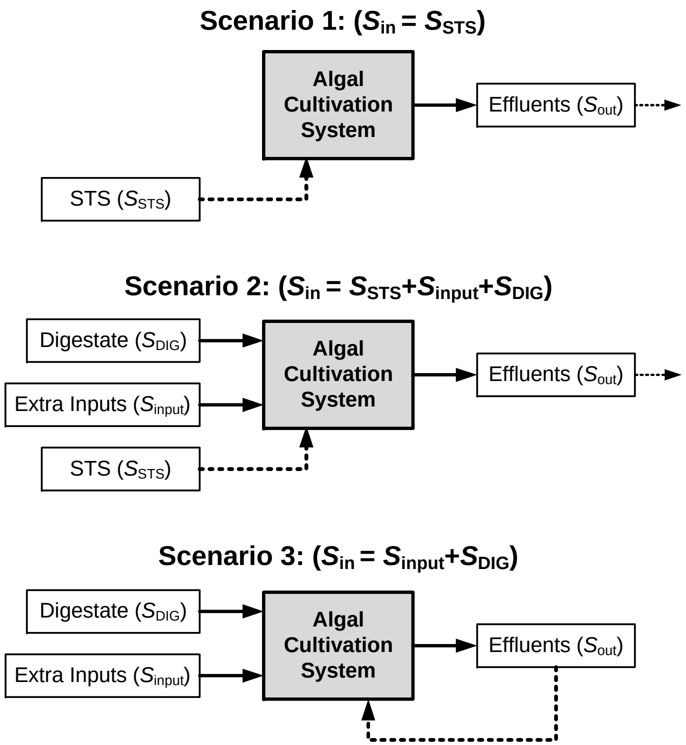

Fig. 2. Scenarios for different nutrient supplies.

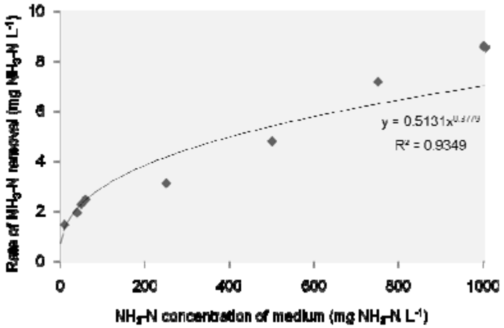

Fig. 3. Average daily NH_3-N removal efficiency of *C. vulgaris.*

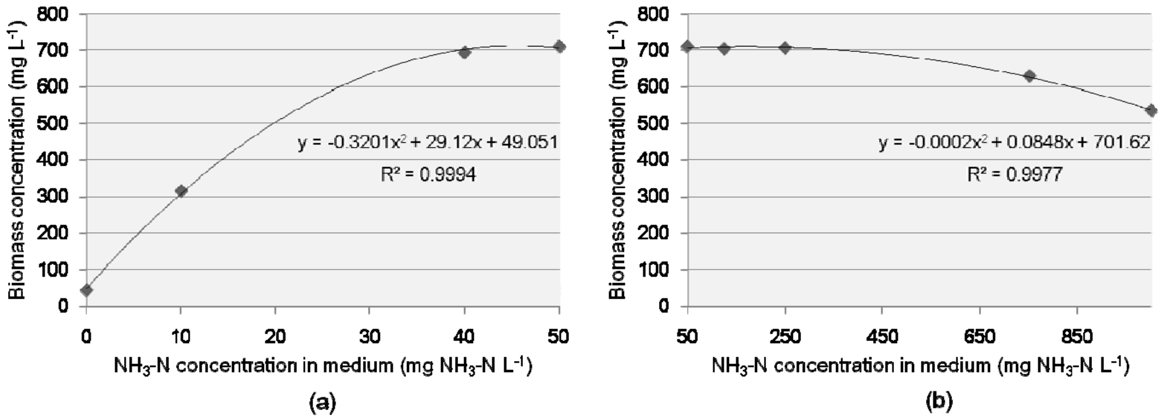

Fig. 4. Biomass concentration of *C. vulgaris* under different NH_3-N concentration (a) 0-50 mg NH_3-N L^{-1}; (b) 50-1000 mg NH_3-N L^{-1}.

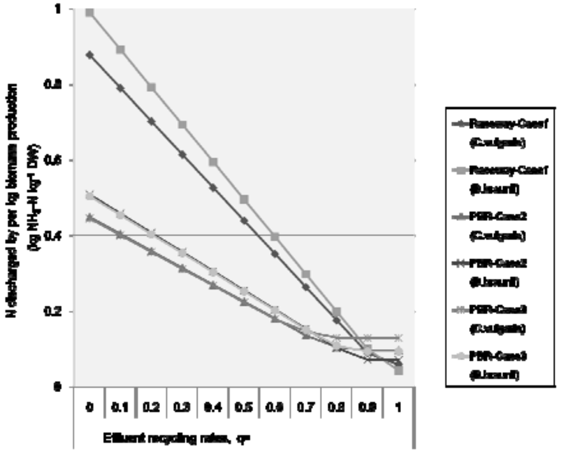

Fig. 5. Nitrogen discharged by per kilogram algal biomass production in medium of S_n= 150 mg N L^{-1}.

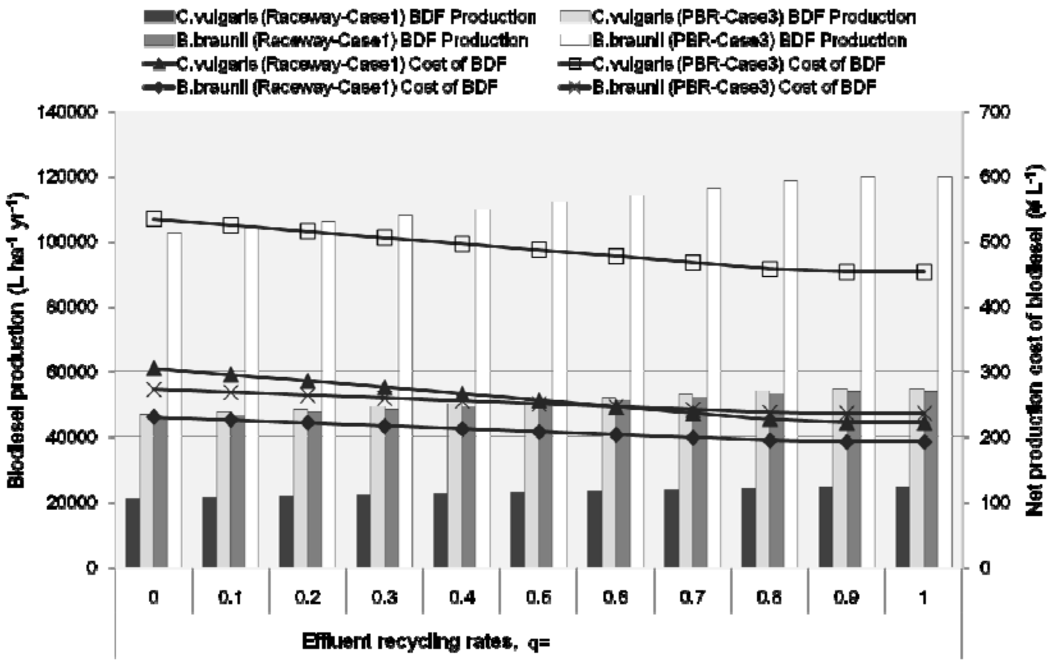

Fig. 6. Variation of biodiesel productivity and the net production cost of biodiesel for different ratios of effluent reuse under a nitrogen discharge limit of 150 mg N L^{-1}.