

Bio-mitigation of carbon dioxide using microalgal systems: Advances and perspectives



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ABSTRACT

Carbon sequestration is an important strategy in combating rising carbon dioxide concentration in the atmosphere. Differing from carbon emission reduction, carbon sequestration offers the possibilities of reducing or avoiding CO₂ emission if CO₂ is to be captured from large stationary sources and utilization of the captured CO₂ for production of chemical and energy. Biological sequestration or bio-mitigation of carbons through microalgal systems, despite in its early stage, represents a promising and sustainable alternative to current carbon mitigation methods. Microalgae consist of a group of highly diverse and fast-growing microorganisms, capable of photoautotrophy, heterotrophy, and mixotrophy. They can be cultivated on non-fertile land with unit CO₂ fixation capacity 10–50 times higher than terrestrial plants. Production of food, feed, fine chemicals, and biofuels from microalgal biomass could further enhance the benefits of microalgae-based CO₂ fixation. This present review is aimed to gain understanding how microalgae assimilate different forms of carbons and provide a comprehensive overview of the current advances in utilizing microalgae for CO₂ fixation, with focus on strain screening and improvement, mass cultivation practice, and effects of environmental and nutritional factors on CO₂ fixation performance. Economic viability, challenges and perspectives of microalgae-mediated CO₂ bio-mitigation are also discussed.

1. Introduction

CO₂ represents 68% of greenhouse gases (GHGs) emission into the atmosphere [1] and is a major contributor to the global warming. The Kyoto Protocol and the Paris Agreement set ambitious goals and responsibility for participating countries to curb GHGs emission. While these agreements are to limit CO₂ emission, there is another aspect in the reduction of CO₂ in the atmosphere, i.e., CO₂ sequestration [2–4]. The technical benefits of CO₂ sequestration are three folds: first, it reduces CO₂ concentration in the atmosphere; second, it reduces or avoids CO₂ emission if CO₂ is to be captured from large stationary sources [5,6]; third, the captured CO₂ can be used as a feedstock or substrate for production of chemical and energy products

[5,6]. In addition to these technical benefits, CO₂ sequestration and utilization can generate new economic and job opportunities.

There are many techniques for CO₂ sequestration, which may be classified into the physical, chemical, and biological categories. Each of them has advantages and disadvantages. The focus of this review is biological platform, specifically, microalgae-based approach. [5,6] (Detailed in Table 1). Physical storage refers to the processes that directly inject highly concentrated CO₂ into deep ocean, aquifers or depleted oil/gas wells [7]. By contrast, chemical fixation involves CO₂ immobilization using adsorption material (such as lithium hydroxide) followed by alkaline-mediated neutralization leading to the formation of carbonates or bicarbonates. Both have their own advantages and shortcomings [7]. Physical methods such as direct CO₂ injection are

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Table 1
Comparison of various CO₂ sequestration methods [123–131].

Category	Method	Mechanisms	Prospects	Limitations
Physical	Membrane separation	Isolation of CO ₂ from the main stream by passing mixed gas through a membrane	1) increased mass transfer	1) energy inefficient 2) membrane fouling and blockage 3) high cost
	Geologic injection	Injection of CO ₂ into geologic reservoirs, depleted oil/gas wells, and coal seams	1) make use of abandoned space 2) relative easy operation 3) possible recovery of oil/methane	1) requirement of particular geological and geomorphological environment 2) gas leakage over time (several thousand years) 3) high cost
	Oceanic injection	Injection of CO ₂ into deep ocean	1) large CO ₂ holding capacity	1) gas leakage over time (several hundred years) 2) threaten the lives of non-swimming marine organisms 3) requirement of high-cost injection techniques
	Adsorption	Using molecular sieves or zeolites	1) minimal waste generation 2) flexible to different CO ₂ sequestration schemes	1) energy inefficient 2) co-adsorption of other components (SO _x)
Chemical	Chemical absorption	Neutralization of carbonic acid to form carbonates or bicarbonates	1) safe and permanent sequestration 2) rich supply of required base ions (Na ⁺ , K ⁺)	1) large equipment size requirement 2) high energy requirements 3) high cost
	Mineral carbonation	Reaction of CO ₂ with metal oxides to form stable carbonates	1) abundantly available metal oxides (MgO, CaO) 2) safe and permanent sequestration 3) utilization of stable carbonates after sequestration	1) requirement of large amount of reagent 2) not cost-effective
Biological	Forestation	Incorporating atmospheric CO ₂ into biomass over the lifetime of trees	1) chemical-free	1) limited CO ₂ sequestration 2) large land area requirement 3) potential threat to biological diversity and food supply
	Oceanic fertilization	Triggered growth of photosynthetic organisms by extra iron sources	1) significant increase in CO ₂ sequestration	1) high cost 2) high level of uncertainty 3) impact on ocean eco-system (change in plankton structures) 4) possible trigger of methane production
	Microalgae-based sequestration	Utilization of CO ₂ via microalgal photosynthesis	1) high photosynthetic efficiency 2) efficient in low-concentration CO ₂ sequestration 3) faster sequestration rate than higher plants 4) do not compete with crops for arable land 5) co-production of food, feed, fuel, fine chemicals, etc.	1) sensitive to toxic substances in exhaust gases 2) not very cost-effective for photobioreactors construction and algal biomass harvesting

suitable for large-scale CO₂ sequestration; however, they require certain geological and geomorphological structures, expensive separation equipment and technologies to collect and concentrate CO₂, uncertainties, and risk with long term leakage, etc. [7]. Chemical neutralization methods are a relatively safe and long-term CO₂ fixation process but not cost-effective, as large amounts of reagents are necessary for neutralization [7]. Furthermore, both physical and chemical methods are faced with challenges in capturing CO₂ from low concentration and diffused- or non-point sources [8,9].

This review is intended to demonstrate the potential of microalgae based approach to tackle these challenges. Carbon is the main component of microalgae cells, accounting for about 50% of cell dry weight. It is estimated that 100 tons of microalgal biomass production is equal to around 183 tons of CO₂ fixation [4,10]. Microalgae have the ability to sequester low concentration CO₂ from air or high concentration CO₂ from stationary sources such as coal burning power plants, and inorganic and organic carbons in wastewater. Furthermore, algae can effectively utilize N and S containing pollutants, suggesting a potential of reducing NO_x and SO_x, potent GHGs. [5,11].

Microalgae are photosynthetic cell factories that possess many unique characteristics well suited for CO₂ sequestration. Microalgae are able to use natural sunlight as energy for CO₂ fixation with high photosynthetic efficiency that is 10–50 times higher than terrestrial plants [12]. They can tolerate extreme environments such as saline-alkali land, desert, and beaches without competing with crops for arable land [10,13]. They grow much faster than higher plants with

doubling time within 2–4 h [14]. They can feed on flue gas from power plants as inorganic carbon source and wastewaters from municipal, industrial and agricultural activities as nutrient source (N, P) [3,15–20]. Microalgae can serve as carbon neutral single-cell bio-factories for the production of food, animal and aquaculture feed products, cosmetics, nutraceuticals, pharmaceuticals, fertilizers, bioactive substances, and biofuels [4,11]. Moreover, microalgae have been used in indoor air purification, exhaust gas treatment from automobile, power plants and other industries and have the potential for CO₂ removal and O₂ generation for life protection system control in confined spaces such as nuclear submarines and manned spacecrafts [21].

Carbons may emit to the atmosphere from natural and human sources such as decomposition of organic materials, respiration of living organisms, and burning/combustion of plant and fossil fuels during manufacturing and agricultural activities, transportation, and other human activities. Carbons may be in the form of atmospheric CO₂ (0.03–0.06%, v/v), soluble inorganic carbonate (HCO₃⁻ and CO₃²⁻) and organic carbons (simple sugars and short fatty acids) [4,12,21]. Fig. 1 schematically illustrates a concept of integrating microalgae-based carbon sequestration with utilization of the captured carbons. In this concept, nitrogen (N) and phosphorus (P) rich wastewaters replace the artificial media to support microalgae growth, nutrients and CO₂ are recycled during downstream refining processes, and the harvested algal biomass is converted to renewable bioproducts and bioenergy. The realization of the concept is expected to enhance the economic viability and environmental friendliness of microalgae-based CO₂ fixation systems.

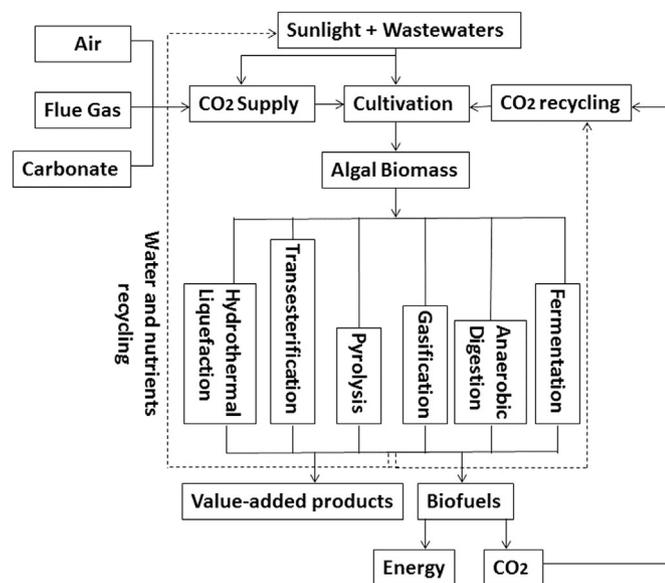


Fig. 1. Process diagram of microalgae-mediated CO_2 bio-mitigation and system integration.

However, microalgae-mediated CO_2 fixation technology is also faced with many challenges in practical applications. In this review, we discuss how microalgae assimilate different carbons and deal with high concentration of CO_2 , how CO_2 transport and diffusion affect CO_2 availability in culture media, and how strains and cultivation methods and conditions affect algae growth and carbon sequestration. We provide several representative application scenarios and their economic outlooks. The need for future research is suggested.

2. Carbon metabolisms in microalgae

Microalgae, generally considered autophototrophs, are the primary oxygen-evolving photosynthetic microorganisms on the earth, whose carbon consumption accounts for almost 50% of the global CO_2 fixation [2,12]. However, some microalgae species possess heterotrophic metabolism and are able to grow in dark environments. Under certain circumstances, some algae strains are able to grow mixotrophically. The ability of microalgae to grow heterotrophically or mixotrophically is significant and important because this allows microalgae to sequester organic carbons present in wastewaters, which can eventually emit to the atmosphere if broken down by bacteria. In the next few sections, the forms of carbon assimilable by microalgae, the mechanisms involved in CO_2 capture by microalgae, and high concentration CO_2 stress will be discussed.

2.1. Auto-phototrophic assimilation of inorganic carbons

2.1.1. Forms of inorganic carbons

Microalgae can acquire dissolved inorganic carbon (DIC) from the aquatic environment in forms of CO_2 , H_2CO_3 , HCO_3^- , and CO_3^{2-} (Fig. 2). By contrast, terrestrial plants are much less diversified in DIC assimilation [22–24]. DIC forms vary greatly with pH, mixing rates, microalgae concentrations, etc. [22,23]. Different microalgae strains may have different preferences for DIC forms. For example, *Chlorella miniata*, *Chlorella vulgaris* 11h, and *Monodus subterraneus* can take up only gaseous CO_2 (CO_2 has unique property of membrane permeability), and two strains of marine eustigmatophyte algae *Nannochloropsis gaditana* and *Nannochloropsis oculata* can only actively transport HCO_3^- [2,25]. On the other hand, some species, such as *Chlamydomonas reinhardtii*, *Scenedesmus obliquus*, *Dunaliella tertiolecta*, *Chlorella pyrenoidosa* and *Chlorococcum lit-*

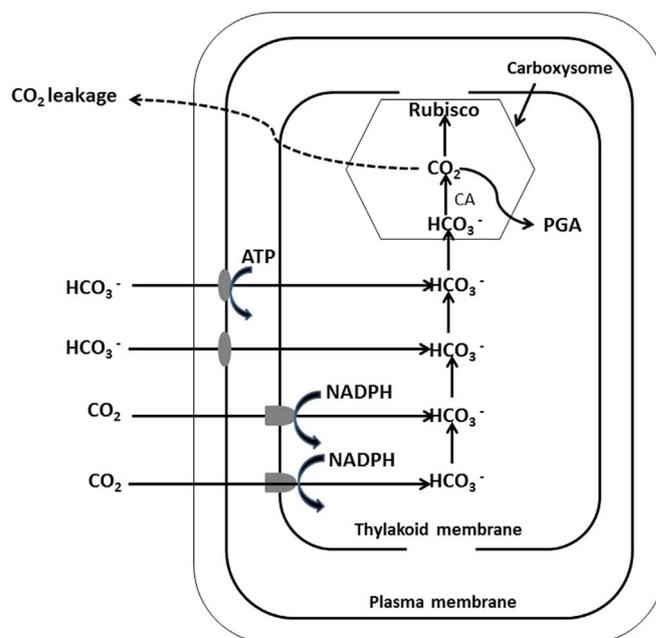


Fig. 2. A typical schematic model for inorganic carbon transport and CO_2 accumulation in microalgal cells. Redrawn after Giordano et al. [2]. CA: Carbonic anhydrase; Rubisco: Ribulose Bisphosphate Carboxylase/Oxygenase; PGA: Phosphoglyceric acid.

torale, harbor an external carbonic anhydrase (CA) and are capable of utilizing both CO_2 and HCO_3^- . Certain strains lack external CA but can still utilize both CO_2 and HCO_3^- , such as *Chlorella ellipsoidea* and *Chlorella kesslerii* [25–27]. The amount and location of CA in microalgae are strain dependent and may determine the forms of DIC for assimilation.

2.1.2. Auto-phototrophic assimilation of CO_2

Microalgal CO_2 fixation refers to the process of converting CO_2 and water into organic compounds powered by ATP and NADPH, which are generated through photosynthesis [28]. Similar to terrestrial plants, microalgae capture CO_2 via Calvin cycle, which consists of three phases: carboxylation, reduction, and regeneration. Briefly, in carboxylation phase, CO_2 is incorporated into ribulose-1, 5-bisphosphate (RuBP) catalyzed by ribulose-1, 5-bisphosphate carboxylase (RuBisCo), resulting in 2 molecules of 3-phosphoglycerate (3-PGA). Then 3-PGA undergoes phosphorylation and reduction, catalyzed by 3-phosphoglycerate kinase and glyceraldehyde phosphate dehydrogenase respectively, to produce glyceraldehyde 3-phosphate (G-3-P). Finally, RuBP is regenerated via a series of reactions and enters the next fixation cycle. In microalgae, CO_2 is transferred to RuBisCo via successive crossing of cell wall, cell membrane, cytoplasm, chloroplast membrane, stroma and extracellular boundary layer [28,29]. During the transfer process, resistance of CO_2 transportation and diffusion are the main limiting factors influencing CO_2 fixation. Microalgae normally take up gaseous CO_2 as the substrate for RuBisCo. However, a few microalgae strains can assimilate HCO_3^- which is then converted to CO_2 by carbonic anhydrase [2,25].

2.2. Heterotrophic assimilation of organic carbons

Heterotrophic metabolism usually relies on light-independent uptake of organic compounds available in the media to acquire energy and carbons. Organic compounds must be small enough to move across the cell walls and are then converted to lipids and other metabolites through respiration pathways such as pentose phosphate pathway (PPP) [2,25]. In some strains, heterotrophy can occur in the presence of light. This process, involving light as an energy source, is termed photoheterotrophy. In contrast to autophototrophy, heterotrophy over-

comes the limitation of light requirement as seen in autophototrophic growth, enables faster growth to higher biomass, lipid, and protein productivity, and facilitates simpler operations [2,25]. However, strains with high heterotrophic capacity are limited, and the impact of bacterial activities on culture vitality could be detrimental [2,25].

Glucose is the most commonly used organic carbon for heterotrophic cultivation of microalgae. This makes the feedstock cost a major limiting factor for production of target metabolites, and on the other hand, has no practical implication in terms of carbon sequestration. Organic compounds in wastewaters, however, are a cheap carbon source and a significant target of carbon sequestration. More details on organic carbon sources are given in 2.3.3.

2.3. Carbon source as a limiting factor

Carbon source may limit microalgae growth in a number of ways. A too low carbon concentration obviously cannot sustain microalgae growth and dissolving sufficient CO₂ in water can be costly; on the other hand, most microalgal strains cannot grow well at CO₂ concentration above 5% (v/v) [22,26,27], which is below flue gas CO₂ concentration of 10–20%; and finally some organic carbons may not be readily available for microalgae to use due to too large molecular size or being locked in large solid particles [30].

2.3.1. CO₂ tolerance

The optimal CO₂ level for growth and highest CO₂ tolerance level vary among different microalgae strains. Microalgae can survive in a wide range of CO₂ environments. In microalgae, RuBisCo is the rate-limiting enzyme involved in the Calvin cycle and has a very low CO₂ affinity. In order to raise the CO₂ level at the carboxylating sites, some single cell organisms including microalgae are able to adapt the “CO₂ concentrating mechanism” (CCM) in low CO₂ environments [31]. However, CCM in microalgae remains elusive and regulating CCM through genetic modification needs further study [2,24].

On the other end of the extreme, high CO₂ concentration has an “anesthetic” effect on microalgal cells, leading to inhibited photosynthesis and algae growth. Yoon et al. [32] reported that *Anabaena variabilis* did not show obvious lag phase when CO₂ concentration was in the range of 4–13%, but when CO₂ concentration was 18%, the growth was completely inhibited. In addition, the specific growth rate decreased with increasing CO₂ concentration. The observed growth inhibition was probably due to the decreased pH of the medium at CO₂ concentration greater than 18%. Some strains exhibited extremely high tolerance to CO₂. For example, Yue et al. [30] isolated a high CO₂ concentration tolerant strain *Chlorella* ZY-1 from soil. Their results showed that the maximum growth rate was obtained when CO₂ concentration was 10% or 20%. When CO₂ concentration was 30% and 50%, relatively high growth rate and high cell density were still remained. Although slower growth was observed when the CO₂ concentration was 70%, the algae concentration still reached 0.776 g/L after 6 days cultivation. Some strains were able to tolerate high CO₂ when CO₂ was injected through gradual concentration increment rather than starting at high concentrations [33]. However, the number of strains tolerating high CO₂ concentrations is limited. Therefore, in addition to screening and selecting CO₂-tolerant strains, it is worthwhile to employ genetic engineering techniques to develop CO₂ tolerant strains, which is the topic of Section 3.2.

The initial CO₂ concentration not only affects the growth, but also determines the lipid yield and composition. Low CO₂ concentration was found to inhibit the synthesis of fatty acids, while high CO₂ concentration benefited fatty acid accumulation, despite the inhibition of carbon chain desaturation and elongation [34–36].

2.3.2. Gaseous mass transfer

Gas-liquid mass transfer of CO₂ is the main limiting step in the growth of photosynthetic microalgae [37]. Accumulation of O₂ to a

certain level during photosynthesis can inhibit microalgal growth, and therefore removal of O₂ is beneficial to algal growth. For example, Becker [38] conducted experiments to investigate the effect of different O₂ concentrations on photosynthetic efficiency of algae and found that photosynthetic efficiency was reduced to 35% when supplied with 100% O₂ while 14% increase was achieved with no O₂ addition. Generally speaking, in order to improve the efficiency of gas-liquid mixing and concentration of dissolved CO₂, mass transfer rate, an effective approach is to increase the gas flow rate or to provide a turbulent mixing zone [39]. In addition, high velocity of mixing in microalgae culture broth will shorten the light/dark cycle, thus improve the efficiency of carbon fixation and enhance total algal biomass accumulation. However, it should be noted that too high turbulence could generate shear stress and damage the microalgal cells [3]. In order to further enhance the mass transfer efficiency of CO₂ and O₂, Cheng et al. [40] studied the carbon fixation efficiency of *Chlorella vulgaris* by using a hollow fiber membrane bioreactor, aiming at removing O₂ through membrane during photosynthesis. The results showed that CO₂ fixation increased up to 3-folds using this unique membrane, suggesting great potential of utilizing hollow fiber membrane reactor for efficient CO₂ mitigation. One main drawback of this technique is the high cost that restricts its application.

2.3.3. Organic carbons availability

Capturing organic carbons in wastewaters through utilization is an important aspect of overall carbon sequestration efforts because wastewaters are sources of GHG emissions [41]. CO₂, CH₄, and N₂O gases emit from the wastewaters when organic compounds are broken down during the treatment or application processes; additional amounts of CO₂ and CH₄ are produced from the energy use for the treatment or application processes. Some microalgae strains are able to assimilate small organics such as acetate, citrate, fumarate, succinate, glycine, glucose, mannose, sucrose, fructose, glycerol, pyruvate, lactate, straight-chain fatty acids and alcohols (C2-C6) [42–44] and at the same time fix nitrogen and phosphorus as well. However, not all organic carbons in wastewaters could be utilized by the algae strains [20], which may be attributed to the physical and chemical properties of the wastewaters. The complete chemical composition profiles of organic carbons in wastewaters are largely unknown, and therefore what can be or cannot be assimilated by algae are also unknown. If processes or management strategy could be developed to break down large organic compounds or to liberate organic compounds locked in solid particles, carbon utilization efficiency is expected to be significantly improved. Pretreatment techniques such as chemical hydrolysis, thermal hydrolytic treatment, ultrasonic treatment, high pressure homogenization, ozonation, hydrothermal treatment, and anaerobic digestion have been reported in the literature. Most of these techniques are energy intensive and not cost effective. Some researchers are exploring the feasibility of modifying anaerobic digestion to limit the generation of methane and facilitate production of small organic molecules [45]. Anaerobic digestion converts organic matters into biogas in four reaction steps: hydrolysis, acidogenesis, acetogenesis and methanogenesis. Volatile fatty acids (VFAs) produced during the first three steps and are rapidly consumed by methanogens in the last step. If the methanogenesis step is suppressed through heat treatment and low-pH treatment of the inoculums. Hu and her colleagues demonstrated that acidogenic fermentation of liquid swine manure produced 36.8% more VFAs than conventional anaerobic digestion [45]. The practical application of this technique has yet to be demonstrated.

3. Microalgal strain selection and improvement

Microalgae are a group of highly diverse, single or multi-cellular photosynthetic microorganisms. They inhabit world widely and are recognized as one of the oldest forms of original life. Currently, more than 40,000 microalgae strains have been catalogued [4]. Microalgal

strains vary in growth rate, ability to assimilate different forms of carbon sources and other nutrients, requirement for light, tolerance to high CO₂ level and other adverse environment conditions, and cellular composition. CO₂ fixation efficiency can be improved by using high-performance algae strains, which can be obtained through screening and/or genetic engineering. Finding strains suitable for specific applications is critical to maximizing carbon sequestration efficiency.

3.1. Microalgae isolation and selection

Many microalgae species such as cyanobacteria, green microalgae, diatoms, and golden microalgae have great potential in CO₂ biomitigation as well as production of biofuel and value-added bioproducts [4,46]. The first step in utilizing microalgae for carbon sequestration is to select strains which not only have a high growth rate, be easy to mass cultivate, and contain valuable ingredients for post-harvest applications but also should be able to adapt to harsh environments and tolerate concentrated carbon and nutrient sources. Bioprospecting microalgae from local habitats is a logical way to isolate strains for easy adaptation to applications which share environments similar to the habitats where the strains were isolated [17]. A wide range of microalgal species exist in diverse and sometimes extreme natural environments [15], offering the possibility of finding strains with characteristics uniquely suitable for certain applications. The Aquatic Species Program (ASP) sponsored by the US Department of Energy collected more than 300 strains over a more than 15 years' time span from different regions of the continental US and Hawaii in order to isolate high-oil-content autophototrophic strains to be grown in open ponds [10]. Strains isolated from hot springs were found to tolerate high temperature, high CO₂, high NO, high SO₂, and low pH [47,48]. These strains are suitable for the treatment of hot flue gas containing high level of CO₂, NO, and SO₂. Chang and Yang [49] isolated more than 200 strains from lakes, ponds, sediments, hog wastewater, paddy fields, hot springs, and seawater in Taiwan. After screening, they found two unicellular microalgae, *Chlorella* sp. NTU-H15 and *Chlorella* sp. NTU-H25, isolated from hog wastewater, were able to grow up even in aeration containing CO₂ up to 40% in laboratory. *Chlorella* species consist of by far the most versatile strains highly tolerant to high CO₂ concentration. The high CO₂ tolerance of these algal strains is probably due to their broad range response to pH variation and low activity of carbonic anhydrase [29,50].

Bioprospecting starts with collecting wild strains from local habitats, followed by isolation and screening [50]. Considering the vast number of microalgae strains worldwide and the wide variations in their response to growing environment, screening remains to be a great challenge. Techniques that enable high throughput screening have practical significance to finding desirable strains from huge pools of microalgae strains in wild and algae bank collections. With the aid of more efficient strain screening technique such as 96-well microplate swivel system (M96SS) [51], critical issues in miniature culture systems like intra-well mixing and sample evaporation could be addressed, allowing up to 768 algae samples for processing at the same time. Another approach to the development of environment tolerant strains is acclimation. Many microalgae have the ability to undergo physiological acclimation to changes in environment conditions such as light [52], CO₂ level [53], osmotic pressure [54], sulfur [55], metals [56], and harsh conditions of concentrated wastewater [17]. Acclimation is usually a gradual process and the results may be long term or short term depending on the strains and methods of acclimation [56,57]. More research is necessary to develop techniques to take the advantages of microalgae's ability to acclimate to flue gas and concentrated wastewater conditions.

3.2. Genetic engineering of microalgae

Genetic engineering of microalgae represents one of the most

effective approaches for trait improvement. Research in the past decades has shown that *Chlamydomonas* is an important model of eukaryotic algae for genetic manipulation. In the mid-1990s, with the development of genetic technology, researchers started to interpret the CCM and manipulate 1, 5, - ribulose biphosphate carboxylase/oxygenase (RuBisCo) activity to enhance CO₂ fixation capability [2,24]. Wakasugi et al. [58] sequenced the chloroplast genome of the unicellular green alga *Chlorella vulgaris* C-27 and had it compared with those of red and brown algae, and terrestrial plants. The results showed that *C. vulgaris* was genetically closer to the terrestrial plants than to the red and brown algae. Beuf et al. [59] isolated partial DNA region encoding RuBisCo activase (*rca*), which was considered to be linked to high CO₂ stress, from marine green algae *Chlorococcum litorale*. It was found that the level of *rca* mRNA increased upon high CO₂ concentration (up to 60%, v/v). Kang et al. [60] expressed two genes encoding fructose-1, 6 -biphosphate aldolase (ALD) and triose phosphate isomerase (TPI) in *Anabaena* sp. 7120 and found that expression levels of the two enzymes were much higher than those of wild-type under all experiment conditions. In addition, it demonstrated that the engineered *Anabaena* sp. 7120 exhibited a significantly enhanced CO₂ fixation efficiency. The above mentioned studies clearly demonstrated that overexpression of the key enzymes closely related to photosynthetic CO₂ fixation pathway could be one of the most promising strategies to enhance microalgal CO₂ fixation. Information on specific genetic modification techniques is beyond the scope of this review and would not be discussed in detail.

4. Cultivation practice

Microalgae based carbon sequestration is realized through mass cultivation of microalgae, which has been practiced all over world for various purposes for decades. Many cultivation systems have been developed and used. There are substantial research advances in understanding and optimizing cultivation conditions. In this section, we will discuss how cultivation systems and conditions affect the growth of microalgae and hence carbon sequestration by microalgae.

4.1. Photobioreactor (PBR)

Photobioreactors (PBR) are physical structures used for mass cultivation of phototrophic microorganisms [61,62]. Depending on whether or not they are directly exposed to the atmosphere, PBRs can be classified as open or closed systems [63]. Fig. 3 shows the configurations of commonly used PBRs. Raceway-based open pond with paddle-wheels (Fig. 3A) is a typical open system. In an open-pond system, CO₂ is supplied through direct injection or by exchanging with ambient air through mixing, through which CO₂ is dissolved in culture media. For improved open ponds, a sump close to the paddle wheel with a depth of about 1.5 m is adopted to maximize the solubility of CO₂ in water when CO₂ is added by direct injection, thus enhancing the CO₂ fixation efficiency [64]. The most prominent features of open-pond systems include simple construction, low cost and easy operation. However, disadvantages of such systems are also obvious, such as large footprint, unstable culture conditions, difficulties for operation controlling, easy contamination, high evaporation loss, and the fact that light intensity decays rapidly with medium depth [13,61,65]. Overcoming the above shortcomings are future research directions for developing advanced open pond systems. Recently, researchers at the University of Minnesota developed a stacked multi-layered hybrid bioreactor, which is designed to overcome the shortcomings of ordinary open and closed reactors to a certain extent (Fig. 3E and Fig. 4) [66,67]. This multi-layered structure renders the system a very small footprint. Therefore, it is feasible to co-locate such a small footprint system with a facility where wastewater is generated and spare land is limited. The open shallow trays could significantly reduce the impact of wall fouling on light transmission, thus maintenance (cleaning) of the

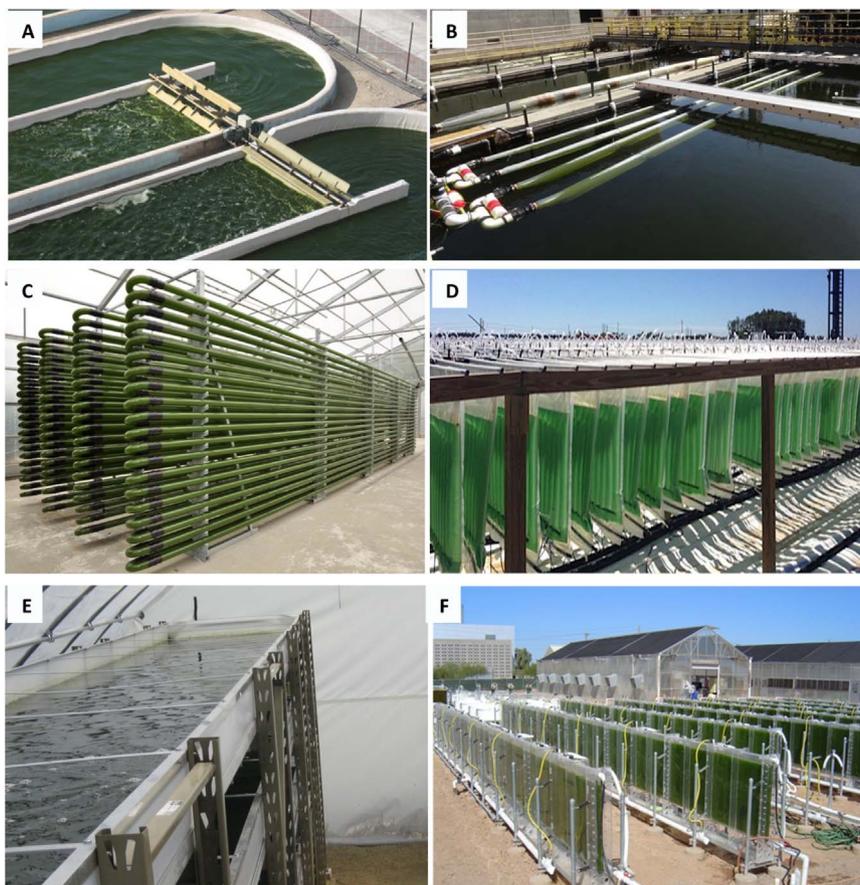


Fig. 3. Reactor configuration for microalgal cultivation: (A) raceway pond (from Sapphire Inc, America); (B) Floating photobioreactor (from OMEGA system-NASA); (C) tubular bioreactor (Wolfsburg, Germany); (D) Plastic Bag bioreactor (from Algenol Inc, America); (E) Multi-layer bioreactor (from center for biorefining, University of Minnesota); (F) Flat-panel Bioreactor (from Nanovoltaics technologies, America) [122].

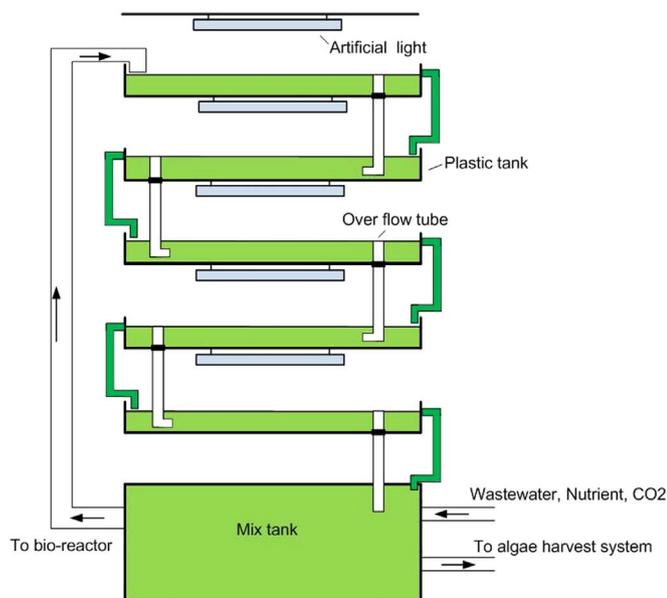


Fig. 4. Design scheme of the newly developed multi-layer bioreactor system [122].

system is minimal [66–68]. Moreover, the entire algae cultivation system can be rendered more sustainable by coupling microalgal biomass production with wastewaters treatment and power generation [66–68].

Closed PBRs are designed to have larger optical cross sectional areas to receive natural or artificial light [69,70]. Microalgae can be

cultivated year round in continuous or semi-continuous culture mode, and achieve high cell density per unit area or volume as well as high CO_2 fixation rate through using PBR [3,65]. Closed PBRs have many advantages over open ponds, including: 1) easier control of parameters that affect algae growth; 2) relatively stable culture conditions, 3) aseptic operation; 4) capable of high density cultivation; 5) high area/volume ratio to increase mass transfer efficiency with less land occupation, which greatly improves CO_2 fixation efficiency; 6) natural (or artificial) light source can be collected and distributed to the interior of bioreactor using collector and optical fiber to obtain much higher light utilization; 7) prevented or reduced water evaporation [13,65,71]. The commonly used closed PBRs (as shown in Fig. 3) include tubular, flat panel, cylindrical airlift photobioreactors, stirred fermenters, and floating and bag reactors, etc. [13,19,61,66,72], as shown in Fig. 3. CO_2 is generally supplied from the bottom of the bioreactor through gas distributor with direct injection. Recently, Wiley et al. [72] reported a novel microalgae cultivation system, named “offshore membrane enclosures for growing algae” (OMEGA) system (Fig. 3B and Fig. 5), in which microalgae were cultivated in floating PBRs deployed in protected marine bay environment with nearby municipal wastewater outfalls and sources of CO_2 -rich flue gas on shore. The advantages of the OMEGA system included uniform temperature maintenance due to “water bath” effect of sea water, low fertilizer cost due to usage of nutrient-rich wastewater and CO_2 -rich flue gas and low energy input for mixing due to waves of ocean, etc. [72]. Nevertheless, wall fouling is a serious problem of this system and needs to be addressed.

Currently, closed PBRs with different configurations are widely used for microalgae-mediated CO_2 fixation, and especially for flue gas bio-mitigation due to the advantages discussed above [73,74].

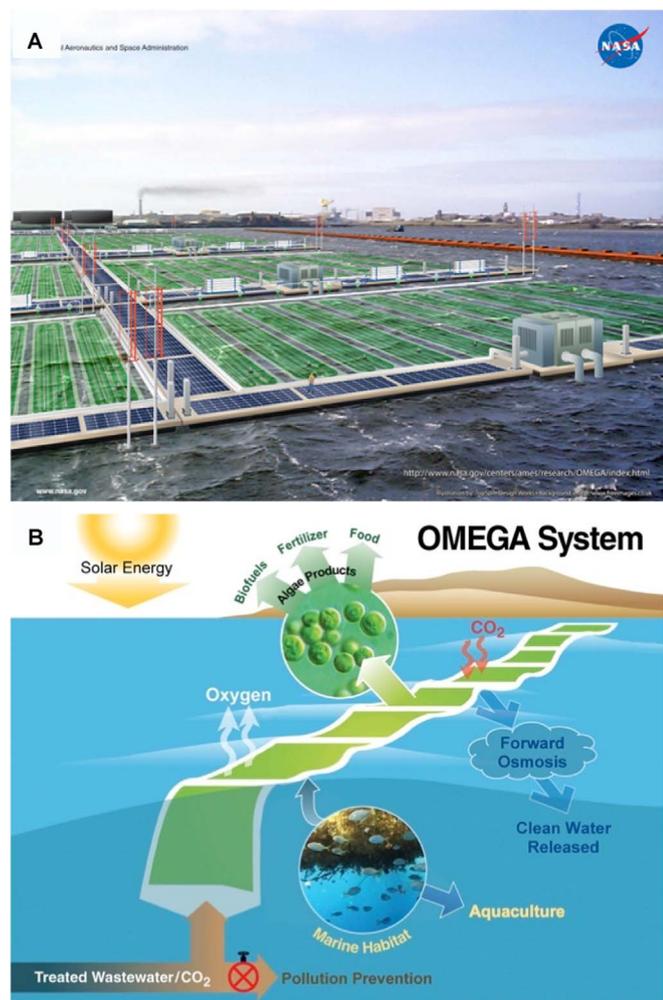


Fig. 5. OMEGA system developed by NASA. (A) Large-scale demonstration of OMEGA system; (B) OMEGA system for effective wastewater treatment and CO₂ bio-fixation. <http://www.nasa.gov/centers/ames/research/OMEGA/#.WAre0NxvBFU>, 2009.

Recently, active research fields on microalgae-based CO₂ removal from flue gas using closed PBR include: 1) investigation of microalgal physiological characteristics using lab-scale closed PBRs; 2) kinetics study of microalgae growth; 3) pilot-scale cultivation of promising microalgae (especially those reconstructed microalgae) and high-value byproducts development [63,75–80]. At present, a major focus on mass cultivation of microalgae is the combination of closed and open cultivation systems, i.e., closed PBRs for screening and optimizing promising microalgal strains, and open ponds for scaling-up cultivation of the prepared seeds. With closed PBR as the main research direction for large-scale sequestration of industrial-derived CO₂, biofixation of CO₂ from flue gas using large-scale closed PBR as well as large-scale production of microalgae for value-added byproducts may be achievable in the near future. Nevertheless, the cost of scaling up the entire closed PBR-based production system will drastically increase mainly due to the need of PBR reconstruction and affiliated light collection, transmission and distribution systems, as well as cooling and heating systems.

Although development of large scale closed PRBs have received considerable progress, many challenges are still required to be addressed in the future. Life cycle assessment (LCA) studies have identified PBR as one of the key energy intensive operations [81,82]. Closed PBRs have yet to become economically competitive against open pond systems. The hybrid PBR model is one way to take the advantages of open and closed systems with acceptable comprise. There has been significant interest in bionic design of PBRs, much of which arose from

the desire to ornament cityscape and landscape with sustainable symbols. This concept should be researched with the intent to maximize light reception and minimize energy consumption associated with flow and mechanical movements during operations. Finally, low cost renewable energy from wind, sun, and biomass may be used in PBR operations, which may lower the carbon footprint and improve the overall LCA impact.

4.2. Cultivation conditions

Microalgae growth and carbon fixation efficiency are influenced by strain, carbon and nutrient level, light intensity, light/dark cycle, temperature, and pH. Some of these factors are managed in order to provide optimal conditions for algae to grow. Understanding the effects of these factors on algae growth and biological carbon sequestration helps guide development of new technologies and process optimization. Strain and carbon source have already been covered in Section 2 and will not be repeated in this section.

4.2.1. Light intensity and light/dark cycle

Light is a prerequisite for the auto-phototrophic growth of microalgae. Light sources can be natural sunlight, artificial light or a combination of both. Growth rate of microalgae increases with increasing light intensity, until it reaches a certain threshold, a phenomenon called light saturation [83]. When light becomes the only limiting factor, microalgal biomass is proportional to light conversion efficiency [62]. Molina et al. [84] proposed several models to describe the relationship between light intensity and growth rate of microalgae. Light intensity also affects lipid accumulation and fatty acid composition of microalgae. Low light intensity can promote polar lipid synthesis, whereas high light intensity can reduce the amount of polar lipid and increase neutral lipid accumulation. Excessive light intensity, however, would hinder the accumulation of lipids, thus increasing light intensity and maintaining it at optimal level could be helpful for neutral lipid synthesis [62]. Li et al. [85] reported that percentage of saturated fatty acids such as C16:0 and C18:0 increased with light intensity, while the proportion of unsaturated fatty acids such as C18:2, C16:1, C16:2, C16:3 decreased accordingly. Composition of algal lipids dictates the end applications of the harvested lipids.

Light/dark cycle also plays an important role in algae growth. Eduardo [86] studied the effects of different light/dark cycles on growth and CO₂ fixation capacity of *Aphanothece microscopica* Nägeli cultivated in an airlift bioreactor. Results showed that light/dark cycle could greatly influence CO₂ sequestration. Under continuous illumination, carbon fixation capacity of microalgae reached 99.69%. Sun et al. [87], however, reported decreased growth rate with the increase in light/dark cycle for the marine microalga *Tetraselmis tetrahele*. It was found that only in light cycles ranged from 6 to 18 hours, the cell density, chlorophyll a and protein content reached high level. Too long or too short light cycles caused cell growth inhibition, leading to a significant decline in cell density, chlorophyll a and protein content, suggesting the importance of optimized light cycle to the metabolism and algae growth. Tor [88] compared the growth of *Chaetoceros gracilis* and *Isochrysis galbana* with light/dark cycle of 24:0 and 12:12, respectively, and the intensity of later was 2 times of the former. The results showed that the growth rate was similar between the two microalgal strains, indicating algae growth is not only affected by light/dark cycle, but also by light intensity.

4.2.2. Temperature

The optimum temperatures for algae growth are between 15 and 30 °C. However, there are some strains which can tolerate high temperatures. For example, Sung et al [89] cultivated *Chlorella* sp. KR-1 with 10% CO₂ at varied temperatures and found that growth rate at 40 °C was lower than those at 25–35 °C, but the algae still maintained high growth rate and high cell density. Maeda et al. [90]

screened one high temperature tolerant strain *Chlorella* sp.T-1 for the treatment of flue gas from power plant (containing 15% CO₂). Their results showed that although the optimum temperature for *Chlorella* sp.T-1 growth was 35 °C, cell density only slightly decreased at 45 °C. Yue et al. [30] isolated one high temperature tolerant strain *Chlorella* ZY-1, and observed that at 20 to 25 °C, its growth rate increased with increasing temperature. When temperature was set between 25 °C and 30 °C, growth rate did not change significantly. When temperature reached 40 °C, cell growth was inhibited, but the cell density can be remained.

4.2.3. pH

The level of pH significantly affects the growth and CO₂ fixation rate of autotrophic microalgae by affecting the activities of metabolic pathway related to enzymes and absorption of ions by microalgal cells [91]. The pH value of a culture medium is determined by the nature of the culture medium and the growth of algae. In addition, industrial waste gas often contains impurities such as NO_x and SO₂, which can lower the pH of the culture medium. The susceptibility to pH changes and optimal pH values vary greatly among different strains. Sung et al. [89] reported that *Chlorella* sp. KR-1 grew well in pH range from 4–7; however, its growth was significantly inhibited at pH below 3.5. *Synechococcus* PCC7942, reported by Kajiwara et al. [92], experienced slow growth at pH of 5.4 and maximum growth at pH of 6.8, indicating a much narrower pH range. Studies also showed that under different pH conditions, the content of dissolved free CO₂ changed accordingly. Furthermore, for different microalgal strains, even under the same pH conditions, photosynthetic efficiency is different due to varied CO₂ affinity coefficient. These factors must be considered when evaluating the response of algae strains to pH changes.

There are successful examples in selecting acid tolerant microalgal strains for waste gas bio-mitigation. Cuaresma et al. [93] isolated an acidophilic *Chlamydomonas acidophila* from river, which exhibited 50% higher photosynthetic efficiency at pH of 2.5 than at pH of 6. Interestingly, this strain was able to sustain pH as high as 9. It was found that the carbon was present in the water mainly in the form of HCO₃⁻, and *C. acidophila* was still capable of converting HCO₃⁻ to CO₂ at pH of 9. By injecting 15% CO₂, the CO₂ removal rate increased with the increase of pH.

4.2.4. Nutrient requirement

Major nutrients in the culture medium such as N, P and other mineral elements such as Mg, K, Fe, Co, Vitamin, are essential for the growth and carbon sequestration of photoautotrophic microalgae. Optimal N and P levels and C:N:P ratios will facilitate fast growth and efficient CO₂ fixation. For example, Ota et al. [94] found that by regulating the ratio of CO₂ and N₂, *Chlorococcum littorale* had attenuated photorespiration and synthesized β-oxidated fatty acids under aerobic conditions. Under nitrate replete conditions with 5 to 50% CO₂ concentrations, fatty acid content of microalgae remained unchanged. However, under nitrate deficient conditions, fatty acid content decreased with increasing CO₂ concentration. The study also found that the ratio of HCO₃⁻/CO₂ was the limiting factor for fatty acid accumulation under nitrate deficient conditions.

Waste streams derived from a variety of wastewater sources. e.g., agricultural run-off, concentrated animal feed operations, and industrial and municipal wastewaters contain large amounts of N, P and other trace elements, and have the potential to replace artificial media for low cost carbon sequestration as well as algal biomass production while obtaining credits for wastewater treatment at the same time [17,19,20,95]. It has to be noted that, however, high concentrations of nutrients (especially ammonium and ammonia) and some refractory substances such as heavy metals, drugs, and disinfectant by-products in wastewater streams, are toxic for microalgal growth, and would impact the performance of microalgae-based systems for CO₂ mitigation, posing a serious challenge to the efforts in using wastewater

streams as microalgae cultivation medium [96–100]. Strategies to address the above mentioned issues include better understanding microalgae-based nutrients removal mechanism, improving capabilities of locally isolated or genetically-engineered microalgae to tolerate various wastewater streams, and optimizing wastewater pretreatment procedures as well as environmental conditions, etc.

5. Value-added practice in microalgae based carbon sequestration

Most physical and chemical carbon sequestration methods are lack of ability to utilize the captured carbons for the production of value-added products, which may be attributed to the extremely low reactivity of CO₂. This puts physical and chemical methods in a disadvantageous position in terms of economic feasibility. On the other hand, carbons captured in microalgal biomass are immediately available for conversion to energy and value-added products via many pathways. Innovative utilization of microalgae biomass can play an important part in the economic equation of microalgae-based carbon sequestration technologies.

5.1. Algae harvest

The lack of economically viable and highly efficient harvesting technologies is one of the major challenges impeding the commercialization of microalgae production. In general, intensive energy input is required for microalgae harvesting due to the tiny size (1–70 μm) and negative charges of cell wall surface which accounts for 20% to 30% of total production costs [101]. So far, various harvesting techniques are used, including sedimentation, centrifugation, flocculation, air flotation, filtration, electrophoresis, and a combination of the above methods [101,102].

The simplest and easiest harvesting method is gravity sedimentation. Major drawbacks of this method are time-consuming and low-cell density of harvested algal biomass [101]. Centrifugation is considered as the most efficient method with efficiency higher than 95% and is widely used in small- and medium-scale microalgae culture systems [103]. However, it is not suitable for large-scale microalgae harvesting due to relatively high capital investment as well as high energy consumption and operation cost. Therefore, centrifugation is only used to harvest microalgae containing high value metabolites, such as polyunsaturated fatty acid (PUFA), pharmaceutical and cosmetic products, etc. [104]. Flotation can also effectively realize microalgae-water separation, however, the main disadvantages are its intensive energy requirement and lack of environmental sustainability [105]. Flocculation requires the use of chemicals and/or a certain amount of synthetic polymers. Addition of chemicals will increase the possibility of toxicity, high cost, corrosion and secondary pollution, significantly reducing the feasibility for large-scale operation [102]. An alternative method is to develop a natural metal cation-assisted self-flocculating method [19]. Filtration is generally applied to harvesting microalgae species with unique properties, such as microalgal cells of very long size and with tendency to form large aggregates (e.g., *Spirulina* sp. and *Micractinium* sp.) [103]. A novel fungi-assisted bio-flocculation method has been reported [20]. By adding filamentous fungal spores into algae broth and optimizing co-cultivation conditions, algae-fungi pellets were formed within two days of cultivation. The diameter of pellets was about 3–5 mm and thus can be harvested through simple filtration (Fig. 6) [20]. The efficiency of this method was up to 98% [20,106,107]. The method was regarded as a "game changing" technology and is expected to fundamentally solve the problem of large-scale microalgae harvesting [106].

5.2. Production of biofuels and bio-based byproducts

Harvested algal biomass can be converted to different biofuels (e.g.

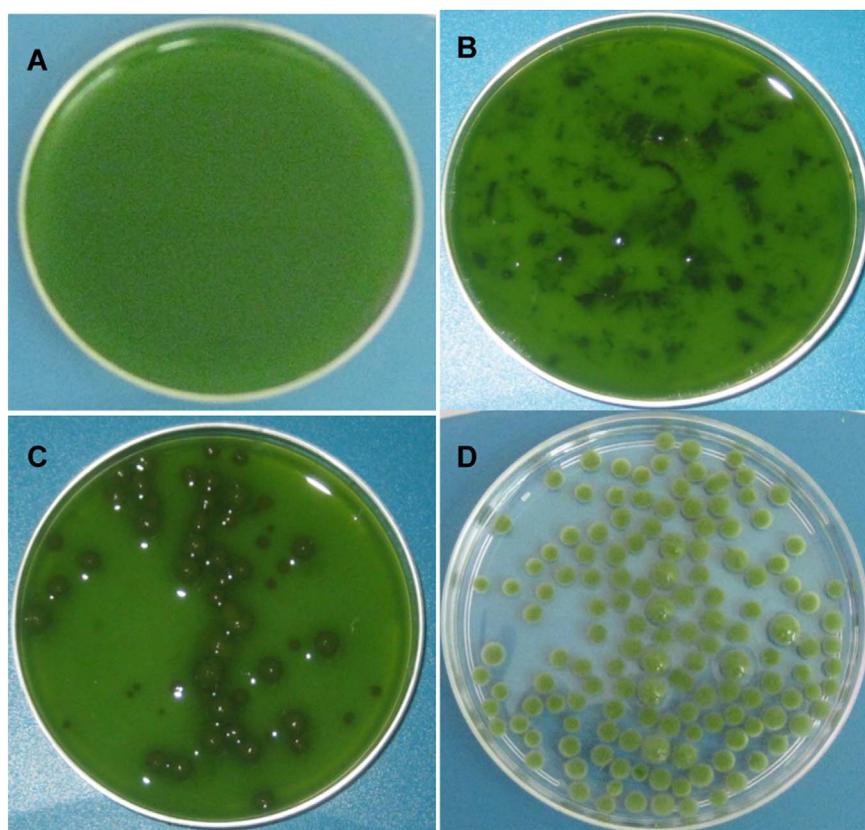


Fig. 6. The process for fungi-algae pellets formation. A, algae alone; B, after adding fungi spores; C, some of algae entrapped by fungi; D, all microalgal cells entrapped and harvested by forming fungi-algae pellets [20].

biodiesel, green diesel, green gasoline, green jet fuel, bio-crude oil, ethanol and hydrogen, etc.) or other value-added chemicals (e.g. food, cosmetics, pharmaceuticals, etc.) through different downstream processes. The harvested wet algal biomass slurry can be directly converted to biogas through anaerobic fermentation or to bio-crude oil through hydrothermal liquefaction. Lipids can be extracted from dry algal biomass for biodiesel production through lipid transesterification. Ethanol fuel, bio-oil, and bio-hydrogen can be produced from dry algal biomass via different pathways [20,108,109]. Algal biomass can also be used for the production of long chain omega-3 fatty acid (DHA/EPA), biological active substances, cosmetics, pharmaceuticals, fertilizer, and animal and aquaculture feed [17,110–112].

6. Economic consideration of different CO₂ capture technologies

Despite the perceived benefits of microalgae based technologies, commercial applications of these technologies are yet to be realized because there is still lack of understanding of the economic viability of microalgae-based technology and how competitive this technology is when leveled against other technologies. Table 2 shows the estimated costs of some chemical, physical, and microalgae methods from the literature. Chemical reaction-based CO₂ capture and storage methods involve several steps, some of which can be quite costly. These methods uses strong chemical reagents, such as monoethanolamine (MEA), amine and potassium hydroxide (KOH) [14] to capture CO₂ and require large amounts of energy for CO₂ separation (high temperature and high pressure are needed for regeneration process) [113], transportation and storage. In general, the cost of CO₂ separation and compression to 110 bars (for transportation) is estimated to be \$30–50 per ton of CO₂, and transportation and sequestration costs are estimated as \$1–3 per 100 km and \$1–3 per ton of CO₂, respectively [12,114]. To minimize the cost of separation and transportation, CO₂

Table 2
Economic evaluation of different CO₂ sequestration methods.

CO ₂ capture methods	Estimated Cost (\$/Ton)	References
<i>Chemical methods</i>		
Amine scrubbing	55	[115]
Monoethanolamine (MEA) for 50 MW plant	57.1	[116]
Monoethanolamine (MEA) for 500 MW plant	40.5	[116]
<i>Physical method</i>		
CaO-based calcium looping process	20	[118]
<i>Microalgae based methods</i>		
^a Microalgae sequestration Scenario 1	753.84	[119]
^b Microalgae sequestration Scenario 2	1,698.86	[119]
^c Microalgae sequestration Scenario 3	1,616.37	[119]
^d Microalgae sequestration Scenario 4	500.73	[119]

^a Microalgal cultivation for biodiesel production and CO₂ bio-mitigation.

^b Microalgal cultivation for biodiesel production and CO₂ bio-mitigation, integrated with anaerobic digestion of biomass residuals after lipid extraction;

^c Microalgal cultivation for biogas production and CO₂ bio-mitigation;

^d Microalgal cultivation for mixed gas (supercritical gasification) production and CO₂ bio-mitigation.

content in waste gas should be maintained as high as possible. However, the actual CO₂ content in industrial flue gas is relatively low (10–20%), which inevitably increases the costs of CO₂ collection and capture. A comprehensive techno-economic analysis conducted by Kierzkowska et al [115] showed that the costs of CO₂ capture using amine scrubbing was approximately \$55 per ton. In addition, capture costs also depend on power plant capacity (corresponding to CO₂ production capacity). For instance, Kadam [116] shows that the delivered CO₂ cost for a 500-MW plant that used MEA extraction

Table 3
Energy and cost requirements of microalgae-based biofuel production and CO₂ bio-mitigation [119].

Scenario No.		1	2	3	4
Scenario description		Microalgal cultivation for biodiesel production and CO ₂ bio-mitigation	Microalgal cultivation for biodiesel production and CO ₂ bio-mitigation, integrated with anaerobic digestion of biomass residuals after lipid extraction	Microalgal cultivation for biogas production and CO ₂ bio-mitigation	Microalgal cultivation for mixed gas (supercritical gasification) production and CO ₂ bio-mitigation
Energy input (kWh/yr)	Cultivation	274,004	273,994	274,007	273,958
	Harvesting	193,204	193,149	193,172	193,149
	Drying	82,980	83,000	-	-
	Lipid extraction	112,871	112,821	-	-
	Biodiesel synthesis	115,440	115,406	-	-
	AD+CHP*	-	83,430	109,799	-
	SWG+CHP*	-	-	-	663,082
	Total	778,420	861,886	576,798	1,130,190
Net energy production (kWh/yr)	Electricity	170,380	426,770	-129,810	497,890
	Heat	-93,620	416,150	670,760	784,530
	Total	76,760	842,920	540,950	1,282,420
Capital costs (\$/yr)	Cultivation	30,390	30,386	30,386	30,385
	Harvesting	6393	7550	7548	6394
	Drying	253	256	-	-
	Lipid extraction	4895	4,894	-	-
	Biodiesel synthesis	3312	3,313	-	-
	AD+CHP*	-	5,776	6,074	-
	SWG+CHP*	-	-	-	7,529
	Total	45,243	52,174	44,013	44,313
Operation costs (\$/yr)	Nutrients	148,822	149,282	144,962	148,832
	Flocculant	202	202	231	189
	Extraction solvent	12,356	12,343	-	-
	Esterification solvent and catalyst	16,592	16,592	-	-
	Labor	158,353	158,337	158,326	158,340
	Heat	11,145	11,129	-	40,070
	Electricity	34,646	39,508	33,663	27,250
	Overhead	95,012	95,002	94,977	94,995
	Maintenance	1,059	1,214	1,017	993
	Solid waste treatment	26,123	22,208	29,224	2,413
	Total	504,309	505,868	462,400	473,082
	Net costs (\$/yr)	Expenditure	826,275	839,096	782,367
Revenue		136,125	190,682	37,431	141,394
Net		-690,150	-648,414	-744,936	-658,774
Net CO ₂ production (ton/yr)	Uptake	1,578.41	1,578.41	1,578.41	1,578.41
	Generation	662.90	1,196.74	1,117.54	262.78
	Net	-915.52	-381.68	-460.87	-1,315.64
CO ₂ removal/ton algae (ton CO ₂ /ton)		0.92	0.38	0.46	1.32
CO ₂ removal cost (\$/ton CO ₂)		753.84	1,698.86	1,616.37	500.73

*AD + CHP = anaerobic digestion + combined heat and power.

*SWG + CHP = supercritical water gasification + combined heat and power.

was \$40.5/ton but rose to \$57.1/ton for a 50-MW plant, indicating a 41% increase in cost. A significant drawback of chemical-based CO₂ capture methods is that the captured CO₂ can hardly be used to produce value-added by-products.

Physical carbon trapping technology, e.g., deep ocean injection and underground storage methods, should be used in sparsely populated areas to reduce the impact of leakage on humans and the environment. In addition, these methods can only slow down the release of CO₂ in practical sense. Thus it is generally believed that the traditional physical carbon capture and storage technology is not sustainable and environmentally friendly because of the risks of leakage during transportation and storage [117]. Moreover, solid physical adsorption technology is based on the affinity of CO₂ to the surfaces of the capturing material such as zeolite, activated carbon, CaO, and metal-organic frameworks (MOFs) and its ability to bind through weak interactions without any chemical bonding. Similar to chemical methods, physical methods also suffer the drawback that the stored CO₂ is unsuitable for utilization. Mackenzie et al. [118] calculated the capture costs of approximately \$ 20 per ton of CO₂ using CaO-based calcium

looping process (Table 2), which is relatively low compared with current solid physical adsorptions technologies and most of the chemical methods from economic point of view and deserve further study.

The costs of microalgae-based CO₂ mitigation with biofuel production are generally higher than the chemical and physical methods (Table 2). It is generally recognized that sequestration of CO₂ through algal biomass accumulation without value added utilization of the algal biomass is economically disadvantaged. Ventura et al. [119] conducted an analysis of CO₂ uptake and emission, energy input and output, costs, and revenue of four different scenarios where microalgae based CO₂ sequestration was combined with different bioenergy production routes such as lipid extraction and esterification (biodiesel), anaerobic digestion (biogas), and thermochemical conversion (syngas). The CO₂ removal costs ranged from \$500 to \$1690 per ton of CO₂. A closer look at the cost factors such as energy input and output, CO₂ uptake and emission, and capital and operation costs shows that the energy input and capital and operation costs for algae cultivation and harvest were very much the same among the four scenarios (Table 3), indicat-

ing that the variations in overall CO₂ removal costs were due to the post-harvest operations. Scenario 4 had highest energy input but also highest energy output. The net costs were in the range of \$675 to \$745 per year for a 1000 dry tons of algae plant. The data show that the high CO₂ removal costs for Scenarios 3 and 4 were attributed to the high emission of CO₂ during the anaerobic digestion process and combustion of the biogas, which significantly reduced the net CO₂ removal, raising the unit CO₂ removal cost by more than 100–300% compared with Scenarios 1 and 4. Nonetheless, these costs are very high compared with those of traditional chemical and physical methods. Fernández et al [120] considered six different cases where CO₂ source (pure or flue gas), water source (fresh or wastewater), nutrient source (fertilizer or wastewater), and biomass productivity (g/m² day) were varied. Their analysis shows that use of flue gas and wastewater significantly reduced the cost of CO₂ removal. In the best case where flue gas and wastewater were used and a productivity of 60 g/m² day was assumed, the cost was estimated at \$75 per ton of CO₂. Even at this cost, which is comparable with other traditional carbon sequestration processes, it remains unfeasible because carbon tax rate is below \$30 per ton. However, these case analyses provide attainable technical targets associated with algae cultivation. In order to improve the economic outlook, we need to develop efficient ways to utilize flue gas and wastewater, take cost saving from waste treatment into account, develop high performance algae strains and efficient cultivation processes to boost biomass and specific component productivity, convert biomass to value added products including fuels and commodities, and recycling residues and wastes to lower cost [3,12,33,119–121]. There is more room for substantial improvement in microalgae-based carbon capture process economics through fundamental biological breakthroughs for microalgal strain re-construction and engineering innovation for microalgal culture system design.

7. Concluding remarks and perspectives

Microalgae can grow in various environments with high growth rate and carbon fixation efficiency and are renewable biomass feedstock for the production of a wide range of value-added products. Research reported in the literature has demonstrated that many microalgae strains are capable of assimilating inorganic and organic carbons from concentrated and non-point sources. Many of the current microalgae cultivation operations can be adopted for applications tailored to carbon sequestration. Nonetheless, implementation of large-scale microalgae-based carbon sequestration technologies has yet to be realized. Innovation and improvement in following areas are necessary before microalgae based technologies can be commercialized: development of technologies to address carbon sources with different chemical forms and distribution characteristics, screening and genetic engineering of high performance strains, improving utilization of industrial waste gases, better understanding of microalgae-based carbon fixation mechanisms, improving CO₂ transfer and oxygen desorption, cultivation process optimization and scaling up, cost-effective photobioreactors, high efficient microalgae harvesting and conversion technologies, value-added products development, and system integration. The understanding and improvement of economic feasibility must be achieved through techno-economic analysis using production facilities with reasonable scale. Life cycle assessment models must be developed to evaluate the environmental impacts of microalgae-based carbon sequestration. With further rigorous research efforts in these areas, microalgae-based carbon mitigation will be advanced and gain broad applications, providing significant economic and environmental benefits in the foreseeable future.

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