

The Role of Phytoplankton in Carbon Dioxide Fixation

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Marine phytoplankton account for more than half of the carbon dioxide fixation of Earth. The export of carbon is highest at the photic zone of the ocean, which is dominated by phytoplankton. The term phytoplankton comes from the Greek words phyto (plants) and plankton (wanderers or drifters). Plankton can also be zooplanktons that feed on phytoplankton and release fecal pellets that are made of dissolved carbon particles.

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1. Phytoplankton

Oceans are the largest reservoirs of carbon on the planet. It is strongly believed that the ocean's capacity to store carbon is not infinite and the pH will change by 0.3 to 0.4 units by the turn of this century ^[1]. In oceans, 90% of the inorganic form of carbon is in bicarbonate, with the release of a proton that contributes to lowering the ocean pH causing acidification ^[2].

Marine phytoplankton account for more than half of the carbon dioxide fixation of Earth. The export of carbon is highest at the photic zone of the ocean, which is dominated by phytoplankton. The term phytoplankton comes from the Greek words phyto (plants) and plankton (wanderers or drifters). Plankton can also be zooplanktons that feed on phytoplankton and release fecal pellets that are made of dissolved carbon particles. Phytoplankton constitute organisms across multiple kingdoms such as Kingdom Monera, e.g., Cyanobacteria; Kingdom Protista, e.g., fire algae, diatoms, dinoflagellates, etc.; and Kingdom Plantae, e.g., Brown algae (Sargassum); while Kingdom Animalia consists of zooplankton ^[3].

The growth of the phytoplankton mainly depends upon three factors, i.e., nutrients, sunlight, and carbon dioxide. Phytoplankton-like plants have chlorophyll that fixes carbon dioxide to glucose using the Rubisco enzyme. Phytoplankton-like plants are sensitive to nutrient availability, and hence, depending on the species, their growth is limited in the ocean based on nutrient availability. Other factors that affect phytoplankton growth are water depth and temperature, pH and salinity, and the diversity of predators that feed on them. When all factors are optimal, there phytoplankton boom over the surface of the ocean to an extent that can be captured by satellite images, as they cover hundreds of miles over the ocean, jeopardizing other oceanic flora and fauna ^{[4][5]}.

2. Cyanobacteria

Cyanobacteria are a class of prokaryotic microorganisms that are blue-green photo-autotrophic Gram-negative bacteria or algae. These are the predecessors of green plants and possess carboxysomes, which are carbon dioxide concentration mechanisms. The enzymes used for carbon capture are the same as in plants, namely, RubisCO, and carbonic anhydrase. Carbon-concentrating mechanisms exist in cyanobacteria, similar to algae that increase the carboxylase activity of RubisCO enzyme by increasing carbon dioxide concentration near the fixing enzyme.

Among all the naturally found carbon-sequestering organisms, algae seem to be most efficient in converting carbon dioxide to biomass and fuel. Microalgae include microorganisms such as diatoms, euglenoids, and green, blue, golden, red, brown, and yellow algae, which fix carbon dioxide using RubisCO and are mostly single-celled. With a biomass accumulation rate that is 100-fold faster than terrestrial plants, microalgae are a very promising candidate for carbon-sequestering units ^{[6][7]}. Algae are high in lipids and could have direct use in the production of biofuels ^[8]. Biochar derived from algae has been studied and found to have excellent properties to be used as a fertilizer ^[9]. Microalgae have carbon-concentrating mechanisms, which increase the concentration of carbon dioxide near the RubisCO enzyme by separating it into a different membrane compartment, into which carbon dioxide and bicarbonates are transported. Uluva and Laminaria species of microalgae are prime choices for bioenergy production ^[10].

Every 1 kg of algae can fix 1.83 kg of carbon dioxide in ideal conditions ^{[11][12]}. Atmospheric carbon dioxide concentrations range from 0.03–0.06% (v/v), and algae can grow efficiently at 2% or higher. Certain species such as *Scenedesmus* sp. have a carbon dioxide tolerance even at 10–20% (v/v) concentration of carbon dioxide. An increase in biomass is the most significant pointer to indicate carbon dioxide sequestering ^[13].

3. Dinoflagellates

Dinoflagellate are a major part of oceanic phytoplankton that use Rubisco form II as a primary enzyme for carbon fixation ^[14]. The form II has both lower specificity and affinity for carbon dioxide/oxygen. This suggests that they have an advanced mechanism to increase carbon dioxide concentration, since they cannot fix carbon dioxide at lower ambient carbon dioxide concentration ^[15]. Of the various isomers of carbonic anhydrase (α , β , γ , and δ), δ -carbonic anhydrase has been implicated in lowering carbon dioxide concentration ^[14]. The enzyme carbonic anhydrase (CA) catalyzes rapid conversion of carbon dioxide and bicarbonate ion ^[16]. Many dinoflagellates have carbon-concentrating mechanisms (CCMs) to transport carbon into the cell ^[14]. One of the biggest bio-indicators of global warming is the bleaching of coral reefs. Coral reefs that belong to Kingdom Animalia are a complex system of multiple species belonging to phylum coelenterates. The base of the ecosystem is built upon dinoflagellates that live as intracellular photosynthetic symbionts. Loss of these dinoflagellates is believed to be the primary cause of coral bleaching that eroded 85% of the Great Barrier Reef in Australia in 2016 ^{[17][18]}. How do coral reefs become bleached? A slight change in water temperature (2 °F) disturbs the photosynthetic electron transfer and in the process damages the *PsbA* (D1), a photosystem II reaction center protein. This disruption of photosynthesis triggers the bleaching of coral reefs ^{[19][20][21][22][23]}.

While genetically engineering cyanobacteria has some success, engineering genes to improve the carbon fixation in diatoms and dinoflagellates has been a challenge due to the lack of critical gene transformation strategies. Initial studies of transformation using silicon carbide whiskers were suggested, but the experiments were difficult to reproduce ^{[24][25]}. Experiments with glass beads were performed for transient expressions, but even these were hard to reproduce ^{[17][26]}.

Recent studies showed that the stable transformation of dinoflagellates is reproducibly possible when they bombarded microparticle-containing plasmid-like minicircles that carried chloroplast gene *psbA*. They successfully transformed two minicircles providing resistance to chloramphenicol and astrazine and in the process assisting the selection of successful transformants. Inability to stably transform has less to do with the inherent genetics of protists than it has to do with the DNA's inability to cross the membrane barrier ^[17].

4. Diatoms

Diatoms exhibit a wide range of metabolic diversity due to their evolution that includes endosymbiosis among diverse lineages. This is why they can be used to make a wide range of compounds. Previous studies have shown that manipulating carbon dioxide levels of *Navicula pelliculosa*, *P. tricornutum*, *T. pseudonana*, and *Asterionella formosa* ^[27] can increase the accumulation of both lipids and carbohydrates. Diatoms *T. weissflogii* and *P. tricornutum* have exhibited excellent absorption systems for carbon dioxide and bicarbonate at quantities typical of ocean surface waters. It has also been observed that their absorption rates can adjust to a wide range of inorganic carbon supplies ^[28]. Mixotrophic cultivation regimes, in addition to photoautotrophy, can aid in the production of higher biomass concentrations and productivities ^{[29][30][31]}.

Marine photosynthetic organisms such as diatom and other microalgae uptake carbon dioxide using a mechanism called carbon-concentrating mechanism or CCM ^{[29][30]}. In diatoms, the carbon dioxide travels inside the cells similar to plants and concentrates near the site where Rubisco is located for fixation. This mechanism is important because it helps regulate the carbon flux and raise it to higher concentrations inside the cell compared to the outside for optimal Rubisco activity ^{[29][30][31][32][33]}. Very few mechanisms are known for the majority of diatoms. For the ones that are known, they use the Rubisco Calvin–Benson–Bassham (CBB) cycle in their chloroplasts, carbonic anhydrases, Glyceraldehyde-3-phosphate dehydrogenase (GAPDH), Phosphoglycerate kinase, Fructose 1,6 bis phosphatase, Sedoheptulose-1,7-biphosphatase, and various transporters ^{[25][26][30][31]}.

Presently, there is a dearth of literature with regards to carbon fixation in diatoms. Studies have shown that the assimilation of carbon dioxide is not linearly proportional to the availability. *P. tricornutum* showed a higher fixation rate with supplemented carbon dioxide concentration but had reduced biomass productions. The carbon fixation rate did not change when subjected to diverse pH ranges. There is diversity among diatoms for carbon substrate specificity for fixation purposes. While *P. tricornutum* assimilates in the form of bicarbonate, *T. pseudonana* takes up carbon dioxide predominantly ^{[27][28][29][30][31][32][33]}.

5. What Are the Challenges in Genetically Modifying Diatoms?

Though diatoms are being used as bioreactors for environmentally sustainable manufacturing of valuable metabolites, there is little research to explore their potential for carbon sequestration and fixation [30][32][33]. There is a huge opportunity available for research on this aspect; hence, it is surprising that this has been less explored. The one of the reasons is the environmental impact of genetically modified microorganisms. Diatoms, like other microalgae, provide huge potential as promising candidates for carbon sequestration and fixation. Genetically modified diatoms have the potential for explosive growth like other microalgae and are hard to predict. They can have a huge environmental impact, jeopardizing the marine ecological balance [34]. The genetically modified diatoms also need to be tested for the new metabolites that they may start producing post-modification that may prove toxic in large quantities to the grazers that are at the top of the food chain. Hence, marine ecosystem modeling need to be used to test and simulate the challenges associated before introducing a new species in the wild. New experimental marine models need to be developed that can assist in predicting and simulating the underlying challenges in a marine ecosystem.

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