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Chapter

Algae Based Bio-Plastics: Future of Green Economy

Arathi Sreenikethanam and Amit Bajhaiya

Abstract

Plastic has become one of the most crucial requirements of the modern-day living. The continuous reliance on the petroleum-based, non-biodegradable plastics has resulted in increased global environmental damage and rapid depletion of fossil fuels. Bioplastic, with remarkably similar properties to petroleum-based plastics is a promising alternative to overcome these emerging challenges. Despite the fact that algae and cyanobacteria are feasible alternative source for bio-plastic, there have been limited studies on strain selection and optimization of culture conditions for the bio plastic production. Naturally, algae and cynobacteria can accumulate higher amount of metabolites under stress conditions however one of the recent study on genetic engineering of *Synechocystis sp.* coupled with abiotic stresses showed up to 81% of increase in PHB level in the transformed lines. This chapter provides summary of various studies done in the field of algal bio-plastics, including bioplastic properties, genetic engineering, current regulatory framework and future prospects of bioplastic. Further the applications of bioplastics in industrial sector as well as opportunities and role of bio plastic in green economy are also discussed.

Keywords: algae, bioplastics, biodegradable, sustainability, green economy

1. Introduction

Plastics are synthetic carbon-based polymers that offer a variety of applications in everyday life. Petroleum-based plastics are widely used due to their desirable characteristics, which include low-cost, transparency, light-weight, strong heat resistance with a good weight-to-strength ratio. It can be easily shaped into different forms to produce a variety of materials. Thus, it has a wide range of applications in the expanding industrial sector. Overuse of these fossil-based, non-biodegradable polymers has shown serious impact on the environment, resulting in pollution, global warming and fossil fuel depletion. They have a poor recyclable potential and moreover, produce toxins in the recycling process. Furthermore, the plastic recycling process is quite challenging, as different plastics require different recycling techniques. Only around 10% of the total plastic manufactured every year is recycled, with the rest dumped into the water bodies and landfills [1, 2]. The indigenous microorganisms do not have inherent potential to degrade these plastic wastes. As a result, both terrestrial and aquatic ecosystems are severely harmed. Bioplastic made from renewable sources with similar qualities of fossil-based polymers is a viable alternative to overcome these major challenges. They are biodegradable and environment friendly, which can lead to a more sustainable and circular economy. In addition, bioplastics can be entirely decomposed by soil microorganisms without

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producing any harmful by-products [3]. A range of polymers utilised in bioplastic synthesis can be derived from key metabolites such as lipids, proteins, and carbohydrates. There are microbes that can utilise these polymers as a source of carbon and energy for their metabolism, and there are species that can produce exoenzymes to degrade them [4]. Given the fact that bioplastics can be made from a variety of renewable sources such as higher plants, bacteria, starch, and cellulose-based materials, algal plastics remain a high-demand research due to multiple advantages of algae as a feedstock.

Algae and cyanobacteria, also known as prokaryotic microalgae, are photoautotrophic organisms that grow at a faster ratewith high biomass. They are popular in the field of bioplastics because of their limited nutritional requirements, harvest regardless of the season and ability to thrive in non-arable environments, including waste waters [5]. Algaecan assimilate carbon dioxide into various organic compounds, which can then be transformed into useful biopolymers, resulting in reduced CO2 emissions, and ultimately leading to a safer environment [6, 7]. Algal biomass, in addition to algal metabolites, can be utilised directly in plastic production, where it can be blended with petroleum-based plastics to improve its mechanical qualities [8]. Algal biomass can also help to speed up the decomposition process of plastic, owing to its high nitrogen concentration, which improves microbial adhesion and promotes the biofilm formation [9]. Each of these beneficial attributes indicates algae as a potential future feedstock for bioplastic production. This chapter summarises all aspects of algae bioplastics and their function in ecological sustainability. This aims to assist and realise the relevance of bioplastic research, as well as the obstacles it faces and the necessity to overcome them in the future.

2. Properties of bioplastics

Bioplastics, as the name implies, are bio-based, biodegradableand compostable polymers containing mechanical and barrier attributes comparable to petroleumbased plastics. There are a variety of starch-based, cellulose-based, and proteinbased bioplastics in the market today, but most of them are derived from food crops, which compete with human consumption. Poly Lactic Acid (PLA) and Polyhydroxyalkanoates (PHAs) are two typical algal biopolymers that have advantages over plant-based bioplastics as algae are easy to cultivate, non-competitive with human food, and can be harvested throughout the year [10]. Biodegradability of these polymers isdetermined by their structures, and strong mechanical qualities make them suitable for industrial uses. Bioplastics also have the potential to be customised in terms of properties, making them far preferable to conventional plastics. **Table 1** shows some of the advantages and disadvantages of bioplastics. Thermal stability, tensile strength, viscosity, elasticity, oxygen permeability, and water resistance are some of the significant characteristics of bioplastics.

Advantages of bioplastics	Disadvantages of bioplastics	Reference
Energy efficient	Expensive	[10–13]
Flexible to be modified	Brittle	
Do not generate toxic by products	Low melt strength	
Biodegradable	Weak barrier properties	

 Table 1.

 Advantages and disadvantages of bioplastics.

Numerous studies have proven that adding additives such as plasticizers and fillers to bioplastics strengthen both their structural and mechanical features [14]. When used as fillers, algal biomass itself has the potential to improve biodegradability. Kalita et al. [9] studied the biodegradation abilities of PLA material with algae biomass as a filler, and found that it increased the bioplastic's biodegradability. Hydrolysis of ester groups into hydroxyl or carboxyl groups occurs during PLA degradation. The algal biomass and PLA composites were extruded into a film and subjected to abiotic and composting degradation conditions. Water hyacinth compost set up was constructed for compost degradation experiments, and the films were cut into pieces and placed in 1 M NaOH for abiotic degradation. The sudden drop in molecular weight under abiotic stress conditions symbolises the molecule's degradation, which was seen in the presence of algal bio fillers. In the test setup with algal fillers, the degradation in the compost conditions was also noticeable, with days required for total biodegradation decreased from 95 ± 7 to 60 ± 2 days. These experiments clearly demonstrate the effectiveness of algae in improving degradation due to its high nitrogen concentration, which attracts microbes [9]. When no additives are used, Starch-based bioplastics have strong biodegradability, but poor mechanical stability compared to traditional plastics [11]. Methods like Coating, blending, as well as physical and chemical alterations can all be used to enhance their properties making them a complete sustainable alternative. Coating is the process of applying a topcoat of materials such as polycaprolactone and polyethylene oxide to assist improved barrier properties, tensile strength and elasticity. Nanomaterials, cellulose, thermoplastic starch, polycaprolactone are common compounds used in blending. When polymers are blended with nanomaterials, the polymer becomes confined between the nanoparticles, resulting in better barrier properties. Cellulose and thermoplastic starch combines well with other biopolymers, lowering water permeability while increasing mechanical qualities such as tensile strength. Polycaprolactones decrease polymer brittleness while simultaneouslyimproving heat stability [15].

Thummala et al. [12] examined the effects of glycerol and sorbitol as plasticizers on protein-based polymers. The findings show that sorbitol enhanced tensile strength whereas glycerol and a combination of the two showed intermediate tensile strength, indicating that bioplastics can be altered to meet specific requirements [12]. Studies on the effect of mould temperature on the viscosity of algal biopolymers demonstrate that increase in mould temperature improves viscosity and water resistance [13, 16, 17]. Plasticizers such as glycerol, water, and latex can enhance antibacterial properties of bioplastics, overcoming bioplastic's limitations in the medical and food packaging industries [18]. Antimicrobial additives like Nisin and cinnamaldehyde increase mechanical qualities and do not interfere with biodegradability, which was formerly a major concern [19, 20]. Although many of the approaches for improving characteristics are performed on plant-based plastics, they can all be applied to algal biopolymers, thus expanding the scope of algal research.

3. Algae used in bioplastic production

Algae are diverse group of photosynthetic organisms that range in size from single-celled microalgae to multicellular macroalgae which play a key role in ensuring a balanced ecosystem. They produce high metabolite content which can be processed into a number of value-added products, offering them to obtain wide range of market opportunities [21]. Bioplastics are one such products derived from algal metabolites, with the most notable being polylactic acid (PLA), polyhydroxyalkanoates (PHAs). These metabolites are naturally formed by algal cells, however the addition of particular chemicals, changes in culture conditions, can help to enhance the metabolite production.

One of the most commonly studied PHAs in bioplastic research is polyhydroxy butyrate (PHB). Sodium acetate in the culture mediumcan enhance PHB accumulation in cells without interfering with cell multiplication [22]. Kavitha et al. [23] optimised culture conditions for PHB production using wastewater cultured *Botryococcusbraunii*. According to the data, the maximum production of PHB $(247 \pm 0.42 \text{ mg/L})$ was obtained at a 60% concentration of sewage water as culture medium at 40°C and pH 7.5 [23]. Mathiot et al. [24] performed study on selection of microalgal strains for the production of starch-based bioplastics. They selected ten microalgal strains, including Chlamydomonas, different species of Chlorella and Scenedesmus. The algal strains were grown in a sulphur-depleted TAP medium with an 18-hour light: 6-hour dark photoperiod with a light intensity of 125 µmol.m⁻¹. s⁻¹ at 25°C and pH between 7.2–7.4 They conclude that *Chlamydomonasreinhardtii* is the best strain for bioplastic production among the 10 strains tested, with a starch to biomass ratio of 49 percent in a sulphur-depleted medium and excellent plasticization with 30 percent glycerol at 120°C [24]. In comparison to synthetic media, a study on optimising culture conditions on Chlorella Salina to discover the best photoperiod, CO2 concentration, and nutrient limitations suggests wastewater under controlled conditions as a feasible medium for optimum starch and carbohydrate production. The findings shows that a 12 h: 12 h light–dark cycle, 5% v/v CO2 concentration, and a combination of nitrogen and phosphorous limits is the ideal culture condition with highest starch and carbohydrate content. However, as compared to culture in synthetic medium, wastewater aerated with 5% CO2 is considered more sustainable, with double metabolite accumulation [25]. According to Das et al. [26] the leftover algal biomass after extracting lipids can also be used in bioplastic production and the result shows 27% PHB content. Chlorella pyrenoidosa was the strain under study in Fogg's medium with 80 Lux light intensity and UV spectroscopy analysis confirms PHB accumulation [26]. Since microalgae are low-cost substrate, Khomlaem et al. [27] used defatted Chlorella biomass and used as a substrate for three bacterial strains to accumulate PHAs. *Cupriavidusnecator*, a bacterial strain employing 75.4 percent defatted chlorella biomass, demonstrated the highest PHA accumulation (7.51 + 0.20 g/L) among the studied strains [27]. Another analysis revealed that adding glycerol as a plasticiser with defatted Chlorella biomass (DCB) to make Chitosan-based biodegradable films improved mechanical characteristics. Higher DCB concentrations resulted in increased tensile strength, reduced water vapour permeability, and reduced transparency. According to FTIR and SEM analysis, the increased attributes were because of the uniform distribution of DCB, which establishes strong hydrogen bonds throughout the matrix [28]. When employed as a carbon source, DCB can also be used to accelerate PHA and carotenoids production [29]. Scenedesmussps., that has not been studied extensively in the field of bioplastics, could also be a source of PHA in modified nutritional conditions. Since it has a rigid cell wall, it can withstand high temperatures, pH, copper concentrations, and a certain amount of salt [30, 31]. The majority of the studies described are recent findings, and the most of them focused on *Chlorella sps.* Because of its rapid growth rate and ease of cultivation.

4. Cyanobacteria used in bioplastic production

Cyanobacteria, commonly knownas blue green algae, are gram-negative prokaryotes with a wide range of species. In comparison to microalgae, they are well recognised for their PHA accumulation under stress conditions and are extensively

studied for bioplastic production. In combination with microalgae and heterotrophic bacteria, cyanobacteria can be grown in municipal and industrial waste waters. They can grow rapidly in wastewater systems and serve a variety of different functions such as heavy metal removal, reduction of Chemical Oxygen Demand (COD) and Biological Oxygen Demand (BOD). For commercial production, a two-stage cyanobacterium cultivation can also be employed, with a photoautotrophic phase followed by a heterotrophic stage with a sole source of carbon [32]. In addition, filamentous cyanobacteria such as *Pseudanabaenasps*. and *Aphanocapsasps*. can survive in nutrient deprived conditions and can also outcompete rapidly growing green algae like Chlorella [33]. Cyanobacteria can be blended with other bioplastics, their smaller size facilitates uniform dispersion, which helps to form cross linkages and increase mechanical qualities. Spirulina is one of these species, which has been widely used in the food industry for many years due to its high protein content and has recently been exploited in the bioplastic research with promising results [34]. The addition of Polyvinyl alcohol as a compatibilizer and glycerol as a filler to a spirulina-based bioplastic increased the tensile strength by 1.89 kgf/cm2 and the elongation by 4.17 percent over commercial plastic [35]. When cultivated in modified Zarrouk medium, where the manufacturing cost was quite

Strain	Culture conditions	Biopolymer	Yield (% dry cell weight) R	eference
Nostocmuscorum	Acetate in medium +Dark incubation	РНВ	43%	[42]
Synechococcus sp. MA19	Autotrophy, Phosphate deprivation	РНВ	55%	[43]
Synechocystis sp. PCC6803	glucose containing BG11(Pre-grown) medium+Acetate+ Phosphhate deprivation	РНВ	29%	[44]
Synechococcussubsalsus	Nitrogen deprivation	PHAs	16%	[45]
Spirulina sp. LEB-18	Nitrogen deprivation	PHAs	12%	[45]
Spirulina subsalsa	Increased salinity+ Nitrogen deprivation	РНА	7.45%	[46]
Calothrixscytonemicola	Photoautotrophy in nitrogen limitation	РНВ	25.4 ± 3.5%	[47]
Aulosirafertilissima	Acetate and citrate supplemented medium	РНВ	66%	[48]
Aulosirafertilissima	Acetate supplementation+ phosphate deprivation	РНВ	77%	[48]
Anabaena cylindrica	Acetate supplemented BG11 medium	РНВ	2%	[49]
Spirulina maxima	Acetate supplemented mixotrophic conditions	РНВ	3%	[50]
Microalgal consortium	Waste water	PHA	43%	[51]

Table 2.

Microalgae and cyanobacteria used in bioplastic production.

high, spirulina-based plastic had higher biodegradable properties with 6.2 percent PHB content [36]. In contrast, a research of thermoplastic blends of bioplastics with spirulina grown in wastewater found that spirulina outperforms Chlorella as a blend. As a result, it is considered as more suitable for commercial purposes [37]. *Spiriulina platensis* showed increased thermal stability without influencing water vapour transmission rate when used as a filler in wheat-gluten based bioplastic [38]. Arias et al. [39] studied the effect of nitrogen and phosphorous starvation in two photoperiods, one with full light and the other with 12 h alternate light and dark, on a mixed culture dominated by cyanobacteria. According to their findings, Nitrogen limitation under alternate light illumination yielded the maximum carbohydrate concentration of 838 mg/L [39]. Synechocystissps. Showed increased PHB accumulation by up to 38 percent when cultivated in the presence of fructose and acetate in phosphorus deficient and gas-exchange limited conditions, which was eight times higher than accumulation under autotrophic conditions [40]. However, a study on Arthrospira platensis found that limiting nitrogen and phosphorus in the medium at the same time resulted in lower PHB and Phycocyanin synthesis. This research was carried out in both autotrophic and mixotrophic conditions, in both normal and nutrient-limited environments. Although autotrophic conditions with more CO2 resulted in increased PHB production of 33%, there was no significant increase in other conditions. The results were justified by stating that the effects of nutrient limitation conditions may differ from species to species, and that limiting both nitrogen and phosphorous at the same time would not be a good option in the case of Arthrospira [41]. The very first research on PHA synthesis in Synechococcus elongates was successful and exhibited maximum production of 17.15 percent of PHA under nitrogen starvation and phototrophic conditions with 1 percent sucrose as external carbon source. However, its yield was lower than bacterial systems and can be enhanced by applying alternative nutrient deprivation conditions and by using genetic tools (**Table 2**) [52].

5. Biopolymers derived from algal metabolites

Algal biomass, in addition to algal metabolites, can be directly moulded into bioplastic beads or sheets. When actively growing cultures are centrifuged, TAG accumulates and settles in the pellet. The pellet can be combined with additives like glycerol before being formed into the appropriate bioplastic shape [53].

5.1 Polyhydroxyalkanoate (PHA)

PHAs are biopolyesters produced as intracellular inclusions by a variety of microorganisms, especially in the presence of abundant carbon and limited essential nutrients. They accumulate in the cell as it enters the stationary phase and can account for up to 80% of the cell's weight. These inclusions are protein and lipid-based membrane bound inclusions. They serve as energy reserves for the cells, allowing them to endure oxidative stress, UV irradiation, temperature shock, and osmotic imbalance. PHAs are made of polyhydroxyalkanoic acid monomer units in which the ester bond is between the carboxyl group of one monomeric and hydroxyl group of the next monomeric unit. The R group in each monomeric molecule forms an alkyl side chain. Monomers can differ depending on the organism's substrate, resulting in the formation of various polymers and copolymers [5, 54]. They are UV resistant, insoluble, and have low oxygen permeability. Melting temperatures range from 40 to 180°C, with a glass transition temperature of -50 to 40°C. The temperature ranges stated here differ depending on the R-group [55]. Cyanobacteria

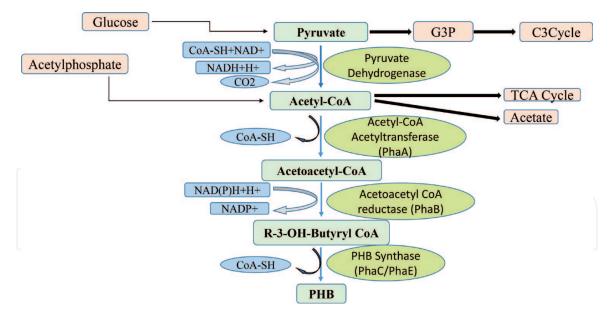


Figure 1.

Metabolic pathway of PHB synthesis showing acetyl-CoA as a branch point.

are known as best producers of PHA and are being constantly studied for ways to boost synthesis in nutrient-restricted conditions or through genetic engineering experiments. Polyhydroxy butyrate (PHB) is the well-known and widely marketed PHA which has qualities similar to petroleum-based polymers [56]. It's made up of repeating units of 3 carbon atoms and a methyl group [55]. **Figure 1** depicts the PHB synthesis metabolic route with acetyl-CoA as a branch point. PHB production can be improved by altering the enzymes in the pathway, either by boosting PHB synthesis enzymes or by deletions of other Acetyl-CoA consuming enzymes to increase the substrate for PHB synthesis.

Acetyl CoA is a branch point in microalgae's central metabolism. In the cells, glucose is transformed to pyruvate, which can then be converted to Glyceraldehyde 3-Phosphate (G3P) and enter the Calvin Cycle (C3 cycle) to produce carbohydrates. The enzyme pyruvate dehydrogenase transforms pyruvate to acetyl CoA, which is the primary substrate for PHB synthesis. Conversion of Acetyl phosphate is another source of Acetyl CoA synthesis. Acetyl CoA synthesised can be used by the cells in PHB synthesis, Tri carboxylic acid (TCA) cycle, or converted to acetate. A set of enzymes, including PhaA, PhaB, and PhaC/E, convert acetyl CoA to PHB in the PHB production pathway [57, 58].

5.2 Polyurethane (PU)

Polyurethane is commercially available biopolymer widely used in adhesives, coatings, elastomers, and foams [59]. They can be made from oils, in which algal oils, such as triglycerides are sustainable sources. The composition of fatty acids varies from species to species. Pawar et al. employed *Chlorella* to produce polyols through oxidation. The oil had a 10% saturates composition and a 2% unknown fatty acid content. Algal oil epoxidation yielded good results, with conversion rates comparable to other vegetable oils. The epoxide ring opening of epoxidized algal oil with ethylene glycol and lactic acid was successfully achieved for the manufacture of rigid polyurethane foams. The synthesis resulted in the manufacture of polyurethane with characteristics similar to those of commercially available polyols [60]. Patil et al. [61] prepared nanocomposite coatings using Polyol from algal oil and ricinoleic acid by combining eggshell based Silver doped nanoparticles in a Polyurethane matrix. The qualities of PU nanocomposite coatings were compared

to PU without nanoparticles and according to the findings, PU coatings had good physico-mechanical properties [61]. Furthermore, the polyurethane coatings made from algal oils exhibited antibacterial and anticorrosive qualities [62].

5.3 Polylactic acid (PLA)

PLA is usually made from algal feedstocks that is fermented to produce lactic acid and then polymerised [63]. PLA is one of the most efficient plastics since it uses a less amount of feedstock (sugar) to generate a biodegradable plastic. In comparison to other biopolymers, the rate of CO2 release is also lesser. It's one among the polymers whose stereochemical structure can be easily changed to get a higher molecular weight. Other characteristics, such as amorphousness and semi-crystalline nature, can also be altered by varying isomers [64]. Since it is an FDA-approved biopolymer, it is commonly utilised in food packaging. Because the monomer is chiral and exists in two optical isomeric states, various PLA structures can be formed. Poly (L-Lactide) (PLLA), Poly (D-Lactide) (PDLA), and Poly (D, L-Lactide) are the three. Packaging made of poly (D, L lactide) with 90% L-Lactide is commonly utilised. Increased D-Lactide in the composition results in a polymer with a high crystalline structure with good mechanical and barrier properties, but it is not economically successful due to its high cost [65].

5.4 Cellulose acetate

Cellulose is the most prevalent natural polymer on the planet, making it a limitless source of raw material for creating environmentally beneficial products without interrupting the food chain. It is made up of glucose monomer units linked together by a β -1,4 glycosidic linkage that compacts them and forms strong inter-chain hydrogen bonds. The structure's alternating side chains contribute to its high crystalline nature, which causes brittleness, poor flexibility, and weak tensile strength. But, in combination with appropriate plasticizers, cellulose derivatives such as ethers and esters act as bioplastics with a range of applications. One such polymer is cellulose acetate, which is used in the production of items such as spectacle frames, combs, cigarette filters and disposable jewellery. Due to its dimensional stability, rigidity, and printability, cellulose acetate was also recognised for its usage in food packaging, where it was used to wrap baked products. However, the packaging industry no longer prefers cellulose acetate due to its weak moisture and gas barrier attributes, as well as the fact that it will be hydrolysed to produce acetic acid. The production of cellulose acetate on a large scale is normally done under regulated conditions, particularly the temperature, which impacts the degree of polymerisation (DP). The product quality will be affected if the DP is too low [66, 67]. Although there are numerous studies on the manufacturing of cellulose acetate from plant sources, research on the production of cellulose acetate from algal cell walls are still under progress. However, because the cell walls are not entirely formed of cellulose, the yield of cellulose-based polymers will be low. But the large-scale production in collaboration with other biopolymers using the remaining biomass will be a viable option to use the complete biomass.

6. Genetic engineering for improved metabolite production

Genetic engineering is a sophisticated method for gene manipulation that has been utilised in a number of research studies. In terms of bioplastic manufacture, there have been various investigations on plant gene manipulations, which are referred

to as first- and second-generation bioplastics. However, research has now shifted to third-generation bioplastics generally known as algal bioplastics. Genetic studies on algae and cyanobacteria are easier due to their lower complexity compared to plants, and they have a high potential for producing bioplastic. Synechosystis sp. PCC6803 is the first photosynthetic organism to be completely sequenced, which is one of the reasons it is being explored in gene manipulation for increased PHA production [54]. PHB accumulation is known to be increased in nitrogen-deficient conditions, and the Sigma factor *sigE* is known for its potential to activate numerous carbohydrate metabolic pathways, including the PHB synthesis pathway. Under nitrogen-deficient conditions, overexpression of sigE in Synechocystis sp. PCC 6803 resulted in increased PHB synthesis. Importantly the molecular weight and monomer units of the produced PHB are identical to those of wild-type PHB [68]. Transformation experiments on Synechocystis sp. PCC 6803 containing pha genes were also effective, with the resultant cells accumulating 12-fold higher PHB under nitrogen stress than the wild type strain [69]. Acetyl-CoA is a branch point in algal cell's core metabolism, and it can be transformed into a number of substances based on the cell's need via various enzymes, including PHB synthesis. Phosphotransacetylase and acetyl-CoA hydrolase are enzymes that convert acetyl-CoA to acetate and are encoded by the pta and ach genes, respectively. This reduces the amount of substrate available for PHB synthesis. Phosphoketolase, on the other hand, is an enzyme produced by the *xfpk* gene that can increase acetyl-CoA levels in the cell. Carpine et al. [70] employed a different approach to boost PHB production, instead of overexpressing the enzyme in the synthesis pathway, they aimed to increase the substrate concentration for PHB synthesis. This experiment was designed by engineering Synechocystis sp. PCC6803. Seven mutants were constructed with three different gene alterations, including deletions of the pta and ach genes and overexpression of the xfpk gene. They were effective, and their findings demonstrate that the mutant with all three modifications accumulated the most PHB (232 mg/L) [70]. Orthwein et al. [71] discovered a novel protein, pirC (PII-interacting regulator of carbon metabolism), and investigated its function for PHB production. The PirC deficient mutant strain of Synechocystis found to have higher phosphoglycerate mutase activity, leading to increased PHB production. The strain was modified even more by transferring PHA metabolism genes (phaA and phaB) from known PHB producing bacteria, Cupriavidus necator, which also showed good results and was termed PPT1. The strain produced 63 percent PHB in nitrogen and phosphorus limitation. PHB level increased to 81 percent in the presence of acetate under the same culture conditions, making it the highest PHB content to be reported in any known cyanobacterium [57]. Although cyanobacteria are widely used in genetic engineering studies, Hempel et al. [72] incorporated a Ralstoniaeutropha bacterial PHB synthesis pathway into the diatom Phaeodactylumtricornutum, demonstrating microalgae as a workable model for PHB production. These bacterial enzymes were sufficient to synthesise PHB in the cells, accounting for up to 10% of the dry weight of the algae. This research was one of the first to utilise genetic engineering to produce PHB in microalgae, and it cleared the path for further research [72]. CRISPR/Cas systems are gene editing tools that can produce a variety of mutant, knock-out, and knock-in strains with desired characteristics. This approach has yet to be thoroughly investigated in algal systems, particularly in bioplastic research [73].

7. Applications of bioplastics

Plastics have a range of applications, and bioplastics have the potential to replace conventional plastics in all of them. Bioplastics can be moulded in a variety of ways, from fibre to thin film, and can be designed in any size, shape, or dimension. PHB

is most likely to be widely used in food packaging, whether it is for fresh or longterm storage. Green house films, protection nets, and grow sacks are examples of bioplastics used in agriculture to maintain appropriate conditions and protect the crop from physical and biological risks. Unlike synthetic polyethylene, these grow bags do not cause deformity, making them root friendly. Since PHA is a biomaterial, it can interact with biological system and elicit a favourable response from the host. As a result, it has applications in medicine, including in the engineering of biological tissues such as bone, cartilage, and skin. It can be utilised to regenerate dental tissue [2], employed for fraction fixationas well as surgical sutures. The medicament can be loaded into PHB-based wound dressings, and the fibrous nature of the material facilitates the drug's release into the wound. The property of cancer cells adhering to PHB sheets has been documented, and contact angle techniques can be used to identify it. In comparison to biopsy, which is often used for medical examinations, this approach is painless [74]. PHB with a high molecular weight can be employed as a drug carrier [23, 75]. Because of its non-adhesive qualities, PHB can be utilised as an effective antibacterial agent in aquaculture, preventing pathogens from forming biofilms and thereby inhibiting infection. PHB is an antifouling compound that can be blended with other metals and applied to the hull to prevent undesired marine creatures from settling [58]. PLA blends are used in a variety of applications, including computer and mobile phone casings, medical implants, and various packaging materials such as cups, tins, and bottles [64]. Algal plastics can also be used to make plastic beads, which have applications in fishing, ornament crafting, and shooting sports. However, it is not much explored due to the high expense of extraction and purification of biopolymers. But research on *Chlamydomonas reinhardtii* showed that triacylglycerol can be directly moulded into 7 mm beads without the use of extraction or purification [53].

8. Challenges in the field of bioplastics

Microalgae are well-known for their ease of cultivation, capacity to grow in waste waters, and ability to survive in adverse environments, but producing a marketable product from this biomass has numerous hurdles at each step along the way, from cultivation to market release. Not all species are capable of adapting to a wide range of cultural settings. As a result, the strain chosen for research may not be adapted to the designed conditions, making the cultivation phase challenging. Following cultivation, the harvesting process, which is highly costly, presents the next hurdle. Major research focuses on ways to boost metabolite synthesis, but there is also an urgent need to identify cost-effective harvesting methods. In addition, the biomass and metabolites produced will be insufficient for industrial production. This difficulty can be solved by inducing heterotrophic conditions with an external carbon source, however there is a high risk of contamination [76]. Genetic engineering is well known for its excellent outcomes in terms of increased metabolite production but, it also offers several challenges, including the need for a genome sequence, the difficulty in gene alterations, and the maintenance and genetic stability of transgenic strains [77]. Moreover, as transgenic cyanobacteria can pose an ecological damage, they cannot be grown in open systems [54]. Mutagens can be used to cause random mutations, which can be a suitable alternative to genetic engineering. But this necessitates extensive screening in order to find the mutant with the desired properties [73].

Cyanobacteria-dominated mixed cultures are also known for their high PHA production, although maintaining their dominance without contamination is difficult prior to purification. Because Cyanobacteria can tolerate high nitrogen

concentrations, maintaining high N: P ratios is considered to be a viable solution. But, mixed cultivations have only been used in laboratory and pilot scale manufacturing, and scaling up production will probably take longer [33]. When compared to currently available polymers, pure algae bioplastics have lower mechanical strengths, which limit their uses. However, employing sustainable biomass as additives such as compatibilizers, fillers, and plasticizers is a possible approach for addressing this problem [78]. Biodegradability is the significant property of bioplastics due to which, it is on high demand. But this requires a set of conditions that may not be present in landfills, where they are usually disposed. As a result, bioplastics that degrade under normal conditions will need to be tailored in the future. Furthermore, if not properly disposed of, bioplastics might emit a small amount of greenhouse gases. These, gases on the other hand, can be collected and used for other purposes, such as biogas production [79]. Future algae research should focus on finding alternatives to all of the aforementioned issues, with the objective of enhancing large-scale production.

9. Role of algal bioplastics in green economy

Unlike conventional plastics, where the whole process remains linear and continuously emit harmful gases including CO2, Algal bioplastics play a significant role in building a future green economy by using emitted CO2 as the carbon source for their survival making it a circular process. Only around 1% of the world's plastics are biodegradable, while the rest are fossil-based, posing a threat to significant flora and fauna in both terrestrial and aquatic habitats. If this condition persists, it will harm the species on the globe and may even result in the extinction of species, reducing biodiversity. This shows the worldwide impact of plastic use, which can be mitigated by the use of sustainable algae bioplastics.

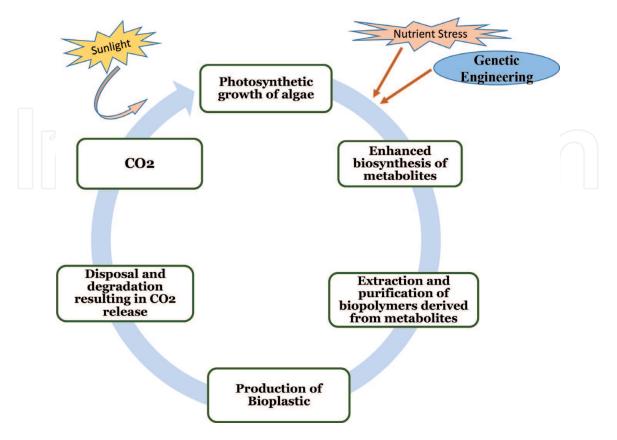


Figure 2. *Role of algal bioplastics leading to a circular and green bioeconomy.*

Figure 2 is a representation of an eco-friendly cycle as a result of using algal bioplastics. Algae use sunlight and CO2 as raw materials for photosynthesis, producing and accumulating a variety of metabolites that can be improved by manipulating nutritional conditions, using gene editing, or inducing mutations. These metabolites can be transformed into biopolymers, which can then be isolated and purified to make bioplastics. After disposal, these bioplastics degrade to produce CO2, which is then used by algae, and the cycle continues. Further, study and improvements onbioplastics and eventually replacing plastics entirelywill result in a green and safer planet.

10. Future prospects of bioplastic production

Despite the fact that this chapter summarises the negative effects of conventional plastics, the benefits of bioplastics, the research framework for bioplastic production and the role of algal bioplastics in the green economy, the majority of the studies are conducted at the laboratory or pilot scale. Only large-scale production, as well as other species characterisation studies will be able to meet the increasing demand. To begin with, algae are highly diverse and there are many species and classes of microalgae that are yet to be identified. The selection of species for an experiment is critical, and further research on evolutionary divergence and species categorisation should be done beforehand to explore the properties and efficiency of each species. Increased metabolite accumulation in response to abiotic stress is well-known, and nitrogen and phosphorus deficiency are particularly well-studied. Although the stress-induced increase in metabolite accumulation is true, it has an adverse influence on the cells and decreases the biomass rate. To overcome this challenge, a two-stage cultivation approach can be used. In this method, algal cells are first grown in optimal conditions before being stressed to accumulate metabolites [32]. This results in higher biomass and metabolite yields at the same time. Adopting these strategies in large-scale production, on the other hand, is less prevalent. Genetic engineering is another cutting-edge method for enhancing metabolite production. To target a transcription factor or a gene involved in metabolic pathway, a variety of genome editing approaches can be applied. This is a highly successful strategy that has yielded positive results in laboratory trials [80, 81]. Nevertheless, since algae are complex eukaryotic organisms with few genomes sequenced, most researchers are limited to studying only those organisms that have had their genomes sequenced. To explore algal genetics, independent research on algal genome sequencing is required. These genetically engineered organisms can also be grown in nutrient-deficient conditions to enhance yields even further. However, algal genetic engineering experiments are limited to laboratory study, and these genetically engineered organisms are not permitted to be cultivated for industrial metabolite production. This is in regards to the stability and maintenance of the strain, as well as the possibility of lateral gene transfer. As a result, techniques for maintaining stability while also safely disposing of remaining genetic material are required. Mixotrophic cultures are now being studied for metabolite production, with promising results. This can also be utilised in large-scale cultivations to overcome the challenges of maintaining pure cultures. This method is particularly useful in waste water treatment, as these mixotrophic cultures can develop by utilising the extra nutrients in the waste waters while also result in waste water treatment. Finally, using the above mentioned strategies for improving metabolite production can either be employed separately or coupled with different combinations for large scale production with profitable yields.

11. Conclusion

Excessive usage of plastics results in pollution, causing harm to the earth and its existing species. Despite the numerous benefits of algae bioplastics, research in this area still need to be progressed. There is also a critical need to take advantage of modern genetic technologies to boost the metabolite synthesis for bioplastic production. Because there have been proven results of employing various algal and cyanobacterial strains as additives that demonstrate increased mechanical qualities of bioplastics equivalent to adding other synthetic components, using solely these renewable sources helps to develop highly compostable bioplastic.

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Conflict of interest

Authors declared that there is no conflict of interest with respect to either authorship, affiliation or in any part of the writing of this chapter.

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Author details

Arathi Sreenikethanam and Amit Bajhaiya^{*} Algal Biotechnology Lab, Department of Microbiology, School of Life Sciences, Central University of Tamil Nadu, Thiruvarur, India

*Address all correspondence to: amitkumar@cutn.ac.in

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