Clean Energy Production Technologies
Series Editors: Neha Srivastava · P. K. Mishra

Neha Srivastava
P. K. Mishra *Editors*

Technological Advancement in Algal Biofuels Production

Springer

Clean Energy Production Technologies

Series Editors

Neha Srivastava, Department of Chemical Engineering and Technology, IIT (BHU) Varanasi, Varanasi, Uttar Pradesh, India

P. K. Mishra, Department of Chemical Engineering and Technology, IIT (BHU) Varanasi, Varanasi, Uttar Pradesh, India

The consumption of fossil fuels has been continuously increasing around the globe and simultaneously becoming the primary cause of global warming as well as environmental pollution. Due to limited life span of fossil fuels and limited alternate energy options, energy crises is important concern faced by the world. Amidst these complex environmental and economic scenarios, renewable energy alternates such as biodiesel, hydrogen, wind, solar and bioenergy sources, which can produce energy with zero carbon residue are emerging as excellent clean energy source. For maximizing the efficiency and productivity of clean fuels via green & renewable methods, it's crucial to understand the configuration, sustainability and techno-economic feasibility of these promising energy alternates. The book series presents a comprehensive coverage combining the domains of exploring clean sources of energy and ensuring its production in an economical as well as ecologically feasible fashion. Series involves renowned experts and academicians as volume-editors and authors, from all the regions of the world. Series brings forth latest research, approaches and perspectives on clean energy production from both developed and developing parts of world under one umbrella. It is curated and developed by authoritative institutions and experts to serves global readership on this theme.

Neha Srivastava • P. K. Mishra
Editors

Technological Advancement in Algal Biofuels Production

Springer

Editors
Neha Srivastava
Department of Chemical Engineering and Technology
IIT (BHU) Varanasi
Varanasi, India

P. K. Mishra
Department of Chemical Engineering and Technology
IIT (BHU) Varanasi
Varanasi, India

ISSN 2662-6861 ISSN 2662-687X (electronic)
Clean Energy Production Technologies
ISBN 978-981-19-6805-1 ISBN 978-981-19-6806-8 (eBook)
https://doi.org/10.1007/978-981-19-6806-8

This Springer imprint is published by the registered company Springer Nature Singapore Pte Ltd.
The registered company address is: 152 Beach Road, #21-01/04 Gateway East, Singapore 189721, Singapore

Preface

Rapid and continuous industrialization and the increasing consumption of fuels by people are major concerns associated with the existing energy crises and need to be resolved by adopting any potential alternative sources. Apart from energy insecurity, harmful impacts of fossil fuel contribute to pollution, and global warming decreases environmental sustainability. To overcome these challenges, several biofuel options are emerging as potential clean forms of energy and are developed from green technologies. Among different existing biofuels, algal biofuels are known as a very efficient mode of biofuels production which can be easily adopted in the near future as a low-cost commercial fuel. Though the third-generation biofuels, algal biofuels, have tremendous advantages, there are parameters that need to be considered for practical applications of these biofuels. Therefore, this book provides information about advancements in algal biofuels production to be implemented as low-cost advanced and sustainable biofuels technology in future. Chapters 1 and 2 present latest trends and advancements of biotechnological contributions and OMICS technology application for technical improvement in algal biofuels production. Chapters 3 and 4 explore algal butanol as a biofuel and gold nanoparticle synthesis from algal biomass for bioenergy applications, and both approaches are being used as new advanced and potential tools for biofuels production. Further, Chaps. 5 and 6 discuss critical challenges in algal biofuels production for technological advancement and engineering-based process parameter strategies to improve algal biofuels production. Additionally, Chapters 7–10 provide new insights on algal biofuels and diversity based on technical advancement ground for practical improvement as well as applications. A detailed understanding of the technological and latest research concepts may help to set goals and help to plan innovative strategies to develop sustainable as well as low-cost algal biofuels production.

Varanasi, India

Neha Srivastava
P. K. Mishra

Acknowledgements

The editors are thankful to all the academicians and scientists whose contributions have enriched this volume. We also express our deep sense of gratitude to our parents whose blessings have always prompted us to pursue academic activities deeply. It is quite possible that in a work of this nature, some mistakes might have crept into the text inadvertently and for these we owe undiluted responsibility. We are grateful to all authors for their contribution to this book. We are also thankful to Springer Nature for giving this opportunity to editors and the Department of Chemical Engineering & Technology, IIT (BHU) Varanasi, U.P., India, for all technical support. We thank them from the core of our heart.

Contents

About the Editors

Neha Srivastava has received her PhD in Biotechnology from the Department of Molecular and Cellular Engineering, SHIATS, India, in the area of bioenergy. She works as Research Scientist in the Department of Chemical Engineering and Technology, IIT (BHU) Varanasi, India. She has published more than 37 research articles in peer-reviewed journals of SCI impact factor. She has filed three patents with one technology transfer. She has 14 published books and two book series with renowned international publishers. She is working on bioprocess technology and biofuels production (microbial screening and enzymes; production and enhancement, biohydrogen production from waste biomass, bioethanol production).

P. K. Mishra is currently Professor and Head of the Department of Chemical Engineering & Technology, Indian Institute of Technology (BHU), Varanasi, India. He has authored/coauthored over 60 technical papers published in reputed national/international journals and supervised more than 20 doctoral students. He has received several awards and honors and has five patents with one Technology Transfer. He is Fellow of Institution of Engineers India. He has received several awards and honors at national/international levels.

Chapter 1
Biotechnological Approaches to Enhance Algae Biofuel Production

Umar Shahbaz, Sidra Zubair, Amna Younas, Xiao bin Yu, Nazra Fatima, Shahzal Babar, Samra Basharat, Asma Bibi, and Muhammad Iftikhar Hussain

Abstract Algae are aquatic species that may reproduce quickly and have over 3000 different breeds, making them more abrasive than terrestrial plants. They may be able to convert CO_2 from the air into oxygen and remove it from the breaking cells of algae plants, which is how they produce wonderful oil. A viable source of feedstock for biofuels, oleaginous microalgae have a number of advantages over terrestrial plants. Due to the lack of robust algal strains with increased lipid content and biomass and methodologies for economically viable oil extraction, the microalgal fuel business is still in its infancy. By carefully targeting important metabolic nodes, microalgal metabolic engineering demonstrates the huge potential to improve lipid

U. Shahbaz (✉)
Dipartimento di Genomica e Biologia delle Piante da Frutto, Fondazione Edmund Mach, San Michele all'Adige, TN, Italy

C3A-center agriculture food and environment, University the Trento, Trento, Italy
e-mail: Umar.shahbaz@fmach.it; https://www.fmach.it/

S. Zubair
Department of Bioinformatics and Biotechnology, Government College University, Faislabad, Pakistan

A. Younas
Center of Agribiotechnology and Biochemistry, University of Agriculture, Faislabad, Pakistan

X. b. Yu · S. Basharat · M. I. Hussain
Key Laboratory of Carbohydrate Chemistry and Biotechnology, Ministry of Education, Jiangnan University, Wuxi, China

N. Fatima
Institute of Atherosclerotic Disease, Xi'an Jiaotong University, Xi'an, Shaanxi Province, China

S. Babar
School of food science, Ministry of Education, Jiangnan University, Wuxi, China

A. Bibi
The Key Laboratory of Microbiology and Parasitology, Anhui School of Basic Medical Sciences and Laboratory Diagnostics, The First Hospital of Anhui Medical University, Hefei, China

N. Srivastava, P. K. Mishra (eds.), *Technological Advancement in Algal Biofuels Production*, Clean Energy Production Technologies,
https://doi.org/10.1007/978-981-19-6806-8_1

accumulation without compromising cell growth. The genetic enhancement of microalgae without impairing cellular biomass remains an underutilized option for large-scale biofuel production, despite recent advances in synthetic biology. To improve microalgae as a biofuel platform for the production of biohydrogen, alcohols generated from starch, substitutes for diesel fuel, and/or alkanes, we consequently present a thorough overview of numerous biotechnological techniques in this chapter.

Keywords Biofuel · Genetic engineering · Microalgae · Nano-additive · Bio-bricks · Biorefinery

Abbreviation

ABE	Acetone-butanol-ethanol
DHA	Docosahexaenoic acid
EPA	Eicosa pentaenoic acid
EST	Expressed sequence tags
FAME	Fatty acid methyl ester
GHG	Greenhouse gases
HBV	Hepatitis B virus
LED	Light-emitting diodes
LHC	Light antenna complexes
PC	Phosphatidylcholine viation
ROS	Reactive oxygen species
SCP	Single-cell proteins
TAG	Triacylglycerols

1.1 Introduction

Algae are the aquatic species that have the fastest capability to reproduce with over 3000 different breeds, therefore more adverse than land plants. They can have the capacity to extract CO_2 from the air and turned into oxygen and have the ability to yield great oil, i.e., the breaking cells of algal plants are extracted (Adeniyi et al. 2018). Renewable fuels are bioalgal fuels derivative of algal feedstock that can be related with its capability to abundantly photosynthesize. The capability to convert all feedstock's energy into various varieties of valuable biofuels is the main advantage (Demirbas 2010). Their other application adds the creation of energy cogeneration (electricity and heat) afterward oil extraction, deduction of CO_2 gases through industrial chimney (algal bio-fixation), bio-fertilizer, animal feeds, and further food products. The important and imaginable effects that algae exist and can survive in thrilling heat, scarcity, salinity, and radiation. Therefore, ecological conditions will affect the country's crop growing ways. Freshwater and marine algae, for example, Chlorophyceae (green algae), Cyanophyceae (blue-green algae), and Pyrrophyceae (fire algae), could be cultured as expected happening in the UK (Adeniyi et al. 2018). Phaeophyceae a brown alga with synthetic cultivation techniques of photobioreactors could be genetically modified. Aimed at the creation of fatty acid methyl ester (FAME), Chlorophyceae, Cyanophyceae, and Pyrrophyceae were recommended. Because of its excessive sugar content, for ethanol production, Phaeophyceae tend to be the greatest suitable algal feedstock (Islam et al. 2015). Owing to growing response for transportation fuel, worldwide, algae have appeared as an appropriate candidate due to their sustainable and renewable characters together with financial reliability to compete by international request aimed to conveyance fuels (García-Olivares et al. 2018). The algal biofuel conversion processes, like fermentation, transesterification, and hydro treatment, are economically expensive and more complex as compared to fossil fuels. The possible earth of optimum established are sustainability for the feedstock, or the enhanced possibility of products and innovative applications because of ranges (Culaba et al. 2020). Due to algal positive characters, it tends to be construed that this feedstock is the world's one of the furthermost valuable renewable and sustainable fuel resources that could show an important role in controlling environmental contamination (Bharathiraja et al. 2018). The major threat is the emission of CO_2 from varied fossil fuels through atmosphere. Worldwide economy in the past is out of petroleum funds, whereas it became rich and switch the unfamiliar trade marketplace in light by petroleum reserves (Mohsin et al. 2019).

To all these abovementioned challenges, the only viable solution for both the worldwide economy and greenhouse gas (GHG) emission of fuel formed from various materials (plant). However, renewable energy has different bases, for example, geothermal, breeze, and solar which probably won't be economically practicable as biofuels; still these GHG play a meaningful part of resolving the concerns of global warming. Biofuels are yields of various sustainable and biodegradable feedstock that can be changed of producing opportunities for advances in agriculture as a result of direct contribution to agriculture plants (Gielen et al. 2019).

1.2 Microalgal Species for Producing Biofuels

Recent biofuel manufacturing exploiting microalga biomass is not cost-effective. To make microalga yields and by-products economically feasible, the growing research is keen to observe new microalgal candidates (Koyande et al. 2019). Various advantages of microalgal species must gain methodical concept as to present a profitable source of extreme worth of products (chemicals) like antioxidant, polysaccharides, β-carotene, natural dyes, bioactive, docosahexaenoic acid, and efficient pigment, astaxanthin, algal extracts, antioxidants, and eicosapentaenoic acid. Those classes show a fundamental function of nutraceuticals, human food, cosmeceuticals, fodder, bioremediation, and pharmaceutical aquaculture (Koyande et al. 2019).

Table 1.1 Some major microalgal species, products, and applications (Mobin and Alam 2017)

Species/groups	Products	Areas of application
Arthrospira (Spirulina) platensis	Phycocyanin, biomass	Health food, cosmetics
Arthrospira (Spirulina)	Protein, vitamin B_{12}	Antioxidant capsule, immune system
Aphanizomenon flos-aquae	Protein, essential fatty acids, β-carotene	Health food, food supplement
Chlorella sp.	Biomass, carbohydrate extract	Animal nutrition, health drinks, food supplements
Chlorella vulgaris	Biomass, carbohydrate extraction	Health food, feed, food supplement

Economically viable is to produce algal biomass, production of extraordinary worth of byproduct pull out are utmost. Normally microalgal organisms are naturally single-cell photosynthetic autotrophic microscopic located at marine and garden-fresh environments. Production of numerous multipart complexes like carbohydrates, protein, and lipids, consuming basic elements situated in their surrounding environment (Trivedi et al. 2015) (Table 1.1). Microalgae are "photosynthetic plants" like microorganisms deprived from land pland possess by particular cell and organ types. For energy production, which take carbon from surrounding airborne. By consuming organic carbon, some microalgae yield energy. Microalgae have more than 300,000 species, and around 30,000 species stay renowned. The living locals are complex or may adjust rapidly in severe environmental circumstances (e.g., heat, UV irradiation, variable salinity, and nutrients) (Ramaraj et al. 2015). However, they can yield an increased variability of interesting subordinate metabolites (biological energetic) through unique structures and biological actions which is usually not found in other candidates. The microalgae also yield certain valuable bio goods comprising polysaccharides, carotenoids (specifically β-carotene), antioxidants, docosahexaenoic acid (DHA), astaxanthin, natural dye, eicosapentaenoic acid (EPA), functional or bioactive stains, and algal extracts (Sathasivam et al. 2019) (Fig. 1.1). On a commercial level, microalgal cultivation has begun five decades ago. The global marketplace worth of the microalgae are considered around US$ 2.5 billion created via health food sector. US$ 1.5 billion are yielding DHA or then US$ 700 million by way of aquaculture. Yearly microalgal production is almost 7.5 million tons.

1.3 Promising Microalgal Species and High-Value Applications

Microalgae are divided into four main collections: (1) cyanobacteria (blue and green algae), (2) chlorophytes (green algae), (3) rhodophytes (red algae), and then (4) chromophytes (wholly remaining algae). Every set consists of more than

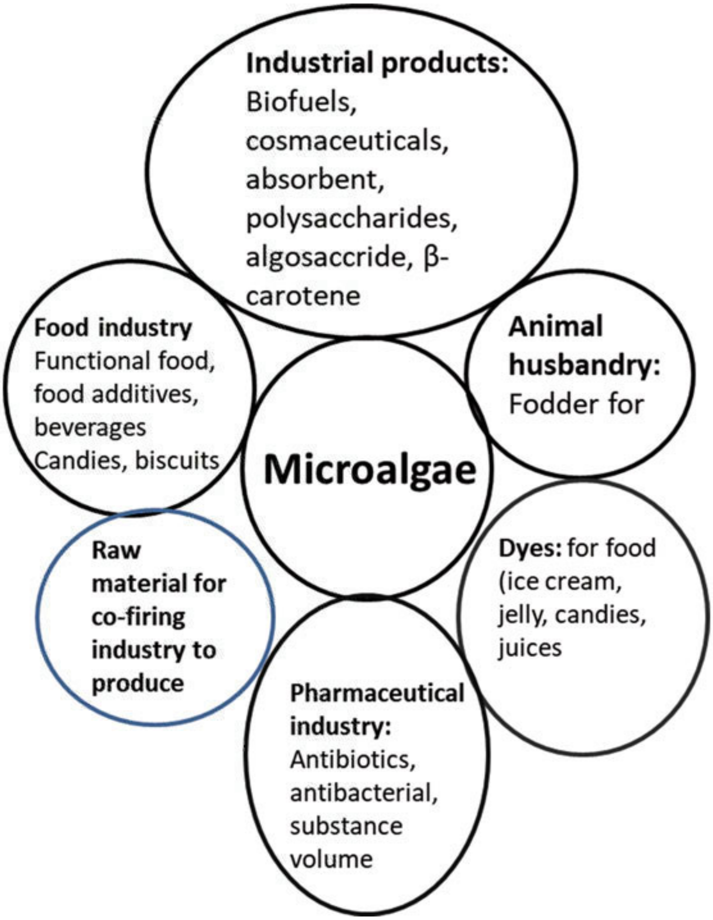

Fig. 1.1 Application of microalgae from numerous areas

hundreds of algae types, and altogether species have thousands or more of strains. For possible beneficial use, only a small variety has been studied. Bacillariophyceae (as well as diatoms), Cyanophyceae (blue-green algae), Chlorophyceae (green algae), and Chrysophyceae (including golden algae) stand generally used microalgae. Some are main microalgal types, their goods, or their application for biotechnological (Sathasivam et al. 2019).

Over the last two decades, four major microalgae are focused on biotechnological applications: (1) *Haematococcus pluvialis*, (2) *Chlorella vulgaris*, (3) *Dunaliella salina*, and (4) *Spirulina* (*Arthrospira*). The characteristics, composition, and production mechanism and, respectively, microalgal type is explained now at the following section (de Morais et al. 2015).

1.3.1 Spirulina

Spirulina (*Arthrospira*) is multicellular, blue and green algae, symbiotic, and filamentous that use nitrogen from the atmosphere. *Spirulina* has two unique shapes; disk-like or spiral rod. Phycocyanin (blue color) is the main photosynthetic pigment in *Spirulina*. The *Spirulina* microalgae likewise hold carotenoids and chlorophyll a. The presence of Phycoerythrin makes the color of microalgae pink or red. Autotrophic contain *Spirulina* then they replicate via binary fission (Mobin and Alam 2017). In 2014 the global manufacture of *Spirulina* sp. was 86,000 tons, as indicated by a report by FAO. It is some of cells considered algae used for extensive outside culture. The high pH (9–11) is best to grow with an extraordinary bicarbonate concentration. To culture *Spirulina*, paddle wheel-driven raceway ponds are used (Mobin and Alam 2017). In the ponds, water varies from 250 to 500 mm because of the microalgae density and seasons. Depth of water pond is also dependent upon pond size, optimal light absorption, and flow velocity by the microalgal culture. Temperature plays a vital role in *Spirulina* productivity (Mobin and Alam 2017). It grows well in between 35 and 37 °C. *Spirulina* filamentous mowing is informal. The starting focus to reached using an inclined gravity screen going on vibrating screefilter (Zhang 2015). Its dewatering remains attempted through vacuum belt filter building a solid pate about 15%; further detail on *Spirulina* is that it is rich in protein, minerals, essential amino acids, vitamins, or required polyunsaturated fatty acid and pigments comprising Zeaxanthin, Myxoxanthphyl, and Phycocyanin. It carries (46–71%) proteins, (8–16%) carbohydrates, and (4–9%) lipids (dry weight). *Spirulina* vital amino acid contains leucine, valine, and also isoleucine. It contains comparatively extraordinary meditation of vitamin K, provitamin A, vitamin B_{12}, and also β-carotene. Fatty acid of *Spirulina* also includes Llinolenic and γ-Linolenic acid and $\Omega - 3$ with $\Omega - 6$ polyunsaturated fatty acids. DHA is the natural source for *Spirulina platensis* demonstrating up to 9.1% of the whole fatty acid (Hemantkumar and Rahimbhai 2019). It furthermore encloses an antioxidant-rich combination more than carotenoids. *Spirulina* mineral content hinge on water, grown and this one content of calcium, iron, and magnesium delivers great nutritional-value (Bensehaila et al. 2015). According to studies, *Spirulina* powder covers provitamin A (2.330 × 03 IU/kg), vitamin E (100 mg, 100/g), β-carotene (140 mg, 100/g), riboflavin B_2 (4.0 mg, 100/g), niacin B_3 (14.0 mg, 100/g), thiamine B_1 (3.5 mg, 100/g), vitamin K (2.2 mg, 100/g), vitamin B_6 (0.8 mg, 100/g), inositol (6 mg/g), vitamin B_{12} (0.32 mg, 100/g), biotin (0.005 mg, 100/g), folic acid (0.0 mg, 100/g), and pantothenic acid (0.1 mg, 100/g) (Ragaza et al. 2020).

1.3.2 Chlorella

Chlorella is spherical-shaped, single-cell (2–10 μm) in diameter, and phototrophic green microalgae without flagella. In its chloroplast, both chlorophyll a and chlorophyll b (green photosynthetic pigment) are present. It multiplies speedily and needs sunlight, water, CO_2, and a slight volume of minerals (Simosa 2016). *Chlorella* was grown in photobioreactor large round tanks and paddle wheel mixed open and circular-open ponds. Though making of microalgae used for aquaculture is mostly at smaller scales, or at most of situations, it is produced within (20–40 L) carboys and huge plastic bags (~1000 L in volume) (Kunjapur and Eldridge 2010). *Chlorella* is harvested by auto-flocculation or centrifugation. Following the collection of biomass sprayer exists drum dried and dust is wholesaled straightforwardly for used to proceed tablets. Chemical configuration of *Chlorella* indicates which comprises (12–28%) carbohydrate, (111–58%) protein, and (2–46%) lipids via dry weight. The usual configuration of *Chlorella* residue may be present at Borowitzka. It also encloses several vitamins like that provitamin A (55,500 IU/kg), vitamin E (1 mg 100/g), inositol (165.0 mg 100/g), thiamin B_1 (1.5 mg 100/g), biotin (191.6 mg 100/g), riboflavin B_2 (4.8 mg 100/g), vitamin B_6 (1.7 mg 100/g), vitamin B_{12} (125.9 mg 100/g), folic acid (26.9 mg 100/g), β-carotene (180 mg 100/g), and pantothenic acid (1.3 mg 100/g). *Chlorella*'s worldwide profitable market worth esteem is above a billion US dollars (Hemantkumar and Rahimbhai 2019).

1.3.3 Dunaliella

Dunaliella is edible, nutrient-rich, single-celled, flagellated, and extremophile green microalgae. *Dunaliella* is brought into being at countless fresh waters or marine water habitats. *Dunaliella salina* (*D. salina*) has gained ample care from scientists by the intense stages of antioxidant actions (Dolganyuk et al. 2020). It is the best source of β-carotenoids, which contains a high extent of β-carotene (above 14% of dry biomass) contrast with further well-known cradles. *Dunaliella* may be grown over 30% NaCl saturation though the ideal development salinity is 22% NaCl saturation. There is only a rare reported challenging type and predators for *Dunaliella*; thus, modest open pond culture is sufficient for it. *Dunaliella salina* (*D. salina*) is full-fledged at each shallow, extensive areas (5–200 ha), in light paddle wheel race way ponds and unstirred ponds which is more than 1000 m^2 in area (Dolganyuk et al. 2020). For *D. salina* using technique is the semi-continuous culture. It has been discovered that at lower salinity level is the best to grow *Dunaliella* closely (22% (w/v) NaCl) saturation however β-carotene content (nearly 33% NaCl). *Dunaliella* shows promising growth at lower salinity conditions, but there are risks of contamination by brine shrimp or even by *Artemia* or protozoa. There are several manufacturing tactics researched by Ben-Amotz for *Dunaliella*. The challenging culture was using nitrogen inadequacy, extreme salt concentration, or penetrating solar radiation which boost fruitful biomass of *Dunaliella* and then β-carotene fabrication (Pourkarimi et al. 2020). By using the centrifugation technique, harvesting of *Dunaliella* has been completed, further flocculating and employing centrifugation methods manipulating the cell membrane at hydrophobic nature. Certain complications coupled to the reaping of *D. salina* are:

1. Cells are of jaggedly to the comparable density like as culture medium.
2. Lower cell concentrations in the culture (typically less than 0.1 g dry weight per liter).
3. Cells are very faint because of no cell wall.
4. Instability of cell (uncertainty of cells are broken in the period of harvesting, so the rapidly dissolving of β-carotene) (Hosseini Tafreshi and Shariati 2009).

Immediately after being harvested, the biomass might be sprayed through drum dried, and the β-carotene may be detached straightly by means of hot oil or any additional solvents. The biochemical configuration of *Dunaliella* is protein (49–57%), lipids (6–8%), and carbohydrate (4–32%) of its total dry weight (Wu and Chang 2019).

1.3.4 Haematococcus pluvialis

Haematococcus pluvialis (*H. pluvialis*) present as unicellular bi-flagellate freshwater Chlorophyta microalga dispersed around the world. Under different stress conditions, the accumulation of a wide variety of strong antioxidant astaxanthin is renowned as *Haematococcus pluvialis* (more or less 2–3% on dry weight). A commercially producing organism is astaxanthin of *H. pluvialis* (Shah et al. 2016). Heterotrophic, photoautotrophic, or mixotrophic centered on progress state, internal or secure photobioreactors, or sweeping channel ponds are mostly recycled for *H. pluvialis* refinement. Standard photobioreactors castoff for refinement cover simmer column, tubular, plus airlift photobioreactors (Narala et al. 2016). A twice step agriculture approach tactics are generally adapted for marketable nearer efficiency. The initial step includes the mounting of algal green biomass at motile stage in a secured system (photobioreactors) and exposed skill (like that pond raceways), and the following step comprises manufacturing of astaxanthin covering aplanospores enlargement through the insufficiency nitrite or phosphate and then improved light intensity and temperature (Kim 2015). Then accretion of astaxanthin is squeezed via natural features, for example, salt concentration, nutritional stresses, light, and pH. Cellular structure of *H. pluvialis* differs remarkably among red-green phases of agriculture. Mowing is attempted by using a grouping of flaccid resolving besides following flotation and centrifugation. Mowing of biomass has got dried out by shower, chilling, sun-oriented drying, lyophilization, or cryodesiccation. To dry high-esteem *H. pluvialis* items, splash drying is the most fitting strategy (Shah et al. 2016). A widespread variety of procedures has been industrialized toward interrupting the *H. pluvialis* cell or recovering the intracellular metabolites. Then mechanical procedures (bead milling expeller pressing) are the most appropriate cell disruption technique. *H. pluvialis* cells are cuddled underneath extraordinary pressure toward breaking the dense pollen ultimately. Later in the disruption of cell walls, the biomass of *H. pluvialis* is essential to be treated within a couple of times to evade damage. However, super critical carbon dioxide (SC-CO_2) and solvent drawing out methods are well-known owing to their proficiency or compatibility. *H. pluvialis* is able to mass more or less 5% dried weight of astaxanthin and is well-thought-out as the good organic cause of the great worth of carotenoid colorant. Due to the huge cost of manufacture, synthetic astaxanthin leads the presently profitable market (yearly production of 130 tons creating a value of US$ 200 million) (Sanzo et al. 2018). Some additional microalgal species areas are as under studying for their appropriate for food or nutritional supplements bases. In the 1970s *Scenedesmus* was not found favorable for human or animal intake due to its huge cost (Ghani et al. 2020). In the 1990s, the dinoflagellate *Crypthecodinium cohnii* has been recognized as the utmost capable entrant for DHA creation. In heterotrophic culture, the cell bulks of *Crypthecodinium cohnii* have been attained up to around 40 g/L with a lipid percentage of 15–30%. The DHA value for this lipid is described to 25–30% (Santos-Sánchez et al. 2016).

1.4 Microalgae

Microalgae have gained considerable attraction worldwide currently, by their wide-spread usage potency in the renewable energy, biopharmaceutical industries, and nutraceutical. Microalgae are renewable, viable, cost-effective homes of biofuels, bioactive medicinal products, and food ingredients. Several microalgae kinds have been examined for their likely function as significant yields with extraordinary biological and pharmacological potentials (Subhadra 2010). As per a biofuel, microalgae are an entire alternative for melted remnant fuels with charge, renew-ability, and environmental interests. It has a considerable capability to change atmospheric CO_2 into valuable products such as carbohydrates, lipids, and additional bioactive metabolites (Hussain et al. 2021). For biopharmaceuticals and bioenergy, the microalga is a viable source, some challenges and problems are remaining, but they must be overcome to advance the equipment from experimental-phase to built-up level. For bioethanol production, pretreating biomass, dewatering algal culture for biomass production is the most essential and challenging feature for improving the growth rate and product synthesis (Tonnaer 2017). For biodiesel production, most microalgae species are appropriate, because of their high lipid contents of 50–70% and may reach 80% such as incase of microalgae *B. braunii*, which accumulate up to 80% of oil in its biomass. Polyunsaturated fatty acids are vital in tissue integrity and have beneficial health effects. Omega-3 and omega-6 fatty acids, in particular, are crucially essential for humans but the human body cannot produce them by itself. Many microalgae species (e.g., *Arthrospira platensis*, *Porphyridium cruentum*, and *Odontella*, *I. galbana*) have been explored for their capability to synthesize these fatty acids. *Pavlova lutheri* produces polyunsaturated fatty acids in large quantities. Microalgae are a rich source of various vitamins; *P. cruentum* produces a high quantity of vitamin C and E, as well as β-carotene (vitamin A); *Haslea ostrearia* is a rich source of vitamin E (tocopherol).

Microalgae have a broad spectrum of industrial applications. Mostly wastewater purification and biofuel production have been reported. Some industrial applications are high-value food, nutraceuticals, pharmaceutical products, health food for humans, fodder additives, polysaccharides, cosmeceuticals, antioxidants, dyes, food for aquaculture, and bioplastic production. Microalgal biomass is also used as a raw material for the co-firing industry to generate power because it has a high heating value other than biomass feedstock. *Spirulina* is used for cholesterol reduction and immune system enhancement. Sulfated polysaccharides of *Spirulina* are broadly used as an antiviral agent. Its tablets are being marketed since 1975 in Japan. *Chlorella* composes of vital substances: β-1,3-glucan. This compound is a free radical scavenger that enhances immune response, also responsible for lowering lipid in the blood. It is also effective in stomach ulcers, wound healing, antitumor activity, and removing toxins from the body. *Spirulina* has a preventive effect in hypercholesterolemia. *Chlorella* is effective on low blood sugar level, enhances hemoglobin concentration, and acts as hepato-protective agent. Astaxanthin from *Haematococcus* is utilized in aquaculture for coloring fish muscles (salmon fish). The antioxidant characteristics of astaxanthin prevent the production of inflammatory compounds and also prevent protein degradation, oxidative stress, and macular degeneration. *Spirulina*, *Dunaliella*, and *Chlorella* all are widely used in the food industry. Their biomass is also utilized for forming a variety of health products including tablets and capsules. Three microalgae are used for bread, noodles, candies, bean curd, and other common food with high health values. *Chlorella vulgaris* stimulates the synthesis of skin collagen; they also help in the regeneration of fibers and make the skin surface free from wrinkles.

Extract of *Spirulina* minimizes the age effects. Purified phycobiliproteins, a product of *Spirulina*, are used in cosmetics, food (colorant), antioxidant treatment, anti-inflammatory, photodynamics of various cancers, leukemia and tumor treatment, and florescent marker production.

1.5 Macroalgae

Algae are distributed in an extreme and diverse environment. Due to their high content in compounds, they are valuable with different biological activities, including both complex organic compounds, both primary and secondary metabolites. It is significant to observe that the majority of these substances include phytopigments (carotenoids and xanthophylls), polyunsaturated fatty acids, phenolic compounds, tannins, peptides, lipids, vitamins, enzymes, terpenoids, and others. Thus algae are viable and economical biomass sources of valuable compounds with potential applications in the pharmaceutical, nutraceutical, chemical, food, and cosmetic industries due to their biological active and regenerative characteristics (Fig. 1.2). Microalgae gained more and more value due to their usage in health aspects. They are promoting properties that can reduce the risks of many chronic diseases and help to extend the life span. They are also used for wastewater treatment or as natural

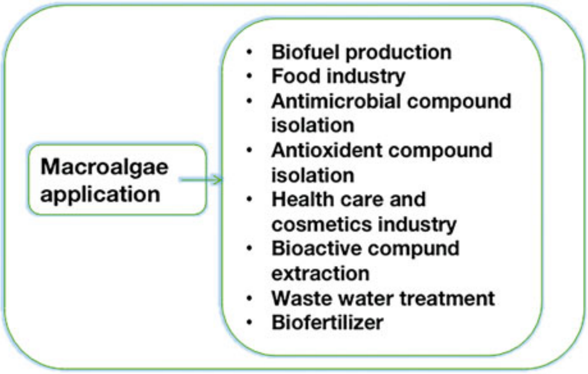

Fig. 1.2 The main application of macroalgae

fertilizer in agricultural areas, therefore improving the quality of products and reducing the need for chemical fertilizers. As a source, the potency of macroalgae of renewable energy is also of considerable interest. These aquatic organisms mitigate carbon dioxide emissions and nowadays are being used as feedstock to form “clean” or so-called third-generation biofuels.

1.6 Chemical Composition

Different marine macroalgae (seaweed) as a source of bioactive and essential compounds had the advantages to use an under-utilized sustainable natural reserve. It had been confirmed that secondary metabolites are made up of biomass (Biris-Dorhoi et al. 2020). The chemical composition has changed due to natural conditions (temperature, candlepower, sea water salinity, and growth habitat) and genetic modifications among species. The protein substance can go from 7% to 31% of total dry weight and a lipid substance varying from 2% to 13% of total dry weight. A big quantity of carbohydrates can range from 32% to 60% of dry weight (Aratboni et al. 2019). Macroalgae are considered a true natural source of vitamin A and E (tocopherol). The abundance of vitamin B_{12} advances the macroalgae created products concerning the dietary supplement for a vegetarian lifestyle taking a risk of deficiency of B_{12} (Biris-Dorhoi et al. 2020). The numerous microelements found in seaweeds are mostly supported by their mineral content; sodium, mechanism, and calcium, which present above 97%, you look after the general mineral content. Additional microelements like iron, zinc, manganese, and copper are discovered in minor quantities (ranging from 0.00 to 0.094) of seaweeds of dry weight (Arguelles 2020). Phenolic compound existence in macroalgae has taken great attraction due to their particular bio-activities and health supporting benefits incorporating antiallergic, antiproliferative, antioxidant, antimicrobial, antidiabetic, and neuro-protective properties (Santos et al. 2019). Secondary metabolite’s presence in macroalgae is encouraging the regular defense system against several injuries, diseases, and environmental aggression. For an extended time, macroalgae are encouraged for their prospective function in protective cancer rate, tumor development, and even health upturn after chemotherapy or radiotherapy treatments (Biris-Dorhoi et al. 2020). Iodine can also be used as an anticancer response, due to its capability to start apoptosis in cancer cells. Similar characteristics are often ascribed to the omega-3 fatty acid like stearidonic acid and hexadecatetraenoic acid discovered in eatable marine algae-like *Ulva* and *Undaria* along with 40% of total fatty acids (Biris-Dorhoi et al. 2020; Fitton et al. 2008).

1.7 Laminarian

Alginate, fucoidan, and lots of other seaweed polysaccharides are proven to possess antitumor activities. A great quantity of polysaccharides (~65% polysaccharides in total dry weight) are often found in many seaweeds like *Ascophyllum*, *Ulva*, *Palmaria*, and *Porphyra* (Ghosh et al. 2021). Alginate filling the intestinal system additionally aids in boosting immunity and intestinal health, reducing the risk of cancer (Eliaz et al. 2007). Laminarian and fucoidan induce apoptosis to prevent cancer, but also unidentified seaweed polysaccharides are able to exhibit straight and unintended antitumor influence. *Sargassum latifolium* reserved cytochrome P450 IA and glutathione S-transferases and prevent cell viability and stimulating apoptosis. Another study about the antiviral potential of algal against foodborne viruses is getting interested about current years; recently the accessible data are still limited (Zorofchian Moghadamtousi et al. 2014). The most compounds from algae that are proved to possess antiviral potency are sulfated polysaccharides, including sesquiterpene hydroquinone, carrageenan, sulphoglycolipids, and fucoidan (Ahmadi et al. 2015). Polysaccharides derived from marine and their lower relative molecular mass oligosaccharides derivatives are displayed to have a kind of antiviral activities and also exercise antimicrobial and antioxidant influence (Zhu et al. 2021). The algal polysaccharides can overturn the DNA replication and leave the host cell colonization by the virus. For instance, the antiviral capability of polysaccharides from brown seaweed exposed substantial inhibiting action against the hepatitis B virus (HBV) and DNA polymerase, accordingly influencing its replication. The antiviral action of those polysaccharides is exerted through suppression of virus adhesion to the host cells (*U. pinnatifida*, bladder wrack, *Cystoseira indica*). Fucoidan inhibits syncytia formation with remarkable selectivity ($IS50 > 2000$) during the early phases of virus infectivity (0–60 min post–infection) (Gheda et al. 2016).

1.8 Algal Biofuel Production

There are an excellent revolution and challenges in biofuel production to exchange fossils fuels. The biological yield of biofuels is apprehending the world's market thanks to the restrictions of petroleum-based fuel. Researchers are more interested in the exploration of latest technologies for biofuel yield. Biomass for biofuel production is one among the alternatives thanks to its sustainability and renewability, low CO_2, fewer greenhouse gases, etc. The most issue regarding biomass usage is the efficiency to exchange complete fossil fuels (Khan et al. 2018). For biomass generation, land use causes ethical issues that are consequences in enhancing food prices. The most focus of scientists is to believe new technologies to beat energy needs, decrease the prices of fuel, and be ready to solve environmental issues too (Fribourg 2008). Photovoltaic technology systems are going to be directly employed by genetically engineered photosynthetic microorganisms of completely synthetic factories. Biofuels are considered the foremost promising within the short term as their market maturity is above other options. Global climate change concerns the buildup of greenhouse gases causing linked regarding the utilization of fossil fuels because it is the major energy source (Hannon et al. 2010). To beat the problems of global climate change, the one solution is to believe the potential of microorganisms to use renewable substrates for biofuel production, thanks to tremendous progress in several industries which are resulting in polluting the environment. The microbial technology gives the simplest solution with an environmentally friendly approach, by identifying the microbial strains, causative agents for the matter, and implementing the useful one for environmental bioremediation (Srivastava 2019). Microalgae have long been known as potential good sources for biofuel production due to their comparatively high oil level and speedy biomass production. Microalgae raise very timely as competed to continental crops; algal mass can grow on non-arable lands using non-potable saline or wastewater (Srivastava et al. 2020).

The microbial-related bioremediation process has benefited society by exploiting the metabolic abilities of microorganisms. Due to the depletion of worldwide petroleum and its value increases, biodiesel is becoming one among the simplest promising worldwide energy markets within the coming future. Growing pattern in the biofuel and high-rate biochemical yield decline the necessity for nonrenewable and artificial sources, to decrease the harmful environmental effects and advance biorefinery (Koenraad et al. 2015). It's likely that the biochemical market will increase from 2% to 20% interest by the year 2025 due to the main growing demands for bio-based products which have directed the R & D efforts to specialize in commercially oriented research styles (Cordova et al. 2020). At the present, biofuels have gotten significant potential among the financial and environmental disasters of fossil fuels. Fuel reproduction molecules as isoprenoids, fatty acid-derived molecules, hydrocarbons, and improved alcohols have a plus over their conventional complement due to engine compatibility, compatibility with present storage, higher density, and transport structure. Furthermore, biologically functional high-value substances containing isoprenoids and fatty acid-derived biomolecules are of environmental, biotechnological, pharmacological, agricultural, and also industrial significance (Adegboye et al. 2021). For the assembly of required titers for subsequent generation, there's a requirement for improvements in gene-splicing techniques that have assisted scientists to develop advanced robust strains (Bharadwaj et al. 2020). Biofuels also require starchy/sugary substrate or lipid-rich biomass for their successive conversion into advanced alcohols: butanol, isoprenoids, bioethanol, and fatty acid-derived substances (Mehmood et al. 2021). First-generation feedstock include food crops, for instance, sugarcane, barley, corn, beetroot, wheat, etc.; however it made the conversation of food deficiency (Hirani et al. 2018).

1.9 The Second-Generation Feedstock

Second generation focused intensely towards the raw and waste materials to account for the issues of first-generation feedstock, yet they're fundamentally expensive and laborious pretreatments for their decomposition into simpler components for their simpler succeeding adaptations to those products (Anto et al. 2020). Microalgae and cyanobacteria are photosynthetic microbes considered because third generation feedstock supply both higher lipid-based pieces of stuff and alcohols. Due to their biochemical composition, microalgae got a standing over lignocellulosic biomass, which contains lipids, protein contents, and carbohydrates (Abomohra and Elshobary 2019). Carbohydrate ingredients are viable to yield bioethanol and alcohols, while the lipid section is employed to supply biodiesels, isoprenoids, and extra lipid-based compounds. The restraints of cultivation, harvesting, and downstream processing are thanks to the commercial execution of third-generation feedstock (Laurens et al. 2017b). The deficiencies of the previous generations were alleged to be reported by GM microbes (designated as fourth generation) to support the growth proficiency and product yield. The earlier decade has understood a rise in extreme value biochemical productions and biofuels engaging microbial cell factories (Shuba and Kifle 2018). To reduce the greenhouse gases and to satisfy the worldwide energy burdens, technological advancement requires specialization in as follows:

1. To boost up the biochemical production, through improving the microbial cell factories.
2. For higher efficiency, optimize the existing production technologies.
3. Development of cell capability to yield biofuel-related designer molecules that advance fuel quality.

Combined methods could support in overwhelming the technological difficulties met during designing a functional, and reliable biofuel production pipeline. For the modernization of metabolic pathways, genome editing (like gene insertion and removal) has developed the reconstruction efforts because it advanced the metabolic engineering of both native and non-native hosts to yield renewable biofuels. By using of strain improvement technique, several approaches are used for the assembly of high titer, e.g., genetic modification, promoter engineering, pathways synthesis, process engineering, enzyme engineering, and competitive pathways blocking (Aro 2016).

Advancement in synthetic biology decreases the struggles, contrasting previously in developing microbial cell factories (Xia et al. 2019). Therefore, the key challenge is the partial number of existing obvious genes and their combinatorial control on metabolomics structure of the host organism. The modification consequence during a complicated phenotype-genotype results in opposing and accidental effects (Manzoni et al. 2018). The purpose for receiving developed titers from microbial cell factories is careful rebuilding and optimization of the metabolic pathway regulation in terms of organic phenomenon, enzyme activity, and metabolic influx (Lee et al. 2018). Difficulties are encountered during a different system of biology and gene-splicing methods to make fourth generation alongside the possible solutions to handle these challenges.

1.10 Heterologous Synthesis of Hydrocarbons

The international marketplace for *n*-butanol is growing at a prompt rate with a predictable market worth of 5 billion USD (Tao et al. 2014). Biological fermentation process (acetone-butanol-ethanol pathway) or petrochemical/chemical methods (for 7.0–8.0 billion USD/year) are usually used for the economic production of butanol. However, the dependence of chemicals on the price of oil makes this alternative unsuitable for the near future. Additionally, *n*-butanol synthesis is taken into account carbon neutrality and sustainability. It's produced naturally by *Clostridia* spp. through acetone-butanol-ethanol (ABE) pathway; however slow ratio of growth, complicated life cycle, complex nutrient need, and demanding genetic modifications constrain the assembly (biological) of butanol (Vogt and Richnow 2013). Furthermore, Clostridium spp. is incapable of directly using low-price substrates like hemicellulose, organic waste, and cellulose and must be dependent on the molasses and starchy materials whose accessibility has been limited by environmental dependence and competition with human food correspondingly (Kucharska et al. 2018). Special engineering strategies are adapted by using non-butanol-producing species, like *E. coli*, *B. subtilis*, *P. putida*, *S. cerevisiae*, and *Lactobacillus brevis* to extend marketable production targets of butanol. Toxicity induced by butanol is the primary major difficulty to become commercial yields. With the chaotropic action of 37.4 kJ/kg/M, butanol interrupts the macromolecular complexes of the host cell and then produces oxidative destruction which finally results in product-induced growth (Cray et al. 2015).

1.11 Production of Microalgae-Biofuels with Nano-Additives

For biofuel generation, microalgae are seen as potential feedback within the current age due to its energy-rich source, cheap culture approaches, inflated rate of growth, the prominent ability of CO_2 fixation, and O_2 accumulation to the atmosphere. Recently, research is constant regarding the improvement of microalgal biofuel technologies (Zhu et al. 2018) (Fig. 1.3). Application of nano-additives has been understand as a prominent improvement to mitigate this experience. At different stages from microalgal cultures, the nano-additive application to end-product use presented a solid possibility into the longer term (Anwar et al. 2020). Currently, the foremost complex technology is that the nanotechnology incorporation with the applications of bioenergy by the nano-energy zone possesses a durative on biofuel transformation mechanism and improvement of engine progress. This technology is described because of the design of a material or device on a nanoscale (10–9 m). To accomplish the biofuel product and develop the effectivity of biofuel consumption in diesel and petrol, nanotechnology has been introduced through nano-additives like nano-crystals, nano-droplets, nano-fibers, nano-magnets, etc. (Hossain et al. 2019).

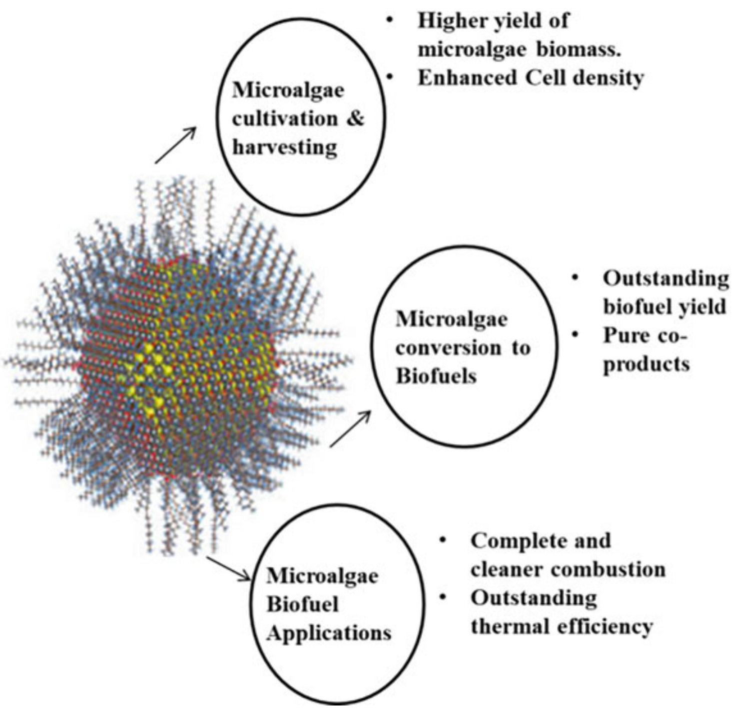

Fig. 1.3 Nano-additive application for the enhancement of microalgae cultivation to biofuel implementation

The thought of microalgae cultivation has appeared to the spotlight for biofuel production due to many positive outlooks like:

1. They don't clash with human and animal food chains.
2. Rich in proteins, carbohydrates, and oil contents.
3. Ability to sprout on aqueous media like wastewater, saline water, and water and assimilate nutrients from brackish water, highly contaminated water.
4. Low tide requirement.
5. Support the power to supply whole year naturally with sunlight existence.
6. Are often grown within the bare land (especially within the cold region), ponds, waste dump area, river, and municipal and industrial waste drainage.
7. Create a sustainable O_2 production system.
8. Use of CO_2 for photosynthesis respiration helps CO_2 elimination from atmosphere (Pal et al. 2019).

Furthermore, microalgae have a really short harvesting biological clock and yield promising biomass that generate greater yield of the specified biofuel. Interesting microalgae have an extraordinary quantity of lipids, protein, and carbohydrates, the only element of biofuel conversion. For the biofuel productions, nanotechnology applications are implemented, meanwhile when the present notorious approaches of usual microalgae culture, biofuel production includes some restrictions like uneven industrial-scale microalgae yield, increased cost harvesting and production, the energy utilization of biofuel construction from microalgae, and therefore the expansion of greenhouse emission concentration within the environment (Work 2014). For various phases of microalgae cultivation, nanotechnology applications are often entitled to microalgae-biofuel application fuel engines due to stability, durability, crystallinity, proficiency, recyclability, catalytic performance, high storing capacity, absorption, and eco-friendly characteristics (Hossain et al. 2019). Due to improvement, nanotechnology improved microalgal cultivation, the larger yield of several microalgae biofuels also as microalgae-biofuel implications in diesel engines and petrol. Numerous nano-materials, e.g., nano-tubes, nano-particles, nano-fibers, nano-sheets, and further nano-structures, are seen as active nanocatalysts in direct and indirect approaches in biofuel (e.g., bioethanol, biodiesel, biomethane, and others) for product improvement (Darwesh et al. 2021). However, magnetic nanoparticles were used as a transporter for enzyme control for biodiesel and bioethanol production effectivity. Due to potent paramagnetic properties, magnetic nanoparticles were also favored for methanogenesis to yield biomethane (Hossain et al. 2019). Bioenergy yields from lignocellulosic biomass (agriculture excesses) and waste of industries (slurry) furthermore as algae (macro and microalgae), with optimization at nanoscale that has just enforced on the instrument of nano-particles, characteristics of biomass, and various application on biomass progression (Kamla et al. 2021). The scientific assent is that by the process of photosynthesis respiration, algae convert O_2 and CO_2 and generate an enormous quantity of cellular energy contents into sugar, proteins, and lipid (Shuba and Kifle 2018). Development and industrialization threaten the present ecosystem due to the dumping of heavy metals, waste containing sulfur, nitrogen, phosphorous, etc. commonly with exhaling a high quantity of CO_2 to the free air (Hossain et al. 2019).

1.12 Nano-Additives for Microalgae Biomass Conversion to Biofuels

Among all microalgae biofuel, biodiesel got high importance because of the commercial and admired biofuel within the oil market. For the production of biodiesel, applications of basic and acidic nano-catalyst spheres can substitute chemical compounds as sodium methoxide by reacting with the oil and free fatty acids. The constructive effect of this nano-catalyst is that reaction occurs with pressure and coldness also as advance reduces the environmental bome by sodium methoxide (Hossain et al. 2019). Another study about biodiesel at industrial level confirmed that commercial CaO-NPs offered 91% biodiesel conversion efficacy throughout scaled-up catalytic transesterification. Microalgal development with sphere-shaped nanoparticles constituted of calcium, and sand silica compounds shown that cellular growth of microalgae boosted significantly exclusive of destroying harvesting also as biofuel production from oil (Akia et al. 2014). The research study about mesoporous silica nano-catalyst Ti-loaded SBA-15 signified ten times advanced free carboxylic acid (FCA) and water tolerance level than another catalyst for production of biodiesel from oil also as this nano-catalyst accomplished three times improved than other effective nano-catalysts titanium silicate-1 (TS-1) and titanium oxide silicate (TiO_2-S) (Chen et al. 2013). Furthermore, Ti-loaded Santa Barbara Amorphous-15 (SBA-15) nano-catalyst application reduced the chemical (alkaline catalyst, NaOH) cost of transesterification process for biodiesel production by recycling the nano-catalyst also as this method is more environment friendly (Hossain et al. 2019). Another study show that sulfate incorporated Ti-SBA also performed as a biocatalyst to vary oil to 100% esterified bio-lubricant. Bio-oil of microalgae with the aid of nano-particle aided to source of bio-lubricant from bio-oil. Another research reported that Niobia (N_2O_5) incorporated with SBA-5 applications on biodiesel production from biomass via esterification characterized a big development for microalgae-biodiesel yield (Chen et al. 2020). A kind of nano-particle, zeolite (an alumina silicate mineral), has been used as a billboard absorbent through transesterification process. Zeolites are able to absorb the unfavorable moisture content (4–6%) and generate pure glycerine as a co-product furthermore of biodiesel (Tran 2018).

1.13 Genetically Engineered Algae for Biofuels

For several years due to the revolution of the global level energy crisis, microalgae have been developed and are using as a replacement of feedstock for the yield of biofuels. Furthermore, for producing key chemicals, microalgae consider having huge strength for bio cell factories like recombinant enzymes, alcohol, hydrogen lipids, and protein (Jagadevan et al. 2018). Microalgae-dependent renewable energy is considered very economic due to the concept of high-value products such as the algal biorefinery approach. Genetic engineering is a new discipline that combine in a sequence of engineering and science to aid support construct and design novel-biological systems and to gain themes that are formulated rationally (Jagadevan et al. 2018). The tools and resources used for artificial gene network construction, nuclear manipulation, and reproduction of microalgae of genome-scale are limited. Microalgae of genetic engineering (GE) are appearing to be a biofuel synthesis commercial release without such ecological studies or public information to inspect the possible risks (Kumar et al. 2020). Green eukaryotic algae and cyanobacteria a blue-green algae are likely to spread-from ponds which are uncluttered and on the subordinate scale with smaller possibilities from unopened photo bioreactors. Cyanobacteria are especially problematic to detect due to the danger of horizontal gene exchange with discrete microbes (Day and Stanley 2012). Before novel, genetically engineered algae transfer to the environment; environmental and major biosafety dangers should usually be addressed by experts' teams like ecologists. For the synthesis of biofuels, biologists assumed that microalgae will be reformed by using a vision or perception from artificial biology and the innovative process of producing biofuels in the form of genetically engineered organisms (GEO) (Ancillotti et al. 2016) (Fig. 1.4). High-throughput sequencing of hybridization, DNA sequencing, metagenomics, and accelerated evolution. Genetic engineering is currently expanding, becoming more accessible, and becoming more effective. Nemours strains of algae have been selected worldwide and collected for raw genetic material (Park and Kim 2016). The pathways and related genes are vital for the biosynthesis of biodiesel in *Dunaliella tertiolecta*, a non-model aquatic flagellate (Rismani-Yazdi et al. 2011). In the microalgae, the composition of DNA which enhances the rate of growth, and Boston the nitrogen efficiency uses, have been studied for Nemours patents. In photobioreactors and in open ponds, the growing algae are very expensive. By research, it is summarized that there is still much need to do innovation including genetically engineered microalgae cultivation (Barry et al. 2016). The novel characters can be accomplished by methods of non-transgenic therefore value assessment, to increase the performance of wild strains, recombinant DNA is used. To introduce microalgae at the commercial level, Sapphire Energy Corporation works together with Monsanto company to investigate innovative genes which converse extra growth and additional positive traits (Snow and Smith 2012). There are spin-off applications in crop plant. The company Joule Unlimited has patented a mix of foreign microbe genes and genetically engineered cyanobacteria that makes more ethanol. Novel GEOs are

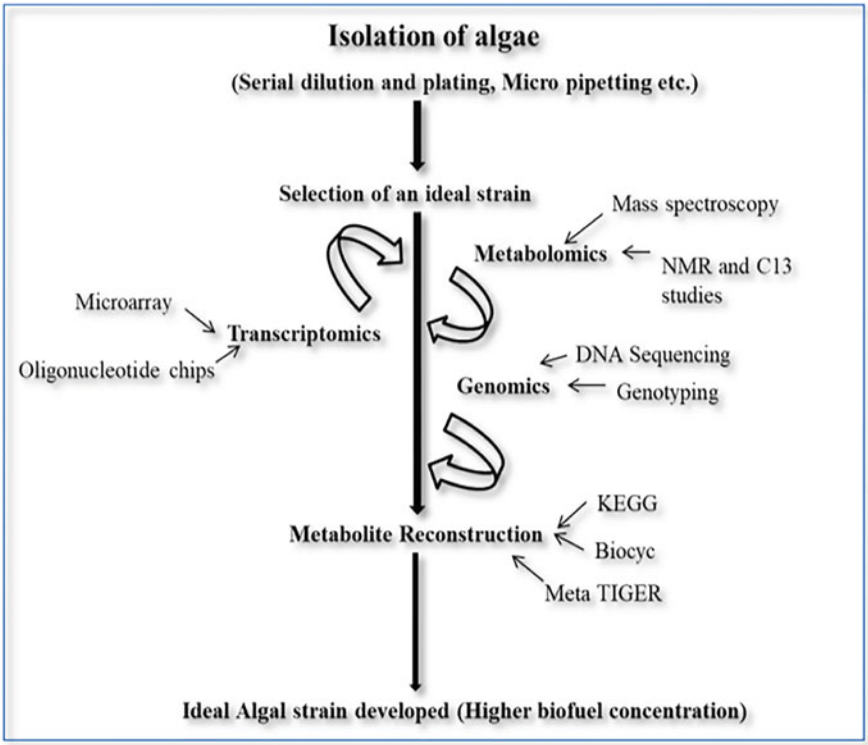

Fig. 1.4 Pictorial representation of the overall process toward biofuel production in microalgae using synthetic biology approach selection of an ideal strain redirecting metabolism to maximize the synthesis of the targeted biofuel

represented within the atmosphere. Moreover, major biosafety dangers should be addressed to invest cause of harm and to the potential stage of exposure (Snow and Smith 2012). To GE algae endurance outside of open lakes or encased bioreactors is essential standard source for hazard investment. The problem has arisen that genetically engineered algae discover lab-created strains must unable to subsist within the rough specially if tamed or kept forming huge volumes of industrial type of by-products. If an unproposed release era to prissiest, the concerned GE algae choose that their developing descendants may have that would allow them to survive and continue in natural surroundings (Henley et al. 2013). Such engineered traits, for example, resistance to severe temperatures or improved growth, may help GE algae survive and develop in a suitable ecosystem.

Significantly more had to think about the condition of lab than their regular habitats (Joutey et al. 2013).