

Algal Engineering for Bioenergy, Environmental Remediation and High-value Biocompounds: A Review

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Abstract: - Sustainable and cost effective ways to produce renewable energy from biomass have generated considerable interest especially the conversion of wastes generated from oil palm industries. These include empty fruit bunch (EFB) (23%), mesocarp fibre (12%), shell (5%) and palm oil mill effluent (POME) (60%), from every tonne of fresh fruit bunches (FFB) processed. POME has high chemical oxygen demand (COD) and biochemical oxygen demand (BOD) and mineral content such as nitrogen and phosphorous which can cause severe pollution to the environment. The demand for improved systems of bioenergy generation, environmental remediation and high value biochemicals co-production have led to the potential use of algae in biomass utilization. A combination of wastewater treatment and renewable bioenergy production would be an added benefit to the palm oil industry.

Key Words: -Microalgae, Bioenergy, Biocompounds, Palm Oil Mill Effluent, Bioremediation

1 Introduction

As the energy demand increases, interests in alternative renewable sources of fuels are increasing worldwide [1]. The challenge in the production of biofuels is the competition between fuels and food production, the effect of which has been an increase in food prices [2; 3]. The solution may lie in the application of microalgae for the production of biofuels. Microalgae have the capability of using inorganic carbon dioxide and wastewater components as nutrients while producing biomass. The microalgal biomass can be used as a source of high-value products including for health, pharmaceuticals, pigments, animal feeds and biofuels [4, 5, 6].

Algae are one of the most abundant and primitive life forms on earth. Categorized as plants because of their photosynthetic ability, algae are now placed

within the diverse kingdom Protista of eukaryotes, predominantly single-celled microscopic organisms [5]. However, they are less complex than the plants. The eukaryotic chlorophyll-containing cells can be a single cell, colonies, filament of cells, or the kelp tissues. A microalgae cell has rapid growth rate-100 times faster than land-based plants and can double its biomass in less than 1 day [7], once every 3–4 h, but mostly every 1–2 days under favorable growing conditions [8].

Microalgae are an ideal alternative as a green energy source, as it is not interfering with world food chain, animal and plant eco-population. The different types of biofuels that can be developed include biodiesel derived from lipid, bioethanol from fermentation of biomass, biomethane by anaerobic digestion of biomass, and photo-biologically produced biohydrogen [6, 9]. Algae adds an advantage to effluent treatment by increasing the performance of degradation,

improving CO₂ balance and lowering energy demand for oxygen supply in aerobic treatment stage. The role of algae is both to assimilate plant nutrients and to support bacteria with oxygen. Bacteria, in turn, are involved in the degradation of organic material in wastewater, the same process utilized in activated sludge [10].

The major challenge to commercialize microalgae biofuels is high energy input required especially in the upstream processes. For biodiesel, the required amount of biomass is huge and the production cost should fall below \$400/tonne of biomass to be economically feasible. This is still far from the price currently reported in a full-scale plant. The costs for a medium-scale plant are still 173 times more expensive [5, 11]. Negative energy balance has been reported in a (life cycle assessment (LCA) of microalgae for biodiesel production essentially in the harvesting and drying process [12].

Malaysia has huge supply of biomass resources mainly derived from palm oil, wood and agro-industries. It is estimated that more than 50 million tonnes of biomass are generated from the palm oil industry. At present, most of palm biomass comes from the oil extraction process such as the mesocarp fiber, shell, and EFB. The mesocarp fiber and shell are burnt within the boiler to generate steam for electricity, while the EFB is being used as fertilizer or soil mulching in the oil palm plantation. POME has not been extensively commercially re-used by the industry but the high content of carbohydrates (29.55%), proteins (12.75%), nitrogenous compounds, and lipids, with a considerable amount of cellulose and non-toxic minerals provide a good source for microbial fermentation [13, 14].

Through microbial activity annually, some 590–880 million tons of methane are released worldwide into the atmosphere and about 90% come from biogenic sources. Methane is over 20 times more effective in trapping heat in the atmosphere than carbon dioxide (CO₂) over a 100-year period [15]. POME can be a good source to produce methane gas. Sustainable energy management in palm oil mill has entered a new dynamic era with the opportunity of culturing microalgae using POME [16]. Most palm oil millers favor the culture of microalgae as a tertiary treatment before POME is discharged due to practicality, low cost and high efficiency. Most of the nutrients such as nitrate and ortho-phosphate that are not removed during anaerobic digestion will be further treated in microalgae pond. The cultured microalgae will then

be used as a diet supplement for live feed culture [17].

The effluent treatment currently used by the Malaysian palm oil industry include anaerobic/facultative ponds, tank digestion and mechanical aeration, tank digestion and facultative ponds, decanter, physicochemical and biological treatment. Treatment of POME using membrane technology, up-flow anaerobic filtration, up-flow anaerobic sludge blanket and up-flow anaerobic sludge fixed film bioreactor has also been reported. At present, 85% of POME treatment is based on anaerobic and facultative pond system, followed by open tank digester attached with extended aeration in a pond [18]. Since POME contains high level of organic matters, implementation of anaerobic digestion in the first stage of the treatment process is necessary to alter the bulk of the wastes to biomethane. The treated effluent is further exposed to aerobic treatment in order to meet the required discharge standards. These treatment steps have been applied either as an open pond or open digesting tank systems in palm oil mills [19].

This review discusses co-cultivation of microalgae using POME for lipid and biomethane production, in combination with wastewater treatments and recovery of valuable biocompounds.

2 Algal Engineering

With more knowledge on algal biology and technological advances, the commercialization of algal biofuel production will be feasible in not too distant future, provided the issues related to large-scale methods to cultivate and harvest algal cells, extraction of lipid and conversion into diesel are properly addressed. Manipulation of processing conditions such as temperature, salinity, light, pH and nutrients as well as culture duration allows modulation of cell growth, biochemical and lipid composition for consequent optimization of overall yield and productivity [20].

2.1 Engineering Considerations

2.1.1 Growth Conditions

Microalgae are naturally acclimatized to a range of aquatic habitats, and it is sensible to use strains isolated from native environments. Autotrophic or photoautotrophic requires photosynthesis to produce complex organic compound from simple inorganic molecules like salts [21]. Heterotrophic microalgae do not carry out photosynthesis but use organic

substrates like glucose or acetate as the carbon and energy source to stimulate growth, while mixotrophic microalgae derive energy from both photosynthesis and chemical oxidation and they combine both photoautotrophic and heterotrophic mechanism to sustain growth [16]. Heterotrophic culture can utilize organic carbon source for growth in the dark thus eliminating the requirement for light [22]. In the presence of a fixed carbon source (glucose), some microalgae, can have higher biomass ($> 20 \text{ g L}^{-1}$) and oil productivity ($> 50\%$ of the dry weight as lipid) than phototrophic production [23].

2.1.2 Cell Productivity

Both biomass productivity and oil content must be considered for oil productivity, and this is most accurately defined by multiplying the areal biomass productivity (or the annual average) and the intracellular oil content at the time of harvesting. Oil productivity as high as $100000 \text{ L ha}^{-1} \text{ yr}^{-1}$ or $27.5 \text{ ml m}^{-2} \text{ d}^{-1}$ for a shallow, open pond system (raceway pond) has been reported [9, 24]. This would actually require a biomass productivity of $36.7 \text{ g m}^{-2} \text{ d}^{-1}$ with 75% oil content, or $50 \text{ g m}^{-2} \text{ d}^{-1}$ with 50% oil content, or $91.6 \text{ g m}^{-2} \text{ d}^{-1}$ with 30% oil content and none of these has been reported. *Botryococcus braunii* accumulates 70-80% of cell weight as lipid, but has low biomass productivity of only $3 \text{ g m}^{-2} \text{ d}^{-1}$ [4, 9]. Other fast growing strains may achieve areal productivity of $12\text{-}47.6 \text{ g m}^{-2} \text{ d}^{-1}$, but the intracellular lipids levels are typically lower than 30%.

2.1.3 Tolerance to Extreme Culture Conditions

Lipids in eukaryotic photoautotrophic cells function as a structural component of cell membranes or to modulate cellular activity, and serve as energy storage compounds. Under stressed conditions, microalgae accumulate high concentrations of carbon in the form of neutral lipids triacylglycerol (TAG) [25, 26]. These high-carbon lipids are the potential source of sustainable oil production. One of the main biological functions of TAG the cells is to provide energy for immediate and delayed metabolic requirements. Once the stationary phase of the growth cycle is reached and nutrients are depleted, microalgae may switch the cell metabolism and store the energy in the form of lipids as an environmental stress response [25, 27]. The switch can be protracted with no net biomass productivity or lowering the productivity. The estimated biochemical composition for single species of chlorophyta *Chlorella*, *Botryococcus braunii*, and *Dunaliella salina* are 30-

50% protein, 20-40% carbohydrate and 8-15% lipids. Under stressed conditions, the species accumulate 80% fatty acid, 80% hydrocarbon and 40% glycerol on dry weight basis [28]. Under nutrient deficient condition, oil productivity may be lesser than that in control at $5 \text{ g m}^{-2} \text{ d}^{-1}$ [25].

Microalgal strains that have been mass cultured in outdoor open ponds, in extreme conditions with consistent productivity are *D. salina* in hypersaline water, *Spirulina platensis* grown at high pH and *Chlorella* sp. grown at high nutrient loading [22]. Altering pH of the entire culture is feasible but requires addition of acids or bases, and extreme eutrophy may require excessive use of fertilizers. An increased salinity represents a favourable option, as daily evaporation loss from the open pond would increase the salinity and a saline-tolerant strain should be able to survive. Metabolic stress agent such as SAN 9785 herbicide, enhances the eicosapentaenoic acid (EPA) production by 28% in *Porphyridium cruentum* [29]. However, the use is limited to indoor, closed culture systems due to the expenses and environment concerns.

2.2 Factors for Optimal Production

2.2.1 Temperature and Light

Light and temperature are probably the most important and well-studied factors influencing the lipid, carbohydrate and biocompounds composition of algae. Carbohydrate and other bioactive contents of microalgae could be enhanced by irradiance and temperature variation [30]. Lipid changes alter the physical properties of membranes which allow unimpaired functioning in important physiological processes including photosynthesis, respiration and membrane transport. Temperature affects the biochemical composition and the quantity of cellular lipid and fatty acid classes [31]. Alterations in environmental temperature change fatty acid unsaturation in membrane lipids [32]. Optimal temperatures for most microalgae strains either freshwater or saline are $16\text{-}28^\circ\text{C}$, although, some survive extremes of -5°C and above 90°C . Temperature lower than 16°C may reduce growth but any higher may result in photosynthetic deficiency [33]. Temperature below optimal range often leads to an increase in unsaturation of lipids in the membrane. This improves stability and fluidity of cell membranes especially thylakoid membrane via increased unsaturated fatty acids to prevent the photosynthetic machinery from photo-inhibition at low temperature [34].

Table 1: Fatty acid profile at optimized photoperiod and light intensity [35]

Experimental		Total fatty acids composition (%)														
Conditions		C14:	C15:	C16:	C16:	C17:	C18:	C18:	C18:	C18:	C20:	C20:	C20:	C20:	C20:	C22:
		0	0	0	1	0	0	1	2	3	0	2	3	4	5	6
<i>N. oculata</i>	PBR	4.4	7.1	22.0	9.8	13.7	5.5	7.3	4.6	3.2	4.0	ND	ND	3.3	6.6	4.5
	Open Tank	3.0	13.0	21.2	7.4	5.2	5.9	5.1	2.8	ND	3.2	ND	ND	ND	ND	4.4
<i>T. suecica</i>	PBR	4.2	17.8	14.8	2.8	2.6	5.4	3.2	5.3	1.6	1.8	1.1	0.4	ND	6.8	3.1
	Open Tank	4.0	16.9	12.6	4.9	3.8	5.6	2.7	2.4	ND	4.2	0.7	ND	ND	ND	0.0
<i>I. galbana</i>	PBR	10.3	8.2	19.1	5.05	9.7	5.4	14.5	2.2	0.6	8.0	2.7	ND	ND	4.1	5.5
	Open Tank	8.9	2.6	13.2	4.5	5.1	3.3	11.5	1.4	ND	2.4	2.1	ND	ND	3.4	1.8
<i>P. lutheri</i>	PBR	2.8	3.7	34.4	21.2	3.4	0.7	9.6	1.4	2.1	2.6	ND	ND	ND	8.4	6.9
	Open Tank	1.5	2.2	26.3	18.7	2.3	ND	7.2	ND	1.2	2.4	ND	ND	ND	7.7	5.4

Stimulation of fatty acid and membrane lipid, mainly chloroplast, synthesis are achieved with increased light intensity. Growth and lipid content of *Pavlovalutheri* has been reported under 24 h illumination with maximum specific growth rate, μ_{max} , of 0.12 day^{-1} and lipid content of 35% as compared to 0.1 day^{-1} and 15% lipid content in the dark [35]. High light intensities may lead to oxidative damage of polyunsaturated fatty acids (PUFA). In *Nannochloropsis* sp., the degree of unsaturation of fatty acids decreased with increasing irradiance, especially the percentage of total n-3 fatty acids (from 29 to 8% of total fatty acids) mainly due to a decrease of EPA [36]. Comparing 5L photobioreactor (PBR) and 300L tank cultivation, the 19.3–24 h illumination and light intensity of $162\text{--}198 \mu\text{mol photons m}^{-2}\text{s}^{-1}$ achieve the highest cell density and biomass with *N. oculata* at 82.6×10^6 and 63.7×10^6 cells mL^{-1} density, with 0.96 and 0.72 gL^{-1} biomass, respectively [35]. However, the lipid content is higher in 5L PBR at 40% as compared to 30.7% in 300 L open tank possibly due to better hydrodynamic condition in the former. Table 1 shows that heptadecanoic acid C17:0 (13.7%), oleic acid C18:1 (7%), palmitic acid C16:0 (22%) and palmitoleic acid C16:1 (9.9%) for *N. oculata* are high in PBR. For *P. lutheri* in PBR, palmitic acid C16:0 (34%) remains high, while both EPA C20:5 (8%) and docosahexaenoic acid (DHA) C22:6 (6.9%) slightly increase, with the total saturated fatty acids (SFA) (47.9%) and monounsaturated fatty acids (MUFA) (30.9%) remain comparable but with PUFA (18.9%) evaluated at optimum illumination and light intensity [35]. A higher lipid accumulation (19.3%) is demonstrated using a 12:12 h light:dark cycle at 25°C as compared to 35°C in *P. cruentum* [37]. A

significant increase in starch content from 8.5% (dry weight basis) to 40% is observed when the mean light intensity is increased from 215 to $330 \text{ mol m}^{-2} \text{ s}^{-1}$ [38].

2.2.2 Salt concentrations and pH

Some algae exhibit an excellent ability to tolerate high salt concentrations. In *D. salina* cells, the lipid content is manipulated by salt stress and nitrogen limitation, and it reaches 38% in cells grown at 16% NaCl combined with 2.5 mM unspecified nitrogen salts [39]. These conditions also increase the relative proportion of PUFAs, in particular the C18:3n-3 and C16:4n-3 fatty acids. An increase in the initial salt concentration from 0.5 M NaCl to 1.0 M results in an increase (from 60 to 67%) of intracellular lipid content in *Dunaliellatertiolecta* [40]. The optimum salinity (30–40 ppt) and pH (8–9) for cell growth and lipid content (34–36%) has been suggested for *P. lutheri*. The alkaline pH stress may increase the TAG percentage accumulation but decrease the relative level of membrane lipids [41].

2.2.3 Nutrients and CO₂ supplementation

Nutrient limitation is an important modulator of algal lipid biosynthesis. When algal growth slows down as a result of nutrient deficiency, there is no requirement for the synthesis of new membrane compounds, and the cells can transfer fatty acids into their storage lipids before conditions improve. The highest lipid cell content (0.40 g g^{-1} dry weight) is obtained at the lowest sodium nitrate concentration (3 mM), whereas a higher lipid productivity of $0.133 \text{ g L}^{-1}\text{d}^{-1}$ is achieved at 5 mM with a lipid content of 0.34 g g^{-1} [42]. However, the maximum lipid content (56% of dry weight) is shown after 6 days of nitrogen depletion without

CO₂ supplementation [43]. The highest lipid accumulation of 37.3, 23.6, 28.3 and 37.2% with slightly reduced cell growth of 0.64, 0.49, 0.54 and 0.38 g L⁻¹ are achieved for *N. oculata*, *T. suecica*, *I. galbana* and *P. lutheri* when cultures are grown under deficiency conditions at 10-65 g L⁻¹ KNO₃, 3-7.5 g L⁻¹ Na₂HPO₄ and 2.5 g L⁻¹ FeCl₃ [35]. The lipid class and fatty acid composition of the green microalga *Chlorella zofingiensis* have been compared under photoautotrophic and heterotrophic cultivation conditions. Heterotrophic cells fed with 30 g L⁻¹ of glucose are shown to increase oleic acid (from 17.9 to 35.2% of total fatty acids) in comparison to photoautotrophic cells. The oils from heterotrophic *C. zofingiensis* appear to be more suitable for biodiesel production [44].

The addition of CO₂ is required for autotrophic growth and a sufficient supply is one of the key factors influencing the accumulation of carbohydrate in microalgae. *N. oculata* grows best in a semicontinuous system aerated with 2% CO₂ and operated by 1-day medium replacement for long-term biomass production and higher lipid yield [45]. Fatty acid synthesis in a highly CO₂ tolerant alga, *Chlorococcum littorale* is increased at low CO₂ after nitrate depletion with a controlled HCO₃/CO₂ ratio [46]. The relative FA content is 34 wt.% at 22°C, light intensity of 170 m mol photons m⁻²s⁻¹ and 5% CO₂ with O₂-free gas and this content is comparable to plant seed oils [46]. Increasing dissolved CO₂ from 3 to 186 mol L⁻¹ in *C. pyrenoidosa* and *C. reinhardtii* cultivation could elevate the carbohydrate content from 9.3 to 21% and 3.2 to 7.4%, respectively [47]. Suitable addition of CO₂ improves the autotrophic growth of microalgal cells and protein content, although it may not directly enhance carbohydrate accumulation in microalgae, unless appropriate stress conditions are employed [48].

3 POME Remediation

Algal treatment replacing conventional tertiary POME treatment can offer an oxygenated effluent and an ecologically safe, less expensive and more efficient mean to remove nutrients and metals. Microalgae as a tertiary treatment for nitrogen and phosphorus not removed during anaerobic digestion can reduce eutrophication at point sources better than can be achieved by conventional treatment [49, 50]. During digestion, bacteria consume the oxygen released by microalgae to decompose the organic matter, giving out carbon dioxide, ammonia, and phosphates, which are assimilated by the microalgae

and methane released as energy. Sludge from wastewater treatment plant can be co-cultured with algae to enhance remediation but unlike activated sludge for secondary effluents treatment, algae can eliminate nitrogen and phosphorus without organic carbon requirement [51]. Cultured microalgae can be used as a diet supplement for live feed culture [52, 53] or harvested for biodiesel.

POME treatment utilizing *N. oculata*, and *Chlorella* sp. achieve the highest removal of COD (95-98%), BOD (90-98%), TOC (80-86%) and TN (80%) after 7 days of anaerobic treatment as compared to treatment without microalgae [54, 55]. POME treated with anaerobic co-cultivation of *Tetraselmis suecica* achieves high removal efficiency of COD, BOD, TOC and TN after 3 and 7 days HRT at 87-95%, 84-95%, 67-90%, 73-80%, respectively. The lowest removal efficiency of COD (53%), BOD (73%), TOC (49%) and TN (48%) are achieved on day 3 of aerobic treatment without microalgae [56].

4 Bioenergy Co-generation

Table 2 shows anaerobic co-digestion of mono-algal species with POME and EFB for biomethane production. With *Chlorella* sp. after 3 days HRT, the highest biomethane yield (5276 mL L⁻¹ POME d⁻¹) and specific biogas production rate (0.129 m³ kg⁻¹ COD day⁻¹) are achieved at 2 mL mL⁻¹ POME and EFB of 0.12 g mL⁻¹ POME. With *N. oculata*, the biomethane is lower (4812 mL L⁻¹ POME d⁻¹) but the specific biogas production rate is consistent (0.126 m³ kg⁻¹ COD d⁻¹) [54, 55, 56]. With reduced amount of EFB (0.06 g mL⁻¹ POME) but high mono-algal *N. oculata* and *Chlorella* sp. (2 mL mL⁻¹ POME), comparable biomethane yield (4443-4524 mL CH₄ L⁻¹ POME d⁻¹) and the specific biogas production rate (0.120-0.122 m³ kg⁻¹ COD d⁻¹) are obtained. At lower amount of *Chlorella* sp. cultured separately (1 mL mL⁻¹ POME) but high EFB (0.12 g mL⁻¹), the biomethane yield (3816 mL CH₄ L⁻¹ POME d⁻¹) and specific biogas production rate (0.105 m³ kg⁻¹ COD d⁻¹) are comparable to *N. oculata*.

At optimum conditions, both at all times register higher production than *T. suecica*. Anaerobic co-cultivation of *T. suecica* with EFB and POME achieve moderate biomethane yield (3965 mL CH₄ L⁻¹ POME d⁻¹) and lower specific biogas production (0.116 m³ kg⁻¹ COD d⁻¹). Without *T. suecica*, high specific biogas production (0.127 m³ kg⁻¹ COD d⁻¹)

Table 2: Biogas production by mono-algal co-digestion with OPEFB, POME and sludge inocula

			Responses					
	Algae (mL mL ⁻¹ POME)	OPEFB (g mL ⁻¹ POME)	Specific biogas production rate (m ³ kg ⁻¹ COD d ⁻¹)			Biomethane (mL CH ₄ L ⁻¹ POME d ⁻¹)		
			<i>N. oculata</i>	<i>Chlorella</i> sp.	<i>T. suecica</i>	<i>N. oculata</i>	<i>Chlorella</i> sp.	<i>T. suecica</i>
Without addition of inocula and algae	0	0	0.094	0.046	0.011	ND	ND	ND
	0	0.06	0.104	0.012	0.014	ND	ND	ND
	0	0.12	0.015	0.014	0.014	ND	ND	ND
With addition of inocula (3 mL mL ⁻¹ POME)	0	0	0.104	0.105	0.105	2703	2704	2704
	0	0.06	0.115	0.115	0.115	3224	3226	3026
	0	0.12	0.125	0.120	0.127	3649	3650	3642
Co-digestion with different concentration of microalgae and inocula (3 mL mL ⁻¹ POME)	1	0	0.094	0.090	0.094	2945	3180	2441
	1	0.06	0.095	0.092	0.099	3030	3266	2854
	1	0.12	0.121	0.105	0.101	4020	3816	3762
	2	0	0.107	0.106	0.109	3874	4132	3669
	2	0.06	0.120	0.122	0.111	4450	4524	3785
	2	0.12	0.126	0.129	0.116	4812	5276	3965

is obtained but with lower biomethane yield (3642 mL CH₄ L⁻¹ POME day⁻¹). These values however are still much higher than the reported methane production of 573-1170 mL L⁻¹ d⁻¹ from co-digestion of *Scenedesmus* sp. and *Chlorella* sp. separately, with 50% waste paper [57].

Without microalgae, the highest biomethane is 3650.3 mL L⁻¹ POME d⁻¹ but equivalent specific biogas production rate 0.1207 m³ kg⁻¹ COD d⁻¹ at high EFB (0.12 g mL⁻¹) POME. Without both algae and sludge inocula, no biomethane is detected although the specific biogas production rate (0.130 m³ kg⁻¹ COD d⁻¹) and CO₂ (190 mL CO₂ L⁻¹ POME d⁻¹) is much higher, and some hydrogen (78 mL H₂ L⁻¹ POME d⁻¹) detected. High microalgae and EFB co-digestion with POME, at the correct ratio of POME and sludge inocula lead to 1.1-1.4-fold higher biomethane production than without microalgae co-digestion. The specific biogas production rate remains consistent between 0.094-0.129 m³ kg⁻¹ COD day⁻¹ [54, 55, 56].

Filtered POME composition in sea water at different levels (1, 5, 10, 15 and 20%) used as an alternative medium obtain enhanced cell growth and lipid accumulation at 10% POME for *N. oculata* and *T. suecica* with maximum specific growth rate (0.21 d⁻¹ and 0.20 d⁻¹) and lipid content (39% and 27%), respectively, after 16 days of flask cultivation. The algal treatment of POME/Seawater also achieve high removal of COD (93.6-95%), BOD (96-97%),

TOC (71-75%), TN (78.8-90.8%) and oil and grease (92-94.9%).

The major fatty acids composition of lipid recovered from *N. oculata* and *T. suecica* cultivated in 10% POME composition with sea water are pentadecanoic acid (C15:0), palmitic acid (C16:0), stearic acid (C18:0) belonging to SFA; and palmitoleic acid (C16:1) and oleic acid (C18:1) belonging to MUFA. The total SFA (59.24%, 68.74%); MUFA (15.14%, 12.26%); and PUFA (9.07%, 8.88%) are obtained for *N. oculata* and *T. suecica*, respectively. *N. oculata* contains high palmitic acid (C16:0) at 28.22% and palmitoleic (C16:1) at 9.37% while *T. suecica* contained high palmitic acid (C16:0) at 36.48% and pentadecanoic acid (C15:0) at 9.21%. In PUFA profile, the highest percentage of linolenic acid (C18:3) is found in *N. oculata* (4.54%) and *T. suecica* (5.11%). The cultivation of *N. oculata* and *T. suecica* in 10% POME composition with sea water therefore is suitable for cell growth as well as MUFA and PUFA production. With high saturated and monounsaturated fatty acids, *N. oculata* and *T. suecica* are potential candidates for the production of biodiesel [35, 58].

Table 3: Antioxidant activities of different microalgal extracts (225 mg L⁻¹)

Algal species	Antioxidant activity (%)						
	Algal extracts						
	Hexane	Chloroform	Ethyl acetate	Ethanol (70%)	Water	Ascorbic acid	BHT
<i>Oscillatoria</i> sp.	47.4±0.6	55.6±0.8	47.4±0.5	69.1±0.4	50.2±0.4	94.6±0.1	85.8±0.1
<i>Nostoc</i> sp.	52.2±0.4	63.8±0.6	63.9±0.8	51.0±0.6	50.6±0.5		
<i>N. muscorum</i>	31.4±0.5	30.2±0.3	27.0±0.5	26.3±0.7	27.8±0.4		
<i>N. piscinale</i>	48.0±0.5	50.5±0.5	49.2±0.7	55.4±1.1	49.5±0.3		
<i>Phormidium</i> sp.	49.7±0.3	50.7±0.3	50.8±0.3	52.6±0.4	50.9±0.1		
<i>A. flos-aquae</i>	44.9±0.2	48.8±0.2	48.7±0.4	44.9±0.2	46.9±0.1		
<i>S. platensis</i>	44.9±0.1	45.9±0.2	47.5±0.3	43.6±0.4	41.4±0.3		
<i>D. splendida</i>	43.9±0.4	46.3±0.3	45.6±0.2	42.6±0.2	45.4±0.2		
<i>Chlorella</i> sp.	41.0±0.3	30.3±0.3	42.6±0.4	42.0±0.4	42.3±0.2		
<i>S. obliquus</i>	59.8±0.2	61.2±0.1	61.3±0.3	60.7±0.1	64.3±0.2		

5 High-value Biocompounds

The antioxidant activity and total phenolic content of three cyanobacterial species (*Phormidium* sp., *Oscillatoria* sp. and *Nostoc muscorum*) evaluated suggest that the ethanolic extracts of all species demonstrate high phenolic contents (mg GAE/g) with *Phormidium* sp. records the highest (12.66±0.16), followed by *Oscillatoria* sp. (7.9±0.11) and *Nostoc muscorum* (5.04±0.15). Phenolic compounds such as flavonoids, phenolic acids, and tannins are considered to be major contributors to the antioxidant capacities of plants. These antioxidants also possess diverse biological activities such as anti-inflammatory, anti-atherosclerotic and anti-carcinogenic activities which may be related to their antioxidant activities. However, there are no direct correlations between the antioxidant capacities and the phenolic contents suggesting that the phenolic compounds are not major contributors to the antioxidant activities of these cyanobacterial species [59].

The ethanolic extract of *Oscillatoria* sp. (Table 2) shows the highest antioxidant activity (69.1 %), while the green *Chlorella* sp. shows higher phenolic content (39.1 mg GAE g⁻¹ dry wt.) and *Scenedesmus obliquus* records higher carotenoid content (3.73 mg L⁻¹), than cyanobacterial species [60]. *S. platensis* shows relatively high phycobiliprotein (0.16±0.01 mg/ml) and total phenolics (21.88±1.67 mg GAE/g), total alkaloids (3.02±0.06%) and terpenoids (0.14±0.00%).

Microalgal biomass therefore has a great potential as the source of natural antioxidants [61].

6 Conclusion

Rapid industrial development, reduction of mineral oil resources, and rise in atmospheric CO₂ require the development of carbon-neutral renewable alternatives. Treatment of POME with microalgae could achieve both waste remediation and bioenergy co-generation. Cells can be harvested for recovery of valuable biocompounds.

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