

## Comparative Study of biomass and biofuel production from different species of micro and macro algae

Hanan. M. Abobaker and Hamida El.salhin

Botany Department, Faculty of Science Omar El-Mokhtar University  
El -Beida-Libya  
Hanan.ma.saleh@Omu.edu.ly.

### ABSTRACT

There has been a lot of focus on renewable fuels as alternative energy sources, and biodiesel is becoming more and more well-liked globally. The process of turning microalgae into biofuels, particularly biodiesel, has become very popular recently. It is believed that because biodiesel is renewable and environmentally friendly, it can take the place of fossil fuels. Phycologists view algae as a promising source of biofuel due to their biomass and fatty acid productivity in the current study we focuses on comparative between the deferent species of algae in production of oil and biomass, it good potential for biodiesel production, the utilization of algae as the resilient source for biofuel specially micro algae *Senedesmus quadricaud*, *Cladophora sauteri* and recorded as mean 48%, 18% respectfully as oil of dry weight. The algae *Cystoseira barbata* was 22% while red algae *Carollina granifera* reached to 16% of oil content in the end of the experiment at pH 10. The algal biomass of *S. quadricaud* and *C. sauteri* was 1.2 g /L , 7.7 g/L respectively whereas seaweeds distinguished by high algal biomass 230g/m<sup>2</sup> for *C. granifera* and 244g/m<sup>2</sup> for *C. barbata*.

**Keywords:** Biofuel Production, Biodiesel, Micro & Macro Algae, Alternative Energy.

## دراسة مقارنة لإنتاج الوقود الحيوي والكتلة الحيوية من أنواع مختلفة من الطحالب الصغيرة والكبيرة.

حنان محمد أبوبكر، حميدة الصالحين الصالحين

جامعة عمر المختار - البيضاء - ليبيا

Hanan.ma.saleh@Omu.edu.ly.

### الملخص:

لقد حظي الوقود المتجدد لمصادر الطاقة البديلة بالكثير من التركيز، وقد اكتسب الديزل الحيوي شعبية عالمية كمصدر بديل للطاقة. اكتسب إنتاج الوقود الحيوي من الطحالب الدقيقة وخاصة وقود الديزل الحيوي شعبية كبيرة في السنوات الأخيرة، ومن المفترض أنه بسبب طبيعته الصديقة للبيئة والمتجددة، يمكن أن يحل محل الحاجة إلى الوقود الأحفوري. وناقش علماء الطحالب باعتبارها واعدة لإنتاج الوقود الحيوي على أساس إنتاجية الكتلة الحيوية والأحماض الدهنية. وفي هذه الدراسة، ركزنا على المقارنة بين أنواع الطحالب المختلفة في إنتاج الزيت والكتلة الحيوية، وإمكاناتها الجيدة لإنتاج وقود الديزل الحيوي، واستخدام الطحالب كوقود حيوي. متجدد المصدر للوقود الحيوي وخاصة الطحالب الدقيقة *Cladophora sauteri* و *Senedesmus quadricauda* وتم تسجيل متوسط كمية الزيت من الوزن الجاف بنسبة 48% و 18% على التوالي. وبلغت نسبة الزيت في الطحلب *Cystoseira barbata* 22% بينما وصلت نسبة الزيت في الطحلب الأحمر *Carolina granifera* إلى 16% في نهاية التجربة عند الرقم الهيدروجيني 10. وكانت الكتلة الحيوية لطحالب *C. sauteri* و *S. quadricauda* 2 جم/لتر، 7.7 جم/لتر على التوالي، بينما تتميز الأعشاب البحرية بكتلة حيوية عالية من الطحالب 230 جم/م<sup>2</sup> بالنسبة الي *C. granifera* و 244 جم/م<sup>2</sup> بالنسبة الي *C. barbata*.

الكلمات المفتاحية: إنتاج الوقود الحيوي، البيوديزيل، الطحالب الصغيرة و الكبيرة، الطاقة البديلة.

## 1. INTRODUCTION

Energy crisis is among the biggest problems, leading the world to be unsafe and non-peaceful. The demand is increasing day by day. The available resources are rapidly decreasing and indication is, soon will be vanished. In such situations, more attention is needed to be given towards renewable energy sources. Fossil fuels are used on a large scale in the world, but unsustainable because they increase CO<sub>2</sub> level and accumulate greenhouse gases which make the environment unhealthy. To keep the environment clean and maintain sustainability, renewable and environmentally friendly fuels are needed to be produced [1]. Biodiesel is biodegradable, nontoxic and a low emission profiles, environmentally friendly biofuel, also contributes no net carbon dioxide or sulfur to the atmosphere and emits less gaseous pollutants than conventional diesel fuel It can be produced in any climate [2]. Many strains of micro and macro algae are known to produce high quantities of lipids that can convert to biodiesel [3].

The word "algae" describes a very broad category of eukaryotic organisms that are harvested from different aquatic environments and are either farmed or wild. Most of these organisms contain chlorophyll. Considered to be among the earliest living forms are algae. So far, between 40,000 and 100,000 species of algae have been identified; however, this figure may even be underestimated [4]. Macro algae can grow up to 60 m in length and are typically fast growing [5], which is located in the uppermost five meters of the surface layer and is referred to as the Photic-zone. The distribution of algae is influenced by a number of natural factors, the most significant of which is light intensity [6]. Heat is another important factor; green algae prefer a temperature of 10-15°C, Bacillaroiphyta prefers a temperature of 15–20°C, and blue-green algae is more common at 25–35°C [7, 8]. Compared to terrestrial crops, which take a season to grow and can only contain a maximum of roughly



5% dry weight of oil, microalgae grow quickly and have a high oil content. Usually, they grow by double every 24 hours. Certain microalgae can double every three and a half hours while they are in their peak growth phase [9].

Oil content of microalgae is usually between 20 percent and 50 percent, while some strains can reach as high as 80 percent [10]. This is why microalgae are the focus in the algae-to-biofuel arena. Phototrophic microalgae require light, carbon dioxide, water, and inorganic salts to grow. The culture temperature should be between 15 and 30°C for optimal growth. The growth medium must contribute the inorganic elements that help make up the algal cell, such as nitrogen, phosphorus, iron, and sometimes silicon [11]. For large-scale production of microalgae, algal cells are continuously mixed to prevent the algal biomass from settling, and nutrients are provided during daylight hours when the algae are reproducing [12].

The present study was focused to comparative the oil and biomass productivity of micro and macro algae species , Isolation, identification and production oil of green alga *S.quadricauda* , *C.sauterian* and Two of sea weeds *C.chilensis* , *C.barbata* using solvent system.

## MATERIALS AND METHODS

### 1.1. Micro algal Isolation, Purification and Identification

The micro algal species *S.quadricaud* has been isolated from Topruk coastal is located in natural area in the north-eastern part of Libya and cultured in the laboratory under suitable culture conditions. The medium used throughout the maintenance and experimental studies was medium (MBL).

The isolated *S. quadricauda* as cultivated with MBL medium and the experiments were carried out in 500 ml Erlenmeyer pyres-glass flasks containing 200 ml of culture under controlled conditions of ambient air at laboratory temperature. Light was provided by cool-white fluorescent lamps at 4000 Lux with a dark/light cycle of 16:8 h for 14 days. After period the culturing the cells of *S. quadricauda*

were harvested by centrifugation at 5000 r p m for 30 min using angle rotor centrifuge. The supernatants were discarded and the remaining pellets were used to extract of biofuel [13,14].

Algal species examined by means of binocular microscope and identified according to the following references:[15],[16], [17], [18], [19], [20], [21], [22].

### 1.2. Collection and identification of macro algae

Manual harvesting of seaweed has been practiced for centuries and it is still common for species naturally growing in coastal area [23]. Seaweeds samples were collected *Cystoseira barbata* C.Agardh, (Phaeophyta, Fam. Sargassaceae), *Corallina granifera* J. Ellis&Solander, (Rhodophyta, Fam. Corallinaceae) from Al-Hamama coast located the northeastern Mediterranean coast of north of the city of Al-Bayda, 25 km from the city of Al-Bayda - libya, during June 2021. Green macro alga samples *Cladophora sauteri* was fresh water of Derna fall and collected and prepared by the same method of macroalgae, in laboratory, all macro algae sample were cleaned from epiphytes and rock debris and given a quick fresh water rinse to remove surface salts. seaweeds were then air dried in the shade at room temperature (25–30 °C) on absorbent paper for estimation of moisture content. Then, they were pulverized in a cereal grinder for 5 min and sieved, using a 100 mesh sieve, to produce a homogenous, fine powder that was kept at -20 °C in hermetic sealed plastic bags pending additional chemical analysis. The taxonomy of all seaweeds was determined using the techniques of [24], [25], [26]. The species names were used in accordance with [27], and the algae base website was utilized to verify them. The Botany Department Laps, Faculty of Science, Omar Al Mukhtar University, identified the samples that were obtained.

### 1.3. Extraction of oil

By used of chloroform–methanol (2:1 by volume) for extraction of lipids from endogenous cells. Briefly, the homogenized cells were equilibrated with one-fourth volume of saline solution and mixed well. The resulting mixture was allowed to separate into two layers and lipids settle in the upper phase [28].

#### 1.4. Determination of algal biomass

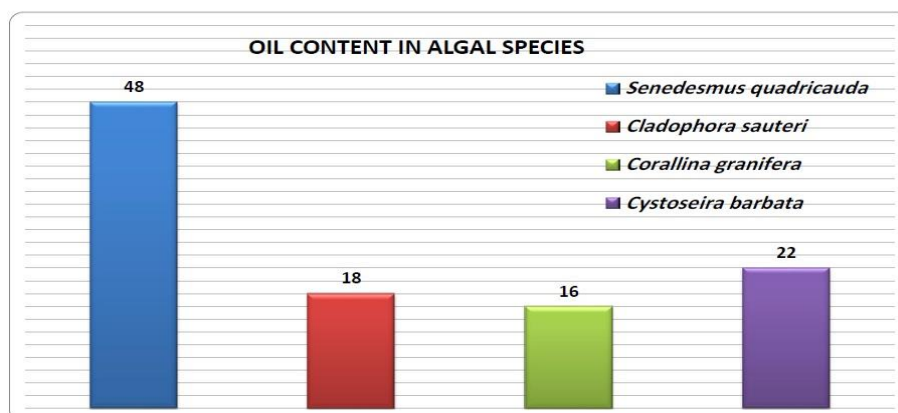
Determination of algal biomass according to [29]. Biomass calculated as grams dry weight per square meter ( $g\ d\ w / m^2$ ).

## 2. RESULTS AND DISCUSSION

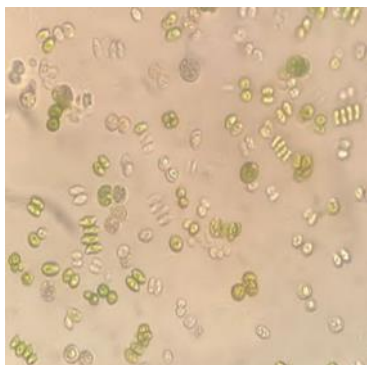
Extraction of the oil from algae experiment designed to investigate produce the oil from the micro and macro algae, the results obtained from this study were Cleared that the maximum value of oil content was from green micro alga *S. quadricauda*, while the value of the rest of the algae converged in terms of the percentage of oil of the dry weight. The minimum value obtained from red alga *C. granifera* the algal biomass recorded the highest values in sea weeds samples *C. barbata* followed by *C. chilensis* as Cleared in table (1) and figure (1).

**Table (1) Algal biomass and oil content in algae species.**

Algal species	Algal biomass g/l	Oil content			Mean
		R1	R2	R3	
<i>Senedemus. quadricauda</i>	2.2g	48	60	50	48
<i>Cladophora. asauteri</i>	7.7g	20	18	25	18
<i>Carollina. granifera</i>	230g	20	18	16	16
<i>Cystoseira. barbata</i>	244g	25	24	22	22



**Figure (1) Oil content in algal species**



*Senedemus. quadricauda*



*Cladophora. asauteri*



*Cystoseira. barbata*



*Carollina. granifera*

Figure (2) Types of some algal species

Algae are renewable and biodegradable source of biofuel less CO<sub>2</sub> and NO<sub>2</sub> emissions. Continuous use of petroleum sourced fuels is now widely recognized as unsustainable because of depleting supplies and the contribution of fuels to the accumulation of carbon dioxide in the environment [30].

By using the solvents on the selective micro and macro algae the results cleared that the green alga *S. quadricauda* contained the highest amount of oil 48% of dry weight 2g at pH 10, the same result Demirbas, 2011 [31] revealed that Under good conditions, green algae can double its biomass in less than 24 hours. Green algae can also have high lipid contents, usually over 50%. This high yield



is ideal for intensive agriculture and can be an excellent source for biodiesel production. Another study on green alga *Odegonium* regarded amount of oil was 22.5% of dry weight 8g and the pH value 8 [32], in agreement with our study [33], cleared that the Productivity of photo bioreactor was 1.2 g/l dry weight algae, and 45% of the dry weight biomass of *Botryococcus braunii* was made up of lipids. Biofuel production is 0.45 ml/g dry weight. that note to algae as promising and economic source of biofuel due to of its availability and low cost , in agree with the results we founded in green alga *Senedemus.quadricauda* produce high quantities of lipids that reached to 48% can be converted into biodiesel. In the similar search the most common microalgae contain oil ranges between 20 and 50% by dry weight of biomass [34]. Identical or very close with the average values that express the amount of biofuel that we founded in this study were 48,18 in *S. quadricauda* and *C.asauteri* respectively.

Conversely, macro algae are the most significant element in marine ecosystems, contributing to the preservation of marine bioresources by averting pollution and eutrophication [35]. Furthermore, macroalgae that have access to sunlight and saltwater's available nutrients can thrive in salinity. Chemical fertilizers are not necessary for them. As a result, significant energy and financial savings are possible. These traits support the long-term viability of the macroalgae-based bioethanol production process. In 2017, Yuvarani *et al.* [36,37] established the green macroalgae *Cladophora sauteri*, which produced 20% of the oil, typically with our result optioned the maximum oil percentage is found to be 18% from methanol chloroform solvent methods. The hydrocarbons, alcohols, carbonyl compounds, acids, and their ester, terbenes, are found in the marine red seaweed *C. granifera* [38, 39].

In the summary of present study the red alga show the high biomass 230g/m<sup>2</sup> and oil content reached to 20%, brown alga *C.barbata* had highest biomass 244g/m<sup>2</sup> and 25% oil content comparative with another study density of bed biomass (221, 332 kg/m<sup>3</sup>), bio-oil yield (29.9-34.8%by. In laboratory scale studies, even if chloroform-methanol blends have been extensively used



with high extraction yields up to 83% (g lipid/g dry weight) ,these results confirmed that increase extraction by using polar and no polar solvents.

#### 4. CONCLUSION

1. The conclusion of the study is that algae are considered source of biofuel, the quantity of oil is greater in small algae than in large algae.
2. The results of the biomass of different types of algae showed that there is no definitive relationship between biomass and amount of oil produced, as high biomass is not necessarily associated with high rate of extracted.

#### REFERENCES

- [1] Schenk, Peer M, Thomas-Hall, Skye R, Stephens, Evan, Marx, Ute C, Mussgnug, Jan H, Posten, Clemens, . . . Hankamer, Ben. (2008). Second generation biofuels: high-efficiency microalgae for biodiesel production. *Bioenergy research*, 1, 20-43.
- [2] Meher, LC., Sagar, D. V, & Naik, SN. (2006). Technical aspects of biodiesel production by transesterification—a review. *Renewable and sustainable energy reviews*, 10(3), 248-268.
- [3] Ahmad, Abdul Latif, Yasin, NH Mat, Derek, CJC, & Lim, JK. (2011). Microalgae as a sustainable energy source for biodiesel production: a review. *Renewable and sustainable energy reviews*, 15(1), 584-593.
- [4] Bruton, Tom, Lyons, Henry, Lerat, Yannick, Stanley, Michele, & Rasmussen, Michael Bo. (2009). A review of the potential of marine algae as a source of biofuel in Ireland. *Sustainable Energy Ireland*, 1-88.
- [5] McHugh, Dennis J. (2003). A guide to the seaweed industry. *FAO fisheries technical paper*, 441, 105.
- [6] Herborne, JB. (1973). *Phytochemical methods. A guide to modern techniques of plant analysis*, 2, 5-11.



- [7] Granhall, Ulf. (1975). 12. Nitrogen fixation by blue-green algae in temperate soils. Nitrogen fixation by free-living micro-organisms, 6, 189.
- [8] Whitton, Brian A, & Potts, Malcolm. (2012). Introduction to the cyanobacteria. Ecology of cyanobacteria II: their diversity in space and time, 1-13.
- [9] Chisti, Yusuf. (2007). Biodiesel from microalgae. Biotechnology advances, 25(3), 294-306.
- [10] Spolaore, Pauline, Joannis-Cassan, Claire, Duran, Elie, & Isambert, Arsène. (2006). Commercial applications of microalgae. Journal of bioscience and bioengineering, 101(2), 87-96.
- [11] Grobbelaar, Johan U. (2004). Algal nutrition: mineral nutrition. Handbook of microalgal culture: biotechnology and applied phycology, 97-115.
- [12] Fernández, FG Acién, Camacho, F García, & Chisti, Yusuf. (1999). Photobioreactors: light regime, mass transfer, and scaleup Progress in industrial microbiology (Vol. 35, pp. 231-247): Elsevier.
- [13] Nichols, HW. (1973). Handbook of phycological methods (pp. 7-24): Cambridge University Press Cambridge.
- [14] Rippka, R. (1992). Pasteur culture collection of cyanobacterial strains in axenic culture. Catalogue and taxonomic handbook, catalogue of strains 1992/1993, 1, 1-103.
- [15] Prescott, GW. (1982). Algae of the western great lakes area, Otto Kaetz Science publishers. W. German.
- [16] Pentecost, Allan. (1984). Introduction to freshwater algae.
- [17] Boney, AD. (1983). Phytoplankton studies in Biology.
- [18] Komárek, J, & Fott, B. (1983). Chlorophyceae (Green algae), Order: Chlorococcales.[in:] Phytoplankton of freshwaters. Eds. Huber-Pestalozzi, G, 7, 1044.
- [19] Prescott, Gw. (1984). Economics of algae. The algae: A Review". Otto koeltz Science publishers D-629 Koenig-Stein, W. Germany.
- [20] Charles, Bold Harold. (1985). Introduction to the algae: structure and reproduction.



- [21] Krammer, K. (1986). Band 2/1. Bacillariophyceae. 1. Teil. Naviculaceae. SuBwasserflora von Mitteleuropa.
- [22] Krammer, K. lange-bertalot, H. 1988. Süßwasserflora von Mitteleuropa. Bacillariophyceae, 2(2).
- [23] Van den Burg, Sander, Bikker, Paul, van Krimpen, Marinus, & van Duijn, Arie Pieter. (2013). Economic feasibility of offshore seaweed production in the North Sea. Paper presented at the Aquaculture Europe Conference, Trondheim, Norway.
- [24] Taylor, William Randolph. (1972). Marine algae of the eastern tropical and subtropical coasts of the Americas: University of Michigan.
- [25] Abbott, Isabella A, Isabella, Abbott, & Hollenberg, George J. (1992). Marine algae of California: Stanford University Press.
- [26] Jha, Bhavanath, Reddy, CRK, Thakur, Mukund C, & Rao, M Umamaheswara. (2009). Seaweeds of India: the diversity and distribution of seaweeds of Gujarat coast (Vol. 3): Springer Science & Business Media.
- [27] Guiry, MD. (2013). AlgaeBase. World-wide electronic publication. <http://www.algaebase.org>.
- [28] Folch, Jordi, Lees, Mark, & Sloane Stanley, Gerald H. (1957). A simple method for the isolation and purification of total lipids from animal tissues. J biol Chem, 226(1), 497-509.
- [29] Fatemi, SMR, Ghavam Mostafavi, P, Rafiee, F, & Saeed Taheri, M. (2012). The study of seaweeds biomass from intertidal rocky shores of Qeshm Island, Persian Gulf. International Journal of Marine Science and Environment, 2(1), 101-106.
- [30] D., Klass L. (1998). Biomass for Renewable Energy, Fuels and Chemicals. . Academic Press, NewYork. , 1-2.
- [31] Demirbas, A., and Demirbas, M.F.(2011)“Importance of algae oil as a source of biodiesel.” “Energy Conversion and Management.” . 52:163-170.
- [32] Muradov, Nazim, Taha, Mohamed, Miranda, Ana F, Wrede, Digby, Kadali, Krishna, Gujar, Amit, . . . Mouradov, Aidyn. (2015). Fungal-assisted algal flocculation: application in

- wastewater treatment and biofuel production. Biotechnology for biofuels, 8(1), 1-23.
- [33] Hossain, ABM Sharif, Salleh, Aishah, Boyce, Amru Nasrulhaq, Chowdhury, Partha, & Naquiuddin, Mohd. (2008). Biodiesel fuel production from algae as renewable energy. American journal of biochemistry and biotechnology, 4(3), 250-254.
- [34] Mata, Teresa M, Martins, Antonio A, & Caetano, Nidia S. (2010). Microalgae for biodiesel production and other applications.
- [35] Notoya, Masahiro. (2010). Production of biofuel by macroalgae with preservation of marine resources and environment. Seaweeds and their role in globally changing environments, 217-228.
- [36] S., Yuvarani M. Kubendran D. Aathika A. S. R. Karthik P. Premkumar M. P. Karthikeyan V. and Sivanesan. (2017). Extraction and characterization of oil from macro algae *Cladophora glomerata*. . Part A: Recovery, Utilization, and Environmental Effects., 1556-7036.
- [37] K.(, De Rosa S. Kamenareska Z. Stevano. (2003). Chemical compstion of *Carollina meditrreinea* Areschoug and *Carollina granifera* . Ell. et 85, 5-6.
- [38] Cioroiu, Doinita Roxana, Parvulescu, Oana Cristina, Dobre, Tanase, Raducanu, Cristian, Koncsag, Claudia Irina, Mocanu, Alexandra, & Duteanu, Narcis. (2018). Slow pyrolysis of *Cystoseira barbata* brown macroalgae. Rev Chim, 69(3), 553-556.
- [39] Yaguchi, T, Tanaka, S, Yokochi, T, Nakahara, T, & Higashihara, T. (1997). Production of high yields of docosaheaxaenoic acid by *Schizochytrium* sp. strain SR21. Journal of the American Oil Chemists' Society, 74, 1431-1434.