

Effect of different pyrolysis temperatures on biofertilizer properties of microalgal biochar and energy analysis

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Abstract

In this study, *Chlorella* sp. (Cs), *Chlorella vulgaris* (Cv), *Neochloris conjuncta* (Nc), *Botryococcus braunii* (Bb), and *Scenedesmus obliquus* (So) microalgae strains were cultivated in channel type ponds. The microalgal biomasses obtained were divided into two groups (350 and 600 °C). The microalgal biomasses in the first group were biocharized at two different pyrolysis temperatures, while those in the second group were untreated crude microalgal biomasses. As a result of the energy input-output analysis of both groups of microalgal biomasses, the highest net energy gain was calculated in the un-treated Cv strain with 52.41, while the lowest value was calculated in the biocharification process of So and Bb strains at 600°C with 13.03. In all groups, the energy efficiency, energy ratio, and net energy gain of the Cv strain were found to be higher than other microalgae strains. When the bio-fertilizer, biostimulant data, and energy data are evaluated together, it's concluded that it's most appropriate to prefer the Cv microalgae strain.

Keywords: Microalgal biomass, Microalgal biochar, Pyrolysis, Biofertilizer, Biostimulant, Energy efficiency

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INTRODUCTION

The increasing trend in the global population is also similar to industrialization. It is known that the increase in industrialization is proportional to the use of fossil fuels. It is stated that the use of fossil fuels and the increase in atmospheric CO₂ levels accordingly exceeds the critical threshold value today. This situation reveals the climate crisis, drought, and the need for efficient use of water resources. Among the many energy alternatives, biofuels are likely to emerge as strategically important sustainable fuel sources in the foreseeable future (Uysal, 2022). Since the amount of C sequestered in the soil (1100 Gt; 1Gt = 1000000 tons) is much higher than that in the atmosphere (750 Gt) (Sundquist, 1993), an average of 60 Gt CO₂ is released annually from the soil to the atmosphere. This CO₂ released into the atmosphere is mostly formed during the respiration of microorganisms during the decomposition of organic matter in the soil. One of the most useful practices to reduce the increasing CO₂ concentration in the atmosphere is the process of converting CO₂ stored in biomass by photosynthesis into a more stable form of C called biochar by pyrolysis (Spokas and Reicosky, 2009). Since biochar contains more and more stable aromatic C than soil organic matter, its degradation is very slow. Biochar has been reported to persist in soil for 100 to 1000 years and has 10 to 1000 times higher organic matter than most soil organic matter. Therefore, unlike organic matter traditionally used in agriculture, biochar is expressed as an additive that will enrich C and positively affect soil quality (Lehmann and Joseph, 2015; Verheijen et al., 2010).

In recent years, studies on the reduction of atmospheric CO₂ have encountered a significant amount of biochar application. Since biochar application causes significant changes in the chemical, physical, and biological properties of the soil, it also affects the C and N dynamics in the soil (Van Zwiiten et al., 2014). There are applications such as green manure, barn manure, compost, and biochar in studies to increase organic matter in soils (Akkeçeci and Özkan, 2022). While some of these applications provide nutrients directly to the soil, some of

them have a healing effect on soil properties. The properties, application amounts, and decomposition degrees of organic materials added to the soil to increase the organic matter are significant for the expected benefits. The balance of decomposition and addition in the preservation of organic matter in soils is an important indicator of sustainable soil management. The good development of plants is closely related to the physical, chemical, and biological properties of the soil's environment. One of the applications made to optimize these properties is the application of organic material to the soil. Biochar applications are considered as a remarkable approach for soils that are tired after the practices during agricultural production. In almost all countries in the world, biochar applications to soils attract a great deal of attention. Biochar is a product resulting from the conversion of organic materials (of plant and animal origin) into materials containing high carbon and mineral substances by gasification at high temperatures, in an oxygen-free or very little oxygen environment (Akgül, 2017). It has been shown that the biochar obtained by the pyrolysis of biomass can make a positive contribution to issues such as reducing the loss of nutrients by washing, reducing the bioavailability of environmental pollutants, enriching carbon in the soil, reducing greenhouse gas emissions and improving soil fertility (Ippolito et al., 2012). In addition to the effect of biochar on C storage in the soil, many studies have been published investigating the effect of N₂O release in fertilized agricultural lands. It is known that N₂O, whose contribution to global warming is 298 times higher than carbon dioxide, is 100 years longer in the atmosphere (IPPC, 2013). In the last century, it has been reported that a significant amount of N₂O has been released into the atmosphere due to the use of nitrogen fertilizers in agricultural lands (Park et al., 2012). N₂O is a thermodynamically strong oxidant, but kinetically refractory towards decomposition and reduction. This kinetic barrier can be overcome through binding and activation with metal ions such as Fe or Cu (Tolman, 2010). The presence of Cu and Fe in the micronutrients required for microalgae cultivation, as well as the presence of microelements in microalgal biomass, is an indication that this kinetic barrier will be overcome. Microalgae is a material with a high potential for biochar as it is a sustainable and renewable resource (Yu et al., 2017). Microalgae's use in the production of biochar is being explored due to various advantages and potential application areas. Carbon Sequestration: Microalgae absorb carbon dioxide from the atmosphere through photosynthesis. This characteristic allows microalgae to potentially serve as a carbon sink during growth, contributing to the formation of biochar. Bioenergy Production: The incorporation of microalgae in biochar production can generate energy during the bioenergy production process. Biochar serves as a biomass fuel in energy production. Soil Improvement: Microalgae biochar can be used to enhance soil quality. The addition of biochar to soil may increase water retention capacity, improve soil structure, and enhance nutrient retention for plants. Carbon Cycling: Microalgae biochar can contribute to controlling the carbon cycle. By providing long-term carbon storage in the soil, it may help reduce carbon emissions into the atmosphere (Cheah et al., 2015; Suganya et al., 2016). Agricultural Applications: When applied to agricultural soils, microalgae biochar may stimulate plant growth and increase yield. It can also support environmental sustainability by reducing pesticide and fertilizer usage. Industrial Applications: Microalgae biochar can find applications in the production of carbon-based materials in industrial processes. Adding microalgal biochar to the soil can have several positive effects. Here are some potential impacts of adding microalgal biochar to the soil (Ayaz et al., 2021; Zhang et al., 2021). Improvement of Soil Structure: Microalgal biochar can enhance soil structure and texture. Especially in sandy or clayey soils, it can increase water retention capacity, combining better drainage and water-holding properties. Increase in Plant Nutrient Retention Capacity: Microalgal biochar can increase the soil's capacity to retain plant nutrients. This can enable plants to access necessary nutrients for a longer duration. Enhancement of Water Retention Capacity: Microalgal biochar can increase the soil's water retention capacity, supporting plant water supply during dry periods. Regulation of Soil pH: Microalgal biochar can balance soil pH and provide an optimal pH level for plant growth. Support for Soil Microorganisms: Microalgal biochar can support the development of beneficial microorganisms in the soil, contributing to the health and balance of the soil ecosystem. Carbon Storage: Microalgal biochar can increase carbon storage capacity in the soil by introducing carbon. This can aid in storing carbon from the atmosphere in the soil. Stimulation of Plant Growth: Microalgal biochar can promote plant growth by making nutrients and water in the soil more efficiently available. Improvement of Seed Germination: Microalgal biochar can enhance seed germination and the development of young plants, supporting the overall process of plant cultivation (Ayaz et al., 2021; Zhang et al., 2021).

The aim of this study is to cultivate five different microalgal (*Chlorella* sp., *Chlorella vulgaris*, *Neochloris conjuncta*, *Botryococcus braunii*, *Scenedesmus obliquus*) strains and to evaluate the potential of the products obtained from the pyrolysis of the resulting biomass at two different temperatures (350 and 600°C) in terms of plant nutrition.

MATERIALS AND METHODS

This study was carried out in the microalgae production greenhouse located in Isparta University of Applied Sciences, Faculty of Agriculture, Research and Application Farm (37°50'28.9" N, 30°32'16.1" E). Open ponds were preferred as a cultivation system. Care was taken to ensure that the treatments were the same during cultivation, and a separate open pond was used for each strain. While the total capacity of each open pond was 1.2 tons, the study was carried out with a volume of 1 T (Figure 1). Cs, Cv, Nc, Bb, and So strains were selected in

the study (SAG, 2024; UTEX, 2024; CCAP, 2024) (Figure 2). Basal media was preferred as the media for all strains and the pH value was recorded between 6 and 8 during the cultivation phase. The cultivation period lasted 12 days for each strain.

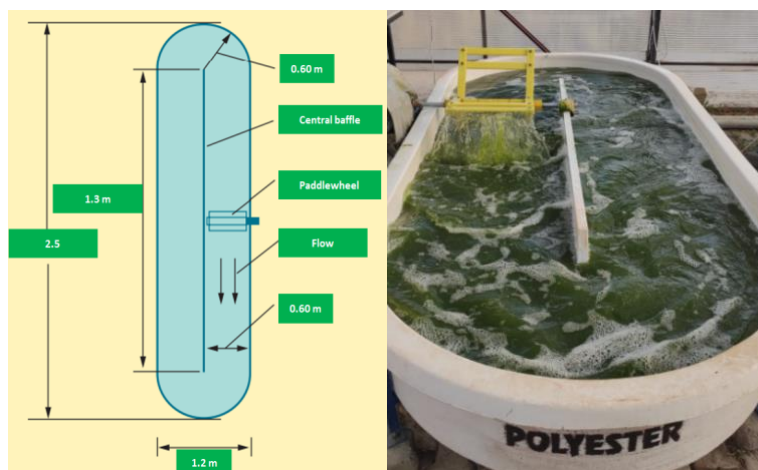


Figure 1. Open channel pond for microalgae cultivation



Figure 2. Microalgal strains in study (a) Cs, (b) Cv, (c) Nc, (d) Bb, and (e) So

Microalgal-based biochar production

Microalgal biomass samples of each strain harvested from open channel ponds were dried in an oven at 65°C until their weights remained constant. In the study, a muffle furnace was used for pyrolysis processes in a laboratory environment, and the pyrolysis duration was set to 2 hours. The dried microalgal biomasses were subjected to pyrolysis process at 350 and 600 °C, each 100 gr.

Harvest of microalgal biomass

At the end of the cultivation periods, microalgal biomass was harvested with an industrial centrifuge. Half of the harvested microalgal biomass was dried in a Memmert oven at 65°C.

Macro and micronutrient elemental analysis

Microalgal microorganisms obtained at the end of cultivation practices micro and macro elements of biomass and biochar-treated sample contents were analyzed by ICP-OES.

Energy analysis

Equation 1 for energy ratio, Equation 2 for energy efficiency, Equation 3 for specific energy, and Equation 4 for net energy gain are considered (Hesampour et al., 2021).

$$\text{Energy ratio} = \text{Output energy (MJ)} / (\text{Input energy (MJ)}) \quad (1)$$

$$\text{Energy efficiency} = \text{Yield (kg)} / \text{Input energy (MJ)} \quad (2)$$

$$\text{Specific energy} = (\text{Input energy (MJ)}) / (\text{Yield (kg)}) \quad (3)$$

$$\text{Net energy gain} = \text{Output energy (MJ)} - \text{Input energy (MJ)} \quad (4)$$

CO₂ removal

During the study period, the amount of carbon dioxide consumed in each channel pond system varied depending on the microalgae strain and biomass yield. At the end of the trials, CO₂ removal was calculated using Equation 5 (Slade and Bauen, 2013).

$$\text{CO}_2 \text{ mitigation} = \text{Dry microalgal biomass (kg)} \times 1.83 \text{ kg CO}_2 \quad (5)$$

RESULTS

Micro and macro elemental analysis

Biochar samples were analyzed for micro and macro element contents are given in Table 1. When the biomasses were subjected to pyrolysis at 2 different temperatures and un-treated biomasses were examined, the highest N value was obtained in the sample pyrolyzed at 350 °C in terms of N values, while the highest value in terms of P was obtained in the sample pyrolyzed at 350 °C. In terms of K, the highest value among the samples was obtained from the sample pyrolyzed at 350 °C. When 5 different microalgae strains were analyzed, the highest N value at 350 °C pyrolysis temperature was obtained at 9.64 % in the Bb strain, while the highest N value at 600 °C was obtained at 8.10 % in the Bb strain.

Table 1. Elemental analysis values of microalgal biochars.

Biochar temp. (°C)	Microalgae	Cv	So	Nc	Bb	Cs
350 °C	N (%)	8.96	9.10	7.85	9.64	9.18
	P (%)	4.58	5.00	2.28	6.27	5.23
	K (%)	1.36	1.08	1.99	2.13	3.86
	Ca (%)	22.59	20.92	11.50	12.55	17.80
	Mg (%)	1.70	1.12	3.48	1.47	0.81
	Fe (ppm)	10560.1	11984.1	3830.39	14274.3	9789.89
	Cu (ppm)	31.572	34.341	9.032	44.933	91.32
	Mn (ppm)	8617.69	11255.7	118.465	12513.2	4839.67
	Zn (ppm)	2650.02	2069.33	75.936	2575.08	2021.74
600 °C	B (ppm)	467.223	274.147	21.924	273.046	168.823
	N (%)	7.05	7.85	5.96	8.10	6.98
	P (%)	3.71	4.22	1.75	4.73	3.67
	K (%)	1.06	0.90	1.48	1.53	2.69
	Ca (%)	17.69	17.09	8.66	9.30	12.66
	Mg (%)	1.35	0.94	2.56	1.07	0.54
	Fe (ppm)	8694.55	9896.93	2802.5	10560.9	6991.39
	Cu (ppm)	26.026	26.822	10.079	39.589	69.856
	Mn (ppm)	6642.69	9261.73	85.49	9312.72	3512.89
	Zn (ppm)	2188.45	1730.37	56.041	1957.2	1423.33
	B (ppm)	392.014	230.937	17.603	198.691	135.832

When evaluated in terms of P value, the highest P value was obtained at 6.27 % in the Bb strain at 350 °C pyrolysis temperature, while the highest P value among the samples at 600 °C pyrolysis temperature was obtained in the Bb strain. When the study was evaluated in terms of K value, the highest values among the strains at both temperature values were obtained in the Cs strain (Table 1). When it comes to sustainable and effective agriculture, the pyrolysis process is observed to be effective for the use of microalgae biochar pyrolyzed at 350°C as a biofertilizer and biostimulant in the soil-water-plant cycle.

Energy Analysis

Energy input and output during the cultivation of biochar obtained from microalgal biomass and biochar obtained by pyrolysis at different temperatures As a result of the analysis; energy ratio, energy efficiency, specific energy, and net energy gain balances of microalgal biochar pyrolyzed at 350°C are given in Table 2.

Table 2. Energy input-output values of microalgal biomass pyrolyzed at 350°C.

	Cv	So	Nc	Bb	Cs
Energy ratio	7.772	6.712	7.595	6.712	7.205
Energy efficiency	0.335	0.289	0.327	0.289	0.311
Spesific energy	2.99	3.46	3.05	3.46	3.22
Net energy gain	21.35	18.01	20.79	18.01	19.56

After the biocharification process of microalgal biomass pyrolyzed at 350 °C, the Cv strain has the highest energy efficiency value, while the So and Bb strains have the lowest energy efficiency values. When the same table in terms of net energy gain, again the highest net energy gain was calculated for the Cv strain. Regarding specific energy, the highest value was calculated in the So and Bb strains, while the lowest value was obtained in the Cv strain (Table 2). 600°C pyrolyzed microalgal biochar energy ratio, energy efficiency, specific energy, and net energy gain balances are given in Table 3.

Table 3. Energy input-output values of microalgal biomass pyrolyzed at 600°C.

	Cv	So	Nc	Bb	Cs
Energy ratio	5.633	4.865	5.505	4.865	5.223
Energy efficiency	0.284	0.245	0.277	0.245	0.263
Specific energy	3.524	4.080	3.606	4.080	3.800
Net energy gain	15.62	13.03	15.19	13.03	14.24

After the biocharification process of microalgal biomass pyrolyzed at 600 °C, the Cv strain has the highest energy efficiency value, while the So and Bb strains have the lowest energy efficiency values. When the same table is analyzed regarding the net energy gain, the highest net energy gain was calculated for Cv strain. Regarding specific energy, the highest value was calculated in the So and Bb strains, while the lowest value was obtained in the Cv strain (Table 3). The energy ratio, energy efficiency, specific energy, and net energy gain balances of crude microalgal biomasses not included in the pyrolysis process are given in Table 4.

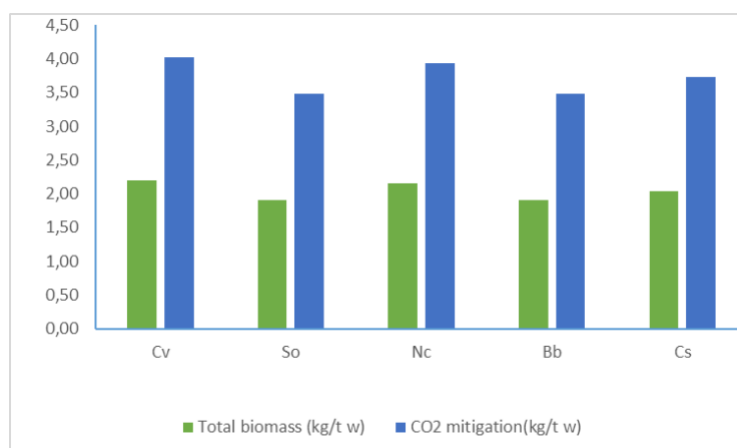
Table 4. Energy input-output values of raw microalgal biomass.

	Cv	So	Nc	Bb	Cs
Energy ratio	39.648	34.242	38.747	34.242	36.765
Energy efficiency	1.622	1.401	1.585	1.401	1.504
Specific energy	0.62	0.71	0.63	0.71	0.66
Net energy gain	52.41	45.08	51.19	45.08	48.50

Energy input and output values of microalgal biomass samples harvested with an industrial centrifuge were calculated. When the table is analyzed in terms of energy efficiency, it is seen that the highest value belongs to the Cv strain, while the lowest energy efficiency value was calculated in the So and Bb strains. While the highest specific energy value was calculated in the So and Bb strains, the lowest value was calculated in the Cv strain, followed by the Nc strain. The highest value as net energy gain was calculated in the Cv strain, followed by the Nc strain. The lowest value was calculated in the So and Bb strains. When the three tables are examined together, it is seen that the highest energy ratio is in microalgal biomasses that are evaluated as raw without processing. Energy ratio, energy efficiency, and net energy gain values decreased with increasing temperature in the pyrolysis process. If the pyrolysis process is to be carried out, it is thought that 350°C temperature will be efficient in terms of energy-biomass relationship compared to other temperatures (Table 4).

CO₂ removal

Thanks to the fact that microalgae consume carbon dioxide as food, it is possible to talk about CO₂ removal in every application. The yield values of the biomass obtained from the applications were calculated. Within the scope of this study, Atmospheric CO₂ removal values were calculated according to the yield values (Figure 3).

**Figure 3.** Biomass and CO₂ removal values of microalgae

Although they were cultivated in the same culture nutrient media, differences in biomass yields and CO₂ removal values were detected. The highest CO₂ removal among the treatments was calculated as 4.03 kg/t w in the Cv microalgae strain, while it was calculated that the same strain would remove 209.35 CO₂/tons year if the cultivation was continued all year round. The lowest CO₂ removal between treatments was determined in So and Bb microalgae strains. CO₂ removal values were also calculated if cultivation continued for 1 year in the same culture nutrient media. CO₂ removal value was determined in the range of 180.80-209.35 kg/t y.

DISCUSSION

Use of microalgal biochar as biofertilizer or biostimulant

Nitrogen and phosphorus are the two main nutrients in the fertilizers. *Scenedesmus obliquus* was used strain as a treatment material in rose industry wastewater (Uysal, 2022). *Scenedesmus obliquus* strain was biocharised both untreated and at 2 different pyrolysis temperatures and the difference in terms of K element is quite significant. When they evaluated in terms of the N element, they obtained a significant increase at 350°C pyrolysis temperature. They reported that high pyrolysis temperature increases the energy input, therefore, when analyzed in terms of yield, 350°C pyrolysis temperature is suitable for the study. In this study, if it is necessary to evaluate in terms of macro elements, it is possible to say that 350 °C pyrolysis temperature is more efficient than 600 °C pyrolysis temperature. When analyzed in terms of microelements, it is possible to say that the content of biochar obtained at 350 °C pyrolysis temperature is more efficient (Uysal, 2022). Researchers compared the biostimulant and secondary metabolite properties of microalgae strains. They reported that the *Scenedesmus obliquus* strain contained auxin, cytokinin, and gibberellin for germination, root, and leaf secondary metabolites for mai bean, cress, and cucumber (Gonzalez-Perez et al., 2022). Drought is known to reduce water turgidity and normal cellular functions by affecting cellular water potential. Many studies have reported reduced photosynthesis, plant growth, and crop productivity under drought conditions (Kambo and Dutta, 2015). Although the drought effect is evident at different stages of the plant life cycle, i.e. seed germination, seedling, vegetative and reproductive stages, several studies have reported the ameliorative effects of biochar on drought-exposed plants (Matovic, 2011; Fahad et al., 2017; Abideen et al., 2020). Researchers have stated that biochar parameters such as higher pH, ash, nitrogen, and extractable inorganic nutrients enhance plant growth under drought stress (Ullah et al., 2021).

Energy analysis

Research reported that microalgal biodiesel and untreated microalgal biofertilizer samples were analyzed for energy input and output and that biofertilizer had the highest values (Uysal, 2022). Also was reported that in microalgae-wastewater applications, after the energy input-output analysis for biomass output, R3 application had the highest energy ratio among the applications with 83.063. It was followed by R2 with 55.201. In terms of energy efficiency, it was reported that the highest value was calculated in the R3 application, while the highest net energy gain was calculated at 40.93 in the R1 application, which did not include the application of GIAS. The always high energy efficiency value of biomass is proportional to the energy requirements of the procedures involved in the processing and conversion of biomass into by-products (Uysal, 2022). It is possible to see similarities in this study. Energy ratio, energy efficiency, net energy gain, and specific energy values were compared at both pyrolysis temperatures. It was observed that the energy analysis values of biochar pyrolyzed at 350°C were higher than those of biochar pyrolyzed at 600°C. It is possible to see this situation more clearly in the raw biomass subject to application without energy input. While specific energy values are low, other energy parameters have the highest values.

CO₂ removal

In a wastewater-microalgae treatment study, it was studied in 3 different application periods and it was reported that the highest removal for CO₂ mitigation in the control treatment, that is, without waste application, belonged to the second treatment and the removal amount was 1.68 kg (Uysal, 2022). In this study, CO₂ removal values were found to be higher even though the same culture medium and even one of the strains was the same. This is explained by the fact that higher biomass was obtained from this study. The biomass value and hence the CO₂ removal value obtained in the wastewater treatment can be attributed to the fact that microalgae are stressed and limit cell growth.

CONCLUSION

It is possible to see that the biofertilizer and biostimulant properties of biocharized bio-mass are further enriched. When evaluated in terms of energy, untreated biomasses have higher energy efficiency values and are therefore preferable. When all the data were evaluated together, it was determined that Cv was the most efficient strain among 5 microalgae strains. When it comes to sustainable and effective agriculture, the use of biochar of microalgae pyrolyzed at 350°C as a biofertilizer and biostimulant in the soil-water-plant cycle is envisaged to be environmentally friendly.

Compliance with Ethical Standards

Peer-review

Externally peer-reviewed.

Conflict of interest

The authors declare that they have no competing interests in this study.

Author contribution

The author contributed in the full study.

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