


Investigation of microalgae isolated from different water resources of Türkiye for their biotechnological utilization

Türkiye'nin farklı su kaynaklarından izole edilen mikroalgelerin biyoteknolojik kullanımlarının araştırılması

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Received date: 22.09.2023

Accepted date: 02.04.2024

How to cite this paper:

Polat, E., & Taştan, B.E. (2024). Investigation of microalgae isolated from different water resources of Türkiye for their biotechnological utilization. *Ege Journal of Fisheries and Aquatic Sciences*, 41(2), 97-104. <https://doi.org/10.12714/egejfas.41.2.03>

Abstract: Microalgae are among the important microorganisms for a sustainable world as a source of renewable energy. In this study, three new microalgae were isolated from different regions of Türkiye and identified by molecular techniques. First isolate was *Chlorella sorokiniana* Shihira and Krauss, 1965 which was isolated from Dim River, second was *Pseudochloris wilhelmii* Somogyi et al., 2013 from Tokat and the third was *Tetrademus obliquus* (Turpin) Wynne and Hallan, 2016 from Tunca River. The maximum biomass of *C. sorokiniana* was 1.02 g/L, 1.86 g/L for *P. wilhelmii* and 0.80 g/L for *T. obliquus*. The chlorophyll (a+b) concentrations were 0.146, 0.278 and 0.181 µg/mL for *C. sorokiniana*, *P. wilhelmii* and *T. obliquus*, respectively. The biotechnological utilization capacities of new isolates were revealed with the support of literature.

Keywords: Biotechnology, *Chlorella sorokiniana*, isolation, chlorophyll, *Pseudochloris wilhelmii*, *Tetrademus obliquus*

Öz: Mikroalgler yenilenebilir enerji kaynağı olarak sürdürülebilir bir dünya için önemli mikroorganizmalar arasında yer almaktadır. Bu çalışmada Türkiye'nin farklı bölgelerinden üç yeni mikroalg izole edilmiş ve moleküler tekniklerle tanımlanmıştır. İlk izolat Dim Nehri'nden izole edilen *Chlorella sorokiniana* Shihira ve Krauss, 1965, ikinci izolat Tokat'tan izole edilen *Pseudochloris wilhelmii* Somogyi ve ark., 2013 ve üçüncü izolat ise Tunca Nehri'nden izole edilen *Tetrademus obliquus* (Turpin) Wynne ve Hallan, 2016'dır. *C. sorokiniana*'nın maksimum biyokütlesi 1,02 g/L, *P. wilhelmii* için 1,86 g/L ve *T. obliquus* için 0,80 g/L olarak bulunmuştur. Klorofil (a+b) konsantrasyonları *C. sorokiniana*, *P. wilhelmii* ve *T. obliquus* için sırasıyla 0,146, 0,278 ve 0,181 µg/mL olarak bulunmuştur. Bu çalışma kapsamında Türkiye'den izole edilen mikroalg türlerinin biyoteknolojik kullanım kapasiteleri literatür desteğiyle ortaya çıkarılmıştır.

Anahtar kelimeler: Biyoteknoloji, *Chlorella sorokiniana*, izolasyon, klorofil, *Pseudochloris wilhelmii*, *Tetrademus obliquus*

INTRODUCTION

The rapid increase in the use of existing energy sources has led researchers to search for renewable energy sources. Microalgae is one of the biomasses used as a renewable energy source. Microalgae are living organisms with biotechnological potential. As a general definition, the term "microalgae" includes both eukaryotic algae such as Chlorophyta, Rhodophyta, Charophyta and the only prokaryotic group Cyanobacteria (Barsanti and Gualtieri, 2014). It is known that cyanobacteria, one of the microalgae, formed about 3 billion years ago and filled the earth with O₂ with their photosynthesis ability. In this way, life forms on earth were formed and diversified. Microalgae are at the forefront of renewable energy studies (Mutaf et al., 2023; Özçiçek et al., 2017). In addition, microalgae are used in food additive production (Jacob-Lopes et al., 2019), wastewater treatment (Taştan et al., 2012a), air pollution prevention (Taştan et al., 2012b), energy production (Perendeci et al., 2019) and many other fields.

The extensive use of microalgae in different industries depends on their growth in both fresh and saline waters, ease and speed of cultivation, and their ability to utilize wastewater

and different substrates (Coronado-Reyes et al., 2022; Lu et al., 2015). The increase in carbon dioxide (CO₂) emissions is one of the consequences of anthropogenic activities that contribute to increased global warming. Microalgae are promising species for preventing the increase in CO₂ emissions (Taştan et al., 2016) and the utilization of microalgal biomass directly or by converting it into related by-products provides added value.

Compared to plants, microalgae have the advantages of growing much faster and requiring less land (Schenk et al., 2008). For example, while the land required for palm oil production is 2 m² year/kg biodiesel, the land requirement for the production of low-lipid microalgae is estimated to be 0.2 m² year/kg biodiesel (Mata et al., 2010). Microalgal biofuels are considered third-generation fuels. Microalgae also have the ability to reduce increasing greenhouse gas emissions and in this context, they fix 40% of global carbon emissions. It is also known that some microalgae species contain 70% lipid (Chu, 2017). Biodiesel from microalgae biomass can reduce 78% of CO₂ emissions on a life-cycle basis compared to conventional diesel fuels (Durrett et al., 2008; Sawayama et al., 1995).

Unfortunately, despite the increase in biodiesel production from microalgae, there is a cost barrier to the commercial use of microalgae in biofuel production, and therefore it has not become practical and cannot replace fossil fuels (Babu et al., 2022; Ghosh et al., 2016).

Nowadays, when the conservation of biodiversity has become even more important due to the increasing loss of species in recent years, the main objectives of our study are to isolate different types of microalgae from different freshwater sources of Türkiye, to determine the species at morphological and molecular levels, to calculate and compare the bioenergy

of these isolates by kinetic methods and to determine in which areas they are used.

MATERIALS AND METHODS

Freshwater sampling

Freshwater samples were taken from 3 different regions of Türkiye (Figure 1), whose coordinates are given in Table 1, during the summer season. Freshwater samples were collected into 50 mL falcon tubes and immediately transferred to the laboratory environment.

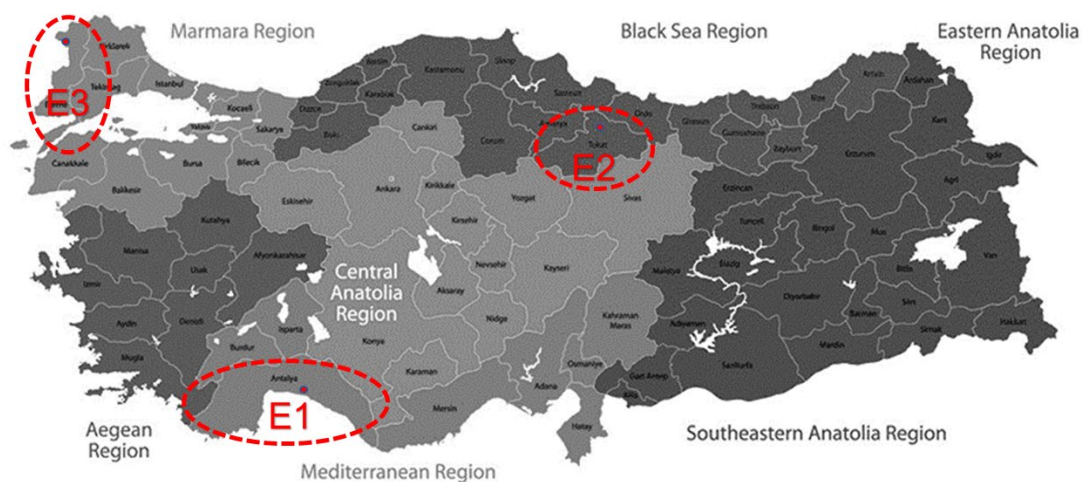


Figure 1. Representation of the regions where water samples were taken on the map of Türkiye

Table 1. Details of the freshwater sources from which microalgae were isolated

Geographical Areas	Freshwater source	Coordinate	Isolate code
Mediterranean Region	Dim Stream	36°33'08.5"N	E1
	Antalya	32°11'11.4"E	
Black Sea Region	Tokat Erbaa village fountain	40°35'31.7"N	E2
		36°40'54.4"E	
Marmara Region	Tunca River Edirne	41°41'44.5"N 26°33'25.8"E	E3

The medium used in the isolation and cultivation studies was BG11 (Rippka, 1988). This medium is generally used for the cultivation of cyanobacteria and is also frequently used for the cultivation of members of the Chlorophyta group (López-Pacheco et al., 2021; Perendeci et al., 2019). BG 11 medium components are summarized in Table 2.

Microalgae isolation and culture conditions

Freshwater samples from three different geographical regions of Türkiye were centrifuged at 5000 rpm for 10 min to precipitate the microbial biomass. 1 mL sample was taken from the pellet obtained and seeded separately into culture media containing BG11 liquid media in 250 mL flasks. They were then incubated at 25±2 °C under 48 µmol/m²s (2400 lx) light (Figure 2). At the end of the incubation, the microalgae were inoculated into agar petri dishes containing BG11 medium by taking 1

loopful sample. Freshwater samples taken from the source were firstly inoculated in liquid media and then isolated in petri dishes in order to strengthen microalgae growth and thus obtain more biomass. Colonies formed in petri dishes were isolated by micromanipulation by planting them in new petri dishes. In the final stage, the purified microalgae cells were transferred to liquid medium. To confirm axenicity, these liquid cultures were also tested for bacterial contamination by plating on bacteriological media.

Table 2. BG 11 medium components

BG11 medium components	
NaNO ₃	1.5 g/L
Stock solutions	g/L
A:K ₂ HPO ₄	0.04
B:MgSO ₄ .7H ₂ O	0.075
C:CaCl ₂ .2H ₂ O	0.03
D:Na ₂ CO ₃	0.02
E: Citric acid	6.00
Ferrous ammonium citrate	6.00
Na ₂ EDTA	1.0
A5 solution	mL/L
H ₃ BO ₃	2.86
MnCl ₂ .4H ₂ O	1.81
ZnSO ₄ .7H ₂ O	0.222
Na ₂ MoO ₄ .5H ₂ O	0.390
CuSO ₄ .5H ₂ O	0.079
Co(NO ₃) ₂ .6H ₂ O	0.049



Figure 2. Cultivation of isolates

The isolates were inoculated into horizontal agar tubes containing agar-BG 11 and incubated at 25 ± 2 °C under 48 $\mu\text{mol}/\text{m}^2\text{s}$ (2400 lx) light and stock cultures were obtained.

Identification of microalgae PCR and sequencing

PCR and sequencing of eukaryotic microalgae samples for molecular identification of microalgal isolates were performed by 18S rRNA gene amplification of cultures in the logarithmic growth phase. The 18S rRNA region was amplified using the following primers; forward p23SrV_F: 5'-GGACAGAAAGACCCCTATGAA -3' and reverse p23SrV_R: 5'-TCAGCCTGTTATCCCTAG-3' (Sherwood and Presting, 2007). PCR was performed in 50 mL of reaction mix containing 0.2 mM of each primer, 0.2 mM of each dNTP, 1.5 mM MgCl_2 and 30 ng of template DNA. Super-HotTaq Taq DNA polymerase (Bioron GmbH, Germany) was the enzyme used for amplification. The initial denaturation step of PCR-amplification was performed at 95 °C for 10 min, followed by 35 cycles of denaturation at 95 °C for 45 s, annealing at 60 °C for 45 s, elongation at 72 °C for 45 s, and final extension at 72 °C for 10 min.

The amplified 5 μL product was analyzed in agarose gel electrophoresis on a 1.2 % agarose gel with 8V/cm ethidium bromide and visualized on a UVP gel imaging system. The amplified PCR product was purified using a QIAGEN gel extraction kit. A total of 18 srRNA amplified products at a concentration of 100 ng/ μL were used for sequencing.

Analytical Methods

a. Cell development analysis

Cellular growth of isolated microalgae was determined by calculating optical density (OD_{600}), total dry weight (X), specific growth rates (μ) and maximum productivity (P_{max}). Optical density was determined by spectrophotometric analysis of the samples at 600 nm; dry cell weight was determined by

weighing the samples after centrifugation at $3421 \times g = 5000$ rpm for 10 min, drying in a sterilizer at 80 °C for 1 night and then weighing. Specific growth rate (μ) was determined according to the following equation (Ip and Chen, 2005);

In this equation, X_2 (g/L) is the dry cell weight at time t_2 and X_1 (g/L) is the dry cell weight at time t_1 .

$$\mu = \frac{\ln X_2 - \ln X_1}{t_2 - t_1}$$

Maximum productivity was calculated according to the following equation, where X (g/L) is the dry cell weight at time t and X_0 is the dry cell weight at time t_0 .

$$P_{\text{max}} = \frac{X - X_0}{t - t_0}$$

b. CO₂ biofixation analysis

CO₂ biofixation rates (F g/g) were calculated according to the following equation (Cheah et al., 2015; Pegallapati and Nirmalakhandan, 2013; Yadav et al., 2015).

$$F = aP \times V$$

a: 1.833 g CO₂, P_x : productivity, V : culture volume.

c. Chlorophyll analysis

Chlorophyll analysis was performed according to the method developed by Porra et al. (1989). Accordingly, chlorophyll a was determined at 646.6 nm and chlorophyll b was determined at 663.6 nm. Chlorophyll (a+b) was calculated as total chlorophyll after calculating chlorophyll a and b separately. Chlorophyll concentrations were expressed as μg chlorophyll per milliliter.

d. Data interpretation and statistical analysis

The recorded results were calculated and interpreted using

descriptive statistical analysis (\pm S.E.). All studies were performed in triplicate. Variances between the data were analyzed in Excel program using %RSD (Relative standard deviation). Before interpreting the data, standard errors were calculated according to the following equation (Kenney and Keeping, 1951);

$$SE = \sqrt{\sigma^2}$$

SE: Standard error, σ : Average of the variable to be analyzed

RESULTS

Identification of microorganisms

At the end of PCR and sequencing studies, according to the results of 18 srRNA, sample E1 was identified as *C. sorokiniana* (NCBI GenBank accession number PP326235) (Shihira and Krauss, 1965); sample E2 as *P. wilhelmii* (NCBI GenBank accession number PP326230) (Somogyi et al., 2013) and sample E3 as *T. obliquus* (NCBI GenBank accession number PP326236) (Wynne and Hallan, 2016). According to the sequences prepared according to the closest species on NCBI

For *C. sorokiniana*;

CTGTTTTACTGTGAAACTGCGAATGGCTCATTAAATCA GTTATAGTTTATTTGATGGTACCTACTACTCGGATACCCG TAGTAAATCTAGAGCTAATACGTGCGCAAATCCCGACTTC TGGAAGGGACGTATTTATTAGATAAAAAGGCCGACCGGGC TTGCCGACTCGCGGTGAATCATGATAAATTCACGAATC GCATGGCCTCGTGCCGGCGATGTTTCATTCAAATTTCTG CCCTATCAACTTTTCGATGGTAGGATAGAGGCCTACCATG GTGGTAACGGGTGACGGAGGATTAGGGTTTCGATTCCGG AGAGGGAGCCTGAGAAACGGCTACCACATCCAAGGAAG GCAGCAGGCGCGCAAATTACCCAATCCTGACACAGGGA GGTAGTGACAATAAATAACAATACTGGGCCTTTTCAGGTC TGGAATTGGAATGAGTACAATCTAAACCCCTTAACGAG GATCAATTGGAGGGCAAGTCTGGTGCCAGCAGCCGCGG TAATCCAGCTCCAATAGCGTATATATAAGTTGCTGCAGT TAAAAGCTCGTAGTTGGATTTCCGGGTGGGGCCTGCCG GTCCGCGGTTTCGGTGTGCACTGGCAGGGCCACCTTG TTGCCGGGGACGGGCTCCTGGGCTTCACTGTCCGGGAC TCGGAGTCGGCGCTGTTACTTTGAGTAAATTAGAGTGTT CAAAGCAGGCCTACGCTCTGAATACATTAGCATGGAATA ACACGATAGGACTCTGGCCTATCCTGTTGGTCTGTA Similarity Rate %99.87;

For *P. wilhelmii*;

GGAATTTCCGCAATGGGCGAAAGCCTGACGGAGCAAT GCCCGTGAAGGATGACGGCCTATGGGTTGTAACCTCT TTTCTCAGAGAAGAATTTGACGGTATCTGAGGAATAAGC ATCGGCTAACTCTGTGCCAGCAGCCGCGGTAAGACAGA GGATGCAAGCGTTATCCGGAATGATTGGGCGTAAAGCGT CTGTAGTTGTGTGACAAGTTTTCTGTCAAAGATCAGGG CTTAACCTGGGCCGCGAGGAAAACTATCATGCTAGAG TTCGGTAGAGGCAGAGGGAATCCAGTGGAGCGGTGA AATCGTAGATATTGGGAGGAACACCAAAGCGCAAAGCA

CTCTGCTGGGCCGAGACTGACACTGAGAGACGAAAGCG AGGGGAGCAAAGGGATTAGATACCCCTGTAGTCTCTGTC TCTTATACACATCTC Similarity Rate: %99.53

For *T. obliquus*;

CTGCTTATACTGTGAAACTGCGAATGGCTCATTAAATCAG TTATAGTTTATTTGGTGGTACCTTACTACTCGGATAACCG TAGTAATCTAGAGCTAATACGTGCGTAAATCCCGACTTC TGGAAGGGACGTATATATTAGATAAAAAGGCCGACCGAGC TTTGCTCGACCCGCGGTGAATCATGATATCTTCACGAAG CGCATGGCCTTGTGCCGGCGCTGTTCCATTCAAATTTCT GCCCTATCAACTTTTCGATGGTAGGATAGAGGCCTACCAT GGTGGTAACGGGTGACGGAGGATTAGGGTTTCGATTCCG GAGAGGGAGCCTGAGAAACGGCTACCACATCCTAGGAA GGCAGCAGGCGCGCAAATTACCCAATCCTGATACGGGG AGGTAGTGACAATAAATAACAATACCGGGCATTTCATGTC TGGAATTGGAATGAGTACAATCTAAATCCCTTAACGAGG ATCCGTTGGAGGGCAAGTCTGGTGCCAGCAGCCGCGGT AATCCAGCTCCAATAGCGTATATTTAAGTTGTTGCAGTT AAAAGCTCGTAGTTGGATTTCCGGGTGGGTTCTAGCGGT CCGCTATGGTGAGTACTGCTATGGCCTTCTTTCTGTC GGGGACGGGCTTCTGGGCTTCACTGTCCGGGACTCGGA GTCGACGTGGTTACTTTGAGTAAATTAGAGTGTTCAAAGC AGGCTTACGCCAGAATACTTTAGCATGGAATAACACGAT AGGACTCTGGCCTATCTTGTGGTCTGTAGGACCGGAGT AATGA Similarity Rate: 99.75% was found.

According to the molecular identification studies, the results of the identification of three different microalgae species isolated from three different geographical regions are summarized in Table 3.

Table 3. Molecular species identification results according to geographical regions

Geographical Regions	Freshwater Source	Coding	Type
Mediterranean Region	Dim Stream Antalya	E1	<i>C. sorokiniana</i>
Black Sea Region	Tokat Erbaa village fountain	E2	<i>P. wilhelmii</i>
Marmara Region	Tunca River Edirne	E3	<i>T. obliquus</i>

Culturing of microorganisms

C. sorokiniana, *P. wilhelmii* and *T. obliquus* microalgae were incubated in 250 mL flasks containing 100 mL BG11 medium at $25\pm 2^\circ\text{C}$ under $48 \mu\text{mol}/\text{m}^2\text{s}$ (2400 lx) light for 7 days. The dry weight values X(g/L) of microalgae recorded during the 3rd and 7th days of incubation period and calculated based on optical density values at 600 nm are shown in Figure 3.

Bioenergy analysis of microalgae

Dry weight (X) (g/L), chlorophyll concentrations chl (a+b) ($\mu\text{g}/\text{mL}$), specific growth rates (μ) (1/d), maximum biomass productivity (Pmax) (g/Ld) and CO₂ biofixation rates (FCO₂) (mgCO₂/d) of *C. sorokiniana*, *P. wilhelmii* and *T. obliquus* microalgae are summarized in Table 4.

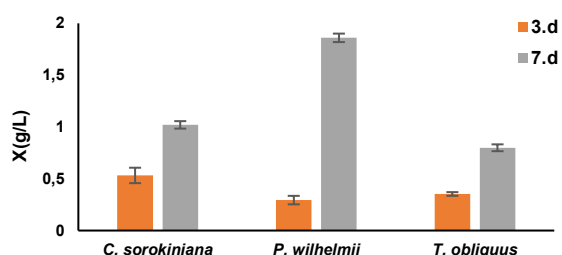


Figure 3. Dry weight values (X (g/L)) of *C. sorokiniana*, *P. wilhelmii* and *T. obliquus*

Table 4. Dry weight (X) (g/L), chlorophyll concentrations chl (a+b) ($\mu\text{g/mL}$), specific growth rates (μ) (1/d), maximum biomass productivity (P_{max}) (g/Ld) and CO_2 biofixation rates (FCO_2) (mgCO_2/d) of *C. sorokiniana*, *P. wilhelmii* and *T. obliquus* microalgae.

	<i>C. sorokiniana</i>	<i>P. wilhelmii</i>	<i>T. obliquus</i>
X (g/L)	1.02±0.036	1.86±0.041	0.80±0.033
Chl (a+b) ($\mu\text{g/mL}$)	0.146±0.0052	0.278±0.0061	0.181±0.0073
μ (1/d)	0.162±0.012	0.465±0.059	0.207±0.019
P_{max} (g/Ld)	0.122±0.063	0.392±0.030	0.111±0.059
FCO_2 (mgCO_2/d) (0.1 L)	0.022±0.0008	0.072±0.0052	0.020±0.0060

According to bioenergy calculations, among the three different geographical regions, *P. wilhelmii* microalga isolated from the village fountain of Tokat Erbaa in the Black Sea Region showed higher growth rate of 1.86 g/L, higher chl (a+b) value of 0.278 $\mu\text{g/mL}$, specific growth rate as 0.465 1/d and productivity as 0.392 g/Ld than the other two isolates under the same environmental conditions. *P. wilhelmii* microalgae was found to be capable of fixing more CO_2 than *C. sorokiniana* and *T. obliquus* microalgae with a value of 0.072 mgCO_2/d .

DISCUSSION

Due to their metabolism, microalgae take CO_2 from the atmosphere through photosynthesis, incorporate it into their structures and produce biomass. Isolation and cultivation of microalgae has been an important area for many years (Ozturk et al., 2019; Atıcı, 2020; Derakhshandeh et al., 2021). Produced biomass is also used in many different areas (Derakhshandeh et al., 2021). Microalgae are therefore a promising feedstock for applications in biofuel production and are recognized as valuable bioproducts. When microalgal fuels are compared to fuels produced from land plants, microalgae can produce 60 times more fuel in the same area. Also, the lack of terrestrial area requirements is an advantage (Skjanes et al., 2013).

In our study, *C. sorokiniana* isolated from water samples taken from Dim Stream in the Mediterranean Region is a very useful microalgae preferred for reducing CO_2 emissions and producing microalgal biomass commercially (Qin et al., 2023). It has also been used in aquatic toxicity assessment studies

caused by pollutants such as tetrabromobisphenol A and Cd (II) from e-waste (Liu et al., 2023). It is also known that this microalgae is a suitable species for recycling studies by obtaining lipids and bioethanol from its biomass when grown in the mixed peel extract of potato, banana and sweet lime (Malakar et al., 2023). In another study, *C. sorokiniana* was used for Cd (II) biomineralization from soil (Xia et al., 2023). It was also used to evaluate the ecotoxicity of some antibiotics on aquatic organisms (Li et al., 2023). *C. sorokiniana* is also considered as a highly efficient source for commercial lutein production (Vadrale et al., 2023). Besides *Chlorella* is also used in heavy metal and lipid extraction studies (Atıcı et al., 2008; Derakhshandeh et al., 2019). Additionally, its ability to grow in many different wastes makes it advantageous (Atıcı and Fidan, 2022).

P. wilhelmii isolated from water samples taken from Erbaa village fountain in Tokat, Black Sea Region is a member of the *Pseudochloris/Picochlorum* genus and has rapid growth rate, extensive range of nutrients and salinity tolerance. (Von Alvensleben et al., 2013; Budiša et al., 2019; Concas et al., 2019). Furthermore, *P. wilhelmii* is a promising microalgae for oil refinery wastewater treatment and high-value biomass production (Blazina et al., 2022).

T. obliquus isolated from water from the Edirne Tunca River in the Marmara Region has been used for lead and cadmium removal (Tafti et al., 2023), bioremediation, CO_2 removal and biofuel production (Selvan et al., 2023) and beta carotene production (Singh et al., 2019).

Among microalgae genera, the genus *Scenedesmus* is the third most studied genus in the world in terms of the number of published literature (Oliveira et al., 2021; Garrido-Cardenas et al., 2018). *Scenedesmus* is one of the most abundant microalgae in freshwater. Species of this genus have single-celled individuals that can form associations of 2 to 32 cells. Mostly, 4-celled ones are common (Oliveira et al., 2021).

T. obliquus has been successfully studied in wastewater treatment and the resulting biomass has been used in renewable energy studies (Oliveira et al., 2021). For example, in a study comparing *T. obliquus* with microalgae such as *C. vulgaris* (34%) and *Oocystis minuta* (27%) in removing sulfate from wastewater, it outperformed other algae by 36% (Ajala and Alexander, 2020). In another study, Ahmad et al. (2019) showed that *T. obliquus* can remove approximately 94% of phosphate from municipal wastewater.

Microalgae cells have also begun to be used in biosensor studies due to their sensitivity to environmental variables (Congur et al., 2022). In this context, the development of biosensors for the detection and monitoring of *T. obliquus* and organic molecules in water has attracted the attention of researchers (Oliveira et al., 2021). Gonzalez and Lorenzo (2019) evaluated the potential of detecting pesticides in water in the cathode they developed using *T. obliquus* cells. As a result, it was determined that *T. obliquus* showed excellent sensitivity and rapid response to environmental changes.

Third-generation biofuels obtained from microalgae, lignocellulosic raw materials, soybeans, corn and other fossil fuels and crops used in biofuel production can be shown as an alternative (Safi et al., 2014; Goh et al., 2019). It has been reported that *T. obliquus* can reach 37.92% ethanol conversion (El-Sheekh et al., 2014) and 90.81% biodiesel conversion (Guldhe et al., 2015) rates and has the potential to produce high amounts of lipids and carbohydrates.

T. obliquus is a microalgae that is also used successfully in the field of health. For example, polysaccharides of *T. obliquus* extracted under different environmental conditions showed antiviral activity against viruses such as *Herpes simplex virus*, *Hepatitis C virus*, *Rotavirus* and *Coxsackievirus* (Singab et al., 2018). Thanks to *T. obliquus* extract, a 40%, 30%, 10% and 40% reduction in *Hepatitis C virus*, *Rotavirus*, *Herpes simplex virus* and *Coxsackievirus* was shown, respectively. It has also been reported that *T. obliquus* extract can also inhibit the growth of 50.4% of human liver cancer cells under in vitro experiments.

CONCLUSION

Microalgae isolation, cultivation and investigation of the use of isolated microalgae in biotechnology studies are important issues in the field of sustainable energy. Within the scope of the study, while microalgae isolated from different

freshwater sources in Türkiye were introduced to the literature, bioenergy calculations of these new isolates were also made through kinetic methods. In conclusion, the isolated microalgae are the species that have the potential for rapid growth and biotechnological approaches in the literature. In this context, they have the potential to be used in future studies.

ACKNOWLEDGMENTS AND FUNDING

The study was supported under the TUBITAK 2209-A program with 1919B012105092 application number.

AUTHORSHIP CONTRIBUTIONS

Eyüp Polat took part in the experiments of the study. Burcu Ertit Taştan was involved in the coordination of experimental studies, interpretation of results, and manuscript writing.

CONFLICT OF INTEREST STATEMENT

The authors declare that they have no conflict of interest or competing interests

ETHICS APPROVAL

No specific ethical approval was necessary for this study.

DATA AVAILABILITY

All relevant data is in the article.

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