Algal Diversity in Biological Soil Crusts of Qom Province, Iran: Insights into Microflora Composition and Environmental Impact

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Abstract

Biological soil crusts (BSCs) result from the close association between soil particles and various microorganisms, including cyanobacteria, green microalgae, and free-living fungi. In this study, we investigated the BSC microflora in soil samples collected from four sites in Qom Province, a semi-arid region in Iran. The study stations included agricultural areas, farms irrigated with saline water, and the edge of Hoz-e-Soltan Lake, a saline water lake. To study algal communities, all soil samples were cultivated in four different culture media including BG11, BG11 with 1.5 M NaCl, nitrate-free BG11 medium (BG110), and BG110 with 1.5 M NaCl. We isolated and identified 37 morphospecies of algae. Cyanobacteria, such as *Phormidium* and *Pseudanabaena*, were dominant genera at most stations. Additionally, green algae and diatoms were present in the studied sites. Our results revealed that algal diversity in BSCs depends on soil physio-chemical characteristics of the soil. For example, *Phormidium* was the dominant genus in all agricultural soils, while in saline soils, the genus *Dunaliella* was reported as the dominant taxon. Therefore, salinity plays a crucial role in shaping species distribution and diversity in these ecosystems.

Keywords: Biological soil crusts, Biodiversity, Salinity, Arid land, *Phormidium*, Qom Province

Introduction

Biological soil crusts (BSCs), which develop in the upper few centimeters of the soil, are a diverse community of photoautotrophic and heterotrophic organisms (García-Carmona et al., 2023). These BSCs can be considered as the result of a close association between soil constituent particles and various organisms, including cyanobacteria, microalgae, micro-fungi, lichens, and bryophytes. The biological soil crust and its various components have the ability to greatly impact the physical and

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chemical properties of soil. In addition, BSCs play a vital role in maintaining soil stability and exert a notable impact on multiple soil factors, particularly in dryland regions (Eldridge et al., 2020). Microorganisms within BSCs are key factors in binding soil particles together, forming a living layer on the ground surface through their presence, interactions, and biological activities (Belnap et al., 2001). Thus, changes in this crucial soil layer can profoundly affect overall environmental health and may contribute to desertification (Nguyen et al., 2022).

Previous studies have demonstrated that some microorganisms found in the soil's upper layer exhibit remarkable resistance to harsh environmental conditions such as dryness, salinity, and high temperature (Li et al. 2021). These groups of microorganisms, called extremophiles, have developed unique adaptations to survive extreme environmental conditions. Among these organisms, extremophilic microalgae play a significant role in soil stabilization and hold potential for applications in biotechnological industries (Varshney et al., 2015).

Cyanobacteria and green algae are pivotal in the formation of soil biological communities among the diverse group of microalgae. Cyanobacteria, as pioneer group of photosynthetic microorganisms, are found worldwide in various environments, including freshwater, saltwater, and arid regions (Rahim et al., 2023; Willis and Woodhouse, 2020). Furthermore, their versatility allows them to adapt to a wide range of conditions (Rahim et al., 2023). Cyanobacteria cells are typically protected by a polysaccharide sheath (EPS), which effectively safeguards them from damage that prevent desiccation. This sheath possesses a great ability to store water and prevent the cells from drying out (Malam Issa et al., 2007). Therefore, EPS plays an important role in connecting soil particles, promoting the cohesion of biocrust inhabitants, and acting as a water-holding biomolecule under drought (Sommer et al. 2020).

The study of microalgae in BSCs, particularly in arid lands, has consistently been a notable and interesting field within algal and environmental research over the past few decades (Chamizo et al., 2016; Miralles et al., 2020; Li et al., 2021). BSCs improve soil stability, regulate soil moisture, and contribute to carbon and nitrogen cycles in arid environments (Li et al., 2021). They impact soil properties such as organic carbon (OC) and pH through their physiological activities (Miralles et al., 2020). Additionally, BSCs enhance soil quality, promote vegetation recovery in semi-arid regions (Gao et al., 2016), and reduce water evaporation from the soil (Chamizo et al., 2016).

Extreme environments, such as drylands, are characterized by severe conditions associated with non-optimal values of various physical and geochemical factors, including temperature, radiation, salinity, and pH (Rothschild & Mancinelli, 2001). Although these areas were once believed to be lifeless, they are now recognized as rich ecosystems supporting highly adapted species (Malavasi et al., 2020). Extremophile microalgae have been the focus of numerous studies worldwide due to their economic advantages (Varshney et al. 2015; Malavasi et al., 2020). Moreover, researchers in Iran have explored extreme habitats extensively, including saline lakes, hot springs, and areas with high radiation backgrounds (Safarpour et al., 2018; Heidari et al., 2020; Salehipour-Bavarsad et al., 2021). Several studies have been carried out on the algal flora of terrestrial ecosystems in Iran; however, these reports are considered insufficient, particularly in high-stress areas. In 2009, Moghtaderi et al. conducted a study on cyanobacteria in the soil desert crust of Chadormalu in the Bafgh region (Yazd province, Iran). Hokmollahi et al. (2016) also described the cyanobacteria found in the soils of Yazd province, Iran, using morphological, molecular, and physiological characteristics. Additionally, in recent years, there have been reports on the morphological and molecular studies conducted on the algal flora of Kavir National Park (Etemadi-Khah et al., 2017). In another study, Irankhahi et al. (2022) studied the diversity and distribution of heterocystous cyanobacteria across solar radiation gradient in terrestrial habitats of Iran. The findings indicate that ecological factors, including solar radiation, relative humidity, and soil salinity, have an impact on the diversity and distribution of cyanobacteria in terrestrial ecosystems. Nostoc was reported to be the prevailing genus across all stations among the identified taxa, particularly in areas with elevated solar radiation, such as Qom Province.

Recent research has explored microalgae inhabiting terrestrial habitats in Qom Province. Studies have documented nitrogen-fixing cyanobacteria such as different species of *Nostoc* (Davari et al. 2018; Irankhahi et al., 2022.), and *Trichoromus* (Davari et al. 2018). In addition, some species of *Oscillatoria* as non-nitrogen-fixing cyanobacteria was reported from this region (Davari et al. 2018). Furthermore, halophile taxa such as *Dunaliella* species have been identified in the arid lands of this province (Salehipour-Bavarsad et al., 2022).

In this study, we identified extremophilic microalgae that form BSCs in semi-arid and saline soils. The study also aims to assess the biodiversity of microalgae at the sampling sites, and determine their distribution based on the prevailing environmental conditions.

Material and methods

Study area and sampling sites

Qom Province, covering an area of 11238000 square kilometers, is located in the central plateau of Iran (Rahmati Zadeh et al., 2014). This Province is in proximity to Iran's central desert, and boasts several salt lakes at its entrance, including the Salt Lake and the Hoz Sultan Lake (Ebrahimivand et al., 2023). This province is affected by the natural factors that dominate the desert areas. Studies show that over 70% of the province consists of desert and semi-desert areas, with approximately 91% of the region experiencing an ultra-arid and dry desert climate (Rahmati Zadeh et al., 2014). This province is affected by the natural factors that dominate the desert areas. Studies show that over 70% of the province consists of desert and semi-desert areas, with approximately 91% of the region experiencing an ultra-arid and dry desert climate (Rahmati Zadeh et al., 2014), and these conditions have made many areas of this province unsuitable for agricultural activities.

In August 2023, soil samples were collected from four distinct locations within Qom Province, Iran, following the methodology outlined by Rangaswamy (1996). The first sampling site was an operational agricultural plot cultivating *Medicago sativa* (alfalfa). The next two sampling sites were barren lands that were previously used as farmlands, but were irrigated with highly saline water for many years and became unsuitable for agricultural purposes (Fig. 2). The fourth site was the dried lakebed of Hoz-e-Soltan Lake, and soil samples were taken from the playa area. In addition to the soil samples, various vegetation samples were also collected from this site for further examination to explore potential coexistence patterns (Figure 1, Table 1).

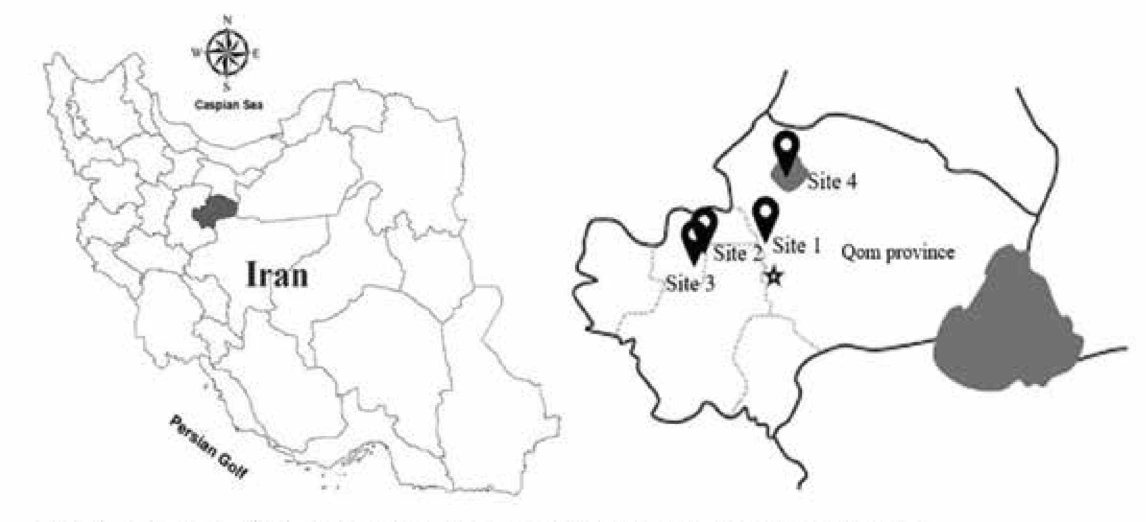

Fig. 1. Location of Qom Province, the area which the samples were collected.

Table 1. Geographical details of the sampling sites and some physiochemical parameters recorded at each site

No.	Location	Latitude Longitude	pH	EC (dSm^{-1})	Total Nitrogen (%)	Total Organic Carbon (%)
1	Qom Province; Central part, Medicago Farmland	34°79'29.19"N 50°85'39.24"E	8.08	22.8	0.09	0.71
2	Qom Province; Mahmoud-Abad	34°80'87.85"N 50°61'22.52"E	7.74	43.1	0.20	2.20
3	Qom Province; Pachian	34°80'04.56"N 50°58'31.22"E	7.67	30.4	0.18	1.67
4	Qom Province; Hoz-e-Soltan Lake	34°96'60.99"N 50°89'73.75"E	7.73	>160	0.08	0.58

Isolation and cultivation of algae

Approximately four grams of each soil sample were individually cultivated in glass containers using four different culture media: BG11, BG11 supplemented with a 1.5 M NaCl concentration (BG11: 1.5 M), nitrate-free BG11 medium (BG110), and BG110 supplemented with a 1.5 M NaCl concentration (BG110: 1.5 M). Employing diverse algal culture conditions can provides deeper insights into the biodiversity of algae in the region and their environmental requirements. BG11 serves as a common culture medium, facilitating the growth of a diverse group of algal species. Cultivating soil samples in BG110 assists in the identifying taxa that exhibit resistance to nitrogen deficiency, particularly in favor of heterocystous cyanobacteria. BG11: 1.5 M enables the identification of algal species thriving in saline conditions. Lastly, the essential role of BG110: 1.5 M lies in distinguishing algae that are resilient to both nitrogen deficiency and saline conditions. Anderson's methodology (2005) was used to isolate species capable of thriving in saline environments. The cool white fluorescent lamps emitting about 74 µmol photons m^{-2} s^{-1} were used to provide a photoperiod consisting of 16 hours of light and 8 hours of darkness for the algal cultures, which were kept under controlled conditions at a temperature of 25 ± 2°C. After approximately two weeks, the presence of algal colonies

Identification of microalgal taxa

To identify different algal taxa, a morphometric study was conducted using light microscopy (Olympus, BH-2 model). Semipermanent slides of the colonies were prepared for identification of taxa. Various references were used to identify algae, including Prescott (1970) for identify green algae species; Komárek and Anagnostidis (1986, 2005) and Komárek (2014) for identification of cyanobacteria; and Eileen J. Cox (1996) for identification of diatoms.

Various morphological characteristics, including color, shape, size of colonies and thallus, presence or absence of mucilaginous sheath, and the shape of apical cells, vegetative cells, and special cells in filamentous taxa, were utilized for identification.

Identification of vegetations

The book Flora Iranica (Rechinger, 2005) was used to identify the collected vegetations.

Soil analysis

Various techniques were utilized to measure physicochemical properties of the soil samples including concentration of organic carbon, nitrogen, pH, and electrical conductivity (EC). The soil samples were directly measured for pH using water extracts (1:5 w/v) by the electrometric method. EC was measured using platinum electrodes. Furthermore, the nitrogen concentration was measured using the Macro Kjeldahl (Juo, 1978), and the organic carbon concentration was determined using the Walkley-Black method (Sikora and Moore, 2014).

Statistical analysis

To investigate the linear relationship between two variables, i.e. EC as a soil physicochemical parameter and the number of algae identified from collected soil crusts, Pearson's correlation coefficient was calculated using IBM SPSS Statistics (version 26). In addition, Microsoft Excel 2019 was used to draw the graphs.

Results

Climate and vegetation of the study area

This study focused on investigating algal communities in BSCs of Qom province, particularly the uncultivable

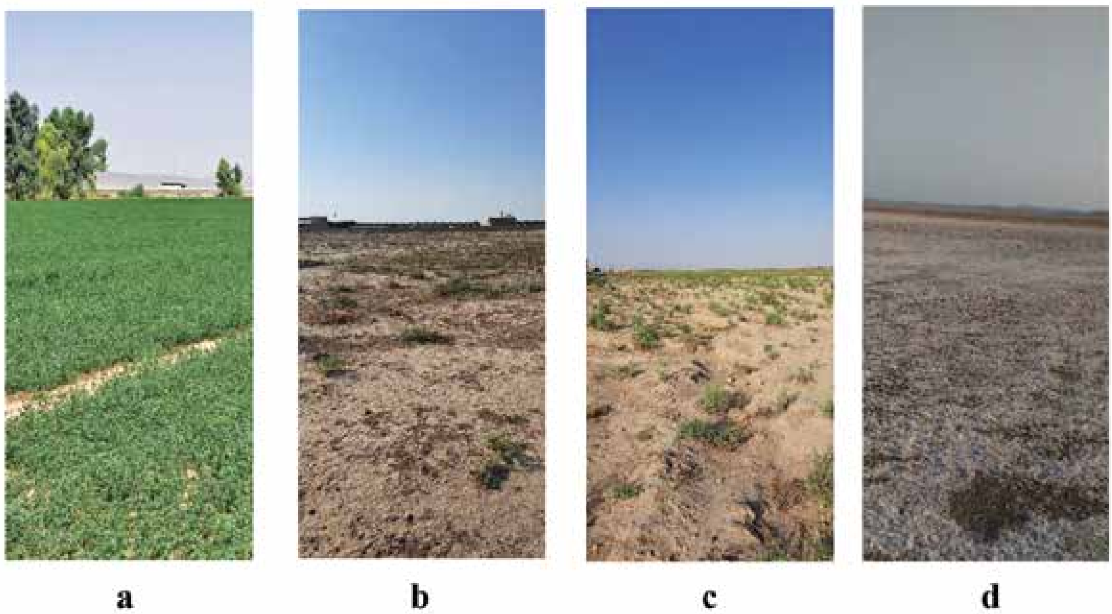

Fig. 2. Pictures of sampling sites: (a) Site 1. agricultural land, (b) Site 2. inactive farmland, (c) Site 3. uncultivated land, (d) Site 4. Hoz-e-Soltan

areas which are irrigated with salt water. Qom Province encounters a semi-arid climate, characterized by very hot summers and extremely cold winters. The average annual precipitation in Qom province is 151.6 mm, with the highest average monthly and annual precipitation recorded at 49.6 mm and 7.99 mm, respectively. The annual mean temperatures of 19.89 °C, with the highest mean monthly temperature reaching 39.5 °C, and the lowest mean monthly temperature dropping to -1°C. The vegetation in the studied sites is described as follows: in Site 1, cultivated *Medicago sativa* L. was predominant vegetation, the main vegetation of site 2 was *Alhagi maurorum* Medik and *Prosopis farcta* (Banks & Sol.) J. F. Macbr, while Site 3 was predominantly covered by *Alhagi*. Moreover, Site 4 was a barren land without any vegetation coverage (Fig 2).

Physicochemical parameters of soil

The physicochemical properties of collected soil samples were measured and represents in Table 1. According to the Table, it can be perceived that the pH of almost all of the samples fell within the normal range of 6.5-8. However, the treated soil in Site 1 exhibited slightly alkaline properties. In addition, all collected soil samples showed high salinity levels based on their EC, which is an indicator of salt concentrations and soil mineralization (Fig 3). The EC

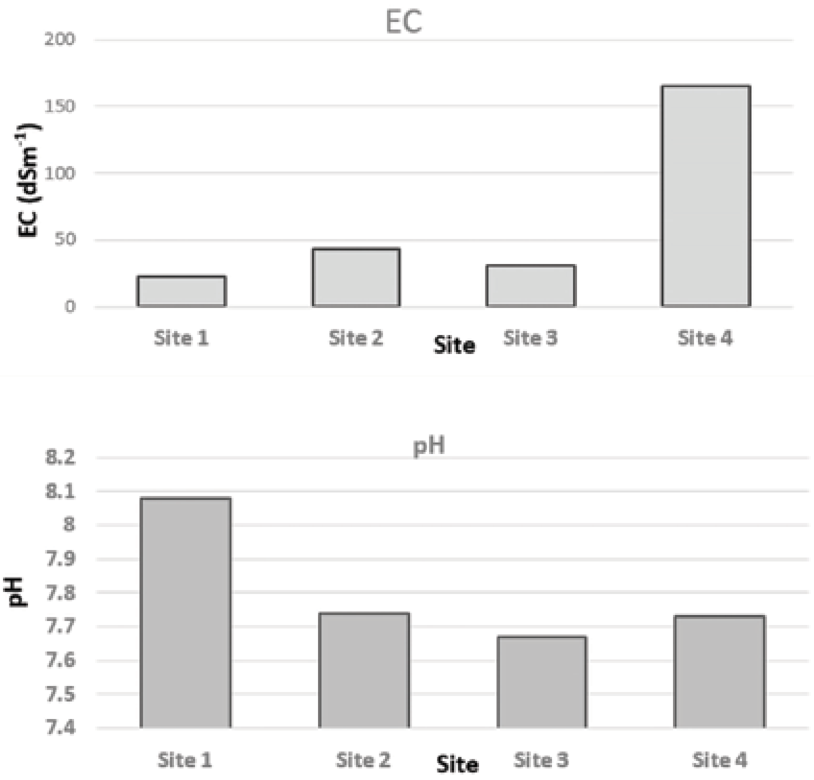

Fig. 3. Comparison of electrical conductivity (EC) and pH levels in sampling sites

values for all soil samples exceeded 4 dS/m. The lowest amount of EC was observed in Site 1 at 22.8 dS/m, while the highest EC value was recorded in Sites 2 and 4 with 43.1 and 160 dS/m. Furthermore, the total nitrogen and total organic carbon (TOC) of soil samples were measured, and the range of these parameters was about 0.08 – 0.20 % for total nitrogen, and 0.58-2.20% for TOC, respectively. The lowest amount of nitrogen and TOC were recorded in Site 4 (0.08%) for nitrogen and 0.58% for TOC), and the highest amount was seen in Site 2 with 0.20 percentage of total nitrogen content and 2.20 percentage in TOC (Fig 4).

Algal diversity

In the present study, a total of 37 microalgal taxa were identified (Table 2, Fig. 5). These taxa included 33 cyanobacterial taxa, three taxa from Chlorophyta, and one diatom species. It should be noted that the genus *Phormidium* appeared most frequently and species diversity among the identified taxa, with a presence rate of 43% (Fig 6). *Pseudanabaena* ranked second with seven species. Further, two species of the genus *Planktolyngbya* were observed among identified taxa. Additionally, some genera were present with only one species among the identified taxa including *Jaaginema pseudogeminatum (G.Schmid)* Anagnostidis and Komárek, *Kamptonema animale* (Gomont) Strunecký, Komárek and J. Smarda, Leptolyngbya foveolarum (Gomont) Anagnostidis and Komárek, *Oscillatoria rupicola* (Hansgirg) Hansgirg ex Forti, *Potamolinea aerugineocaerulea* (Gomont) M.D.Martins and L.H.Z.Branco, *Schizothrix lenormandiana* Gomont, and *Stenomitos frigidus* (F.E.Fritsch) Miscoe and J.R.Johansen. Despite the variety of cyanobacteria taxa, the only nitrogen-fixing cyanobacteria identified in this study was *Nodularia harveyana* Thuret ex Bornet and

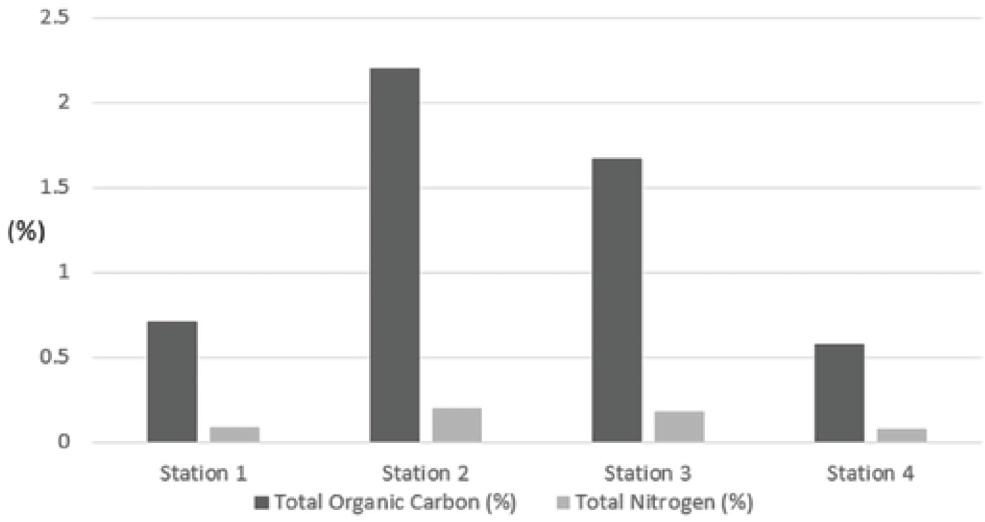

Fig. 4. Comparison of total organic carbon and total nitrogen in sampling sites

Flahault.

Site 1 showed the greatest number of *Phormidium* species totaling nine species. Additionally, Sites 2 and 3 recorded five and six *Phormidium* species, respectively. Conversely, in the final site, there were not any *Phormidium* species (Table 2).

Additionally, three genera of green microalgae were observed from one class, one order, and three families. These microalgae included *Dunaliella* Teodoresco, *Limnomonas* Tesson & Pröschold, and several *Chlorococcum* species.

Accurately classifying different species of *Chlorococcum* genus presents a challenge, thus requiring the use of molecular investigations. Moreover, *Hantzschia* sp. was the only diatom identified in this study; the presence of this species was notably observed in areas irrigated with saline water, particularly at Sites 2 and 3.

In the present study, BSCs samples collected from agricultural sites showed the highest algal diversity. This phenomenon could be attributed to agricultural practices and the utilization of varied water sources for irrigation over time. In Sites 1, 2, and 3, the number of observed microalgae genera was 20, 16 and 18, respectively. However, in the playa collected from Site 4, only two species were identified, including *Dunaliella* sp. and *Pseudanabaena minima* (G.S.An) Anagnostidis.

Data analysis

The correlation between soil properties and the number of algal species in each site was calculated using the Pearson correlation coefficient. The findings indicate a significant correlation between EC and the number of algal species at the 0.01 level. However, no significant correlation was observed between pH level and the number of algae, considering that the range of the mentioned parameter fell between 7.67 and 8.08.

Discussion

The diversity of microalgae within BSCs is intricately influenced by several environmental factors, encompassing both natural processes and human-induced activities across diverse temporal and spatial scales (Büdel et al., 2016; Hakkoum et al., 2021). Among these factors, soil physicochemical properties are decisive determinants that shape the composition and distribution of microalgal communities within terrestrial ecosystems (Hakkoum et al., 2021). Accordingly, our findings suggest that the relationship between ecological parameters and algal diversity is notably complex; nonetheless, certain generalizations can be inferred regarding pH and EC levels.

Cyanobacteria exhibit a remarkable capacity to thrive in extreme environments, including habitats

Table 2. Microalgal diversity in biological soil crusts at different sites of Qom Province

Taxon	Site 1	Site 2	Site 3	Site 4
Bacillariophyta				
Hantzschia sp.		●	●	
Chlorophyta				
Chlorococcum spp.	●	●	●	
Dunaliella sp.		●	●	●
Limnomonas sp.		●		
Cyanophyta				
Jaaginema pseudogeminatum	●		●	
Kamptonema animale		●		
Leptolyngbya foveolarum	●		●	
Nodularia harveyana	●		●	
Oscillatoria rupicola		●	●	
Phormidium breve	●	●	●	
Phormidium caerulescens			●	
Phormidium fragmentosum	●			
Phormidium grunowianum	●			
Phormidium hamelii	●			
Phormidium holdenii	●			
Phormidium incrustatum	●			
Phormidium interruptum		●		
Phormidium kuetzingianum	●			
Phormidium lusitanicum	●	●		
Phormidium nigrum			●	
Phormidium sp.$_1$	●			
Phormidium sp.$_2$		●		
Phormidium sp.$_3$			●	
Phormidium sp.$_4$			●	
Phormidium uncinatum		●	●	
Planktolyngbya holsatica		●		
Planktolyngbya limnetica		●		
Potamolinea aerugineocaerulea		●	●	
Pseudanabaena balatonica	●			
Pseudanabaena minima	●	●	●	●
Pseudanabaena sp.$_1$	●			
Pseudanabaena sp.$_2$	●			
Pseudanabaena sp.$_3$			●	
Pseudanabaena sp.$_4$			●	
Pseudanabaena starmachii	●	●		
Schizothrix lenormandiana	●			
Stenomitos frigidus	●		●	

Site 1 (agricultural land), Site 2 (inactive farmland), Site 3 (uncultivated land), Site 4 (Hoz-e-Soltan)

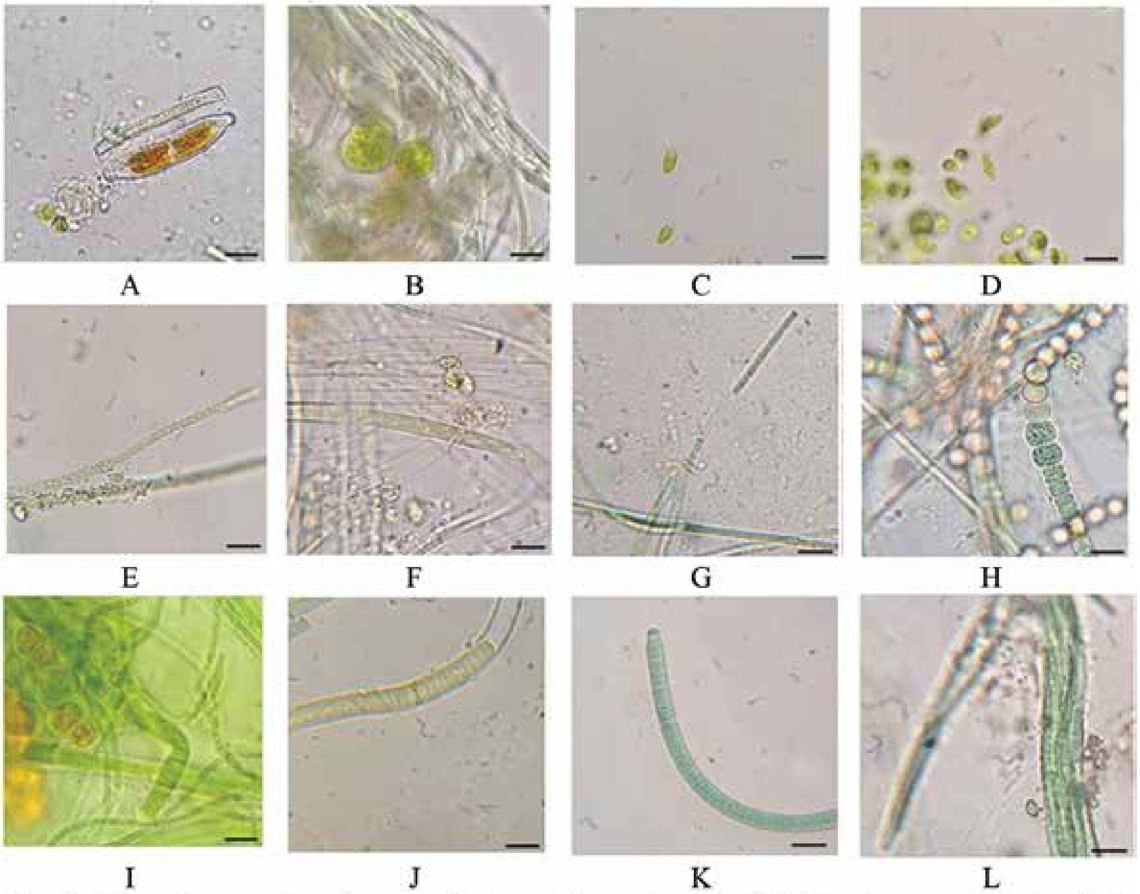

Fig. 5. Microphotographs of some of the algal species identified in the present study: (A) *Hantzschia* sp., (B) *Chlorococcum* sp., (C) *Dunaliella* sp., (D) *Limnomonas* sp., (E) *Jaaginema pseudogeminatum* (F) *Kamptonema animale* (G) *Leptolyngbya foveolarum* (H) *Nodularia harveyana* (I) *Oscillatoria rupicola* (J) *Phormidium interruptum* (K) *Phormidium uncinatum* (L) *Schizothrix lenormandiana* (Bars = 10 μm).

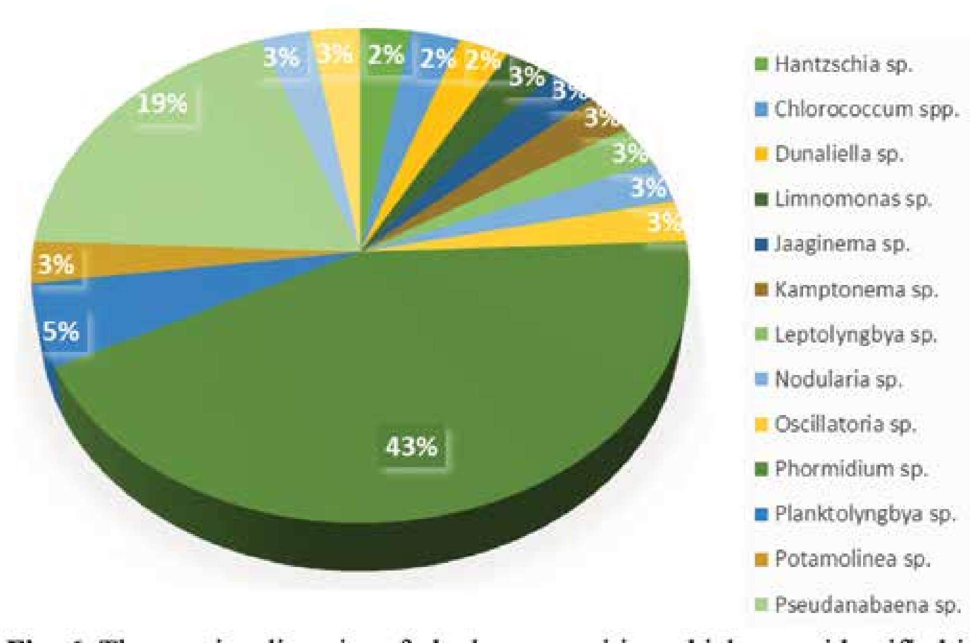

Fig. 6. The species diversity of algal communities which were identified in the studied area in Qom Province

with high radiation, low water availability, and elevated temperatures - characteristics that set them apart from many other biological species. Their autotrophic nature and ability to maintain high metabolic functions with limited moisture requirements enhance their capacity to colonize a diverse range of ecosystems, particularly arid environments (Perera et al., 2018). Moreover, not only do the mentioned features help these microorganisms to survive in saline environments, but also they use mechanisms to survive in ionic stress (Belnap, 2003). Additionally, some cyanobacteria species show specific alternations in their morphological and physiological features while adapting to harsh conditions (Perera et al., 2018).

Site 1 (cultivated land) emerged as the area with the highest algal diversity among the sampling sites. It displayed an elevated pH level (8.08) and lower EC value (22.8) in comparison to the other sites, indicating distinctive environmental conditions that likely exert an influence on the microalgal community structure in that specific area. In addition, in alignment with the observations of Kirkwood and Henley (2006), our study highlights a significant relationship between salinity levels and the diversity of algal species. Higher salinity concentrations (indicated here by high EC values) were associated with a reduced presence of algal species, revealing that excessive salinity could potentially restrict the diversity of microalgae in soil ecosystems. This emphasizes the direct influence of salinity on the overall diversity of microalgae in these environments. Therefore, the research findings enrich our understanding of how environmental factors shape microalgal diversity in soil ecosystems and emphasize the critical role of soil physicochemical properties in driving these ecological dynamics.

As stated, Site 1 is a cultivated area for *Medicago sativa* with the highest algal diversity. However, it is worth noting that *Dunaliella*, as a halotolerant microalga, is not present in this location. The absence of *Dunaliella* in Site 1, and its presence in those three uncultivated sites indicates unfavorable conditions of the other three stations for plant cultivation, as the studied strains of *Dunaliella* showed robust growth in BG11:1.5 M NaCl. Soil salinity is a critical factor that limits plant productivity and quality. Various growth parameters and yield components decline under salinity stress across numerous plant species (Shahbaz and Ashraf, 2013; Ansari et al., 2019). Salinity stress exerts negative effects on photosynthesis, gas exchange, and overall plant growth, attributed to osmotic and ionic stress (Chaves et al., 2009; Ansari at al., 2019). Similar to many other plants, the yield of *M. sativa* decreases at salinity levels exceeding 2 dS m^{-1}, although it is considered moderately saline-tolerant among leguminous crops (Maas and Hoffman, 1977). *M. sativa*, a perennial plant extensively cultivated as forage in large irrigated areas worldwide, holds significant economic and agronomic value due to its high-quality forage and nitrogen-fixing capabilities (Anower et al., 2013).

The presence and wide distribution of *Phormidium* species in collected BSCs suggest that this genus plays a significant role in agricultural soils (Alghanmi et al., 2019). Research has shown that certain *Phormidium* species, such *as P. breve*, exhibit tolerance to salinity, as demonstrated by Tashlykova and Afonina (2022). Similarly, Sepehr et al. (2019) have reported that *P. uncinatum* is capable of surviving and adapting to saline conditions. Other similar reports also emphasize the resistance of this genus to salinity and their presence in harsh environments (Etemadi-Khah et al., 2017; Zafar et al., 2022). Overall, the presence of *Phormidium* in agricultural soils can be beneficial for soil fertility, plant growth, and potentially for disease control (Rezaee et al., 2019; Younesi et al., 2019; Neyshabouri et al., 2021; Righini et al., 2022). There are also reports on the efficient role of this genus in desalination processes (Zafar et al., 2022).

Our findings also show that *Pseudanabaena minima* was the dominant species among the other taxa, suggesting its potential as a saline-resistant species. Notably, previous research by Ahlesaadat et al. in 2017 also showed the presence of *P. minima* across a range of EC levels in agricultural fields. It should be noted that there is limited information on *Pseudanabaena* spp. in the scientific literature, including their physiology, molecular, and metabolic characteristics (Foster et al., 2020). Although these cyanobacteria are associated with blooming events in various environments and are widely present in aquatic and terrestrial habitats, this genus is frequently disregarded. Interestingly, one of the reports shed light on bioremediation potential of the *Pseudanabaena galeata* strain. Ouhsassi et al. (2020) demonstrated that these filamentous cyanobacteria may effectively remove contaminants from industrial effluents during wastewater treatment processes.

While there is currently no documented evidence of *Oscillatoria rupicola* existing in saline water or being able to tolerate saline environments, we have observed its presence in soils with high salinity levels at Sites 2 and 3. The finding indicates that this species of *Oscillatoria* survives in saline environments. Plus, the genus *Planktolyngbya* which was reported from Site 2 showed a high salinity tolerance compared to other taxa isolated from agricultural fields. This resilience aligns with findings by Andreote et al. in 2014, who also noted the salinity tolerance of this genus.

Furthermore, *Leptolyngbya* is known as a halophile genus (Verma and Bhattacharjee, 2015), which serves as supporting evidence for the presence of its species in our research. In addition to the widespread observation of *L. foveolarum* in Sites 1 and 3, it was also found to be the predominant species cultured in BG11 medium with a 1.5 M saline concentration.

Previous studies have reported *Kamptonema* as a genus with saline tolerance (Hokmolahi, et al., 2017). In our research, one species of this genus was identified, with *Kamptonema animale* being found in the highest salinity soil among the agricultural fields.

Despite the presence of nitrogen-fixing species *Nodularia harveyana* in sites 1 and 3, the nitrogen content in the first site was only 0.09, which was one of the lowest among the soil samples studied; this can be the result for the method used in the tillage system (Szostek et al., 2022). Based on the findings of Samylina et al. (2024), *N. harveyana*, when observed in saline soils, i.e., Site 3 in the current investigation, has exhibited the capacity to withstand certain degrees of salinity. Consequently, it can be regarded as a species possessing a degree of salt tolerance.

Moreover, *Hantzschia* species exhibit a broad distribution encompassing both freshwater and marine environments, as well as terrestrial habitats (Joh, 2014). Notably, in the present study, *Hantzschia* was the sole diatom observed in saline agricultural soils at Sites 2 and 3. Given that these regions had experienced past associations with saline water, it can be inferred that this particular diatom has persisted since those earlier periods.

Similar to the mentioned genus, *Dunaliella* was also transferred to the agricultural fields while they were aggregating with saline water (Sites 2 and 3); more over this microalga was the dominant species in the soil samples collected from Site 4. Our findings were also proved by other studies representing the presence of *Dunaliella* species in saline soils (Salehipour -Bavarsad et al., 2022).

Even though the higher number of microorganisms results in higher amounts of TOC in soils (Roncero-Ramos et al., 2020; Hakkoum et al., 2021), it has been proved that different method of tillage systems in agriculture has effects on the TOC of the soils (Szostek et al., 2022). Therefore, the low amounts of TOC in the BSCs collected from site 1 can be the result of the tillage system before planting. Thus, compared to site 4 with the weakest BSC, sites 2 and 3 have more TOC concentrations; so that we can argue the BSCs in site 4 are the weak soil crusts, based on the amounts of TOC, EC, and the fewer number of algal species in the soil. Moreover, it can be mentioned that the high amounts of organic carbon in sites 2 and 3 compared with site 1, is the result of being unused through past years.

The comparison of four different BSCs that were affected by natural stress factors such as salinity and drought, as well as human-made factors like tillage methods for preparing agricultural lands and irrigating soil with high saline water, showed us that these factors have a noteworthy effect on the biological soil crusts; also, previous research by Lan et al. (2010) showed the harmful effects of salinity and dry conditions on BSCs.

In conclusion, this study has identified numerous algae species that are tolerant to high salinity levels. However, further research is needed to explore the salinity resistance and other attributes of these microorganisms in order to determine their potential for restoring biocrusts in arid and saline environments.

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