



Impact of ecological factors on biocrust performance in the stability of loess soils in the Incheh Buron region

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ABSTRACT

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Biocrusts are used as a protective cover to stabilize the soil in dry areas, where the soil is more affected by erosion. In this research, various characteristics of the soil under the biological and physical crust were investigated as indicators of soil performance in the the Incheh Buron region in the study area, from the subsoils of biological (moss, lichen and cyanobacteria) and physical crusts at the depth 0-2 cm were sampled Then, the influence of biological and physical shells on soil properties was analyzed with three replications. A one-way analysis was considered to discover significant contrast among treatments. Compare mean was done with using Duncan's multiple range test. Notable distinction was reported at $p < 0.05$ between biocrust and physical crust. Carbon, nitrogen and phosphorus of microbial biomass, soil-based respiration, substrate-induced respiration, microbial metabolic quotient, mineralization quotient and other characteristics that have a direct impact on the functioning of the ecosystem (soil organic carbon, soil organic matter, mean weight diameter, geometric mean diameter, wind erosion soil stability) were higher in soil under biocrusts compared with the soil beneath the physical crust. Results showed that biocrusts increase the percentage of nutrients, structure and physical characteristics of soil. The biocrusts improved basal soil respiration, substrate-induced respiration, carbon, nitrogen and phosphorus of microbial biomass contents. Biocrusts improve the soil's biological characteristics, and have a crucial function on increasing the stability and resistance of the soil to erosion, which is confirmed by the results of the soil grain stability indicators.

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1. Introduction

A loess is a sedimentary particle, mainly silt-sized, which is composed with dust accumulation caused by the wind (Vasiljevic *et al.*, 2011). Loess areas are one of the sensitive regions to erosion in the world (Wang *et al.*, 2017; Li *et al.*, 2017a; Best, 2019). Therefore, Stabilization of these soils is very important in order to increase their stability from environmental and ecological aspects. Some of these areas are enveloped with different types of biological shells. (Concostrina-Zubiri *et al.*, 2021). Biocrusts have a key effect on maintaining function as well as preventing environmental hazards (Concostrina-Zubiri *et al.*, 2021). Especially, different sequences of biological crusts are increasing the fertility of dry lands. (Liu *et al.*, 2018). Therefore, they are effective in maintaining and increasing the efficiency of the soil due to their biological productivity (Bastida *et al.*, 2014). Indicators of soil performance in soil include biological, physical and chemical characteristics (Wang *et al.*, 2017; Delgado-Baquerizo *et al.*, 2016). Moreover, Biocrusts have a crucial function on enriching the natural environment, which increases the component and diversity of microbial population of soil (Lucas-Borja *et al.*, 2012; Atashpaz *et al.*, 2023).

The investigations conducted in the field of biological crusts have shown that biological soil crusts carry out numerous actions in the environment (Martínez *et al.*, 2022). These shells have an effective character on various actions including breathing of soil (Miralles *et al.*, 2018), improving nitrogen and carbon fixation and soil fertility (Muñoz-Rojas., 2018), increasing organic matter, increasing the adhesion of soil particles which cause the formation of coarser, more stable and erosion-resistant soil grains (Roncero-Ramos *et al.*, 2020), moreover improving the cation exchange capacity (CEC) and soil moisture (Roncero-Ramos *et al.*, 2019).

Temperature and rainfall are among the environmental elements that are important, changeable, and have impact on biological activity, the microbial population of soil and soil biological and chemical producers (Ladwig *et al.*, 2015). Therefore, in areas with low rainfall and higher temperature, the soils have generally lower fertilization and are susceptible to water and wind erosion (Cui *et al.*, 2021). The presence of biocrusts in these regions, especially loess soils that are highly susceptible to wind erosion effectively increases soil quality and stability, and reduces the environmental mucus caused by the erosion of these soils by improving the different characteristics of the soil (Dacal *et al.*, 2020). The biological soil crusts, fungi and actinomycetes have the greatest impact on soil stability (Duchicela *et al.*, 2013). The filaments of fungal hyphae in the lower part of lichen connect soil particles to each other and construct stable soil grains (Eldridge & Greene, 1994; Dou *et al.*, 2023). Basal soil respiration is more related to microbial activity, indicating mineralization of the organic matter of soil with microorganisms in the low productivity lands. Basal soil respiration, qCO_2 (microbial metabolic quotient) as well as enzymatic pursuit is an indication of microbial function, that is a susceptible factor to changes in the quality soil characteristics in reaction to ecosystem modifications. (Baldauf *et al.*, 2023). Since there are few reports on the effects of the procedure different sequences of biocrusts on ecosystem sustainability through improved soil quality, this research was done in order to explain the biological, physical and chemical characteristics of bio-shells in of soil. Therefore, this study was conducted with the aim of investigating the effect of biocrusts on various physicochemical and biological characteristics of the studied area in order to improve the quality of the soil and increase its stability against the environmental risks caused by erosion.

2. Materials and Methods

2.1. Study area

Incheh Borun located at the north of Golestan province with a latitude of 37° 28" N and a longitude of 54°42" E and an elevation of -10 m below sea level (Fig. 1). The studied area is in

the Ag Qala basin. This region has different series of biocrusts including physical crust, cyanobacteria, lichen and moss placed on the loess plateau (Atashpaz *et al.*, 2023). Incheh Borun region has a midpoint temperature and precipitation of 19.2 °C, 253 mm respectively. This area has a thermal humidity regime and an arid thermal regime. The parent material of the study area is mainly loess sediments and geomorphological units in the loess plateau of Golestan province also includes lowlands, loess hills, terraces, and alluvial plains (Rahimzadeh *et al.*, 2019).

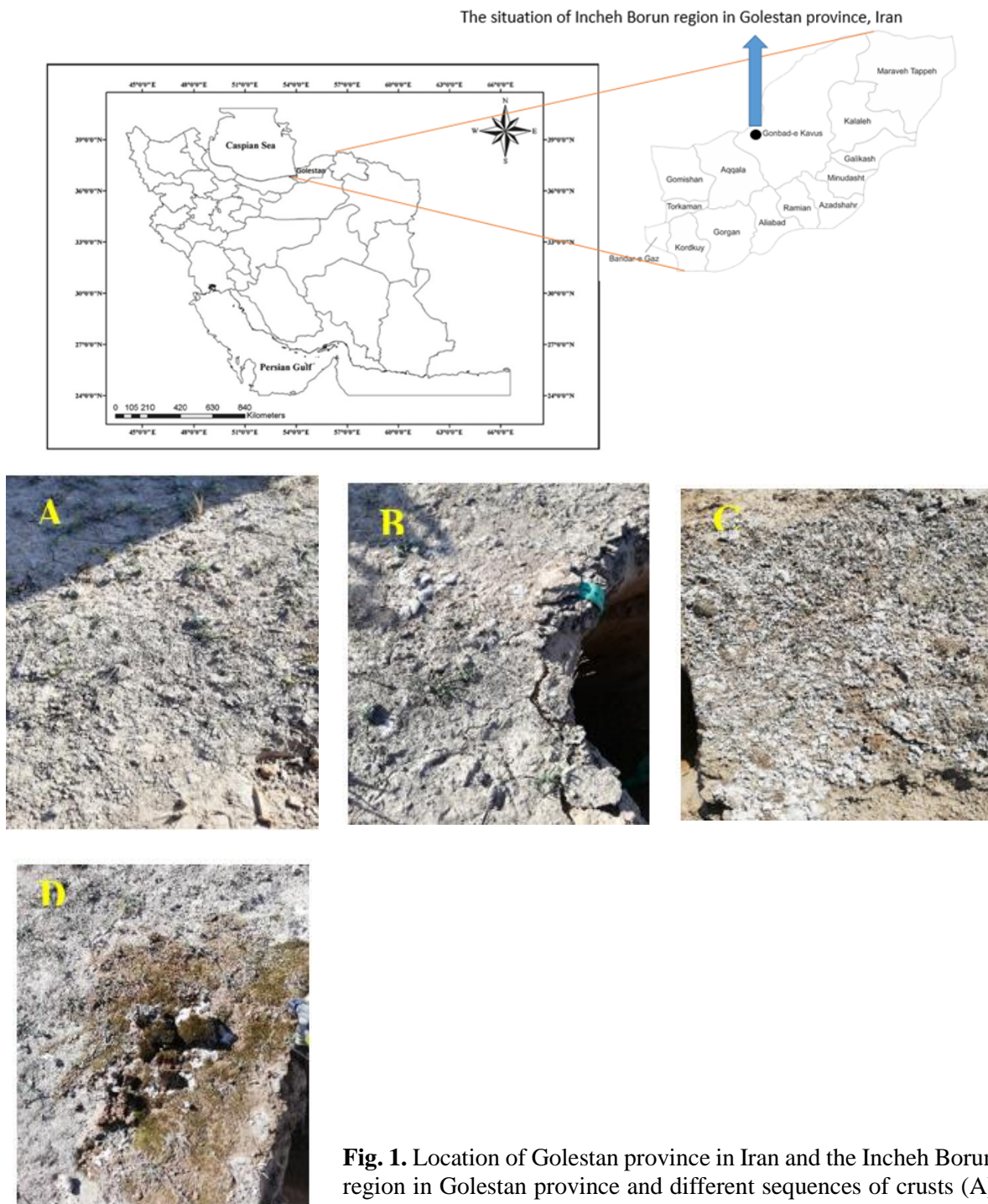


Fig. 1. Location of Golestan province in Iran and the Incheh Borun region in Golestan province and different sequences of crusts (A) Physical crust, B) cyanobacteria, C) lichen and D) moss).

2.2. Experimental design

In the Incheh Borun region, different sequences of soil biocrusts containing physical, cyanobacterial, lichen and moss were identified on the loess soils of this region and shown in figure one. The method of sampling from the subsoil on the surface of the crusts was as follows:

Three plots with dimensions of 25 × 25 m were considered in a place with the most distribution of biocrusts. In these three regions, 10 samples of soil were randomly gathered from subsoils (The depth is 0 to 2 cm) of each shell. Also, samples were taken from the crust-free soil to serve as a control in the same manner as described above. The experiment was in the format of 3 replications.

2.3. Biological properties

Basal respiration of soil was distinguished by static incubation with triplicate (Gutián and Carballas, 1976). Substrate-induced respiration was analyzed with the procedure stated in pervious instructions. Microbial biomass carbon measurement was done by the chloroform fumigation method, using 0.5 M K₂SO₄ as extraction. (Shibahara and Inubushi, 1995). Microbial biomass nitrogen (MBN) in all samples was specified with chloroform (Shibahara and Inubushi, 1995). The total nitrogen was acquired with reducing the K₂SO₄ by fumigated soil and distributed to a deduction content of 0.54 (Shibahara and Inubushi, 1995). Microbial biomass phosphorus (MBP) was fumigated with chloroform and then exploited with ammonium molybdate-stannous chloride (Shibahara and Inubushi, 1995).

The microbial metabolic quotient (qCO₂) shows the value of CO₂-C constructed and was accounted for basal soil respiration in the unit of mg CO₂-C. mg MBC-1 d-1 (Liu *et al.*, 2018). The mineralization quotient (qM) is defined in percentage as well as deliberated like a proportion of microbial soil respiration to soil organic carbon. The qM demonstrates the output of microflora in metabolizing the organic carbon of soil (Mganga *et al.*, 2016). The microbial quotient (qMic) was acquired with the proportion of MBC and total of soil organic carbon (MBC/TOC) × 100 that stated as a percent (Sparling, 1992).

2.4. Physico-chemical properties

Soil pH was evaluated with a proportion of 1:2.5 soil: water. (Kome *et al.*, 2018), and the measurement of EC in solution was done with an EC meter (Hanna Instruments, Model HI5321-02). Also, soil texture was determined (Mohajer and Salehi, 2018). Sodium absorption ratio and sodium exchangeable percentage were calculated with the method of Richards (1954) The Walkley and Black (1934) procedure was done to appraise the organic carbon of soil (SOC) and the van Bemmelen factor (=1.724) was applied to evaluate the content of soil organic matter (SOM) with the following equation (Martínez *et al.*, 2018):

$$\text{SOM} = \text{SOC} \times 1.724 \quad (1)$$

The mean weight diameter (MWD) and geometric mean diameter (GMD) were acquired to explain the particle size distribution (Besalatpour *et al.*, 2013) that were calculated as:

$$\text{MWD} = \sum_{i=1}^n W_i \bar{X}_i \quad (2)$$

$$\text{GMD} = \exp(\sum_{i=1}^n W_i \log \bar{X}_i) \quad (3)$$

Bulk density was investigated with Czyż and Dexter (2015) procedure. Wind Erosion Soil Stability (WESS) based on the difference in silt and clay values was calculated using the dry method (Zobeck and Scott, 2014) through the following equation:

$$WESS = \left[\frac{(A-B)}{(A+B)} + \frac{(C-D)}{(C+D)} \right] \quad (4)$$

Where,

A: Silt percentage of the sample before centrifugation

B: Clay percentage of the sample before centrifugation

C: Silt percentage of the sample after centrifugation, and

D: Clay percentage of the sample after centrifugation

Statistical analysis

A one-way analysis was considered to discover notable contrast among treatments. Compare mean was done with using Duncan's multiple range test. Notable distinction was reported at $p < 0.05$. The experiment was analyzed using the SAS v.9.0 software (SAS Institute, Inc., Cary, NC). Also, the correlations of data were analyzed with the Excel v.2016 software (Microsoft Office 2016 Pro Plus).

3. Results

In order to investigate the possible reaction of various orders of biocrusts on the stability of soil, the biological and physicochemical properties of soils covered with these crusts were compared with each other.

3.1. Impact of physical and biological crusts on soil biological properties

According to Table 1, the content of basal soil respiration (BSR) and substrate-induced respiration (SIR) in biocrusts was greater than those in the physical crust so that the highest amount of these two factors was discovered in the moss crusts and the low one was noticed in the physical crust. Also, the outcomes expressed that the order of MBC content in the biological and physical shells was moss > lichen > cyanobacteria > physical crust. The most amount of microbial biomass nitrogen was detected in the biological crust of moss (11.3 mg N g⁻¹ soil) and the minimum one was discovered in the physical crust (4.02 mg N g⁻¹ soil).

Microbial biomass phosphorus (MBP) in biological crusts was significantly greater than the physical one so that the highest content was seen in the soil under the moss and the lowest one was observed in the subsoil of the physical crust. The highest content of Microbial metabolic quotient (qCO₂) (0.025 mg CO₂-C. mg MBC⁻¹ d⁻¹) was observed in the moss biocrust, which had a notable difference at the probability level of 5% with other biological and physical crusts. We also found statistically remarkable contrasts across soils under the various sequences of crusts in terms of qM. These principles were diverse from 1.6% to 5.2% in the Incheh Borun region. Also, the soil under the lichen biocrust had a significantly higher qMic compared with the soil under the moss crust by 25.20% and 23.56%, respectively. The lowest qMic was observed in cyanobacteria (20.25%) which was statistically similar to the physical crust. Also, the correlation of these biological factors was determined, and according to Figure 2, all factors had a positive and significant correlation with each other.

3.2. Impact of biophysical crusts on physicochemical properties of soil

The outcome of physicochemical characteristics under the several orders of bio-shells are summarized in Table 2. There was no difference between the four crusts regarding the soil texture and pH. Nevertheless, the biological crusts contained a lower pH and a higher silt value than the physical crust. The results discovered that the organic carbon of soil in the bioscrusts was greater than the physical one. SOC was 1.21%, 1.73% and 2.30% in cyanobacteria, lichen and moss, respectively, which was more than that in the physical crust (1.03%) (Table 2).

Table 1. Microbial parameters in soil under the different sequences of biocrusts

Parameter	Unit	Soil depth	Different sequences of bio-shells			
			Physical crust	Cyanobacteria	Lichen	Moss
Basal soil respiration (BSR)	mg CO ₂ -C g ⁻¹ soil d ⁻¹	0-2 cm	0.16 ± 0.05a	0.32 ± 0.032b	0.75 ± 0.019c	1.20 ± 0.024d
substrate-induced respiration (SIR)	mg CO ₂ -C g ⁻¹ soil d ⁻¹	0-2 cm	0.23 ± 0.014a	0.41 ± 0.012b	0.88 ± 0.017c	1.36 ± 0.010d
Microbial biomass Carbon (MBC)	mg C g ⁻¹ soil	0-2 cm	2.1 ± 0.41a	2.45 ± 0.28b	4.36 ± 0.57c	5.42 ± 0.39d
Microbial biomass Nitrogen (MBN)	mg N g ⁻¹ soil	0-2 cm	4.02 ± 0.19a	6.37 ± 0.72b	8.6 ± 0.28c	11.3 ± 0.41d
Microbial biomass phosphorus (MBP)	mg P g ⁻¹ soil	0-2 cm	0.13 ± 0.025a	0.153 ± 0.055b	0.21 ± 0.064c	0.257 ± 0.029d
Microbial metabolic quotient (qCO ₂)	mg CO ₂ -C. mg MBC ⁻¹ d ⁻¹	0-2 cm	0.011 ± 0.002a	0.0153 ± 0.005b	0.020 ± 0.009c	0.025 ± 0.055d
Mineralization quotient (qM)	%	0-2 cm	1.6 ± 0.022a	2.61 ± 0.071b	4.3 ± 0.027c	5.2 ± 0.033d
microbial quotient (qMic)	%	0-2 cm	20.38 ± 0.19a	20.25 ± 0.44a	25.20 ± 0.38b	23.56 ± 0.31b

Mean ± standard error of every factor conformed with a same symbol are notable various relying on the minimum considerable distinction (Duncan's) at $p < 0.05$.

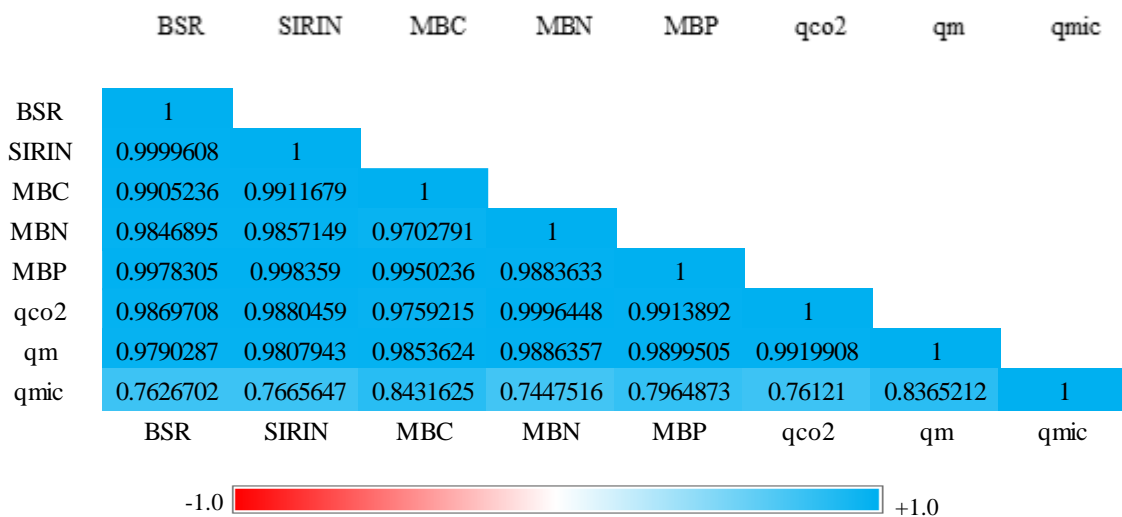


Fig. 2. Heat map of the overall correlations between the soil biological indicators. Blue indicates positive correlation between two factors, red indicates negative correlation between two factors and the number in each cell indicates Pearson correlation coefficient.

Also, the most SOM was seen in the moss crust, whereas the least one was detected in the physical crust (3.97% and 1.78%, respectively). The highest amount of Electrical Conductivity (EC) was found in soil under the physical crust (9.04 dS.m⁻¹), which was more than that in soil

under the cyanobacteria (8.49 dS.m^{-1}), lichen (7.64 dS.m^{-1}) and moss (5.55 dS.m^{-1}), respectively. Soil bulk density was notable greater in the physical crust in contrast with the biological one, respectively (1.53 g.cm^{-3} physical crust, 1.43 g.cm^{-3} cyanobacteria, 1.36 g.cm^{-3} lichen and 1.27 g.cm^{-3} moss). Also, the Sodium Adsorption Ratio (SAR) and Exchangeable Sodium Percentage (ESP) in soil under the different crusts were in order: physical crust > cyanobacteria > lichen > moss. A thermal map of soil physico-chemical attributes under the variety shells is introduced in Fig. 3. The most valuable connections ($p < 0.01$) were shown between pH and EC ($+0.9093$), ESP ($+0.9510$), SAR ($+0.9135$), and sand ($+0.8493$). Also, this consequence was true in the case of MWD, GMD ($+0.9960$), and WESS ($+0.9958$).

Table 2. Physicochemical parameters in soil under the different sequences of biocrusts

Parameters	Unit	Soil depth	Different sequences of biological soil crusts			
			Physical crust	Cyanobacteria	Lichen	Moss
Potential hydrogen (pH)	0-2 cm	-	$7.23 \pm 0.06a$	$7.15 \pm 0.04a$	$7.02 \pm 0.02a$	$6.97 \pm 0.11a$
Electrical conductivity (EC)	0-2 cm	ds.m^{-1}	$9.04 \pm 0.76a$	$8.49 \pm 0.69a$	$7.64 \pm 0.43b$	$5.55 \pm 0.83b$
Sodium adsorption ratio (SAR)	0-2 cm	meq.L^{-1}	$13.37 \pm 0.26a$	$12.59 \pm 0.57a$	$12.51 \pm 0.44b$	$11.75 \pm 0.17b$
Exchangeable sodium percentage (ESP)	0-2 cm	%	$82.13 \pm 0.11a$	$61.02 \pm 0.72b$	$52.75 \pm 0.31c$	$46.8 \pm 0.14d$
Sand	0-2 cm	%	33 ± 0.01	34 ± 0.02	21 ± 0.02	25 ± 0.03
Silt	0-2 cm	%	47 ± 0.04	44 ± 0.02	63 ± 0.01	56 ± 0.01
Clay	0-2 cm	%	20 ± 0.03	22 ± 0.04	16 ± 0.02	19 ± 0.02
Soil texture	0-2 cm	-	Silt loam	Silt loam	Silt loam	Silt loam
Soil organic carbon (SOC)	0-2 cm	%	$1.03 \pm 0.05a$	$1.21 \pm 0.05b$	$1.73 \pm 0.09c$	$2.30 \pm 0.06d$
Soil organic matter (SOM)	0-2 cm	%	$1.78 \pm 0.02a$	$2.08 \pm 0.01b$	$2.98 \pm 0.07c$	$3.97 \pm 0.05d$
Bulk density (BD)	0-2 cm	gr.cm^{-3}	$1.53 \pm 0.05d$	$1.43 \pm 0.07c$	$1.36 \pm 0.04b$	$1.27 \pm 0.01a$

Mean \pm standard error of every factor conformed with a same symbol are notable various relying on the minimum considerable distinction (Duncan's) at $p < 0.05$.

According to Figure 4, the mean comparison of three parameters i.e., mean weight diameter (MWD), geometric mean diameter (GMD) and wind erosion soil stability (WESS) showed that the lichen biological crust had the highest content of these parameters compared with the other biological and physical crusts so that the content of MWD was in order: lichen > moss > cyanobacteria > physical crust. About GMD, the results discovered that the amount of this factor in the lichen crust was significantly greater than its content in the moss, cyanobacteria and physical crust respectively (0.25 mm in the physical crust, 0.33 mm in the cyanobacteria, 0.71 mm in the lichen and 0.46 mm in the moss). Accordingly, the maximum WESS was recorded in the lichen crust, whereas the minimum one was found in the physical crust (1.185% and 0.663% , respectively).

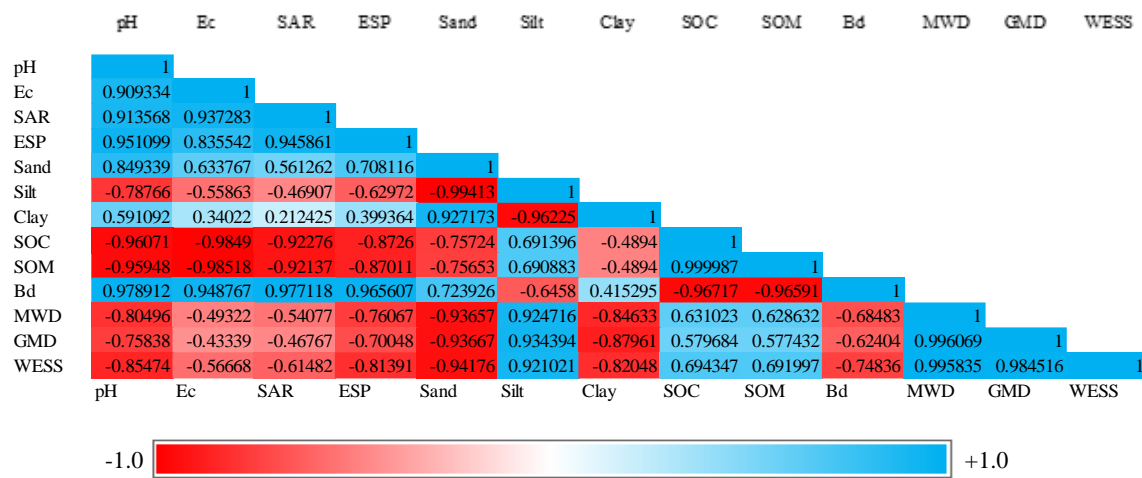


Fig. 3. Heat map of the overall correlations between the soil physicochemical factors. Blue indicates positive correlation between two factors, red indicates negative correlation between two factors and the number in each cell indicates Pearson correlation coefficient.

Discussion

According to the results, the soil under the biocrusts showed a high performance compared with the soil under the physical crust. The biocrusts had the most amount of MBC, MBN, MBP, BSR, SIR and both SOC and SOM, MWD, GMD, and WESS (Fig 2) qCO_2 and qM , which discovers that the presence of biocrusts can improve the biogeochemical cycles and recycling the essential nutrients in these fragile and sensitive ecosystems (Tables 1 and 2), which is consistent with other investigations that were conducted in dry regions (Dou *et al.*, 2023). The outcome of this investigation discovered that the efficacy of biocrusts and physical crust on biological features (Table 1) which can be proportional with the variation of physicochemical characteristics of the soil (Cui *et al.*, 2021). Some researchers reported an increase in soil electrical conductivity (Miralles *et al.*, 2020) and permeability, whereas the findings of other researchers indicate a decrease in electrical conductivity (Hasanzadeh Bashtian *et al.*, 2018). Also, with investigation the impact of biocrusts on soil quality, researchers emphasized the increase of organic carbon, soil stability and soil porosity (Kakeh *et al.*, 2018). One of the important findings in our results was investigation of the effect of biocrusts on MWD and WESS values. The treatment used in the measurement of MWD was soil grain immersion in water according to the De Leenheer and De Boodt method (De Leenheer & De Boodt, 1959), which most researchers have used as an index of soil grain stability in their research (Kelishadi *et al.*, 2018; Armin *et al.*, 2016). The middle of soil stability index (MWD, GMD and WESS) in the soils covered by biological crusts significantly improved (MWD: 0.21mm in the physical crust to 0.55mm in the lichen crust, GMD: 0.25mm in the physical crust to 0.71mm in the lichen crust, WESS: 0.663mm in the physical crust to 1.185mm in the lichen crust). Also, the sodium absorption ratio and exchangeable sodium percentage in different biological soil crusts (cyanobacteria, lichen and moss) were less than in the physical crust. Therefore, biological soil crusts can improve soil fixation as well as soil stability by decreasing the exchangeable sodium and soil dispersion, which is consistent with the outcomes of another researcher (Miralles *et al.*, 2020). According to the obtained results, the main reason for these increases can be the presence of more organic substances in the soils covered with biological crusts. The organic substances secreted from them have caused a stronger bond between soil particles and created more stable

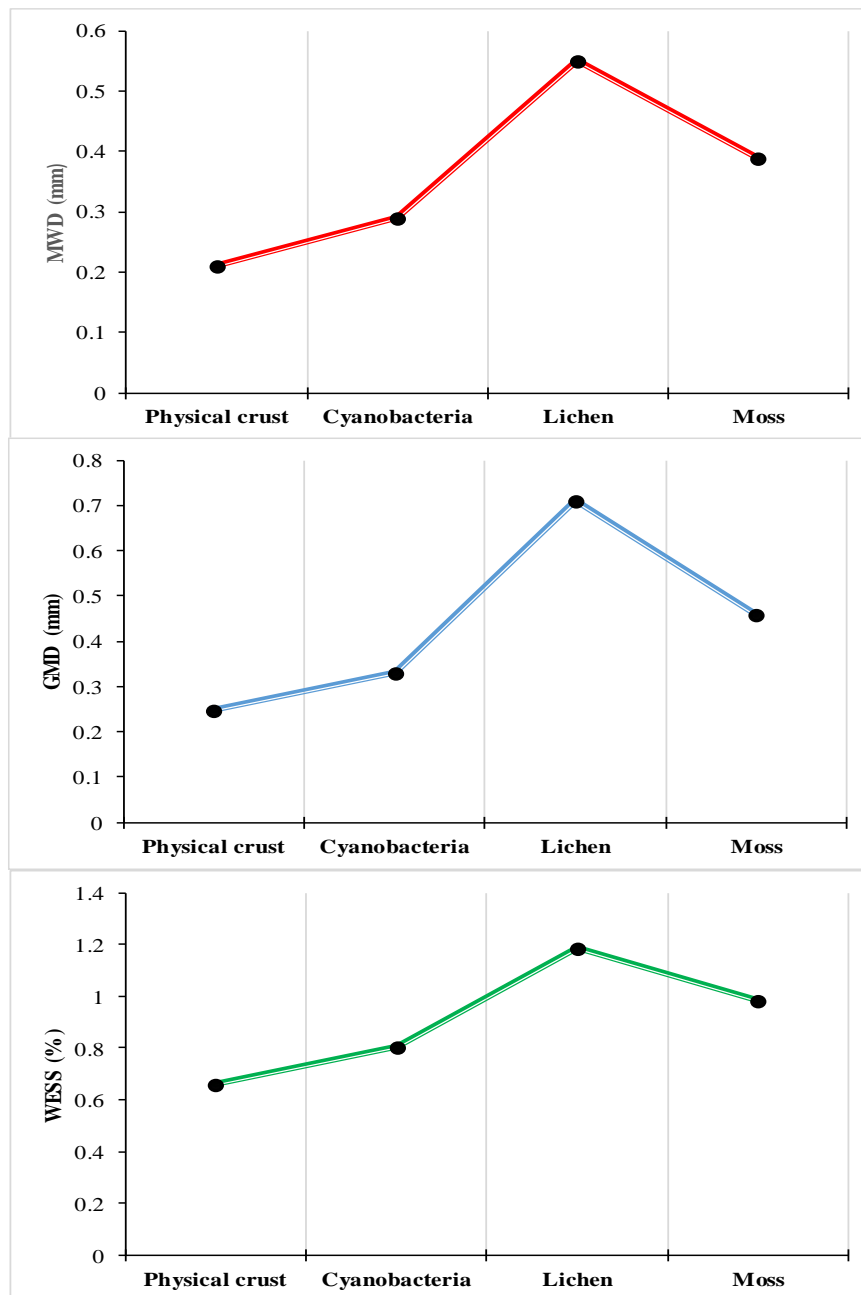


Fig. 4. Impact of various orders of shells on soil mean weight diameter (MWD), geometric mean diameter (GMD) and wind erosion soil stability (WESS)

soil grains that caused a stronger bond among soil particles and created more stable soil grains. Biocrusts are collections of lichens, mosses, algae, cyanobacteria, etc., which all improve soil grain stability in their own unique ways. For example, polysaccharide organic compounds secreted by cyanobacteria act like glue and cause stronger bonds between soil particles and create stable soil grains (Belnap *et al.*, 2016). On the other hand, the physical connection of soil particles with the hyphae of lichen (fungi part of lichen) is another important factor in particle bonding and the formation of stable soil grains (Eldridge & Leys, 2003). Therefore, the stability of soil grains and soil protection is partly due to special physicochemical mechanisms that cover

the soil with biological crusts. Also, they can play a very effective role in protecting and preventing soil losses while improving the stability of soil grains due to specific biological mechanisms. Some researchers have shown that the abundance of nutrients in biological crusts provides rich nutrient sources and a suitable perimeter for microorganisms of soil, which stimulates their function and leads to an increase in the other biological, physical and chemical activities (Liu *et al.*, 2018). Unlike, little nutrient in the physical crust might induce a defined environmental pressure of soil microbial associations as well as reduced their work (Bastida *et al.*, 2014) So, the alterations caused by biological crusts on soil physicochemical, biological attributes and soil microbial populations (Mganga *et al.*, 2016). The more content of qCO_2 was found out in subsoil of moss crust ($0.025 \text{ mg CO}_2\text{-C. mg MBC}^{-1} \text{ d}^{-1}$) and the low one was found in the soil under the physical crust ($0.011 \text{ mg CO}_2\text{-C. mg MBC}^{-1} \text{ d}^{-1}$). Previous studies have shown that stressful conditions may increase qCO_2 (VanWesemael *et al.*, 2019). The most amount of this element can be connected to variations in the proportion of bacteria to fungi (Nannipieri *et al.*, 2003). Also, we found out remarkable contrasts between shells about carbon mineralization quotient (qM). That one was diverse from 1.6 % in the soil under the physical crust to 5.2% in the soil covered by the moss biocrust, i.e., the biological crust had a better microbial C metabolism efficiency (Mocali *et al.*, 2008). The highest content of microbial quotient (qMic) was figured out at the lichen crust which showed a greater microbial quotient than the other biological and physical crusts. Therefore, the biological crust -especially lichen and moss- had a high biological performance in increasing soil aggregation as well as soil stability in sensitive regions like loess soils (Pichel, 2023).

Conclusion

Biological crust can strongly affect soil performance in dry environments, which is explained with (micro) biological indicators. With increasing the biological characteristics, mineralization processes and enzyme production of soil microorganism extension. This can lead to depletion of carbon collected at biocrusts that which can serve as a carbon dioxide source. Therefore, the reduction of metabolic procedures in dry durations shows that biological activity influences the performance of soil and greenhouse gas emissions with these crusts. Therefore, biocrusts are a good indicator to evaluate the degree of soil fertility. Further studies can provide the possibility of using biological indexes to evaluate the quality of sensitive soils to erosion and increase their stability.

Author Contributions

For research articles with three authors, a short paragraph specifying their individual contributions must be provided. The following statements should be used “Conceptualization, B.A. and S.M.; methodology, B.A.; software, B.A.; validation, F.K. and S.M.; formal analysis, B.A.; investigation, B.A.; resources, F.K.; data curation, F.K.; writing—original draft preparation, B.A.; writing—review and editing, F.K.; visualization, S.M.; supervision, F.K.; project administration, F.K.; funding acquisition, F.K. All authors have read and agreed to the published version of the manuscript.

All authors contributed equally to the conceptualization of the article and writing of the original and subsequent drafts.

Data Availability Statement

Data available on request from the authors.

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Ethical considerations

The authors avoided from data fabrication and falsification.

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Conflict of interest

The authors declare no conflict of interest.

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