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Soil C:N and C:P ratios positively influence colonization and development of incubated biocrusts in a sandy desert environment

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Abstract

Over the past few years, incubated biocrusts (IBSC)—the inoculation of soil/sand with cyanobacteria, moss, and lichen—have become one of the most promising biotechnological strategies for preventing soil erosion and restoring soil function in degraded drylands. Soil nutrient content (C, N, and P) is one of the key factors that influences IBSC colonization and development; however, the effects of soil C:N, C:P, and N:P stoichiometric ratios on the colonization and development of IBSC in desert environments are little known. We used four soil substrates, collected from four areas on the southeastern edge of the Tengger Desert, China, to incubate biocrusts. The four substrates differed in particle size and nutrient content. We flattened the dunes so their sand surfaces were level before covering them with soil substrates. We used a fully factorial design with four soil substrates and with and without biocrust additions, generating a total of 12 different treatments. The soil substrates differed in C, N, and P content and C:N, C:P, and N:P stoichiometric ratios. We measured IBSC coverage and thickness to establish relationships between IBSC characteristics and soil C:N, C:P, and N:P stoichiometric ratios. After 12 months of development, all treatments had significantly more cyanobacteria coverage, lichen coverage, and total coverage of IBSC than did control plots, which had little or no IBSC development. C:N and C:P ratios were significantly positively related to cyanobacteria coverage and total coverage of IBSC. Soil C:N and C:P ratios were mainly controlled by soil C; C limitation was greater than N and P limitation. Our study indicates that increasing initial soil substrate C content to improve C:N and C:P ratios will help the recovery of biocrusts and IBSC colonization. We demonstrate that stoichiometric ratios of soil should be a concern when assessing IBSC restoration treatments and when using IBSC to restore degraded land, especially at large scales.

KEYWORDS
arid area, biocrusts, cyanobacteria, soil stoichiometry, Tengger Desert

1 | INTRODUCTION

Desertification and land degradation in drylands are important social, economic, and environmental problems worldwide. More than 2 billion people live in drylands, and the degradation of drylands threatens the livelihoods and health of many people (Reynolds et al., 2007). Desertification is a major cause of soil erosion and soil nutrient loss and contributes to declines in soil fertility and biodiversity (Gisladottir &

Stocking, 2005). Restoration and rehabilitation of degraded lands is a challenge for sustainable development around the world (Reynolds et al., 2007). Berdugo et al. (2020) estimated more than 20% of the global terrestrial surface will be degraded and threatened by desertification by 2100. Many researchers have called for immediate actions to minimize the negative impacts of desertification and land degradation on essential ecosystem services (Chen et al., 2019; Reynolds et al., 2007; Singh & Ajai., 2019).

Current approaches to recovering degraded lands include biological engineering and chemical methods (Zhao et al., 2019; Zhao, Wang, et al., 2021). In recent years, incubated biological soil crusts/biocrusts (IBSC)—the inoculation of soil/sand with cyanobacteria, moss, and lichen—have become one of the most promising biotechnological strategies for preventing soil erosion and restoring soil functionality in degraded drylands (Antoninka et al., 2020; Chamizo et al., 2018; Chock et al., 2019). IBSC has been used to restore soil function at different scales and has been particularly successful at larger scales (Zhou et al., 2020). The most important ecological effect of IBSC is to improve sand surface stability. Zhao, Xu, and Wang (2021) found that IBSC significantly increased threshold friction velocity from 3.70 m s^{-1} (sand) to 10.00 m s^{-1}, and reduced soil surface wind erosion. IBSC also increases soil aggregation, strength, soil organic carbon (C), total nitrogen (N), and enzymatic activity (Lan et al., 2014; Zhou et al., 2020). Additionally, IBSC can enhance soil species diversity (Lan et al., 2014). In short, IBSC accelerates the restoration of ecosystem function in degraded drylands.

Soil nutrient content is one of the key factors that influences colonization and development of IBSC (Antoninka et al., 2020). A previous report showed that soil organic C, total N, and total phosphorus (P) were positively correlated with percentage cover by an incubated cyanobacteria crust (Chamizo et al., 2018). Similarly, Bu et al. (2018) observed that increasing soil nutrients increased IBSC coverage by 40%–50% compared to controls. This result is consistent with our work, in which we found that increasing initial soil substrate nutrient content, especially soil organic C content, helped the recovery of biocrusts (Zhao, Xu, & Wang, 2021). The rapid growth of IBSC can be achieved if soil nutrient limitations are removed (Antoninka et al., 2016). In soil ecological stoichiometry, C:N, C:P, and N:P ratios are key indicators that reflect the nutritional structure of the soil, such as the composition of soil organic C and the availability of soil nutrients (Atkinson et al., 2020). Many studies have examined the relationship between soil stoichiometry and biocrusts. For example, in the Gurbantunggut Desert, Y. Li et al. (2019a) studied the relationship between soil stoichiometry and moss stoichiometry. Another study examined the effects of N addition on C:N:P stoichiometry and moss crust-soil continuum in the same desert (Liu et al., 2020). Meng et al. (2021) explored ecological stoichiometric characteristics of soil-moss-C, N, and P during stages of the restoration of a rocky area that was undergoing desertification. Although most studies have focused on the relationship between soil and naturally developed biocrusts (i.e., moss crusts), the relationships between soil and IBSC, particularly incubated cyanobacteria crusts, C, N, and P stoichiometry, however, remain unclear; therefore, understanding the effects of soil C:N:P stoichiometric ratios on the establishment of IBSC is vital to improving the restoration of ecosystem function in drylands.

Hence, we collected four soil substrates with varying relative C, N and P content from the southeast edge of the temperate Tengger Desert in northern China. We investigated the effects of C:N, C:P, and N:P stoichiometric ratios on the colonization and development of IBSC. The specific objectives were: (1) to estimate the effects of soil C, N, and P content on IBSC properties; (2) to evaluate the effects of C:N, C:P, and N:P stoichiometric ratios of soil substrates on the colonization and development of IBSC; and (3) to optimize the selection of soil substrates for cultivating IBSC in a sandy desert environment. This research provides scientific evidence to inform the restoration of soil function and the management of biocrusts in degraded drylands ecosystems.

2 | MATERIALS AND METHODS

2.1 | Study site

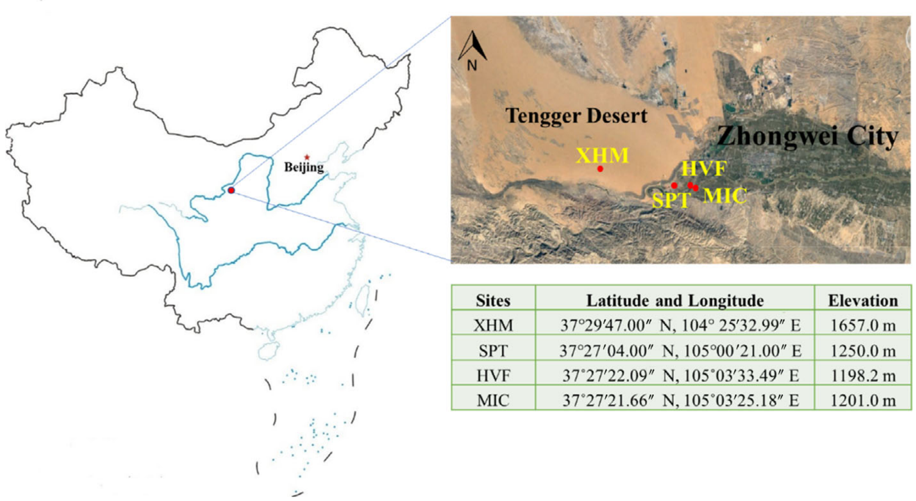
We conducted our experiment within the Shapotou Desert Research and Experiment State Station, Chinese Academy of Sciences (37°27′36.8″ N, 105°00′42.7″ E, elevation 1339 m), which is located on the southeastern edge of the Tengger Desert, China. The annual air temperature is 9.6°C, annual wind velocity is 2.9 m s^{-1}, and annual precipitation is 186.6 mm. Most precipitation occurs from June to September. Common plant species include *Artemisia ordosica* Krasch., *Caragana korshinskii* Kom., *Ceratoides latens* Reveal et, *Carex stenophylloides* Krecz, *Cleistogenes songorica* Ohwj, Holmgren, *Reaumuria soongorica* Maxim, *Salsola passerina* Bunge, *Oxytropis aciphylla* Ledeb. and *Stipa breviflora* Griseb. Total vegetation cover ranges from 1% to 30% coverage on aeolian sandy and orthic sierozem soils, respectively (X. R. Li et al., 2018).

Naturally developed biocrusts are dominated by cyanobacteria, lichens, and mosses. Dominant genera are *Anabaena*, *Hydrocoleus*, *Lyngbya*, *Microcoleus*, *Oscillatoria*, *Phormdium* and *Scytonema*, *Bryum*, *Didymodon*, *Collema* and *Endocarpon* (Wang et al., 2020).

2.2 | Field soil and BSCs collection

We used four soil substrates collected from four different areas in Zhongwei City, Ningxia Hui Autonomous Region, on the southeastern edge of the Tengger Desert (northern China; Figure 1). The four sites from West to East are described as follows: (1) the Xiaohong Mountain (XHM, 37°29′47.00″ N, 104°25′32.99″ E) at an elevation of 1657.0 m; (2) the Shapotou Station experimental area (SPT, 37°27′04″ N, 105°00′21″ E) at an elevation of 1250.0 m; (3) an abandoned farmland in Heilin Village of Zhongwei City (HVF, 37°27′22.9″ N, 105°03′33.49″ E) at an elevation of 1198.2 m; and (4) a Meili irrigation channel next to an abandoned farmland (from which soils were dredged) (MIC, 37°27′21.66″ N, 105°03′25.18″ E) at an elevation of 1201.0 m (Figure S1). All soil substrates (except SPT)

FIGURE 1 Map of soil substrate collection locations with longitude, latitude, and elevation. HVF, abandoned farmland in Heilin village; MIC, a dredged area of the Meili irrigation channel; SPT, the sandy Shapotou Station experimental plot; XHM, Xiaohong Mountain. [Colour figure can be viewed at wileyonlinelibrary.com]



Sites	Latitude and Longitude	Elevation
XHM	37°29′47.00″ N, 104° 25′32.99″ E	1657.0 m
SPT	37°27′04.00″ N, 105°00′21.00″ E	1250.0 m
HVF	37°27′22.09″ N, 105°03′33.49″ E	1198.2 m
MIC	37°27′21.66″ N, 105°03′25.18″ E	1201.0 m

were collected from 0 to 10 cm depth (because 0–10 cm soil organic C, total N, and total P contents were higher at that depth than in deep soil) and sieved to 2 mm to remove herbs, roots, and other fragments.

Naturally, well-developed cyanobacteria and cyanobacteria-lichen crusts were collected from Hongwei (37°27′ N, 104°46′ E, elevation: 1570 m), located on the southeastern edge of the Tengger Desert. In total, 16 natural cyanobacteria-lichen crusts (NCL) and 16 natural cyanobacteria crust (NC) samples were taken, each with 6% coverage (equivalent to 600 cm^2 in a 1 m^2-plot). The average thickness of the NC and NCL fragments were 2.94 mm and 4.85 mm, respectively.

2.3 | Experimental design

In August 2017, experimental plots were established in mobile dunes, where no natural biocrusts had developed on the sand surface. First, we flattened the dunes so their sand surfaces were level, then we covered their surface with the soil substrates (with 100% coverage and a 2 cm thickness). We used a full-factorial design with four soil substrates and with and without IBSC additions, generating a total of 12 different treatments. We randomly assigned treatments to plots and replicated each treatment four times, creating a total of 481-m^2 (1 × 1 m) plots. We spread collected natural cyanobacteria fragments (-NC) and natural cyanobacteria-lichen fragments (-NCL) randomly on each plot once. NCL and NC fragments covered each plot with 600 cm^2 in a 1-m^2 plot (equivalent to 6% coverage). Precipitation during the study (total 316.9 mm) was the only source of water.

2.4 | Field measurements and laboratory data analysis

We measured the coverage and thickness of IBSC in September 2018 using a point sampling frame (X. Li et al., 2010) and vernier calipers, respectively. We measured the soil particle size of each original soil substrate using the pipette method (Loveland & Walley, 2001). The $K_2Cr_2O_7$ method was used to measure the soil organic C; the Kjeldah method was used to measure the total N; and the digestion-molybdenum antimony resistance spectrophotometry method was used to measure the total P, as described by the Nanjing Institute of Soil Research Chinese Academy of Sciences (1980). XHM had clay, silt, and sand contents of 3.10%, 18.85%, and 78.06%, respectively; SPT had clay, silt, and sand contents of 0.00%, 0.00%, and 100.00%, respectively; HVF had clay, silt, and sand contents of 7.34%, 52.64%, and 40.02%, respectively; and MIC had clay, silt, and sand contents of 73.04%, 23.49%, and 73.47%, respectively. Data describing particle size and soil organic C, total N, and total P were published in our previous study; see Zhao, Xu, and Wang (2021) and Table 1 and Figure S1.

2.5 | Data analyses

All statistical analyses were performed using SPSS 16.0 statistical software (SPSS Inc., Chicago, IL). The C:N, C:P, and N:P stoichiometric ratios of soil were expressed by mass ratios. We used a one-way ANOVA to assess differences in soil and IBSC variables among the 12 different treatments. A post hoc test was conducted using Duncan's multiple range test when the assumption of equal variance was met, and the data were analyzed using Tamhane's T2 post hoc test if this assumption was not met (heterogeneity of variance). Pearson correlation analysis was used to determine the relationships between IBSC properties and soil C, N, and P contents, as well as C:N, C:P, and N:P stoichiometric ratios. In addition, we analyzed the relationships between IBSC properties and soil C:N, C:P, and N:P stoichiometric ratios using linear, quadratic, power, and exponential functions. The best model to predict dependent variables was selected based on the determination coefficient (R^2) and Akaike information criterion (AIC), which is a penalized likelihood criterion; the best statistical model minimized the value of AIC (Burnham & Anderson, 2002). Data normality and equality of error variances were checked by Shapiro–Wilk test and Levene's test separately before ANOVA and correlation analysis.

3 | RESULTS

3.1 | Soil C, N, and P content and stoichiometric ratios in the original soil substances

In four original soil substances, abandoned farmland soils (HVF) had the highest C content (9.51 ± 0.01 g kg^{-1}), N content (0.88 ± 0.03 g kg^{-1}), and P content (1.51 ± 0.02 g kg^{-1}), all of which were significantly higher than the other three soil substrates ($p < 0.05$). Soil C, N, and P content in the sandy Shapotou Station soils (SPT) were significantly lower than in HVF soils, dredged soils (MIC), or Xiaohong Mountain soils (XHM; Table 1). The HVF and MIC had higher C:N stoichiometric ratios (10.87 and 11.02, respectively), followed by XHM (6.98) and SPT (5.51). The HVF C:P stoichiometric ratio was 6.30, which was significantly higher than for MIC (5.01), XHM (5.43), and SPT (3.50; $p < 0.05$). The highest N:P stoichiometric ratios occurred in XHM (0.78), followed by 0.68 in SPT, 0.58 in HVF, and 0.45 in MIC (Table 1).

3.2 | Coverage and thickness of IBSC

After 12 months of development, all treatments had significantly more cyanobacteria coverage, lichen coverage, and total coverage of IBSC than any of the control plots (CK), which had little or no IBSC development. Cyanobacteria coverage and total coverage of IBSC were higher in soils with incubated treatments (i.e., HVF, XHM, and MIC with NC and NCL) compared to the SPT with cyanobacterial and lichen treatments (Figure 2a,c). Lichen coverage of IBSC only occurred in –NCL treatments and was 3.25% in HVF, which was significantly lower than for MIC (5.88%), XHM (5.19), and SPT (5.00; $p < 0.05$; Figure 2b).

After 12 months, the thicknesses of the IBSC in the SPT, HVF, XHM, and MIC with NC treatments were greater than the initial thickness of the NC (2.94 mm), although the differences were not statistically significant; however, the thicknesses of IBSC in the SPT, HVF, XHM, and MIC with NCL treatments were lower than the initial thickness of the NCL (4.85 mm; Figure 2d), although the differences were not statistically significant.

3.3 | Relationship between IBSC characteristics and soil C, N, and P stoichiometry

Correlation analysis revealed that soil C, N, and P were significantly positively correlated with each other and were also significantly positively correlated with C:N and C:P ratios ($p < 0.05$). Soil C and P were, however, significantly negatively correlated with N:P ($p < 0.05$). Soil C, N, P, and C:N and C:P ratios were significantly positively correlated with cyanobacteria coverage and total coverage of IBSC ($p < 0.05$); however, lichen coverage was not correlated with soil C, N, P, or C:N, C:P and N:P ratios (Figure 3; Table S1).

Cyanobacteria coverage and C:N ratio showed a significant positive exponential relationship ($y = 4.31 \times e^{(0.03 \times x)}$, $R^2 = 0.449$, $p < 0.001$) with an AIC value of -594.25. Total coverage of IBSC and C:N ratio showed a significant positive power relationship ($y = 1.20 \times x^{0.63}$, $R^2 = 0.451$, $p < 0.001$) with an AIC value of -594.32. Cyanobacteria coverage and C:P ratio showed a significant positive power relationship ($y = 1.40 \times x^{0.44}$, $R^2 = 0.571$, $p < 0.001$) with an AIC value of -594.32. Total coverage of IBSC and C:P showed a significant positive power relationship ($y = 1.04 \times x^{0.51}$, $R^2 = 0.541$, $p < 0.001$) with an AIC value of -608.42 (Figures 4 and 5; Table S2).

4 | DISCUSSION

4.1 | Effects of soil nutrient contents on IBSC properties

C, N, and P are essential components of all organisms and soil. C is produced by photosynthesis and is the substrate and energy source for an organism's physiological activities; N and P play crucial functional roles in organism growth and physiology (Andersen et al., 2004; Elser et al., 2003). Generally, there are two main ways that biocrusts (natural or artificial) access nutrients to support growth: (1) biocrusts such as cyanobacteria, algae, and other autotrophic microorganisms can fix C and N; and (2) soils contain and provide nutrients necessary for biocrust growth (X. Li, 2012). Numerous studies demonstrate that low soil C and low nutrient availability significantly limit the growth and development of natural biocrusts and also limit soil microbial

TABLE 1 Soil organic carbon (C), total nitrogen (N), total phosphorus contents (P), C:N, C:P and N:P ratios (mean ± SE) in soils collected from the sandy Shapotou Station experimental area (SPT); abandoned farmland in Heilin village of Zhongwei city (HVF); a dredged area of the Meili irrigation channel (MIC); and Xiaohong Mountain (XHM).

Soil subtracts	C g kg^{-1}	N g kg^{-1}	P g kg^{-1}	C:N	C:P	N:P
SPT	0.90 ± 0.10d	0.16 ± 0.01c	0.26 ± 0.02d	5.51 ± 0.61c	3.50 ± 0.37c	0.64 ± 0.02b
HVF	9.51 ± 0.01a	0.88 ± 0.03a	1.51 ± 0.02a	10.87 ± 0.41a	6.30 ± 0.07a	0.58 ± 0.03c
MIC	4.56 ± 0.05b	0.41 ± 0.01b	0.91 ± 0.03b	11.02 ± 0.39a	5.01 ± 0.23b	0.45 ± 0.01d
XHM	2.81 ± 0.08c	0.40 ± 0.01b	0.52 ± 0.01c	6.98 ± 0.35b	5.43 ± 0.29b	0.78 ± 0.02a

Note: Different letters indicate statistically significant differences ($p < 0.05$) between soil substrates.

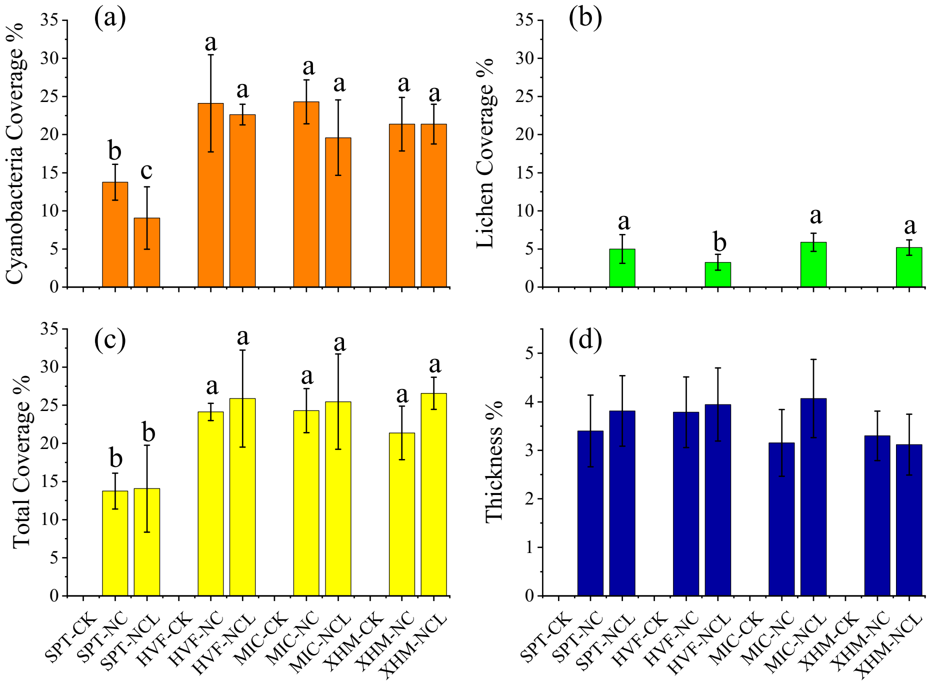


FIGURE 2 Cyanobacteria coverage, lichen coverage, total coverage, and thickness of incubated biological soil crusts on soil substrates collected from the sandy Shapotou Station experimental plot (SPT); abandoned farmland in Heilin village (HVF); a dredged area of the Meili irrigation channel (MIC); Xiaohong Mountain (XHM). Different letters reflect statistically significant differences ($p < 0.05$) between treatments. Bars represent one SE. CK, control plots; NC, natural cyanobacteria crust; NCL, natural cyanobacteria-lichen crusts. [Colour figure can be viewed at wileyonlinelibrary.com]

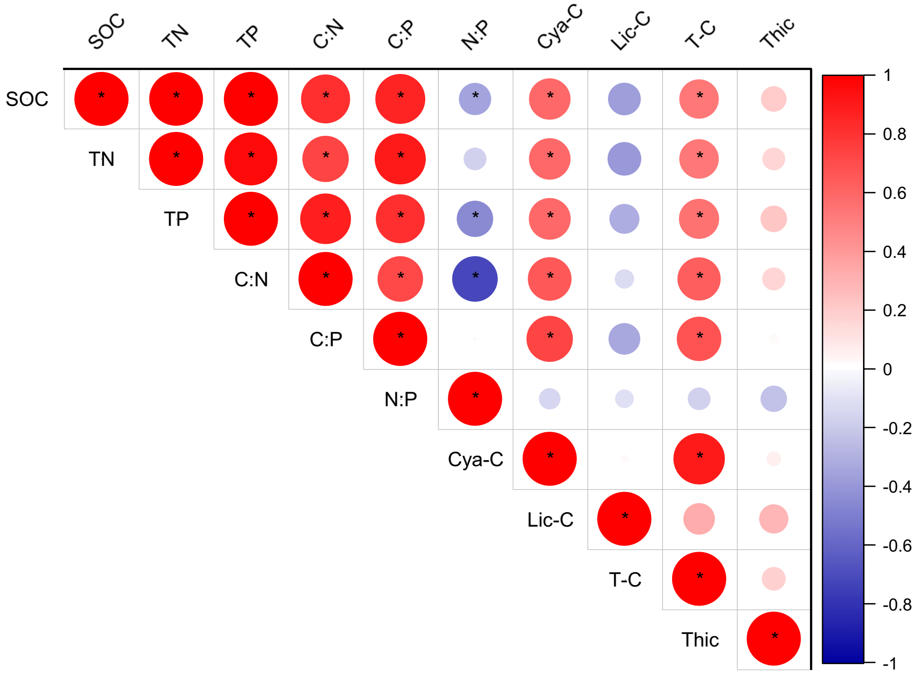


FIGURE 3 Pearson correlation analysis between soil organic carbon (SOC), total nitrogen (TN) and total phosphorus (TP) contents and C:N, C:P and N:P stoichiometry ratios and cyanobacteria coverage (Cya-C), lichen coverage (Lic-C), total coverage (T-C) and thickness (Thic) of incubated biological soil crusts. The "*" means $p \leq 0.05$. [Colour figure can be viewed at wileyonlinelibrary.com]

communities (X. Li, 2012; Yu et al., 2013). Specifically, soil C, N, and P contents are significantly positively correlated with the development of natural biocrusts in some sand desert environments, such as the Negev Desert (Kidron et al., 2010) and Tengger Desert (X. Li et al., 2021). Resource limitations slow growth in biocrusts. Rapid growth can be achieved if soil nutrient limitations are removed (Antoninka et al., 2016).

Our results show that C, N, and P were positively correlated with coverage of incubated cyanobacteria crust and total IBSC coverage. The four soils we tested differed in nutrient content; C, N, and P content were 2–10 times higher in HVF, XHM, and MIC soils than in SPT sandy soils (Table 1). Additionally, C, N, and P content were significantly positively correlated with soil silt and clay. Sandy soils had low nutrient content. Higher soil nutrient content reduced the resource limitation for IBSC growth. Incubated cyanobacteria crust and total IBSC coverage increased as soil nutrients increased. Previous reports support our results. For example, Bu et al. (2018) found that increasing soil nutrients increased IBSC coverage compared to controls.

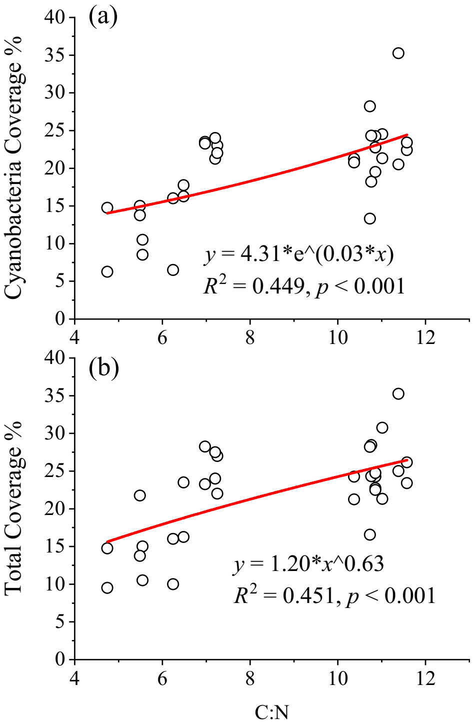


FIGURE 4 Regression analyses showing cyanobacteria coverage and total coverage of incubated biological soil crusts with soil C:N ratio. [Colour figure can be viewed at wileyonlinelibrary.com]

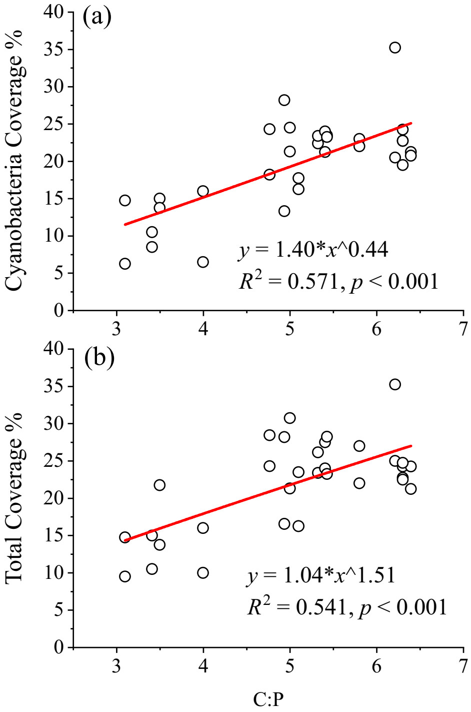


FIGURE 5 Regression analyses showing cyanobacteria coverage and total coverage of incubated biological soil crusts with soil C:P ratio. [Colour figure can be viewed at wileyonlinelibrary.com]

Antoninka et al. (2016) also demonstrated that nutrients promote IBSC development. Competition between cryptogams and heterotrophic microorganisms for limited soil nutrients may inhibit biocrusts growth. Generally, microbial biomass C and N are correlated with soil resource C and N, suggesting that increases in nutrient resources may alleviate the pressure of competition between those biota in biocrusts. This may provide another reason for the positive relationships among soil C, N, and P and IBSC growth.

4.2 | Effects of soil C:N:P stoichiometry on IBSC establishment

The ratios of C:N and C:P we observed suggest that plants assimilate C while simultaneously absorbing N and P. Increased C:N and C:P ratios indicate increases in the efficiency of N and P (Elser et al., 2003). Soil stoichiometry significantly influences plant growth and directly influences the composition, structure and productivity of ecological communities (Andersen et al., 2004; Fan et al., 2015). There were significant differences in the ratios of C:N and C:P among the four soils in our study, which represented four different initial soil microhabitats. Higher C:N and C:P occurred in HVF, MIC, and XHM soils than in SPT sandy soils. Soil C:N:P stoichiometry reflects the composition of soil nutrient and nutrient availability and plant nutrient status (Andersen et al., 2004). Generally, a higher C:N value (>25) indicates faster organic C accumulation than decomposition. C accumulation in soils is constrained by the litter C:N ratio. Also, C:P ratios are an effective indicator of soil P availability (Hessen et al., 2004). Results from the Gurbantunggut Desert suggest that C:N and C:P ratios are important factors influencing cyanobacteria, especially diazotrophic diversity in biocrusts (Xu et al., 2021). The ratios of C, N, and P might indicate nutrient limitation in cryptogam species growth in biocrusts (Y. Li et al., 2019b). Biocrusts can become C limited due to declining biocrust C:N ratios, often caused by declining C content in drylands soil (Mallen-Cooper & Cornwell, 2021). In contrast, Xu et al. (2021) found that C:N ratios of biocrust systems may indicate N limitation, because N fixed by cyanobacteria could accumulate at higher rates than C, reducing C:N ratios.

In our study, IBSC coverage was positively related to soil C:N and C:P ratios. Also, the correlation between C:N and C was higher than that between C:N and N, suggesting that soil C:N is mainly controlled by soil C. Additionally, soil total P is mainly derived from rock weathering, and the correlation of C with C:P was higher than the correlation of P with C:P, indicating that soil C:N and C:P were both primarily controlled by soil C. More important, low C:N ratios can reflect C limitation (Zechmeister-Boltenstern et al., 2015). N and P were low in the four soils we tested, and C:N and C:P increased linearly with soil organic C content; therefore, increased C content may increase C:N and C:P ratios, reduce C limitation, and promote IBSC growth. Our results demonstrate that covering sand with soils with relatively high C:N and C:P ratios may improve soil micro-environments and benefit incubated cyanobacteria crust establishment and development in field conditions. Increasing initial soil substrate C:N and C:P ratios may benefit the recovery of biocrusts in sandy desert environments.

In general, biocrust thickness increases resilience and reduces the vulnerability of biocrust erosion. Additionally, the degree of biocrust restoration or degradation may be affected by the thickness of the overlying sand (Kidron et al., 2017). A long-term (more than 50-year) study by X. Li (2012) in the Tengger Desert found that biocrust thickness increased 0.3–0.5 cm per 10 years, showing that biocrust thickness is restored slowly. In our study, the thickness of the IBSC was not significantly different than the initial thickness of NC or the thickness of the NCL after 12 months of cultivation in any of our treatments (Figure 2). There was no significant relationship between soil C, N, P, C:N, C:P, and N:P and the thickness of IBSC. The short duration of our research may explain the lack of relationship and suggests that long-term additional study should be conducted.

4.3 | Indication of soil C:N:P stoichiometry on biocrust incubation and restoration in the harsh sandy desert environments

Generally, approximately 20% biocrust cover is required to keep sediment transport below an erosion control target of 5 $\mathrm{g\ m^{-1}\ s^{-1}}$ in a restoration area (Eldridge & Leys, 2003). In our study, the coverage of IBSC reached that threshold when C:N and C:P ratios were between 7 ∼ 11:1 and 5 ∼ 6.3:1, respectively. These results suggest that XHM, HVF, and MIC are ideal soil substrates that support cyanobacteria crust cultivation in field conditions. C:N:P ratios are related to biological processes in desert ecosystems; however, imbalances in soil C:N:P stoichiometry can further influence the structures and functions of ecosystems (Hessen et al., 2004). Studies have also reported that nutrient-rich soils may contain more silt, clay, and soil water content than nutrient-poor soils. These soil properties can directly or indirectly affect biocrust establishment and development (Román et al., 2018; Zhao, Xu, & Wang, 2021). In soil with high C content and high C:N (C:P), N or P may, however, become limiting while C availability is high (Mooshammer et al., 2014). We found that C:N and C:P ratios ranging from 7:1 to 11:1 and 5:1 to 6.3:1, respectively, may reflect a relatively balanced status of soil C:N:P stoichiometry, providing suitable soil conditions for cyanobacteria soil colonization and improved ISBC growth and development.

Lab and field experiments suggest incubation of biocrusts with soil substrates works; soil substrates enhance sand surface stabilization and increase silt, clay, and nutrient content (Román et al., 2018; Zhao, Xu, & Wang, 2021). Additionally, soil nutrient content is significantly positively related to biocrust colonization and growth (Zhao, Xu, & Wang, 2021). Traditionally, soil substrates are selected according to differences in soil conditions, including physicochemical properties, such as silt, clay, pH, bulk density, and nutrient content (Román et al., 2018). Higher nutrient content is a standard used to select soil substrates for biocrust incubation (Antoninka et al., 2020). In our study, coverage of IBSC was positively related to soil C, N, and P, and with soil C:N and C:P ratios. These results indicated that soil stoichiometry (C:N and C:P) characters can be used as an index to select soil substrates for biocrust cultivation. Compared to soil C, N, and P contents, soil C:N:P ratios directly reflect the nutrient status in the soil. These ratios reflect the balance of soil C, N, and P, as well as soil quality (Tian et al., 2009). Additionally, if the C:N:P ratio approaches the optimal ratio required for soil microbial cells, and there are no other limiting factors (e.g., pH), then microbial growth will occur and contribute to C storage (Sinsabaugh et al., 2013); therefore, soil C, N, and P stoichiometry provide a powerful tool to select for soil substrates when considering candidates for biocrust cultivation.

Cryptogam growth strategy is influenced by different microhabitats and the need to balance energy and substrate allocation accordingly. Numerous studies have found that the imbalance in the C, N, and P stoichiometry fluctuates with climate, topography, and vegetation type in drylands (de-Bashan et al., 2021; Eldridge et al., 2020). According to Delgado-Baquerizo et al. (2013), based on data from 224 dryland sites across all continents, soil C–N–P stoichiometric imbalance increases with increasing aridity. Additionally, a study from a temperate desert in Central Asia found that soil C, N, and P stoichiometry ratios present strong spatial dependence in linear sand dunes areas. They found that soil stoichiometry is influenced by climate, wind strength, topography, and vegetation. Particularly, topography directly drives the stoichiometry ratios on some sand dunes (Tao et al., 2020). Gong et al. (2017) found that, in the 0–20 cm soil horizon, drought inhibited the uptake of nutrients by the desert plants community, impacting the N–P stoichiometric balance. Climate, topography, and vegetation caused different imbalances of soil stoichiometry, which varied across restoration areas. Given our study results, we strongly suggest that managers consider increasing soil C content in target restoration areas where C:N and C:P ratios are controlled by C in soil substrates. This may improve IBSC colonization rates or biocrust recovery rates in sandy desert environments. In regions where C:N and C:P ratios are controlled by N or P, managers should, however, use different strategies, such as increasing N or P content in soil substrates. Managers should pay attention to stoichiometric balance when using the soil substrates method to incubate biocrusts for degraded land or for damaged biocrusts recovery in large-scale ecological restoration activities in drylands.

5 | CONCLUSION

Our study showed that C:N and C:P ratios were significantly positively correlated with cyanobacteria coverage and total coverage of IBSC. We found significant positive relationships between cyanobacteria coverage and total coverage of IBSCs and C:N ratio. Significant positive relationships occurred between cyanobacteria coverage and total coverage of IBSCs and C:P ratio. Our study also established links between biocrust coverage and soil C–N–P stoichiometry ratios and provided new evidence for the relationship between biocrust and soil nutrients. Our study indicated that increasing initial soil substrate C:N and C:P ratios may benefit the recovery of biocrusts and IBSC colonization. We demonstrated that the stoichiometric ratios of soil should be considered when assessing IBSC restoration treatments and using IBSC to restore degraded lands, especially in large-scale activities.

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CONFLICT OF INTEREST STATEMENT

The authors declare no competing interests.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

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