Cyanobacteria drive community composition and functionality in rock-soil interface communities

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Running title: Cyanobacteria drive food-webs in hypoliths

Abstract

Most ecological research on hypoliths, significant primary producers in hyperarid deserts, has focused on the diversity of individual groups of microbes (i.e. bacteria). However, microbial communities are inherently complex, and the interactions between cyanobacteria, heterotrophic bacteria, protista and metazoa, are likely to be very important for ecosystem functioning. Cyanobacterial and heterotrophic bacterial communities were analysed by pyrosequencing, while metazoan and protistan communities were assessed by T-RFLP analysis. Microbial functionality was estimated using carbon substrate utilization. Cyanobacterial community composition was significant in shaping community structure and function in hypoliths. Ecological network analysis showed that most significant co-occurrences were positive, representing potential synergistic interactions. There were several highly interconnected associations (modules) and specific cyanobacteria were important in driving the modular structure of hypolithic networks. Together, our results suggest that hypolithic cyanobacteria have strong effects on higher trophic levels and ecosystem functioning.

Introduction

In many soil systems, including desert pavements, the ventral surfaces of translucent stones support unique lithic microbial communities. These communities, termed hypoliths, are dominated by photoautotrophic cyanobacteria, frequently in association with heterotrophic bacterial assemblages, as well as lichens and mosses in colder deserts (Cary *et al.* 2010; Makhalanyane *et al.* 2013a). By inhabiting the sublithic niche these communities benefit from physical stability, protection against incident ultra-violet radiation and excessive photosynthetically active radiation, thermal buffering, protection from freeze-thaw events, and enhanced moisture availability over the surrounding soil (reviewed in Chan *et al.* 2012). Hypoliths are often the major primary producers in hyperarid deserts (Pointing & Belnap 2012).

Many ecological aspects of hypolithic communities, including photosynthetic carbon fixation, nitrogen cycling, biodiversity and potential function have been investigated in desert environments (Pointing *et al.* 2009; Cowan *et al.* 2011; Chan *et al.* 2013). Community assembly in hypoliths has also been the focus of some research. For example, hypolithic assemblages are demonstrably different from those of surrounding soils (Makhalanyane *et al.* 2013b), and these assemblages are shaped by both local (e.g. environmental conditions) and regional (e.g. dispersal) factors (Caruso *et al.* 2011; Makhalanyane *et al.* 2013a). In particular, climate regime and salinity have been found to explain a large proportion of the variation in microbial community composition in hypoliths (Pointing *et al.* 2009; Stomeo *et al.* 2013). However, despite comprehensive evidence that microbial communities are inherently complex, most biodiversity research on hypoliths has focused on bacterial communities. To fully understand biological interactions in hypolithic communities, it is essential that the other components of the food-web should be included. This is of importance because food-web structure has

been shown to determine ecosystem stability (Rooney & McCann 2012), ecosystem functions (Cardinale *et al.* 2011) and the services they provide (Cardinale *et al.* 2012).

Here, we report on a study of the food-web structure including cyanobacteria, heterotrophic bacteria, protista and metazoa, and function in hypoliths found in the Namib Desert. We have combined pyrosequencing data (for heterotrophic bacteria and cyanobacteria), T-RFLP profiles (for metazoa and protista), carbon substrate utilization patterns, and environmental (climate and mineralogy) data. Because different cyanobacterial populations coexist in the Namib Desert (Stomeo *et al.* 2013) and cyanobacteria build and modify microhabitats, they act as ecosystem engineers (cf. Jones *et al.* 1994). Hence, we hypothesize that hypolithic cyanobacterial community composition can be linked to heterotrophic bacteria, protistan and metazoan assemblages, as well as to ecosystem functions.

Materials and methods

Sample collection and mineral analysis

In the central Namib Desert water is a scarce resource. The annual mean rainfall at Gobabeb recorded from 1962 to 2010, was 25 mm (Eckardt et al. 2013). Therefore, fog events, which are common in a zone from the coast to ca. 60 km inland, are thought to be the dominant source of bioavailable water in the region (Eckardt et al. 2013). Six sampling locations, three dominated by fog and three dominated by rainfall, were randomly selected in a 25 km radius of the Gobabeb Research & Training Station, central Namib Desert. At each sampling location, 5 colonized rocks (hypoliths) of similar composition (quartz), size (11-14 cm^2) and thickness (5-7 cm) were randomly selected (n=30) and stored in sterile Whirl-Pack sample bags (Nasco, WI, USA).

Simultaneously, sublithic soil samples were transferred into sterile 50 ml tubes. Samples were transported, at 4ºC, to the laboratory within two days of sampling and then stored at -80 °C until further analysis.

Because hypolithic communities are supported on essentially inert substrates with similar features, we characterised soil mineralogy for explaining variation in hypolithic community composition. The elemental content of the sublithic soil samples were determined at the Stellenbosch Central Analytical Facilities (Stellenbosch University, RSA) using standardized procedures. Light element analysis (%N, %C) was determined using a LECO Truspec elemental determinator. Major element analysis (Al_2O_3, CaO, Cr_2O_3, Fe_2O_3, K_2O, MgO, MnO, Na_2O, P_2O_5, SiO_2 and TiO_2) was determined using X-ray fluorescence spectrometry (Philips PW1404 XRF). The metadata (elemental content, geographical coordinates etc.) are given in MIMARKS format (Table S1, Supporting information).

DNA extraction and T-RFLP analysis

DNA was extracted from 0.5 g of each hypolith sample, previously removed from the rock with a sterile razor blade, using the MoBio PowerSoil DNA isolation kit (MoBIO, Carlsbad, CA, USA), eluted in 40 µl of Tris-EDTA buffer and quantified using the Nanodrop 1000 spectrophotometer (NanoDrop Products, Wilmington, DE, USA). PCR amplifications were performed in triplicate with the primer pairs, targeting 18S rRNA genes, and conditions described in Euringer & Lueders (2008) for protista, and Fonseca *et al.* (2010) for metazoa. For all primer pairs, the forward primer was labeled with 6' carboxyfluorescein (6-FAM). Purification, digestion with *Hae*III, separation of fragments, evaluation of electrophoretic signals and subsequent binning into operational

taxonomic units (OTUs) was performed as reported elsewhere (Valverde *et al.* 2012).

16S rRNA gene amplicon barcoded pyrosequencing

Twenty-four sets of barcoded PCR primers were designed to allow direct 454 tag sequencing of all samples on a single run. The PCR primers consisted of three components: 5'-[454 GS FLX adapter A/B] + [4 nt barcode] + [forward/reverse gene-specific PCR primer]-3'. Adapters and barcodes are provided in Table S1 (Supplementary material). DNA amplification with primer pairs 27F (5'-AGRGTTTGATCMTGGCTCAG-3') and 519R (5'- GTNTTACNGCGGCKGCTG-3') was performed in a single-step PCR using HotStarTaq Plus Master Mix Kit (Qiagen, Valencia, CA, USA). The following conditions were used: 94ºC for 3 minutes; 28 cycles of 94ºC for 30 seconds, 53ºC for 40 seconds and 72ºC for 1 minute; a final elongation step at 72ºC for 5 minutes. All amplicon products from different samples were mixed in equal concentrations and purified using Agencourt Ampure beads (Agencourt Bioscience Corporation, MA, USA). Samples were sequenced at the Molecular Research LP next generation sequencing service (http://www.mrdnalab.com) utilizing Roche 454 FLX titanium instruments and reagents, and following manufacturer's guidelines.

Pyrosequencing data were analysed using MOTHUR (version 1.27.0; Schloss *et al.* 2009) following a previously described pipeline (Schloss *et al.* 2011). Briefly, the FASTA quality and flow data were extracted using the sffinfo command. Low quality sequences were removed using the shhh.flows command, which is an implementation of the PyroNoise component of the AmpliconNoise suite of programs (Quince *et al.* 2011). The data set was simplified by obtaining the unique sequences using the unique.seqs

command. An alignment was generated using the align.seqs command by aligning the data to the SILVA reference alignment. The screen.seqs command was used to ensure that there was no overlap between the sequences. Chimeric sequences were removed through the chimera.slayer command.

After quality filtering, sequences were used to construct a distance matrix and grouped into OTUs (cut-off level of 97%) in MOTHUR. The taxonomic affiliations of the OTUs were determined using the naive Bayesian rRNA classifier (Wang *et al.* 2007), at an 80% confidence level. Sequences that had the highest similarity to chloroplast sequences were removed prior to further analysis. In order to compare diversity measures between the different sample types, the number of reads per individual sample was rarefied to 2198, which represents the lowest number of sequences obtained for a single sample.

Ecosystem functioning

We used Biolog EcoPlates (Biolog, Hayward, USA) to assess microbial utilization of carbon substrates. Biolog EcoPlates contain 31 different carbon substrates in triplicate and three carbon-free control wells providing intraplate replication. Plates were inoculated by pipetting 125 µl of hypolith suspensions into the wells. Colour development was measured with a microplate reader (Multiskan Go, Thermo Scientific), at 590 nm and daily for seven days after inoculation. The blank was subtracted from each reading and values divided by the average well colour development (AWCD) when AWCD reached a reference value of 0.5 (Garland *et al.* 2001).

Data analysis

Community structure was explored using Bray-Curtis dissimilarity matrices after Hellinger transformation, for metazoa and protista, and Weighted UniFrac dissimilarity matrices, for cyanobacteria and heterotrophic bacteria. Singletons were included, as their removal did not modify beta diversity patterns. Differences between habitats (fog- vs rainfall-dominted samples) were tested with ANOSIM (Analysis of similarity; Clarke, 1993), performing 9999 permutations. To determine which taxa generated most differences between groups, we used SIMPER (Similarity percentage; Clarke, 1993) analysis. The analyses were run with functions *anosim* and *simper* in the vegan package (Oksanen *et al.* 2013) for R. Correlations between biotic distance matrices were tested using Procrustes analysis with functions *procrustes* and *protest* in the vegan package . Correlations between two biotic distance matrices, while holding environmental distance constant, were tested using the partial Mantel test with *mantel.partial* in the vegan package. Function *daisy* (metric=gower), in the cluster package (Maechler *et al.* 2014), was used to incorporate climate regime (categorical variable) when calculating environmental distance between samples to perform partial Mantel tests. Diversity measures and abiotic raw data were compared by Kruskal-Wallis test. Mineralogy data were standardized. Forward selection of the environmental variables was performed, using function *ordistep* in the vegan package, to find the set of parameters that could best explain the variation in community composition. To evaluate the effects of the environment on community composition distance-based RDA (db-RDA; *capscale* function in the vegan package) was used.

We used the predictive form of co-correspondence analysis (ter Braak & Schaffers 2004), function *coca* in the cocorresp package (Simpson 2009) for R, to quantify the

strength of cyanobacterial community data in predicting heterotrophic bacteria, protista and metazoan community composition. A leave-one-out cross-validation procedure was performed to obtain the cross-validation fit (%) for different number of axes solutions and to select the minimal adequate predictive models. Any cross-validation fit > 0 implicitly validates the model, indicating that prediction is better than that obtained under the null model (Schaffers *et al.* 2008).

To perform network analysis we calculated all possible Spearman's rank correlations between OTUs with at least 0.1% relative abundance (260 of 4635 OTUs). We undertook this filtering step to increase the sensitivity of the network (Barberan *et al.* 2011; Berry & Widder 2014). We considered a valid co-occurrence event to be a robust correlation if the Spearman's correlation coefficient was both >0.6 and statistically significant (P-value <0.01) (Barberan *et al.* 2011). To study the topology of the resulting network we calculated average node connectivity, average path length, diameter, cumulative degree distribution, clustering coefficient and modularity using igraph (Csárdi & Nepusz 2006). Network characteristics were also obtained from random networks, which had the same number of nodes and edges as the empirical networks, generated using the Erdös-Rényi model in igraph. Association networks were visualized in Cytoscape v3.1.1 (Saito *et al.* 2012). Highly connected clusters or modules were defined based on network topology using the MCODE App (Bader & Hogue 2003) in Cytoscape.

All results presented here are for samples where all four microbial community data sets were available (n=24).

Results and discussion

In this study we examined the effect of cyanobacterial community composition in shaping food-web structure and function in hypoliths, with the working hypothesis that changes in cyanobacterial communities due to contrasting environmental conditions would affect both consumers and ecosystem functioning.

After quality filtering and rarefaction, a total of 4,435 OTUs (97% identity) were recovered from 259,640 sequences, of which 517 (37% singletons) OTUs belonged to the phylum *Cyanobacteria* and 3,918 (57% singletons) OTUs belonged to 14 other phyla (hereafter referred as to heterotrophic bacteria). Bacterial communities were dominated by photoautotrophic cyanobacteria, which comprised 34% of sequence abundances, whereas heterotrophic bacteria included members of the Alphaproteobacteria (22%), Actinobacteria (17%), Acidobacteria (6%), Betaproteobacteria (5%), Bacterioidetes (4%), Gammaproteobacteria (3%), *Deinococcus-Thermus* (2%). Another 10 phyla/classes were found in relative abundances below 2% (Fig. S1, Supporting information). Similar results have been reported in other studies investigating hypoliths, both in hot and cold arid environments (Caruso *et al.* 2011; Makhalanyane *et al.* 2013b), suggesting these well-represented phyla/classes are core members of hypolithic communities. Using T-RFLP analysis we detected a total of 90 protistan and 110 metazoan OTUs. Since T-RFLP data do not provide direct taxonomic information no comparisons with other environments were possible. Nevertheless, fingerprinting techniques have been shown valuable tools for the study of food-web structures (Chow *et al.* 2014).

Communities were separated by habitat type (Fig. S3, Supporting information): cyanobacteria (ANOSIM, global $R = 0.20$, $P < 0.01$), heterotrophic bacteria (ANOSIM, global $R = 0.77$, $P = 0.001$), protista (global $R = 0.55$, $P = 0.001$) and metazoa (global $R = 0.77$, $P = 0.001$). No clustering by site was found within habitat type. The most

important OTUs for each habitat type are shown in Table S2 (Supporting information). In general, rainfall-dominated communities showed higher α diversity regardless of the diversity metric (richness, Shannon or inverse Simpson) used, although those differences were not statistically significant for protistan communities (Table S3, Supporting information). We did not measure salinity in our samples, but salinity is usually higher in soils from fog-dominated regions (Stomeo *et al.* 2013), which could explain this pattern. Interestingly, whereas salinity has been identified as the dominant factor driving global patterns of bacterial biogeography (Lozupone & Knight 2007), protistan biogeography seems to be best predicted by moisture availability (Bates *et al.* 2013). The underlying mineralogy (ANOSIM, global R = 0.47, P = 0.001) was different between habitats and both climate regime and mineralogy (i.e. sodium oxide) were important factors explaining changes in communities composition (β diversity), as tested using db-RDA (Fig. 1), and together explained 13%, 11%, 36% and 18% of the variation in cyanobacterial, heterotrophic bacterial, metazoan and protistan community structure, respectively. Overall, our survey adds to the growing body of literature suggesting the dominant role of deterministic processes in structuring microbial communities (e.g. Wang *et al.* 2013).

Because we assumed bottom-up, rather than top-down, regulation on multitrophic interactions, as justified by Scherber *et al.* (2010), we tested for correlations between cyanobacteria-heterotrophic bacteria, cyanobacetria-metazoa and cyanobacteria-protista. Using T-RFLP fingerprinting data (Bray-Curtis dissimilarity matrices, after Hellinger transformation) for metazoan and protistan communities, and pyrosequencing data (Weighted UniFrac dissimilarity matrices) for heterotrophic bacterial and cyanobacterial communities, we found that cyanobacterial OTUs were significantly correlated with all three other communities (Procrustes correlation coefficient=0.6-0.7,

Fig. 1 Distance-based redundancy analysis (db-RDA) biplots of (a) cyanobacteria (weighted UniFrac distances), (b) heterotrophic bacteria (weighted UniFrac distances), (c) metazoa (Bray-Curtis distances) and (d) protista (Bray-Curtis distances). Only the environmental variables that significantly explained variability in community structure are fitted to the ordination (arrows). The direction of the arrows indicates the direction of maximum change of that variable, whereas the length of the arrow is proportional to the rate of change. Black symbols (fog-dominated samples), white symbols (rainfall-dominated samples).

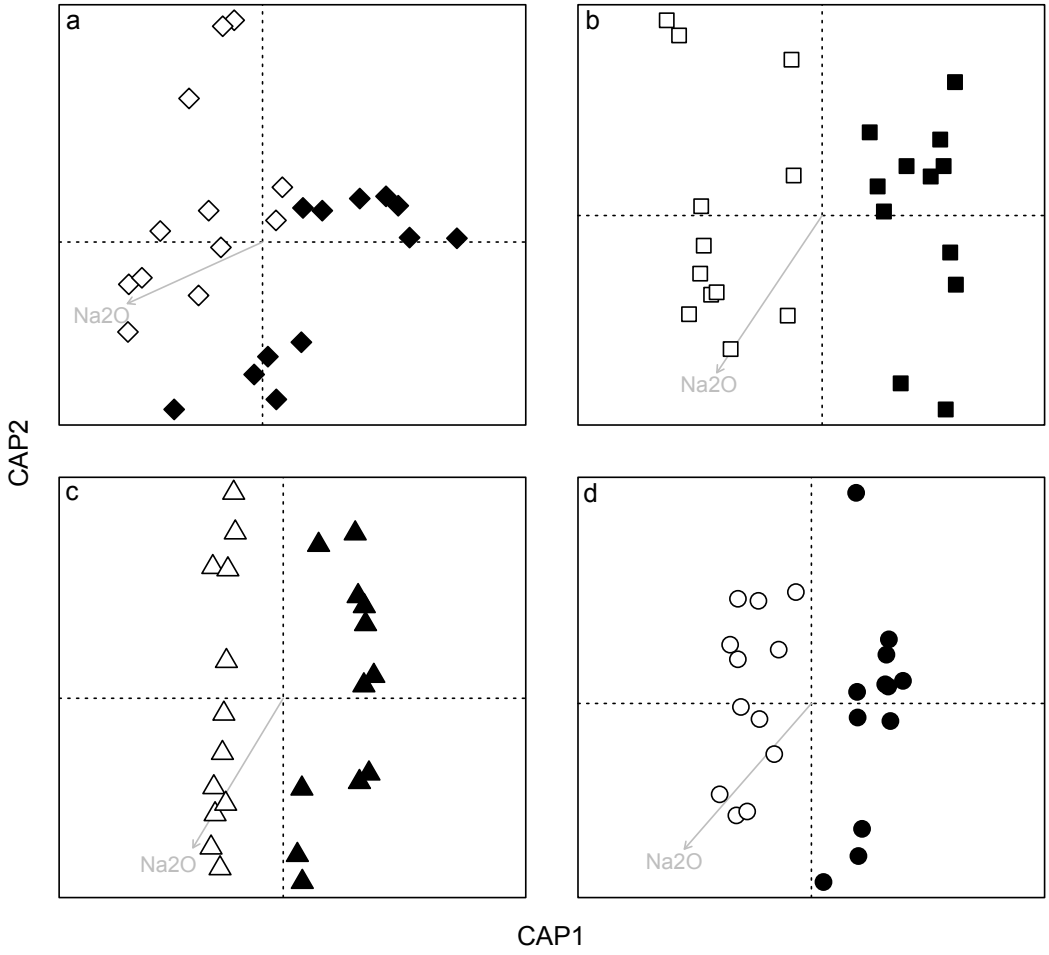
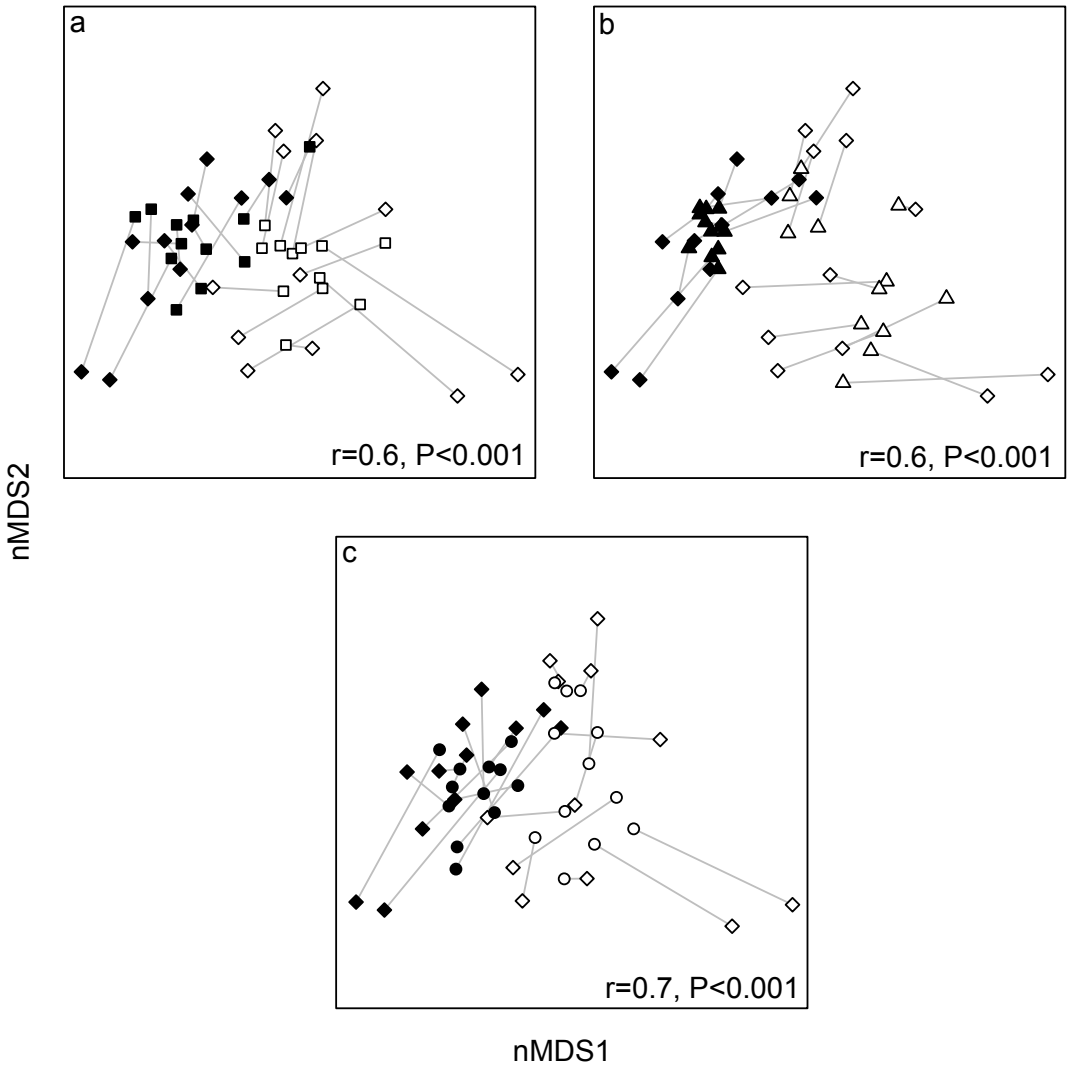


Fig. 2 Procrustes plots comparing nMDS analysis results from (a) cyanobacteria (weighted UniFrac distances) and heterotrophic bacteria (weighted UniFrac distances), (b) cyanobacteria and metazoa (Bray-Curtis distances) and (c) cyanobacteria and protista (Bray-Curtis distances). Cyanobacteria (diamonds), bacteria (squares), metazoa (triangles) and protista (circles). Black symbols (fog-dominated samples), white symbols (rainfall-dominated samples).



P<0.002 based on 999 permutations) (Fig. 2).. These results implied that cyanobacterial community structure is an important factor in shaping heterotrophic bacterial, metazoan and protistan community composition. To determine whether this outcome was due to the differences in the underlying environmental conditions, we tested for correlations, holding environmental distance constant, and using a partial Mantel test. The results remained qualitatively similar (partial Mantel r=0.5-0.6, P<0.01 based on 999 permutations), supporting the concept that cyanobacterial community composition is important *per se* in explaining food-web structure. Given these results, we investigated the amount of variation in heterotrophic bacterial, metazoan and protistan community composition that could be explained by cyanobacterial community composition alone, using co-correspondence analysis (ter Braak & Schaffers 2004). We found that 15%, 13% and 12% of the variance in bacterial, metazoan and protistan community composition was due to changes in cyanobacterial community composition, and therefore conclude that cyanobacteria are major drivers in determining food-web structure.

In order to investigate the coupling of food-web structure and functional performance, we combined all four sample-by-OTUs data tables and calculated the average well colour development (ecosystem functioning) of Biolog EcoPlates. While Biolog assays suffer from the same inherent biases as selective culturing, they are a valuable and inexpensive approach for elucidating selected functional properties of microbial communities (Baho *et al.* 2012). A significant relationship between community composition (Bray-Curtis distances of Hellinger-transformed data) and functioning (Euclidean distances of square root transformed data) was found (partial Mantel r=0.7, P<0.01 based on 999 permutations) while holding environmental distance constant, indicating that changes in community composition were accompanied by changes in

function. A similar pattern has also been observed in other studies (e.g. Fierer *et al.* 2012), emphasising that species composition is important for specific microbial community processes.

As cyanobacterial communities were shown to influence food-webs, we used network analysis to identify putative interactions between specific cyanobacteria and members of the other communities. It is important to note that in our network, as in many others (Chaffron *et al.* 2010; Barberan *et al.* 2011; Chow *et al.* 2014), nodes represent OTUs, and edges co-occurrence and not necessarily interactions. However, network analysis has proven very useful in revealing extensive phylogenetic and functional trait associations among members of terrestrial (Barberan *et al.* 2011) and aquatic (Chow *et al.* 2014) microbial communities, and provides an excellent framework for generating hypotheses regarding potential interactions (Faust & Raes 2012; Bissett *et al.* 2013). Significant correlations between individual cyanobacterial, bacterial, protistan and metazoan taxa were found in the full data set (Table 1). However, because co-occurrence networks lose interpretability when the effects of habitat filtering become significant (Berry & Widder 2014), as we have shown above, we generated separate networks for rainfall and fog-dominated samples (Fig. S3). Several topological properties, potentially relevant for community roles (Faust & Raes 2012) and the type of relationships (positive and negative) between co-occurring OTUs, were calculated to describe the complex pattern of inter-relationships between OTUs (Table 1). Overall, these networks showed statistical features similar to previously described ecological networks (Barberan *et al.* 2011; Steele *et al.* 2011; Chow *et al.* 2014). The observed:random network clustering coefficient ratio of 13 (Fog-dominated) and 14 (rainfall-dominated) is an indication that these association networks have 'small world' properties (that is, nodes are more connected than a random network of similar size ;

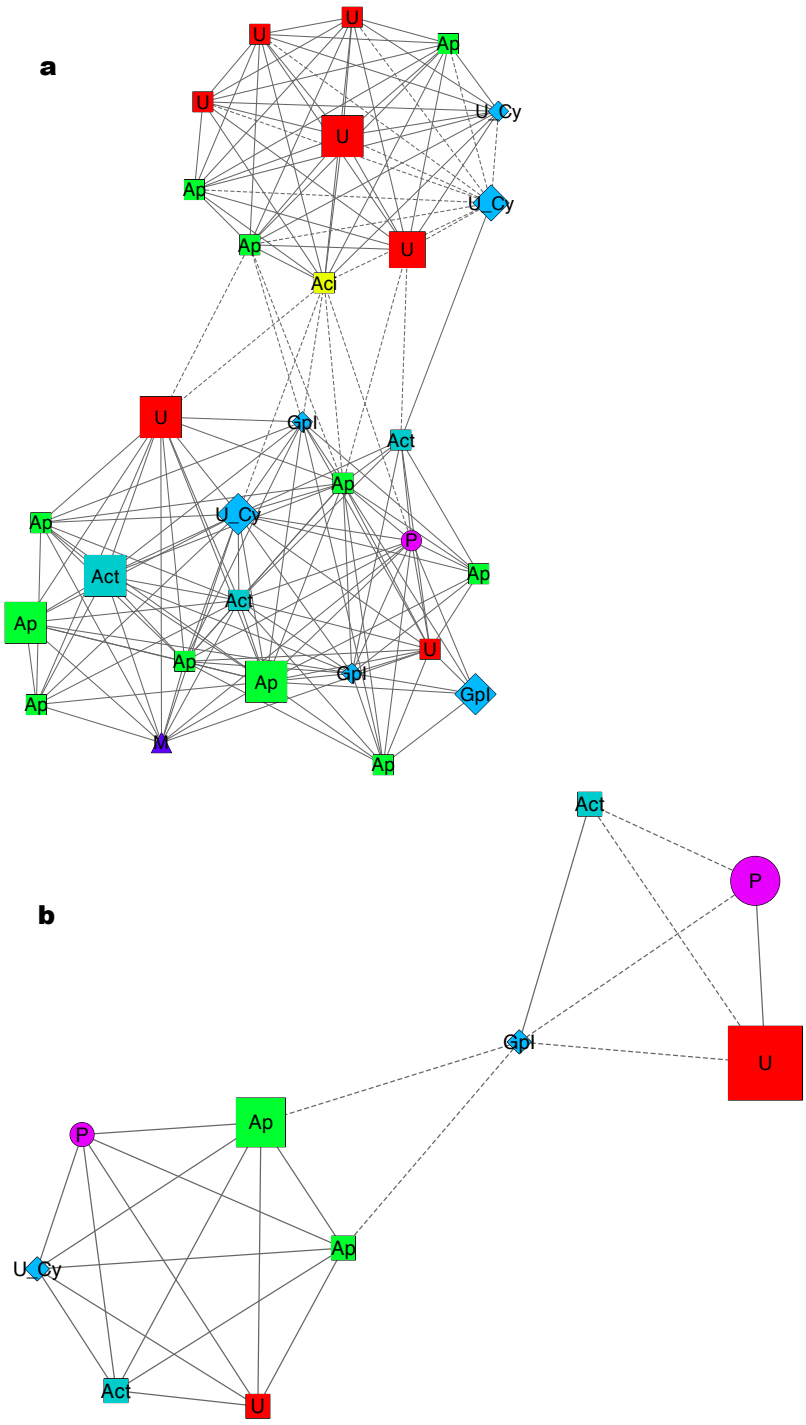
Table 1 Network statistics of hypolith samples

Topological properties	**Fog-dominated**	**Rainfall-dominated**	**Full data set**
Average node connectivity	8.71	7.88	12.98
Average path length	3.58 (2.62)*	4.27 (2.73)	3.52 (2.43)
Diameter	9 (5)	12 (5)	10 (4)
Clustering coefficient	0.52 (0.04)	0.72 (0.05)	0.48 (0.05)
Modularity	0.54 (0.22)	0.66 (0.28)	0.52 (0.20)
Nodes	179	174	248
Edges (total)	780	686	1610
Positive	618	597	1391
Negative	162	89	219
Edges (cyanobacteria)	330	312	679
Positive	269	259	587
Cyanobacteria-bacteria	57 %	59 %	63 %
Cyanobacteria-protista	4 %	5 %	2 %
Cyanobacteira-metazoa	8 %	2 %	7 %
Cyanobacteria-cyanobacteria	12 %	17 %	14 %
Negative	61	53	92
Cyanobacteria-bacteria	14 %	12 %	10 %
Cyanobacteria-protista	2 %	2 %	1 %
Cyanobacteira-metazoa	2 %	1 %	2 %
Cyanobacteria-cyanobacteria	1 %	2 %	1 %

*Values between brackets were obtained from random graphs (see materials and methods)

Steele *et al.* (2011)). Both association networks showed modularity (that is, they were composed of highly interconnected network regions with fewer node connections outside the module than inside), although modularity was lower in the fog-dominated network (Table 1). Modules may arise as a consequence of resource partitioning, habitat heterogeneity, divergent evolution and phylogenetic relatedness (Lewinsohn *et al.* 2006), and are thought to play a major role in ecosystem stability, as disturbances are expected to spread more slowly through a modular than a non-modular structure (Olesen *et al.* 2007). Cyanobacteria were important in explaining the modular structure of hypolithic networks, because a number of them were either module hubs or inter-module hubs (Fig. S3, Supporting information). Inter-module hubs (connectors) interact simultaneously with different modules of the network through transfer of energy and mater and are frequently referred as gatekeepers or keystone nodes (Steele *et al.* 2011). Such nodes have a high betweenness centrality (Fig. S4, Supporting information) and are thought to be crucial for ecological network structure and persistence (Saavedra *et al.* 2011). One cyanobacterial OTU, assigned to the genus GpI following the RDP classifier (Wang *et al.* 2007), was found to be a gatekeeper (betweenness centrality values of 0.45 and 0.5, for the fog and rainfall-dominated networks, respectively) in both climate regimes (Fig. 3). This is notable as this node is not one of the most abundant in the network (0.1 and 0.2 % relative abundance, for the fog and rainfall-dominated networks, respectively), highlighting the ability of the network approach to reveal the potential importance of cryptic groups (Bissett *et al.* 2013). The modules in which this OTU was found consisted of two sub-modules with strong negative co-occurrence between them, indicating that these sub-modules are composed of organisms present in distinct subsets of samples (that is, taxa were patchily distributed within habitats). The positive co-occurrence of Alphaproteobacteria in these sub-modules is

Fig. 3 Sub-networks highlighting position of genus GpI (cyanobacteria) in the network topology (Fig.S3, Supporting information). (a) fog-dominated samples and (b) rainfall-dominated samples. Nodes represent cyanobacteria (diamonds), heterotrophic bacteria (squares), metazoa (triangles) and protista (circles). Lines connecting nodes (edges) represent strong ($R > 0.6$) and significant ($P < 0.01$) positive (black) or negative (dashed black) co-occurrence relationships. Node size is proportional to an OTU's relative abundance. Node's names are the finest level that passed the Ribosomal Database Project classifier (80% confidence threshold): Aci, Acidobacteria; Act, Actinobacteria; Ap, Alphaproteobacteria; Ba, Bacteroidetes; Bp, Betaproteobacteria; U, unclassified heterotrophic bacteria; U_Cy, unclassified cyanobacteria; M, metazoa; P, protista.



also noted. Similar results have been observed in other ecosystems, such as salt marshes (Bissett *et al.* 2013) and the human gut (Faust *et al.* 2012), and might indicate that closely related taxa do not compete with each other, but may act synergistically. Interestingly, most correlations involving cyanobacteria (Table 1) were positive, which may suggest commensalism or a mutualistic relationship between organisms cooperating within the same niche, but also similar preferred habitat conditions. Negative correlations were also detected, which may suggest detrimental effects due to environmental modification or competition. However, microbial competition has been shown to be reduced in desert soils (Fierer *et al.* 2012).

Summarizing, we have shown that environmental variables and cyanobacterial communities modulate food-web composition in Namib Desert hypoliths. Hypolithic cyanobacteria produce carbon- and nitrogen-containing organic compounds such as amino acids, carbohydrates and extracellular polymeric substances (Chan *et al.* 2012). These products, as well as cyanobacterial biomass, are then consumed by protozoa, metazoa and heterotrophic bacteria, which act as trophic links connecting primary producers to the higher trophic levels and form the basis for the essential biogeochemical roles played by microbial food-webs in arid ecosystems (Pointing & Belnap 2012). We have also demonstrated that key physiological functions are related to food-web composition, which have important implications, as climate change is likely to influence the geographic distribution (Garcia-Pichel *et al.* 2013) and metabolic activity (Tracy *et al.* 2010) of soil cyanobacterial communities.

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Author Contributions

All authors contributed to the writing of the manuscript. AV design the study and performed research; AV and TPM led the sample collection; AV analysed the data and, along with DC, led the writing of the manuscript.

Data Accessibility

The sequence data generated in this study were deposited in the NCBI Sequence Read Archive and are available under the project number SRP044178.

T-RFLP data matrices (metazoan and protistan), Biolog data, pyrosequencing derived OTUs (heterotrophic bacteria and cyanobacteria), OTUs included in network analysis and the alignment generated in MOTHUR using the align.seqs command have been uploaded to Dryad doi:10.5061/dryad.nt4g6