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LETTER

Soil microbial communities drive the resistance of ecosystem multifunctionality to global change in drylands across the globe

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Abstract

The relationship between soil microbial communities and the resistance of multiple ecosystem functions linked to C, N and P cycling (multifunctionality resistance) to global change has never been assessed globally in natural ecosystems. We collected soils from 59 dryland ecosystems worldwide to investigate the importance of microbial communities as predictor of multifunctionality resistance to climate change and nitrogen fertilisation. Multifunctionality had a lower resistance to wetting–drying cycles than to warming or N deposition. Multifunctionality resistance was regulated by changes in microbial composition (relative abundance of phylotypes) but not by richness, total abundance of fungi and bacteria or the fungal: bacterial ratio. Our results suggest that positive effects of particular microbial taxa on multifunctionality resistance could potentially be controlled by altering soil pH. Together, our work demonstrates strong links between microbial community composition and multifunctionality resistance in dryland soils from six continents, and provides insights into the importance of microbial community composition for buffering effects of global change in drylands worldwide.

Keywords

Bacteria, carbon, fungi, multifunctionality, nitrogen, phosphorus, resistance.

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INTRODUCTION

Soil microbes are the most abundant and diverse organisms on Earth (Fierer & Jackson 2006; Locey & Lennon 2016). Recent experiments and observational studies have showed that, consistent with reported observations for plant communities (Cardinale *et al*. 2011; Maestre *et al*. 2012; Soliveres *et al*. 2016), soil microbial diversity plays an important role in maintaining multiple ecosystem functions simultaneously (i.e. multifunctionality) in terrestrial ecosystems (Philippot *et al*. 2013; Wagg *et al*. 2014; Delgado-Baquerizo *et al*. 2016). These functions include, but are not limited to, litter decomposition, nutrient cycling, primary production and the regulation of greenhouse emissions (Philippot *et al*. 2013; Wagg *et al*. 2014; Delgado-Baquerizo *et al*. 2016; Liu *et al*. 2017). Conversely, the role of microbial communities in regulating the resistance of multifunctionality (multifunctionality resistance hereafter) to global environmental change drivers remains largely unexplored and poorly understood (Orwin *et al*. 2006; De Vries *et al*. 2012; de Vries & Shade 2013). Identifying the major microbial drivers (composition, diversity or abundance) of multifunctionality resistance is crucial for developing sustainable ecosystem management and conservation policies. Such knowledge will help in prioritising future protection of microbial attributes involved in multifunctionality resistance, with implications to reduce impacts from climate change and land-use intensification on terrestrial ecosystems.

Existing knowledge, based mostly on the results of small-scale controlled experiments, suggests that particular soil microbial attributes (e.g. fungal: bacterial ratio) might regulate the resistance of particular ecosystem functions (e.g. soil respiration or N mineralisation) to global change drivers such as land use intensification and drought (Orwin *et al*. 2006; Downing & Leibold 2010; De Vries *et al*. 2012; de Vries & Shade 2013). However, we lack direct empirical evidence to identify how multiple microbial attributes, including the abundance, richness and composition of soil bacteria and fungi, regulate the response of multifunctionality to global change drivers, particularly at the global scale. Microbial attributes such as abundance, richness and community composition could play important roles in driving multifunctionality resistance to global change (MRGC hereafter), as they constitute important regulators of microbial growth, microbial interactions and key functional attributes belonging to particular taxa (e.g. nitrification). Furthermore, little is known about how changes in the composition of microbial communities

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across such scales (e.g. dissimilarity across sites; β-diversity) affect MRGC, particularly in drylands. These ecosystems already cover ~45% of Earth's land mass (Prăvălie 2016), and are expected to increase by up to 23% by the end of the 21st century due to forecasted increases in aridity under climate change (Huang *et al*. 2016). Achieving a better understanding of how dryland soil microbes drive MRGC is particularly important because: (1) microbial communities are highly affected by changes in aridity (Maestre *et al*. 2015), (2) drylands are overrepresented in developing countries (Huang *et al*. 2016) and (3) 38% of the global population is highly reliant on the primary production of drylands (Powell & Agnew 2011).

Herein we assess the importance of soil microbial community composition and abundance for MRGC, including warming, wetting–drying cycles and N fertilisation. This has never been assessed at the global scale. We aimed to do so using soils from 59 dryland ecosystems from all continents except Antarctica (Fig. 1). Soils were incubated for 21 days under different conditions to simulate expected impacts from temperature (control and 4.5 °C warming), changes in water availability (control and wetting–drying cycles) and N fertilisation (control and 20 kg N ha^{-1} $year^{-1}$), which were used as proxies of two major global change drivers (climate change and N deposition; Fig. 2a). Following incubation, we measured eight soil variables (hereafter 'functions') related to carbon (starch and cellulose degradation and carbohydrate availability), nitrogen (chitin degradation and availability of nitrate and ammonium) and phosphorus (P mineralisation and availability) cycling.

METHODS

Study area and soil sampling

Field data were collected between 2006 and 2014 from 59 dryland sites located in 12 countries from all continents except Antarctica (Fig. 1). All the surveyed sites had an aridity index (AI = precipitation/potential evapotranspiration) between 0.05 and 0.65 (UNEP 1992). Locations for this study were selected to cover a wide variety of natural and semi-natural ecosystem types (including grasslands, shrublands and open woodlands) representative of dryland ecosystems worldwide. Field surveys were conducted according to a standardised sampling protocol (Maestre *et al*. 2012). In brief, a composite topsoil (0-7.5 cm) sample (collected from five randomly selected plant interspaces) was obtained from each site and separated into two portions. One portion was air-dried and used for soil biochemical and functional analyses. The other portion of soil was immediately frozen at −20 °C for molecular analyses. Note that previous studies have found that air drying and further storage of dryland soils from do not alter the biogeochemistry of these soils (i.e. enzyme activities and nutrient contents; Zornoza *et al*. 2009). Similarly, previous studies have found a small effect, or no effect from air drying and further storage of soils on the community composition of bacteria and fungi (Macdonald *et al*. 2008; Lauber *et al*. 2010). For this reason, this storage approach is generally used in large-scale surveys (e.g., Maestre *et al*. 2012, 2015).

Environmental and physicochemical analyses

Air-dried soils were extracted in de-ionised water for 1 h to achieve a 1:5 soil: water solution. Soil pH was then determined using a combination pH electrode. Total soil organic carbon (TOC) was determined using the Walkley-Black method as explained in Maestre *et al*. (2012). AI (mean annual precipitation/potential evapotranspiration) was determined from Zomer *et al*. (2008), who use interpolations from the Worldclim database (http://www.worldclim.org). For clarity, we used aridity [1 − AI] instead of AI (Delgado-Baquerizo *et al*. 2013a). We used aridity instead of mean annual precipitation in our study because aridity includes both mean annual precipitation and potential evapotranspiration, and is therefore a more accurate metric of the long-term water availability at each site.

Characterising soil microbial communities

DNA was extracted using the Powersoil® DNA Isolation Kit (Mo Bio Laboratories, Carlsbad, CA, USA) according to the instructions provided by the manufacturer. qPCR reactions were performed in triplicate using 96-well plates on an ABI 7300 Real-Time PCR (Applied Biosystems, Foster City, CA, USA). The bacterial 16S-rRNA and fungal ITS genes were amplified with the Eub 338-Eub 518 and ITS 1-5.8S primer sets (Evans & Wallenstein 2011). The fungal: bacterial ratio was calculated using qPCR data. Note that calculating this ratio using qPCR may be inaccurate in terms of absolute values; however, it can still be useful for assessing its relationship with MRGC. In addition, we obtained information on the richness and composition of soil bacteria and fungi by performing 16S rRNA and ITS genes amplicon sequencing (Illumina MiSeq platform, Illumina, San Diego, CA, USA) and the 341F/805R and FITS7/ITS4 primer sets respectively (Herlemann *et al*. 2011; Ihrmark *et al*. 2012). Bioinformatic analyses were conducted using the QIIME package (See Maestre *et al*. 2015 for analytical details). Operational taxonomic units (OTUs) were picked at 97% sequence similarity. The resultant OTU abundance tables from these analyses were rarefied to an even number of sequences per samples to ensure equal sampling depth (11789 and 16222 for 16S rDNA and ITS respectively). Bacterial and fungal alpha diversity (i.e. number of phenotypes) was calculated from these OTUs tables. We also obtained the diversity (i.e. number of phenotypes) of common (the top 10% in terms of number of reads) and rare (the bottom 90%) species as described in Soliveres *et al*. (2016). Rare species, which are highly vulnerable to global change drivers, are being increasingly recognised as important drivers of ecosystem functioning (Jousset *et al*. 2017).

Experimental design: soil incubations

Soils were incubated to evaluate the effects of warming, changes in water availability, i.e. wetting–drying cycles and N fertilisation. In parallel, 5 g of soil from each site was placed in four plastic containers, one for each global driver plus an environmental control. The levels of the different treatments were selected to provide a realistic estimation of the response of ecosystem functioning to climate change, and to land-use

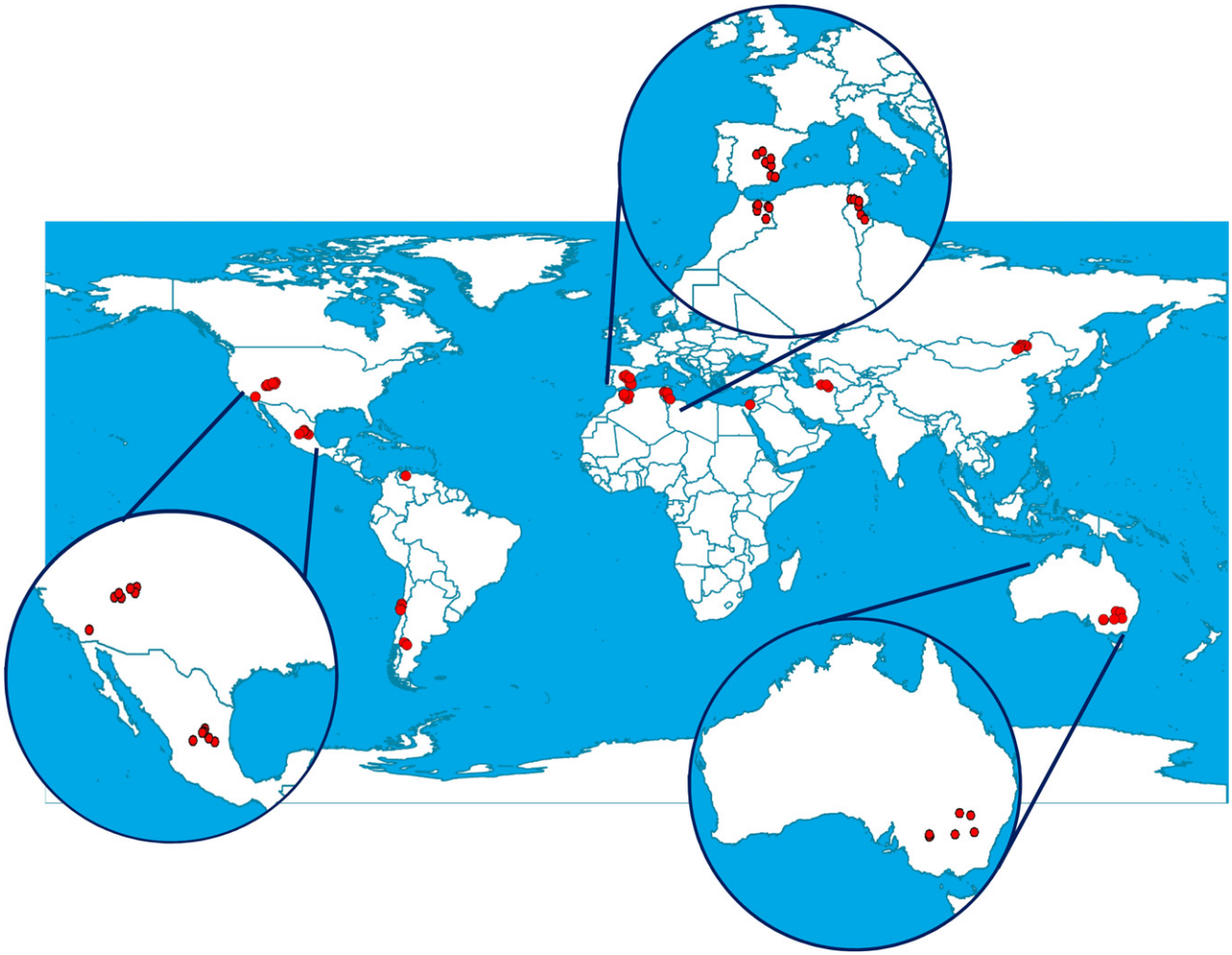

Figure 1 Locations of the 59 sites included in this study.

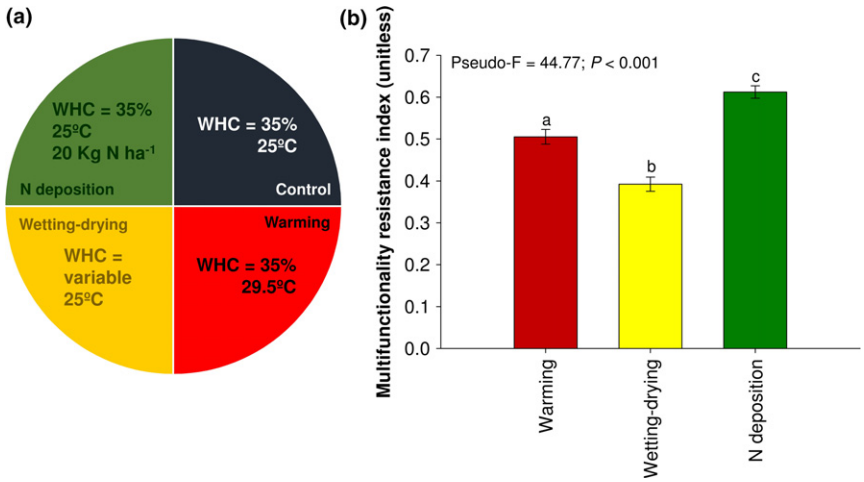

Figure 2 (a) Methodological framework explaining the conditions in all experimental treatments used. (b) Effects of warming, wetting–drying cycles and nitrogen (N) fertilisation on the multifunctionality resistance of dryland soils from across the globe. Data are means ± SE (n = 59). WHC = water holding capacity.

intensification driven by N fertilisation from atmospheric N deposition and livestock dung in global drylands. Thus, the environmental control was incubated at 25 °C, the average land surface temperature for all sites (see https://neo.sci.gsfc.nasa.gov/), and 35% of water holding capacity (WHC). The amount of water in the control was chosen to ensure a

minimum of microbial activity during the incubation period (Fig. 1 in Schwinning & Sala 2004; Delgado-Baquerizo *et al.* 2013b,c). The warming treatment had similar water conditions as the environmental control but with increased temperature (+4.5 °C; Fig. 2a). This temperature increase mimics global warming forecasts by the end of this century (A2 scenario from IPCC 2013). The wetting–drying treatment was incubated at the same temperature than the environmental control, but included four wetting–drying cycles. Each wetting–drying cycle involved wetting until a 35% WHC was achieved and a subsequent natural drying for 5 days. Soil samples were watered the first day of incubation (Fig. 2a). Rapid changes in water availability, such as those from wetting–drying cycles, are expected to increase with climate change in global drylands (IPCC Climate Change 2013). Finally, the N fertilisation treatment includes the same temperature and water conditions as the environmental control plus the equivalent to 20 kg N ha^{-1} $year^{-1}$ (Fig. 2a), which were added in the form of NH_4NO_3 during the first watering. This amount was selected to simulate artificial N loads from N deposition and N in manure from grazing, a major driver of land degradation in drylands worldwide (Eldridge & Delgado-Baquerizo 2017). The levels applied at our study sites (Fig. 1) were predicted using published mapping information (Dentener *et al.* 2006; Potter *et al.* 2011). Moisture content was adjusted and maintained at 35% WHC during the duration of the experiment for all treatments other than the wetting–drying treatment. A total of 236 samples (59 sites × 4 treatments) were incubated under the different treatments for 21 days.

Assessing multiple ecosystem functions

After incubation, we measured in all soil samples eight functions related to C, N and P cycling: activity of β-glucosidase (starch degradation), β-D-celluliosidase (cellulose degradation), N-acetyl-β-glucosaminidase (chitin degradation) and phosphatase (organic phosphorus mineralisation) and four measurements of C (dissolved carbohydrates), N (ammonium and nitrate) and P (inorganic P) availability. Extractable carbohydrates, ammonium and nitrate were obtained from K_2SO_4 extracts as explained in Delgado-Baquerizo *et al.* (2013a). Soil P availability was estimated from sodium bicarbonate extracts as described in Maestre *et al.* (2012). Extracellular soil enzyme activities were measured from 1 g of soil by fluorometry as described in Bell *et al.* (2013). Overall, these variables constitute good proxies of processes driving nutrient cycling, biological productivity and the build-up of nutrient pools (Maestre *et al.* 2012). In brief, carbohydrates are an essential source of energy for soil microbes and are used as an indicator of organic matter biodegradability (De Luca & Keeney 1993). Extracellular enzymes such as those we measured are produced by soil microorganisms and are involved in the processing, stabilisation and destabilisation of soil organic matter and nutrient cycling in terrestrial ecosystems (Bell *et al.* 2013). They are also considered a good indicator of nutrient demand by plants and soil microorganisms (Bell *et al.* 2013). Ammonium and nitrate are important N sources for both microorganisms and plants, and are produced by important ecosystem processes such as N mineralisation and nitrification (Schimel & Bennett 2004). Inorganic P is the main P source for plants and microorganisms, and its availability is linked to the desorption and dissolution of P from soil minerals (Vitousek 2004). We explicitly focused on the bioavailable pools of C, N and P (usually <1% of the total of their respective forms) because the total pools of these elements may not be relevant for the MRGC within our short-term incubation experiment.

Assessing the resistance of multiple ecosystem functions to global change drivers

We used the Orwin & Wardle (2004) index (RS) to evaluate the resistance of multiple functions as:

$$\mathrm{RS} = 1 - \frac{(2 \cdot (D_0))}{((C_0) + (D_0))}$$

In this equation, D_0 is the difference between the environmental control (C_0; value of each functional variable in the absence of global change treatments) and the disturbed (P_0, warming, wetting–drying cycles and N fertilisation treatments) soils after the incubation period. This index has the advantage of being: (1) standardised by the control and (2) bounded between −1 (lowest resistance) and +1 (maximal resistance) even when extreme values are encountered (Orwin & Wardle 2004). We calculated the resistance of each function independently for each global change driver. After this, and to evaluate MRGC, we averaged the resistance of the eight functions measured to obtain a standardised index of multifunctionality resistance. Similar approaches have been used to obtain multistability (Durán *et al.* 2017) and multifunctionality (Maestre *et al.* 2012; Wagg *et al.* 2014; Delgado-Baquerizo *et al.* 2016) indexes, as well as response ratios in meta-analysis (Eldridge & Delgado-Baquerizo 2017). Note that our study focuses on the simultaneous responses of multiple functions to global change rather than on the response of single functions that might not be representative of the overall functioning of a particular ecosystem.

Statistical analyses

Relationship between microbial community composition and multifunctionality resistance

We first explored the overall relationship between the β diversity of microbial communities and MRGC. To do this, we calculated microbial β-diversity using Bray–Curtis dissimilarity matrices at the OTU level independently for bacterial and fungal communities. Similarly, the Euclidean distance was used to create three independent distance matrices from the resistance of eight single functions. A matrix was constructed for each of the three global environmental drivers: warming, wetting–drying cycles and N fertilisation. We then independently correlated the β-diversity of bacteria and fungi to the dissimilarity matrices from resistance measurements using Mantel correlations (Pearson). We also assessed all possible Mantel correlations (Pearson) among resistance multifunctionality to warming, drying–wetting cycles and N fertilisation.

Random Forest modelling

To gain a mechanistic understanding of the drivers of MRGC, we conducted a classification Random Forest analysis (Breiman 2001) as described in Delgado-Baquerizo *et al.* (2016), which allowed us to identify common microbial predictors across sites. We used class-level information in these analyses for two main reasons (1) information on microbial functional traits has become increasingly available at this taxonomic level (Fierer *et al.* 2007; Trivedi *et al.* 2013); and (2) unlike high taxonomic rank information (OTU/genus), class-level taxa are shared across all soil samples at the global scale, allowing us to infer general patterns in the role of microbial composition in predicting MRGC at this spatial scale. In addition to class-level predictors, we included in our models other microbial attributes such as abundance (qPCR), fungal: bacterial ratio and alpha diversity (richness of all, common and rare fungi and bacteria). The importance and statistical significance of each predictor were computed using the rfPermute package (Archer 2016) of the R statistical software, version 3.0.2 (http://cran.r-project.org/). We also used Spearman correlations between selected major microbial attributes from Random Forest analyses and the resistance of single functions to global change. The aim of this approach was to obtain insights into the relationships between the relative abundance of particular microbial taxa and the resistance of specific functions, complementing results from MRGC analyses.

Structural equation modelling

We used structural equation modelling (SEM; Grace 2006) to evaluate the direct and indirect relationships between geographical location (latitude and longitude), aridity, soil properties (pH and soil total organic carbon) and microbial attributes on MRGC based on expectations under an *a priori* model (Fig. S1). Microbial drivers included pre-selected major significant MRGC predictors from Random Forest analyses described above. Aridity and soil properties such as total organic carbon and pH are major drivers of microbial community composition in drylands (Fierer & Jackson 2006; Fierer *et al.* 2012; Maestre *et al.* 2015). These same drivers have been reported to strongly influence multifunctionality in global drylands (Delgado-Baquerizo *et al.* 2016). Geographical location was included in our models to control for spatial autocorrelation (Delgado-Baquerizo *et al.* 2013a). In our study, aridity does not represent a lack of available water because soils were watered during incubation. Rather, we included it to illustrate the legacy effects of aridity on soil properties and microbial communities. Microbial drivers and geographical location were included as composite variables in the SEM. The use of composite variables does not alter the underlying SEM model, but collapses the effects of multiple conceptually related variables into a single composite effect, aiding to interpret model results (Grace 2006).

As some of the variables introduced were not normally distributed, the probability that a path coefficient differs from zero was tested using bootstraping. Bootstrapping is preferred to the classical maximum-likelihood estimation in these cases because probability assessments are not based on the assumption that the data conform to a specific theoretical distribution. Bootstrapped data were randomly sampled, with replacement, to derive estimates of standard errors associated with the distribution of the sample data. Following these data manipulations, we parameterised our model and tested its overall goodness-of-fit. There is no single universally accepted test of overall goodness-of-fit for SEM (Schermelleh-Engel *et al.* 2003). We used three metrics to quantify the goodness of fit of our model: (1) Chi-square test (χ^2; the model has a good fit when $0 \leq \chi^2/df \leq 2$ and $0.05 < P \leq 1.00$) (Schermelleh-Engel *et al.* 2003), (2) The root mean square error of approximation (RMSEA; the model has a good fit when $0 \leq RMSEA \leq 0.05$ and $0.10 < P \leq 1.00$) (Schermelleh-Engel *et al.* 2003) and (3) Bollen–Stine bootstrap test (the model has a good fit when $0.10 <$ Bollen–Stine bootstrap *P*-value ≤ 1.00). The different goodness-of-fit metrics used indicate that our *a priori* model was satisfactorily fitted to our data, and thus no post hoc alterations were made.

Finally, to aid interpretation of the SEM, we calculated the standardised total effects (STEs) of geographical location (latitude and longitude), aridity, soil properties (pH and soil total organic carbon) and microbial attributes on MRGC. The STEs, the net influence that one variable has upon another is calculated by summing all direct and indirect pathways between the two variables. If the model fits the data well, the total effect should approximate the bivariate correlation coefficient for that pair of variables.

RESULTS

On average, multifunctionality showed the lowest and highest resistance values to wetting–drying cycles and N fertilisation respectively (Fig. 2b; $P < 0.001$). The resistance of single functions to global change drivers followed similar patterns to those observed for MRGC (Table S1; Fig. S2). Mantel tests revealed that the more similar the microbial communities between two sites, i.e. the more similar their β-diversity, the more similar their functional resistance to warming, wetting–drying cycles and N fertilisation is (Fig. 3; $P < 0.05$). Interestingly, we also found significant positive relationships among multifunctionality resistance to warming and to wetting–drying cycles and N fertilisation (Fig. S2; $P < 0.05$). Conversely, we failed to find any significant relationship between the richness of fungi and bacteria and MRGC (Table S2). The abundance of bacteria was positively related (Spearman $\rho = 0.26$; $P = 0.05$) to multifunctionality resistance to warming (Table S2).

In general, the composition of fungi and bacteria were selected over other microbial drivers as the main predictors of MRGC (Fig. S4). We found that a relatively small proportion of bacterial and fungal taxa (2–10%) were major drivers of MRGC in our studied drylands (Fig. S4). Microbial attributes selected by Random Forest analyses as major predictors of MRGC were also significantly correlated with the resistance of single functions to the global change drivers evaluated (Table S3). The fungal: bacterial ratio was never selected as a major predictor of MRGC by our Random Forest models. Even so, we still found a positive correlation between this ratio and the resistance of particular functions such as nitrate (Spearman $\rho = 0.27$; $P = 0.04$) and carbohydrate availability (Spearman $\rho = 0.23$; $P = 0.08$).

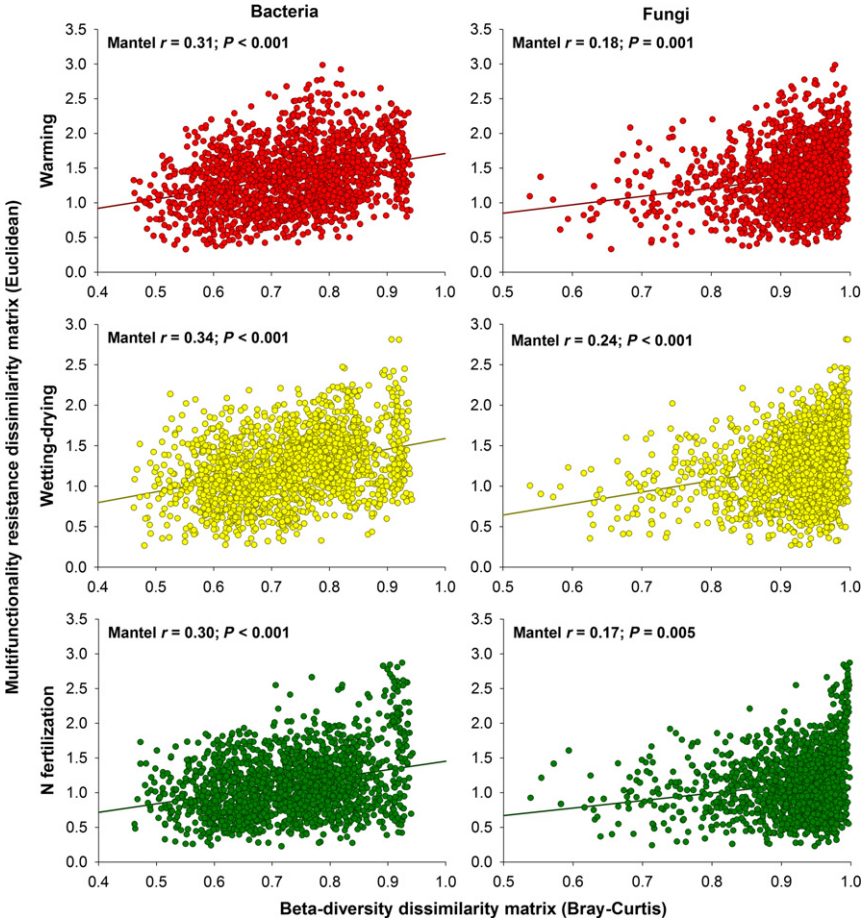

Figure 3 Relationship between community dissimilarity for community composition of bacteria and fungi and multifunctionality resistance to warming, wetting–drying cycles and N fertilisation in soils from global drylands. The solid lines represent the fitted linear regressions.

Our SEM analyses provided further evidence that microbial taxa can have both positive and negative effects on MRGC via direct effects and that these effects are maintained after accounting for important drivers of soil microbial communities and ecosystem multifunctionality (Fig. 4; Appendix S1; Table 1). For example the relative abundance of class Saprospirae (Bacteroidetes) was negatively related to the resistance of multifunctionality and labile C availability to warming (Fig. 4 and Tables 1 and S3). Conversely, the relative abundance of the classes Solibacteres and Spartobacteria (phyla Acidobacteria and Verrucomicrobia) were both positively related to the resistance of multifunctionality and starch degradation to drying–wetting cycles and warming respectively (Fig. 4, Table 1; Appendix S1). Selected examples of specific effects from microbial taxa on MRGC are given in Table 1 and explained in detail in Appendix S1.

We also found that, compared with geographical location, soil carbon and aridity, only pH had a consistently net positive effect on MRGC (Fig. 4). This was an indirect effect driven via changes in the soil microbial composition induced by this variable (Fig. 4). For example pH had a negative direct effect on the relative abundance of Spartobacteria and Saprospira, which were both negatively related to multifunctionality resistance to warming (Fig. 4; Table 1). Moreover, soil pH had a positive effect on the class Gitt-GS-136, which promotes multifunctionality resistance to drying–wetting cycles, and negatively related to the class Solibacteres, which reduced multifunctionality resistance to wetting–drying cycles (Fig. 4; Table 1). Finally, pH had a positive effect on the relative abundance of class Fibrobacteria, which increased the resistance of multifunctionality to N fertilisation (Fig. 4; Table 1).

DISCUSSION

Our study provides strong evidence for a link between the composition of bacterial- and fungal-communities and multifunctionality resistance to warming and fertilisation in dryland soils from across the globe. Most importantly, we identified particular microbial taxa that are likely to be major drivers of the resistance of multifunctionality to these major global change drivers. In the short-term – while improvements in microbial isolation and culturing techniques take place, our results suggest that MRCG could be promoted by altering soil properties such as pH, a major driver of microbial community composition (Fierer & Jackson 2006; Lauber *et al.* 2009). Notably, multifunctionality had a lower resistance to wetting–drying cycles than to warming or N deposition. This is an interesting point, as we should expect that wetting–drying

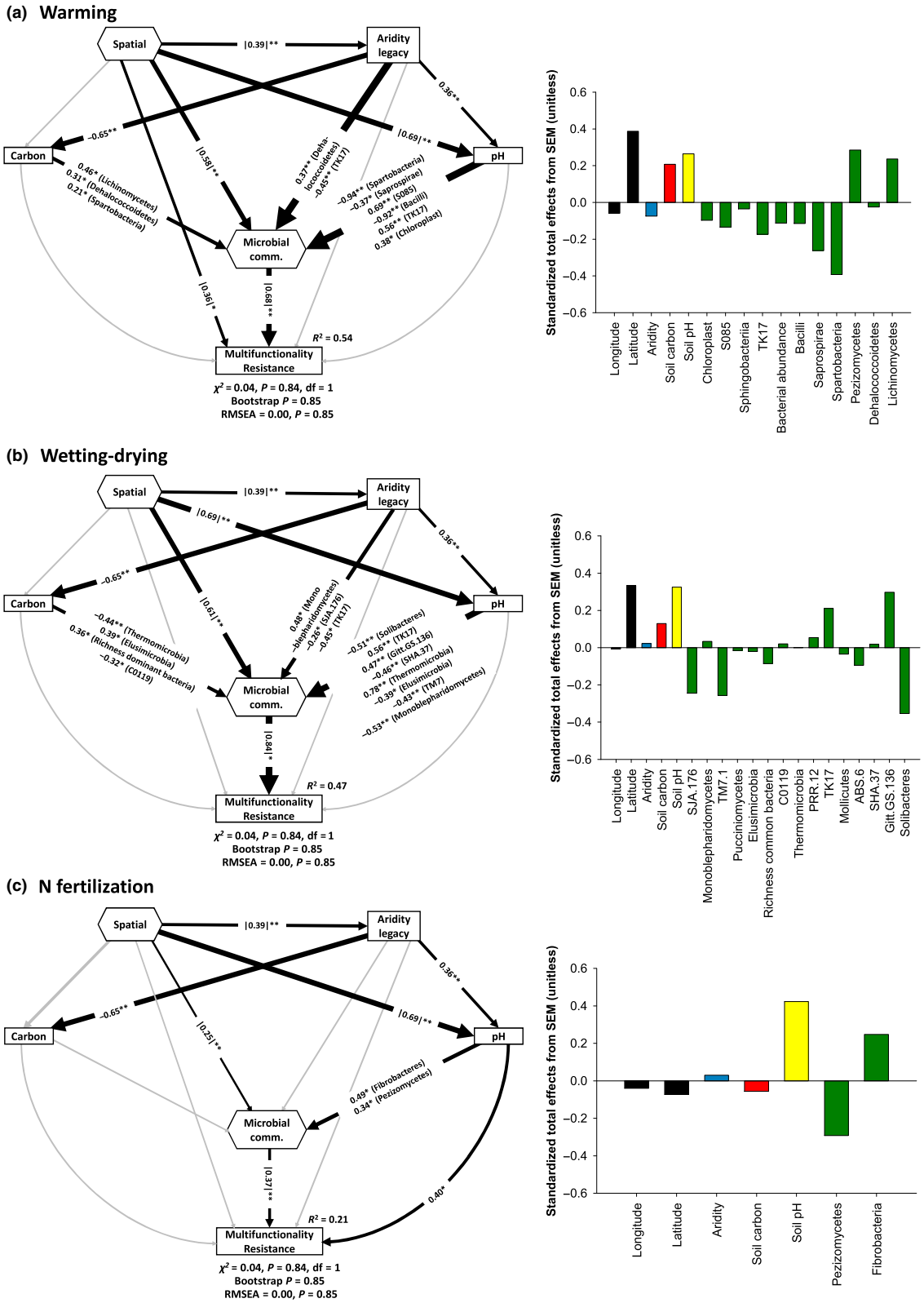

Figure 4 Structural equation model describing the effects of multiple drivers on multifunctionality resistance to warming, wetting–drying cycles and N fertilisation. Numbers adjacent to arrows are indicative of the effect size of the relationship. For simplicity, only significant direct effects are plotted ($P < 0.05$; see *a priori* model in Fig. S1). ($P < 0.05$; see *a priori* model in Fig. S1). Brackets include information of the particular taxa related to multifunctionality resistance to global change. R^2 denotes the proportion of variance explained. Significance levels of each predictor are $*P < 0.05$, $**P < 0.01$. Bar graphs include total standardised effects (sum of direct and indirect effects) from SEM on multifunctionality resistance to warming, wetting–drying cycles and N fertilisation. Grey lines represent tested, but not significant, paths.

Table 1 Selected examples of the positive and negative effects of differential microbial drivers on the resistance of multiple and single ecosystem functions

Global change driver	Microbial driver	Effect	Function	Microbial trait
Temperature	Lychinomycetes		Multifunctionality, NH_4^+ availability, P mineralisation	Ascomycota. Dominant phylum in dry environments. Highly adapted to extreme temperatures conditions (physical protection)
	Pezizomycetes		P mineralisation	
	Spartobacteria		Starch degradation (+) Multifunctionality, Chitin degradation & P mineralisation (−)	Verrucomicrobia –Saccharolytic. Oligotroph: slow C cycling.
	Saprospirae		Multifunctionality, Labile C availability, Chitin degradation & P mineralisation	Bacteroidetes – copiotroph: fast C cycling.
Wetting–drying cycles	Gitt-GS-136		Multifunctionality, Labile C availability, cellulose and chitin degradation & NH_4^+ and NO_3^- availability (+) Starch degradation (−)	Chloroflexi – Prefer dry to humid ecosystems. Structural adaptations to desiccation. Resistant– life strategy vs. wetting–drying cycles. Slow-growing bacteria.
	TK17		Multifunctionality, NH_4^+ availability (+) & Starch degradation (−)	
	Solibacteres		Starch degradation (+), Multifunctionality, chitin degradation & NH_4^+ and NO_3^- availability (−)	Acidobacteria – Prefer humid to dry ecosystems. Oligotroph: slow C cycling. May need to immobilise/release large amounts of N (in osmolytes) to survive desiccation
N fertilisation	Fibrobacteria		Multifunctionality, NH_4^+ and P availability	Obligatory anaerobic. Slow-growing bacteria in dry conditions.
	Pezizomycetes		Multifunctionality, Starch degradation	Dryland fungi – N use efficiency. Use N to produce C degradation enzymes.

This table is derived from results in Fig. 4 and Table S3. An extended version of these examples with further explanations is available in Appendix S1.

cycles are the disturbances that these dryland soils are more likely to be adapted to. However, our results accord with the largely accepted notion that water availability is the principal driver of ecosystem functioning in drylands (Maestre *et al.* 2012). It further indicates that more intense wetting–drying cycles will reduce MRGC in drylands worldwide (Evans & Wallenstein 2014). Overall, our work provides new insights into the importance of microbial composition for buffering the negative effects of global change drivers.

Interestingly, we also detected significant positive relationships between multifunctionality resistance to warming and to wetting–drying cycles and N fertilisation, suggesting some commonalities in the processes driving MRGC across the globe (Fig. S3). The importance of soil microbial communities as drivers of multifunctionality is supported by a number of small-scale experiments showing that total abundance of microbes controls the resistance of particular functions such as soil respiration or N mineralisation to drought (Downing & Leibold 2010; De Vries *et al.* 2012; de Vries & Shade 2013). However, to the best of our knowledge, our results provide the first empirical evidence, based on experimental manipulation, that microbial community composition and multifunctionality resistance are linked at the global scale. Our findings indicate, therefore, that microbial community composition can be critical for maintaining MRGC, and that changes in this composition resulting from land use intensification (Gossner *et al.* 2016) or climate change (Maestre *et al.* 2015) will likely alter the resistance of critical ecosystem functions to global change drivers in drylands across the globe.

Our Random Forest analysis allowed us to identify particular microbial taxa (class level) as major predictors of MRGC over other microbial attributes such as abundance, diversity and fungal: bacterial ratio. In particular, we found that a relatively small proportion of bacterial and fungal taxa (2-10%) were major drivers of MRGC. These included specific classes within phyla *Verrucomicrobia*, *Bacteroidetes*, *Chloroflexi*, *Acidobacteria*, *Firmicutes* and *Ascomycota*, which are globally distributed (Ramirez *et al.* 2014; Maestre *et al.* 2015). The same microbial taxa were also correlated with the resistance of single functions to global change (Table S3). These results imply that different microbial drivers govern multifunctionality and MRGC in dryland soils worldwide. Thus, while multifunctionality *per se* is likely to be driven by multiple microbial attributes (Appendix S2; Figs. S4 and S5), the effects of microbial attributes on MRGC are mostly limited to those from microbial composition via key microbial taxa. These results are consistent with novel soil ecological theories suggesting that key microbial taxa may control the resistance of soil functioning to global change (de Vries & Shade 2013). Conversely, we failed to find any significant relationship between abundance and richness (rare and common species) of fungi and bacteria and MRGC. Similarly, our results further suggest that the fungal:bacterial ratio, previously suggested to be a major predictor of ecosystem functions (De Vries *et al.* 2012), may be a poor predictor of MRGC. Note that, unlike De Vries *et al.* (2012), we used a qPCR approach to calculate the fungal: bacterial ratio. Thus, we would like to acknowledge that the use of different

methods might also partially explain differences between De Vries *et al.* (2012) and our results. Nevertheless, we still found a positive correlation between this ratio and the resistance of particular functions such as nitrate, a proxy for nitrification rates and carbohydrate availability. This finding supports results of a previous study demonstrating strong relationships between the fungal:bacterial ratio, and both N mineralisation and soil respiration (De Vries *et al.* 2012).

Our SEM revealed a direct and significant relationship between the composition of microbial communities and MRGC after accounting for multiple drivers of this resistance. These results further support the notion that key microbial taxa play critical roles in supporting MRGC in dryland soils worldwide. We found that different microbial taxa were involved in the multifunctionality resistance of each global change factor. Given that multiple global change drivers will occur simultaneously, our results suggest that preserving the diversity of soil microbial communities may be crucial to sustain the provision of ecosystem services in the future. Furthermore, we found both direct positive and negative effects from particular taxa on MRGC. We argue that many of the effects can be understood by drawing on our current knowledge of soil microbial communities. Of special interest is the role that microbial life strategy (i.e., r- vs. k- strategists) might play in driving MRGC, with special references to C cycling (de Vries & Shade 2013). For example the relative abundance of class Saprospirae (Bacteroidetes), classified as r-strategist or copiotrophs (Fierer *et al.* 2007) directly and negatively affected multifunctionality resistance and labile C availability resistance to warming, presumably due to their rapid growth. Conversely, the greatest net negative effect of a microbial taxon on the resistance of multifunctionality (i.e. to wetting–drying cycles) came from Solibacteres (Fig. 4; Table 1), which was positively related to functions associated with the C cycle (e.g. starch degradation) but negatively related to functions from N cycle (e.g. chitin degradation and N availability; Table S3). The positive effect of Solibacteres on the resistance of labile C mineralisation is consistent with results from previous studies suggesting that oligotrophic communities (*sensu* Fierer *et al.* 2007; Trivedi *et al.* 2013) promote the resistance of functions related to C cycle (de Vries & Shade 2013). The negative effect of class Solibacteres may be related to the necessity of certain bacteria to immobilise/release large amounts of N in osmolytic forms to survive desiccation in response to wetting–drying cycles (Schimel & Bennett 2004; Tables 1 and S3; de Vries & Shade 2013). The resistance of starch degradation appears to behave differently to the other functions. Thus, microbial taxa that are positively correlated with the resistance of starch degradation seem to be negatively correlated with the resistance of other functions. This intriguing result suggests that C preferences from microbial communities (labile vs. more recalcitrant) might influence the resistance of particular ecosystem functions to global change drivers.

Our SEM analyses further suggested that by adjusting soil pH we could potentially unleash the positive effects of microbial community composition on MRGC. Thus, pH was the only environmental predictor having a consistent net positive effect on MRGC either by suppressing or promoting taxa that were negatively (Spartobacteria, Saprospira and Solibacteres) and positively (Gitt-GS-136 and Fibrobacteria) related to MRGC, respectively. The importance of soil pH as a major driver of the composition of bacterial and fungal communities in terrestrial ecosystems is well known (Fierer & Jackson 2006; Lauber *et al.* 2009). However, our study provides evidence, for the first time, that soil pH also indirectly regulates the effects of microbial community composition on MRGC. These results have implications for the understanding and management of MRGC in the field, as they suggest that we could still potentially increase MRGC by changing soil pH, thereby driving the composition of soil microbial communities in a specific direction. Future endeavours exploring the role of microbial composition in driving multifunctionality resistance may further test this hypothesis using experimental approaches including soil pH manipulations.

Altogether, we found a strong link between soil bacterial and fungal communities and MRGC in soils from global drylands. Our results suggest that key microbial taxa, rather than the richness, abundance and the ratio of bacteria and fungi, control MRGC. They also point to the potential role that manipulations in soil pH could have to buffer negative effects of global change drivers on multifunctionality resistance. Our findings imply that climate- and/or management-induced changes in the composition of soil bacterial and fungal communities may alter multifunctionality resistance, with concomitant effects on the provision of key ecosystem services than rely on them.

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AUTHORSHIP

MD-B conceived this study and designed experiments. MD-B, DJE, VO and BG were involved in the collection of the soils used in this study, under the coordination of FTM and DJE. Laboratory analyses were done by VO, BG and MD-B. BKS provided the Illumina data and bioinformatics analyses. MD-B conducted statistical modelling. The manuscript was written by MD-B, with contributions from FTM, DJE and BKS.

DATA ACCESSIBILITY STATEMENT

The primary data have been deposited in figshare: https://figshare.com/s/8892a0ab3cfff186458e (https://doi.org/10.6084/m9.figshare.5089942). The raw sequence data have been deposited in the GenBank SRA database (BioProject accession no. PRJNA301533).

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