

The Biogeography of Dust in the Eastern Mediterranean

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Abstract

The atmospheric microbiome, the aerobiome, represents a complex mixture of suspended microorganisms originating from different environments, *e.g.*, soil, marine environments, and plants. Aerobiomes sampled around the globe show distinct compositions, yet some common features might be observed. Defining these similarities and differences will contribute to our understanding of this understudied environment. In this study, we sequenced the metagenomes of atmospheric dust collected in Israel, representing different particulate matter (PM) concentrations, dust origins and atmospheric trajectories. The metagenomes were compared to publicly available atmospheric metagenomes, as well as to marine and soil metagenomes from the Eastern Mediterranean region. This study helps elucidate the role of dust storms in replenishing the atmosphere with bacteria, fungi and archaea, which facilitate essential biogeochemical processes, by investigating microbiomes and functional genes from dust metagenomes. Our results reveal a high level of similarity between metagenomes of dust sampled in Israel and metagenomes of dust collected in Saudi Arabia asynchronously. Moreover, by identifying taxa and genes that are correlated with PM concentrations, we suggest a possible "core" atmospheric microbiome composed mainly of fungi. Application of the SourceTracker algorithm revealed an inverse correlation between the "core" aerobiome and the fraction of soil-derived bacteria and archaea in the dust metagenomes. Marine microbiomes contributed very little to the dust microbiome, even in dust that traversed the Mediterranean and Red Seas. The dust metagenomes showed a high relative abundance of genes associated with antibiotic resistance, sporulation and the ability to degrade aromatic pollutants, which could not be traced to any of the natural microbiomes that were examined. This study represents an important step toward integrating the knowledge accumulated on the global aerobiome and reveals the important role of dust storms in contributing to the presence of various environmentally relevant microbial functions.

Introduction

For the past decade, data on global airborne microbial communities have slowly accumulated¹⁻⁷. In these studies, aerosols and airborne bacteria were collected using various sampling methods, covering different aerosol sizes and types, and were genetically analyzed by different DNA sequencing platforms. This effort has created a growing knowledge base on the microbial community in the atmosphere and has provided a few clues regarding the variability, viability and functionality of this community.

However, many questions remain unanswered; although the studies were performed at different locations around the globe (*e.g.*, Europe and the Alps,⁸⁻¹⁰ Asia,^{2,11-13} the Mediterranean,^{7,14} North America,^{15,16} South America,¹⁷ the Amazon rainforest,¹⁸ and Antarctica¹⁹), little is known about the residence time of microorganisms in the atmosphere; the connectivity of microbial communities between regions; the presence of a "core" microbiome in the atmosphere; anthropogenic effects on the atmospheric microbiome; and the role played by the atmosphere in the introduction of new bacteria and functions to different environments, and therefore about the global dissemination of pathogens and antibiotic resistance genes. Finally, the viability and functionality of airborne microorganisms in different niches

within the atmosphere, such as clouds vs. dust storms, remain unknown. These questions are of great importance for understanding the global microbiome and could be essential for global ecosystem functioning, human health and the climate.^{20,21}

To date, most atmospheric microbiome studies have applied amplicon sequencing of a taxonomic proxy, such as the gene encoding the small subunit (SSU) of the ribosome, *i.e.*, 16S rRNA or 18S rRNA for prokaryotes and eukaryotes, respectively. This approach enabled sequencing despite the relatively low biomass collected from air samples and provided important insight into the microbial community structure. It also provided some hints about the possible functionality of these microorganisms. However, only a limited number of studies have succeeded in collecting sufficient material to obtain the DNA concentrations required for metagenomic and metatranscriptomic sequencing, including aerosols from anthropogenic environments such as farms and cities^{13,22-24} and natural environments such as clouds⁸ and dust aerosols³. Each of these studies explored different aspects of the atmospheric microbiome, including metabolic processes, environmental epidemiology and microbial viability, in addition to documenting the microbial community structure and functional gene abundance and distribution. However, these rare studies provided only a glimpse into the complexity of the airborne microbiome, *i.e.*, the aerobiome, and its potential functionality.

Israel is at a crossroads of air masses from different sources situated in the Eastern Mediterranean basin in the vicinity of the Sahara, Arabian and Syrian deserts.²⁵ As such, it experiences dust events originating from all these sources and exhibits a relatively high daily average loading of particulate matter (PM), with the concentration of particles under 10 μm (PM₁₀) averaging approximately 30–50 $\mu\text{g}\cdot\text{m}^{-3}$.²⁶ Dust sampled in Israel has been shown to display significant source variability, with distinct signatures for local aerosol and desert dust microbiomes.^{4,7} This variability enabled us to differentiate among the contributions of different aerobiome sources: terrestrial, marine and anthropogenic.⁴ We assume that dust storms in the Middle East, as well as a significant portion of the general aerosols in this region, originate from soil and are perhaps affected to some extent by the Mediterranean Sea and the Red Sea and by anthropogenic sources such as farms, industrial sites and cities. Under this assumption, we conducted a comparative analysis of shotgun metagenomes of dust samples collected at a single location in Israel, covering different atmospheric conditions, with publicly available atmospheric, soil and marine metagenomes, addressing differences and similarities in both taxonomic and functional compositions.

Methods And Materials

Aerosol sampling:

Particulate matter was collected on the roof of a four-story building at the Weizmann Institute campus in Israel (31.9070N, 34.8102E) using a high-volume sampler (Ecotech, model: HiVol 3000, PM₁₀ inlet, flow rate: 67.8 $\text{m}^3\cdot\text{h}^{-1}$) onto prebaked quartz fiber filters (Whatman, 203 mm \times 254 mm) for 6 to 48 h at a

time, depending on the load of particulate matter with an aerodynamic diameter under 10 μm (PM_{10}). The filters were then wrapped in sterile aluminum foil and kept at -20°C until DNA was extracted. The samples are described in Table 1. The source of each sample was determined using the hybrid single-particle Lagrangian integrated trajectory model (HYSPLIT) via a web-based interface (READY, http://ready.arl.noaa.gov/HYSPLIT_traj.php). The dust sources were also verified using the Giovanni online data system, developed and maintained by the NASA GES DISC.²⁷ Back trajectories for all sampling events detailed in Table 1 are presented in Table S1.

DNA extraction:

DNA was extracted using a DNA PowerSoil Kit (Qiagen, Germany), with modifications to the manufacturer's protocol, as follows: four strips, 30 cm^2 each, were cut from each filter using a surgical scalpel, and the upper layer of each strip was scratched off and inserted into a single PowerSoil bead tube, to which 250 μl of PBS and 150 μl of a phenol:chloroform:isoamyl solution (ratio 25:24:1) were added. Next, the tubes were shaken in a Mini-Beadbeater-16 (Biospec, USA) for 2 minutes, removed and placed on ice for 15 seconds and then shaken for an additional 2 minutes. Extraction was continued according to the protocol up to the membrane binding step. In this step, the contents of the four tubes were loaded onto two spin filter tubes. Next, the samples were washed twice with C5 solution to rid the DNA of all traces of phenol and chloroform. Elution was conducted by loading 50 μl of solution C6 on one tube, leaving it for 5 minutes at room temperature, centrifuging for 1 minute at 16,000 $\times g$, loading the eluent onto the second spin filter, and repeating the described elution step.

Following extraction, samples with similar characteristics (PM_{10} concentration and source of aerosols according to back-trajectories and satellite images) were pooled together (see Table 1), and DNA was precipitated using 100% ethanol and 5 M NaCl solution. The precipitated DNA was resuspended in 25 μl of TE buffer. We thus created a set of nine samples, five representing two major dust events originating in Syria and in the Arabian Desert, three composite samples representing minor to moderate dust events from the Sahara, Sinai and Jordanian Deserts and a single composite sample representing days with low PM_{10} concentrations. Pooling of samples was necessary due to low extracted DNA concentrations.

Table 1
 Sample description of dust collected in Israel. Samples Dust_2 to Dust_5 were created by pooling DNA from several sampling events.

| Sample | Aerosol origin | Sampling date | PM ₁₀ (µg m ⁻³) |
|----------------|----------------------------------|---------------|--|
| <i>Dust_2</i> | Local | 7/18/2016 | 33 |
| | North Africa | 6/6/2016 | 35 |
| | North Africa | 8/15/2016 | 35 |
| | Mix | 8/31/2016 | 37 |
| | Arabia | 8/26/2016 | 40 |
| <i>Dust_3</i> | North Africa | 1/17/2017 | 75 |
| | North Africa | 5/9/2017 | 84 |
| | North Africa | 1/7/2017 | 96 |
| | North Africa | 11/20/2016 | 109 |
| <i>Dust_4</i> | Mix (North Africa and Arabia) | 11/25/2016 | 121 |
| | North Africa | 1/9/2017 | 144 |
| | North Africa | 3/12/2016 | 150 |
| | Mix (North Africa and Arabia) | 11/24/2016 | 186 |
| <i>Dust_5</i> | North Africa | 2/28/2016 | 209 |
| | Mix (Sinai Peninsula and Jordan) | 11/23/2016 | 232 |
| | North Africa | 4/12/2017 | 118 |
| <i>Dust_6</i> | Syria | 9/8/2015 | 844 |
| <i>Dust_7</i> | Syria | 9/9/2015 | 1859 |
| <i>Dust_8</i> | Syria | 9/10/2015 | 928 |
| <i>Dust_9</i> | Arabia | 11/3/2015 | 422 |
| <i>Dust_10</i> | Arabia | 11/4/2015 | 810 |

Shotgun metagenome sequencing:

Sequencing was conducted at the Genome Research Core of the University of Illinois, Chicago, as follows: library preparation was performed with acoustic shearing using a Swift 1S Kit. The libraries were pooled, followed by Pippin-prep (Sage Science) size selection (375–425 bp insert size). MiniSeq Q/C was

performed to ensure an even distribution of sequencing output. The samples were repooled, followed by repeating Pippin-prep size selection. The samples were sequenced using a HiSeq2500 (Illumina) with 2x250 cycles using a single lane.

Metagenome dataset composition and analysis

Table 2 shows the reference metagenomes that were compared to the dust metagenome collected in this study. Raw and assembled reads were uploaded to the MG-RAST metagenomic analysis server²⁸ and were analyzed using the default parameters for shotgun metagenomic sequencing. Taxonomic data were acquired by comparison to the RefSeq database (NCBI²⁹⁻³¹), and functional genes were annotated against the KO database (KEGG³²⁻³⁴), both available on the MG-RAST server. Taxonomic data were filtered to exclude multicellular organisms as well as viruses. Functional annotation data were filtered to exclude organismal system and human disease categories. All counts were transformed using the center log-ratio (clr) transformation method prior to data analysis. Downstream analysis, statistical analysis and figure preparation were conducted using R v.4.0.3³⁵ and mothur v.1.44.0.³⁶ General data formatting was conducted with tidyverse;³⁷ statistical significance tests for differentially abundant genes and taxa, as well as nonmetric multidimensional scaling (NMDS) ordination, were conducted using Vegan,³⁸ broom³⁹ and purr;⁴⁰ correlation matrices were produced using Hmisc;⁴¹ gene and taxon counts were clr-transformed using the compositions package;⁴² analysis of molecular variance (AMOVA)⁴³ tests were conducted using the *amova* command in mothur; fitting of functional categories, taxa and NMDS ordination axes was conducted using the *corr.axes* command in mothur with the method set to Spearman; plots were produced using the R packages ggplot2,⁴⁴ and Superheat.⁴⁵ The presented q-values are p-values fitted according to Benjamini-Hochberg correction for multiple comparisons.

To compare dust to probable sources, as well as to other atmospheric metagenomes, we divided the different datasets according to their type: “Dust” included the dust samples collected in this study (Table 1) and dust samples collected over the Red Sea by Aalismail *et al.*³; “Smog” represents samples collected during a smog event in Beijing¹³; “Air” includes cloud condensation samples collected in France⁸ and urban aerosol samples from two locations in North America²²; “Soil” represents different soils collected over a precipitation gradient in Israel⁴⁶; and “Sea” includes samples from the Eastern and Western basins of the Mediterranean Sea⁴⁷ and various locations along the Red Sea.⁴⁸ All datasets and sample types are described in Table 2.

Table 2
Metagenomes used for comparison with the dust collected in this study.

| Metagenome dataset | Description | Sample type | Reference and accession no. |
|---------------------|--|-------------|---------------------------------|
| <i>Dust IL</i> | 9 dust samples, Weizmann Institute, Israel | Dust | This study |
| <i>Dust Red Sea</i> | 8 dust samples, Red Sea, off the shore of Saudi Arabia | Dust | ENA, PRJEB31563 ³ |
| <i>Beijing Smog</i> | 7 samples, a single smog event in Beijing, China | Smog | mgp3756 ¹³ |
| <i>Clouds</i> | 3 cloud samples, Puy-de-Dome, France | Air | NCBI, PRJEB25763 ⁸ |
| <i>Urban Air</i> | 4 samples, urban aerosols, USA | Air | NCBI, PRJNA218551 ²² |
| <i>IL Soils</i> | 12 soil samples, different locations in Israel | Soil | mgp14608 ⁴⁶ |
| <i>Red Sea</i> | 4 Red Sea water samples, different locations | Sea | mgp20413 ⁴⁸ |
| <i>Med Sea</i> | 6 Mediterranean Sea water samples, different locations | Sea | mgp20252 ⁴⁷ |

Results

Community structure and functional profile

Figure 1 presents NMDS ordination based on taxonomy (1a) and functional genes (1b) correlated with microbial genera or KEGG pathways. We divided the samples into three categories: atmosphere, including all atmospheric sample types, *i.e.*, dust, air and smog; marine, for the sea sample type; and terrestrial, for the soil sample type. All presented taxa and pathways were significantly correlated (Spearman's correlation, p -value < 0.05) with either the MDS1 or MDS2 axis and displayed a vector length greater than 0.8. AMOVA results indicated that all five sample types presented, *i.e.*, air, dust, sea, smog and soil, differed significantly from one another (q -value < 0.0005), both taxonomically and functionally. However, when comparing the different datasets, urban air and clouds did not significantly differ in their functional gene profiles according to the AMOVA test (q -value = 0.058). All other datasets differed significantly in their community composition as well as functional gene profile (Tables S2 and S3).

According to Fig. 1a, the variance along the MDS1 axis was correlated with the abundances of the fungal phylum Ascomycota, the bacterial phylum Proteobacteria and the archaeal phylum Euryarchaeota. The variance along axis MDS2 was affected by the abundances of Actinobacteria and Firmicutes.

NMDS functional ordination, based on KEGG annotated genes, did not differ much from that based on taxonomy, yet it displayed a greater variance between the different sample types in the marine and atmospheric environments (Fig. 1b). Screening against the KEGG pathway database revealed that the variance along MDS1 was correlated with the abundance of metabolic pathways compared with the abundance of genetic information processes.

In both ordinations, dust and smog samples clustered between soil and air sample types, suggesting that these microbiomes are composed of microbial components from both air and soil. Marine microbiomes did not seem to contribute microbial components to the dust, smog, soil and air samples. To test this hypothesis, we assessed the source apportionment of soil, air and sea microbiomes in the dust samples using SourceTracker⁴⁹ (Fig. 2). Beijing smog was not included in this analysis since it did not form an end member on either taxonomic or functional ordination. As expected, over 60% of the dust storm samples, *i.e.*, *Dust_6* to *Dust_10*, were predicted to be soil derived. The highest air contribution was observed for samples *Dust_2* to *Dust_5* and samples *Dust_9* and *Dust_10*, with a range of 3.8–12.0% microorganisms resembling those in air samples. In all samples, including those sampled over the Red Sea, the contribution of sea microorganisms was at most 3.1%.

Functions and taxa correlated with PM₁₀ concentrations in dust

To further understand the contribution of soil microorganisms to the dust microbiome, we performed correlation analysis (Spearman's correlation⁵⁰) between PM₁₀ concentrations and taxa or functional genes with clr-transformed counts of *Dust_IL* samples. Assuming that taxa and genes that were positively correlated with PM₁₀ are likely to originate from soil, especially in the Mediterranean area, whereas negatively correlated pathways and genera originated from unknown sources, we could distinguish between the local, terrestrial contribution to the dust aerobiome and the taxa and functional genes that were common to all atmospheric samples examined here. The taxa and functional genes that displayed a significant correlation with PM₁₀ concentrations, Spearman's $|r| > 0.75$, and $q\text{-value} < 0.05$ were then further clustered (k-means) based on clr-transformed counts in all datasets examined in this study, with the optimal number of clusters determined by the total within-sum value of squares. The list of significantly correlated taxa and genes is presented in Tables S4a and b, along with their clusters.

PM₁₀-correlated genera

A total of 137 genera were negatively correlated with PM₁₀ concentrations. Most of these genera were bacterial (80 genera), a significant amount were fungal (56 genera) and only one was archaeal. The genera that were negatively correlated with PM₁₀ concentrations were of the phyla Proteobacteria, Firmicutes, Bacteroidetes and Ascomycota. These genera formed 6 clusters, as presented in Figs. 3 and S1. Clusters 1, 2 and 6 represent fungal genera that were more abundant in atmospheric samples, *i.e.*, clouds, urban air, Beijing smog, Israeli dust and Red Sea dust. They consisted entirely of fungi, mostly Ascomycota. Cluster 1 represents genera that were found only in atmospheric samples, and very often in all of them; these genera were absent or rarely found in most soil and marine samples, suggesting that

their main source was not local soils or seas. This cluster consisted of the following 13 fungal genera: *Allomyces*, *Cordyceps*, *Dissoconium*, *Epidermophyton*, *Glomus*, *Hypocrea*, *Lecanicillium*, *Metarhizium*, *Millerozyma*, *Mycosphaerella*, *Phycomyces*, *Rhizopus* and *Tilletia*. Cluster 2 genera were found in some marine samples, as well as atmospheric samples; accordingly, they included some marine fungi, e.g., *Debaryomyces* and *Meyerozyma*. The fungi included in cluster 6 were found in all examined environments but were relatively more abundant in atmospheric samples than in marine and soil samples. These included *Neosartorya*, *Aspergillus*, *Gibberella*, *Phaeosphaeria* and *Penicillium* (Table S4a). Some of these pathogens are animal and human pathogens (e.g., *Neosartorya* and *Aspergillus*), and some are plant pathogens (e.g., *Gibberella*).

Cluster 5 included Bacteroidetes, Firmicutes and Gammaproteobacteria taxa that are ubiquitous in soil and aquatic ecosystems, and clusters 3 and 4 were composed almost entirely of Firmicutes, which were less abundant in urban air and clouds. The latter three clusters included almost solely bacterial genera. A single archaeon, *Methanobrevibacter*, a methanogen often found in the digestive systems of a diverse array of animals, ranging from ruminants to termites⁵¹, was found in cluster 4. Many of the bacteria included in these clusters were human or animal commensals or pathogens, such as *Escherichia*, *Bacteroides*, *Helicobacter*, *Histophilus*, *Salmonella* and *Enterobacter*. In general, more human-associated bacteria and more fungi were linked to low PM₁₀ concentrations than to high PM₁₀ concentrations, suggesting that they were from local sources.

Most of the 321 genera that were positively correlated with PM₁₀ concentrations were bacteria (260 genera), some were archaea (52 genera), 6 were algae and 3 were fungi (Figs. 4 and S1). Proteobacteria was the most prevalent phylum of bacteria (126 genera), with genera such as *Burkholderia*, *Bradyrhizobium*, *Rhodopseudomonas*, *Sinorhizobium* and *Roseobacter* that are ubiquitous in soil and marine environments. Many actinobacterial genera were also found to be positively correlated with the PM₁₀ level, including mesophilic and thermophilic terrestrial-associated genera such as *Thermomonospora*⁵², *Frankia*⁵³, *Thermobispora*⁵⁴, and *Conexibacter*⁵⁵ and halotolerant, radiation-tolerant and desiccation-tolerant genera such as *Kineococcus*^{56,57} and *Geodermatophilus*.⁵⁸⁻⁶⁰ This was also true for specific extremophile archaeal genera such as *Aeropyrum*, *Methanothermococcus* and *Desulfurococcus*.

Genera that were positively correlated with PM₁₀ concentrations in *Dust IL* formed three distinct clusters (Figs. 4 and S1). Most of the positively correlated genera were less abundant in cloud and urban air samples than in all other samples. Cluster 1, consisting of 188 genera, represents genera that were generally highly abundant in all samples but to a lesser extent in cloud and urban air. This cluster included only bacteria, mostly from the Proteobacteria and Actinobacteria phyla, e.g., *Streptomyces*, *Mycobacterium*, *Burkholderia*, *Frankia* and *Arthrobacter*. Many of these bacteria are soil bacteria, and some are extremophiles.

Cluster 2, consisting of 124 genera, included a majority of the archaea that were positively correlated with PM₁₀ concentrations, i.e., 50 archaea out of 52 archaeal genera. Many of these species were

extremophiles, *e.g.*, thermophilic *Aeropyrum* and *Thermoplasma* and halophilic *Halalkalicoccus*. Methanogens such as *Methanopyrus* were also found. Bacteria in this cluster were mostly of the *Proteobacteria* and *Firmicutes* phyla. This cluster represents genera that were found in dust, soil and marine samples but were rare in smog, cloud and urban air samples. Thus, this cluster might best represent the contribution of desert soil microbiomes to the dust aerobiome.

Last, cluster 3 represents 9 genera that were rarely found in all samples. Only three of these genera were bacteria: *Fluoribacter* (*Legionella*), which contains species that often reside in protozoa, and some that can cause Legionellosis⁶¹; the freshwater cyanobacterium *Leptolyngbya*; and *Candidatus Hodgkinia*, a cicada endosymbiont. Three other genera were of the algal phylum Chlorophyta, and three were fungi.

PM₁₀-correlated functional genes

The same analysis was applied to functional annotation, *i.e.*, KEGG annotation. The results are presented in Figs. 5, 6 and S2. A total of 1516 genes were positively correlated with PM₁₀ concentrations, compared with only 565 genes that were negatively correlated with PM₁₀ concentrations. Most of the positively correlated genes, *i.e.*, 985 genes, were related to metabolism, whereas most of the negatively correlated genes were related to genetic information processing (278 genes). The negatively correlated genes included many eukaryotic genes and pathways, such as ribosome biogenesis in eukaryotes (ko03008), RNA transport (ko03013), mRNA surveillance pathway (ko03015), spliceosome (ko03040) and cell cycle - yeast (ko04111).

In general, the functional profile of genes correlated with PM₁₀ concentrations was in accordance with the community composition: low PM₁₀ concentrations were accompanied by increased fungal abundance and eukaryotic functional genes, whereas increasing PM₁₀ concentrations associated with desert dust storms resulted in increasing bacterial and archaeal taxon and gene abundance.

Functional genes that were negatively correlated with PM₁₀ concentrations formed four distinct clusters (Figs. 5 and S2); clusters 1 and 2, comprising 377 genes, represent genes that were usually more abundant in clouds, urban air, Beijing smog and most Israeli dust samples, except for *Dust_6* to *Dust_8*, and less abundant in Israeli soils and Mediterranean and Red Sea samples. These included mostly eukaryote-specific genes, such as those related to cell cycle - yeast (ko04111) and meiosis - yeast (ko04113), as well as prokaryotic genes encoding the transport of trehalose, sucrose, maltose galactitol, lactose and cellobiose via the phosphotransferase system pathway (ko02060). Stress adaptation genes were also found in these clusters: SSK1 (K11233), SOH1 (K11246) and YPD1 (K11232) were related to osmotic stress in fungi; *bceA* and *bceB* (K11631, K11632) were associated with bacterial resistance to bacitracin; and *vraS* (K07681) was associated with bacterial resistance to the antibiotic vancomycin.

Clusters 3 and 4 represent genes that were found in all samples but were usually more abundant in clouds, urban air, Beijing smog and most samples of Israel dust (except for high PM₁₀ samples *Dust_6* to *Dust_8*). Cluster 3 was dominated by metabolism-associated genes, whereas cluster 4 was dominated by genetic information processing-associated genes. These clusters included eukaryotic genes related to

pathways such as endocytosis, phagosome and RNA transport. In addition, prokaryotic genes encoding the transport of mannose, cellobiose, and beta-glycoside via the prokaryotic phosphotransferase system pathway (ko02060). The *spo0A* gene, which is associated with sporulation, especially in *Bacillus*, and the SLN1 fungal gene, which is associated with osmotic stress, were also found in cluster 3.

Genes that were positively correlated with the PM₁₀ level formed 3 clusters (Fig. 6 and S2). Cluster 1 comprised 717 genes that were highly abundant in all samples but less abundant in the cloud and urban air samples. Many of these genes were associated with general functions, *e.g.*, ribosome (ko03010), DNA replication (ko03030), and aminoacyl-tRNA biosynthesis (ko00970). They also included many bacterial- and archaeal-specific genes, such as genes involved in the bacterial secretion system (ko03070), flagellar assembly (ko02040), bacterial chemotaxis (ko02030), and peptidoglycan biosynthesis (ko00550).

Cluster 3 comprised 604 genes that were found in most samples, excluding clouds and urban air, but to a lesser extent than genes included in cluster 1. Many of these genes were associated with xenobiotics biodegradation and metabolism *e.g.*, chlorocyclohexane and chlorobenzene degradation (ko00361) and benzoate degradation (ko00362). ABC transporter genes and two-component system genes were also abundant in this cluster, with 85 and 45 genes, respectively.

Finally, cluster 2, comprising a total of 197 genes, represented genes that were rare in most samples. These included mostly metabolism-associated genes, *e.g.*, amino acid metabolism and carbohydrate metabolism genes, as well as genes associated with environmental information processing, *e.g.*, two-component system and ABC transporter genes.

The genes that were positively correlated with PM₁₀ were usually similarly abundant in most samples, excluding cloud and urban air; thus, pathways that were highly abundant in Israeli dust were also highly abundant in Israeli soils, Mediterranean and Red Sea water, Red Sea dust and Beijing smog. This implies that these pathways are common to bacteria everywhere and that their low abundance in clouds and urban air is in accordance with the significantly higher abundance of fungi than of bacteria and archaea in these samples. No unique contribution of the marine microbiome to dust collected in Israel or over the Red Sea was observed in this analysis.

Functional differences between dust and other environments

We assigned KEGG gene annotations to environmentally relevant functional categories (Table S5)^{62,63} and calculated their relative abundances as percentages of the total gene counts. These functional categories were specifically chosen since they do not include general housekeeping genes but are usually unique to specific environmental functions such as metabolism and adaptation to stress. A full description of the abundances of the selected genes and functional categories can be found in Table S6. Previous studies have suggested that the adaptation of airborne microorganisms to atmospheric conditions would benefit from the presence of functions such as UV resistance, osmotic stress resistance, envelope stress resistance, sporulation, methane oxidation, formate degradation, acetic acid degradation and oxidative stress resistance.^{3,8,64,65} Thus, these studies targeted these functions in metagenomic as

well as metatranscriptomic analyses of different atmospheric samples. Consistent with this, we focused on these functional genes, as well as many other environmentally relevant functional genes, in our analysis to better define the contribution of mineral dust to the potential functionality of the aerobiome.

According to our results, dust samples often differed from soil samples in the composition of the selected functions; out of a total of 58 examined functions, 36 showed significant differences in abundance between soil and dust samples (Kruskal-Wallis test, q -value < 0.05). Dust also differed from air samples, *i.e.*, urban air and clouds displayed 35 functions that were differentially abundant between the dust and air sample types (Kruskal-Wallis test, q -value < 0.05).

Figure 7 shows the relative abundances of chosen functions in the different sample types. Dust samples showed a high relative abundance of antibiotic resistance genes, specifically genes conferring resistance to beta-lactam antibiotics, bacitracin and vancomycin, compared with sea and soil samples. Moreover, dust showed the highest relative abundance of these genes compared with all other atmospheric samples. These genes are often found in *Bacillus* species and other spore-forming bacteria, along with genes associated with sporulation.

Comparison of all atmospheric sample types, *i.e.*, dust, air and smog, suggested that the Beijing smog metagenome had the highest abundance of functions linked with microbial survival in the atmosphere, including temperature, oxidative stress and UV resistance, as well as formate degradation. Dust also exhibited a high relative abundance of relevant functions, such as osmotic stress and envelope stress resistance and sporulation-associated genes. In addition, dust displayed a high relative abundance of antibiotic resistance genes and genes for the biodegradation of aromatic compounds. Clouds and urban air samples exhibited a higher abundance of fungal resistance genes for cell wall stress and starvation, in accordance with their taxonomic composition.

Comparison of the list of functions with PM₁₀-correlated genes revealed that most stress resistance-associated genes were positively correlated with PM₁₀ concentrations, as were genes associated with the degradation of aromatic compounds, whereas fungal genes associated with stress resistance were negatively correlated with PM₁₀ concentrations. Antibiotic resistance genes displayed a mixed trend, where bacitracin resistance genes were negatively correlated with PM₁₀ concentrations and most vancomycin resistance genes were positively correlated with PM₁₀ concentrations.

Discussion

Dust is a well-recognized vector for the long-range microbial transport of microorganisms and pollution.^{12,64,66,67} Recent studies suggest that aerosol microbiomes are most affected by the microbiomes of underlying environments.^{19,64,68} Variations over seasons and in airmass origin were also linked to changes in aerosol microbial community composition.^{1,2,6,9,69,70} The aerobiome community composition displays mixtures of sources, and detecting a constant and stable indigenous microbial community was not yet reported. As such, the identified microorganisms may serve as tracers for the

sources contributing to the aerobiome. Previous studies comparing aerobiomes to possible sources have shown that soils, leaf surfaces, chicken coops, humans and farm animals contribute to the composition of the aerobiome to varying extents. However, most of these studies relied on amplicon sequencing of taxonomic genetic markers, *e.g.*, the 16S rRNA-encoding gene; focused on a single kingdom, *e.g.*, bacteria; and overlooked functional differences between closely related taxa.

In this study, we sampled dust under different atmospheric conditions, representing various sources, transport trajectories and different concentrations of mineral dust expressed by varying concentrations of PM₁₀. The collected dust metagenomes were compared to metagenomes from three major environments: atmospheric (air and smog sample types), terrestrial (soils) and marine. This comparison, based on taxonomic classification as well as functional gene annotation of metagenomes, was conducted to better understand the similarities and differences among the different microbiomes, infer their functional capacities, and potentially elucidate the contribution of different sources to the atmospheric microbiomes.

Our results show that the metagenomes of dust sampled in Israel in this study most closely resembled dust metagenomes sampled in Saudi Arabia, close to the Red Sea, over 1000 km away, on different sampling dates³. A common source that is attributed to soils contributed to the two dust datasets, according to the SourceTracker analysis. However, variability within *Dust IL* and *Dust Red Sea* samples was observed. This variability in *Dust IL* was correlated with PM₁₀ concentrations, *i.e.*, a higher relative contribution of the soil microbiome was observed in samples with higher PM₁₀ concentrations. This is expected for this region, since a major source of PM₁₀ is mineral dust originating in desert soils. *Dust IL* samples with low PM₁₀ loading showed higher similarity to atmospheric microbiomes of cloud condensation samples collected in France (cloud samples) and of urban areas in the USA (urban air samples). Sea sample metagenomes contributed very little to all the dust samples, *i.e.*, both *Dust IL* and *Dust Red Sea*, despite the proximity to the Mediterranean and Red Seas, respectively. Some of the trajectories of the dust sampled in Israel included a marine component, which was previously linked to an effect on the aerobiome composition⁷. Moreover, all *Dust Red Sea* samples were collected in a location adjacent to the Red Sea, and some were even collected on a ship in the Red Sea. Nonetheless, the contribution of the Mediterranean and Red Seas to the dust microbiomes was always below 3.1%. For reference, the contribution of the air microbiomes sampled in France (cloud dataset) and North America (urban air dataset) to the dust microbiome ranged between 3.8% and 12% (Fig. 2). This suggests that the marine environment does not contribute much to transported dust. This conclusion is consistent with another study showing that the marine environment did not significantly contribute to the airborne microbiome over the Atlantic and Pacific Oceans.⁷¹ In contrast, dust deposition can contribute to the composition and functional capacity of the marine microbiome, as shown in a few previous studies.^{14,72-76}

Since elevated PM₁₀ concentrations in Israel usually indicate dust events, we looked for correlations between PM₁₀ concentrations and taxonomic and functional gene composition in *Dust IL* samples. The

results indicated that dust storms changed the aerobiome community composition and the functional profile from a fungal-dominated environment to a bacterial-dominated environment. A significant part of the functions that were previously suggested to be linked to microbial survival in the atmosphere, *e.g.*, UV, oxidative and envelope stress resistance, was positively correlated with PM₁₀ concentrations, suggesting that dust-borne bacteria might be a global source of viable or even active bacteria in the atmosphere.

Genes associated with the biodegradation of organic contaminants such as aromatic compounds were relatively more abundant in dust than in any other environment examined and positively correlated with PM₁₀ concentrations. Most of these genes were also common to soils. Previous studies indicated the cometabolism of organic contaminants along with methane and ammonia, due to the presence of di- or monooxygenases with a broad substrate range⁷⁷. Accordingly, we found that along with genes associated with the biodegradation of organic contaminants, methane metabolism genes were also positively correlated with PM₁₀ concentration, as well as genes associated with glyoxylate and dicarboxylate metabolism, which includes formate metabolism. All or some of these substances might be relevant to metabolic activity in the atmosphere due to their biodegradation, as suggested in other studies,^{65,78} as well as in dust-affected terrestrial and marine regions.

We found that genes encoding beta-lactam and bacitracin resistance were more abundant in transported dust than in other atmospheric samples. Beta-lactam antibiotics such as penicillins and cephalosporins are commonly administered to humans as well as to food-producing animals such as cattle⁷⁹, and the findings suggest that an anthropogenic source contributes to the dust microbiome, as was suggested previously by Mazar *et al* and Gat *et al*, who showed that antibiotic resistance genes are found in aerosols on dusty and nondusty days and that their source is more likely anthropogenic than natural.^{4,7} Bacitracin is also a common antibiotic that is administered mostly as an ointment to prevent skin infections, yet resistance to bacitracin is commonly found in *Bacillus* species and other Firmicutes, suggesting that its source might be natural. The presence of resistance to these two antibiotics in dust metagenomes should raise concerns regarding the spread of this resistance and the possible effects on public health.

Overall, fewer KEGG-characterized functional genes were negatively correlated with PM₁₀ concentrations than were positively correlated with PM₁₀ (566 vs. 1518, respectively). This observation emphasizes the role played by dust storms in changing the microbial community composition from a fungal-dominated community to a bacterial-dominated community in the Mediterranean, as well as in the dispersal of environmental functions. Given the changes in dust storm frequency and severity due to global climatic changes,^{26,80} the possible implications of such transport should be further investigated.

Interestingly, the SourceTracker analysis showed that low-PM₁₀ dust sample microbiomes significantly overlapped with cloud and urban air sample microbiomes. These common taxa and associated functional genes were also negatively correlated with PM₁₀ concentrations. They were more abundant in the aerobiome than in the soil and sea microbiomes and were mostly fungal. We suggest that these

fungal taxa constitute a "core microbiome" for the atmosphere and provide insights into their possible functioning. We found that in general, the relative abundance of fungi, especially those of the phylum Ascomycota, increased when PM₁₀ concentrations decreased, with 13 fungal genera found to be common to all atmospheric datasets and only to these datasets. If we extend our definition of "core microbiome" and include taxa that were negatively correlated with PM₁₀ and were rare in soils and marine samples (clusters 1, 2 and 6, Fig. 3), we obtain 56 fungal genera, mostly belonging to Ascomycota, with highly varied environments and functions. Some of these fungi might be carried by long-range transport, such as *Cordyceps*, *Paracoccidioides* and *Allomyces*, which are often found in tropical regions. Other fungi are animal and plant commensals or pathogens, such as *Malassezia*, *Tilletia*, *Epidermophyton*, *Mycosphaerella*, *Arthroderma*, *Botryotinia* and *Gibberella*. Studies indicate that fungal spores are actively emitted and dispersed into the atmosphere,^{21,81-86} which explains their ubiquity in atmospheric samples. Another source of fungal spores in the atmosphere is biomass burning,^{87,88} as fire smoke is transported great distances in the atmosphere.^{89,90} Both processes could explain the presence of Ascomycota in all atmospheric metagenomes, as well as their negative correlation with high PM₁₀ concentrations.

Often overlooked members of the aerobiome are archaea. In this study, we found a clear correlation between PM₁₀ concentrations and archaeal abundance in *Dust IL* samples. Many of these archaea were anaerobic methanogens; accordingly, some methanogenesis-associated genes were positively correlated with PM₁₀ concentrations. Methanogenesis-associated genes were also more abundant in dust than in other atmospheric samples. Methanogenesis is carried out solely by archaea under strictly anaerobic conditions, usually in highly reduced soils or sediments but also in the digestive system and feces of ruminants as well as chicken and other farm animals.^{91,92} Ruminant feces found in farms or grazing grounds or chicken feces found in chicken coops could have contributed to the dust metagenome. In addition, landfills could also be a source of methanogens in the atmosphere.⁹³⁻⁹⁵ This finding may suggest that atmospheric conditions that result in high PM₁₀ concentrations may also result in the suspension of particles other than soil and mineral dust. It also suggests that some methanogens in the atmosphere can mark anthropogenic source contributions. The results from this study, showing the enrichment of antibiotic resistance genes in the aerobiome, corroborate this suggestion. Previous studies have suggested that a dust storm collects material over its course, and its microbial community composition is affected accordingly.^{7,96} In this manner, a dust storm that passes over an urban, agricultural or industrialized area is likely to increase anthropogenic emissions of microorganisms into the atmosphere and increase their potential impact on aerobiome functionality and composition. This is consistent with the presence of anthropogenic organic contaminants that adhere to transported dust, as shown by Falkovich *et al.*^{97,98}

The importance of anthropogenic influence on the atmospheric microbiome is only now gaining attention.⁹⁹ It is predicted that the aerobiome will be influenced by the changing chemical and physical characteristics of the atmosphere due to anthropogenic activities such as cooking, agricultural practices and land use change. Climatic changes such as increased temperatures, fires and drought are projected

to increase the amount of dust emitted to the atmosphere,²⁶ thus increasing the contribution of soil microbiomes to the composition of the aerobiome. The increasing frequency and intensity of wildfires^{100,101} also increases the concentration of particles in the atmosphere. These serve as nuclei for bacterial transport in the atmosphere,⁹⁹ thereby increasing their dispersal and potentially the functional scope of the aerobiome. Recent evidence on the possible metabolic activity of bacteria in the atmosphere suggests that specific carbon sources are likely to sustain bacteria, *e.g.*, formate, acetic acid and ethanol.^{8,65,78} Moreover, it was suggested that bacterial metabolism might also affect cloud or even atmospheric chemistry.^{65,102} Our results suggest that if dust storms carry viable bacteria, they may be metabolically relevant for atmospheric chemical cycles and may spread these bacteria globally.

In this study, we investigated the links between the surface and atmospheric microbiomes, as well as between different atmospheric microbiomes. We showed that different atmospheric niches display different functional profiles and community structures and that they share a common background dominated by fungi. We also showed that two similar niches, *i.e.*, the dust collected in Israel and over the Red Sea on different, nonsynchronized occasions, were strikingly similar, emphasizing the importance of the source of the dust in determining its composition on a regional and possible global scale. As expected, dust showed high resemblance to soils both in community structure and functional profile, yet the dust samples harbored functional genes that are likely to have been derived from an anthropogenic source.

Declarations

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References

1. Cáliz, J., Triadó-Margarit, X., Camarero, L. & Casamayor, E. O. A long-term survey unveils strong seasonal patterns in the airborne microbiome coupled to general and regional atmospheric circulations. *Proc. Natl. Acad. Sci.* **115**, 12229–12234 (2018).

2. Du, P., Du, R., Ren, W., Lu, Z. & Fu, P. Seasonal variation characteristic of inhalable microbial communities in PM_{2.5} in Beijing city, China. *Sci. Total Environ.* **610–611**, 308–315 (2018).
3. Aalismail, N. A. *et al.* Functional metagenomic analysis of dust-associated microbiomes above the Red Sea. *Sci. Rep.* **9**, (2019).
4. Mazar, Y., Cytryn, E., Erel, Y. & Rudich, Y. Effect of Dust Storms on the Atmospheric Microbiome in the Eastern Mediterranean. *Environ. Sci. Technol.* **50**, 4194–4202 (2016).
5. Mhuireach, G., Betancourt-Román, C. M., Green, J. L. & Johnson, B. R. Spatiotemporal controls on the urban aerobiome. *Front. Ecol. Evol.* **7**, (2019).
6. Bowers, R. M., McLetchie, S., Knight, R. & Fierer, N. Spatial variability in airborne bacterial communities across land-use types and their relationship to the bacterial communities of potential source environments. *ISME J.* **5**, 601–612 (2011).
7. Gat, D., Mazar, Y., Cytryn, E. & Rudich, Y. Origin-Dependent Variations in the Atmospheric Microbiome Community in Eastern Mediterranean Dust Storms. *Environ. Sci. Technol.* **51**, 6709–6718 (2017).
8. Amato, P. *et al.* Metatranscriptomic exploration of microbial functioning in clouds. *Sci. Rep.* **9**, (2019).
9. Lang-Yona, N. *et al.* Links between airborne microbiome, meteorology, and chemical composition in northwestern Turkey. *Sci. Total Environ.* **725**, 138227 (2020).
10. Meola, M., Lazzaro, A. & Zeyer, J. Bacterial Composition and Survival on Sahara Dust Particles Transported to the European Alps. *Frontiers in microbiology* **6**, 1454 (2015).
11. Yamaguchi, N. *et al.* Abundance and Community Structure of Bacteria on Asian Dust Particles Collected in Beijing, China, during the Asian Dust Season. *Biol. Pharm. Bull.* **39**, 68–77 (2016).
12. Hua, N. P., Kobayashi, F., Iwasaka, Y., Shi, G. Y. & Naganuma, T. Detailed identification of desert-originated bacteria carried by Asian dust storms to Japan. *Aerobiologia (Bologna)*. **23**, 291–298 (2007).
13. Cao, C. *et al.* Inhalable microorganisms in Beijing's PM_{2.5} and PM₁₀ pollutants during a severe smog event. *Environ. Sci. Technol.* **48**, 1499–1507 (2014).
14. Rahav, E., Ovadia, G., Paytan, A. & Herut, B. Contribution of airborne microbes to bacterial production and N₂ fixation in seawater upon aerosol deposition. *Geophys. Res. Lett.* **43**, 719–727 (2016).
15. Barberán, A. *et al.* Continental-scale distributions of dust-associated bacteria and fungi. *Proceedings of the National Academy of Sciences* **112**, 5756–5761 (2015).
16. Docherty, K. M., Pearce, D. S., Lemmer, K. M. & Hale, R. L. Distributing regionally, distinguishing locally: examining the underlying effects of local land use on airborne bacterial biodiversity. *Environ. Microbiol.* **20**, 3529–3542 (2018).
17. Souza, F. F. C. *et al.* Uncovering prokaryotic biodiversity within aerosols of the pristine Amazon forest. *Sci. Total Environ.* **688**, 83–86 (2019).
18. Souza, F. F. C. *et al.* Influence of seasonality on the aerosol microbiome of the Amazon rainforest. *Sci. Total Environ.* **760**, 144092 (2021).

19. Archer, S. D. J. *et al.* Airborne microbial transport limitation to isolated Antarctic soil habitats. *Nature Microbiology* **4**, 925–932 (2019).
20. Griffin, D. W. Atmospheric Movement of Microorganisms in Clouds of Desert Dust and Implications for Human Health. *Clinical Microbiology Reviews* **20**, 459–477 (2007).
21. Fröhlich-Nowoisky, J. *et al.* Bioaerosols in the Earth system: Climate, health, and ecosystem interactions. *Atmos. Res.* **182**, 346–376 (2016).
22. Yooseph, S. *et al.* A Metagenomic Framework for the Study of Airborne Microbial Communities. *PLoS One* **8**, e81862 (2013).
23. Be, N. A. *et al.* Metagenomic Analysis of the Airborne Environment in Urban Spaces. *Microb. Ecol.* **69**, 346–355 (2014).
24. Tringe, S. G. *et al.* The Airborne Metagenome in an Indoor Urban Environment. *PLoS One* **3**, e1862 (2008).
25. Lelieveld, J. *et al.* Global Air Pollution Crossroads over the Mediterranean. *Science (80-.)*. **298**, 794–799 (2002).
26. Krasnov, H., Katra, I. & Friger, M. Increase in dust storm related PM10 concentrations: A time series analysis of 2001–2015. *Environ. Pollut.* **213**, 36–42 (2016).
27. Acker, J. G. & Leptoukh, G. Online analysis enhances use of NASA Earth Science Data. *Eos (Washington, DC)*. **88**, 14 (2007).
28. Keegan, K. P., Glass, E. M. & Meyer, F. MG-RAST, a metagenomics service for analysis of microbial community structure and function. in *Methods in Molecular Biology* **1399**, 207–233 (Humana Press Inc., 2016).
29. O’Leary, N. A. *et al.* Reference sequence (RefSeq) database at NCBI: current status, taxonomic expansion, and functional annotation. *Nucleic Acids Res.* **44**, D733–D745 (2016).
30. Brister, J. R., Ako-Adjei, D., Bao, Y. & Blinkova, O. NCBI viral genomes resource. *Nucleic Acids Res.* **43**, D571–D577 (2015).
31. Tatusova, T. *et al.* NCBI prokaryotic genome annotation pipeline. *Nucleic Acids Res.* **44**, 6614–6624 (2016).
32. Kanehisa, M. & Goto, S. KEGG: kyoto encyclopedia of genes and genomes. *Nucleic Acids Res.* **28**, 27–30 (2000).
33. Kanehisa, M., Sato, Y., Furumichi, M., Morishima, K. & Tanabe, M. New approach for understanding genome variations in KEGG. *Nucleic Acids Res.* **47**, D590–D595 (2019).
34. Kanehisa, M. Toward understanding the origin and evolution of cellular organisms. *Protein Sci.* **28**, 1947–1951 (2019).
35. R Core Team. R: A Language and Environment for Statistical Computing. (2020).
36. Schloss, P. D. *et al.* Introducing mothur: open-source, platform-independent, community-supported software for describing and comparing microbial communities. *Appl. Environ. Microbiol.* **75**, 7537–7541 (2009).

37. Wickham, H. *et al.* Welcome to the Tidyverse. *J. Open Source Softw.* **4**, 1686 (2019).
38. Oksanen, J. *et al.* Vegan: community ecology package. R package version 1.17-4. (2007).
39. Robinson, D. *et al.* Convert Statistical Analysis Objects into Tidy Tibbles. (2020).
40. Henry, L., Wickham, H. & RStudio. Functional Programming Tools. (2019).
41. Harrell, F. E. J. Hmisc: Harrell Miscellaneous. (2020).
42. Boogaart, K. G. van den. Compositions: compositional data analysis. (2020).
43. Excoffier, L., Smouse, P. E. & Quattro, J. M. Analysis of Molecular Variance Inferred from Metric Distances among DNA Haplotypes: Application to Human Mitochondrial DNA Restriction Data. *Genetics* **131**, 479–491 (1992).
44. Wickham, H. ggplot2: Elegant Graphics for Data Analysis. (2016).
45. Barter, R. L. & Yu, B. Superheat: An R Package for Creating Beautiful and Extendable Heatmaps for Visualizing Complex Data. *J. Comput. Graph. Stat.* **27**, 910–922 (2018).
46. Tripathi, B. M. *et al.* Trends in Taxonomic and Functional Composition of Soil Microbiome Along a Precipitation Gradient in Israel. *Microb. Ecol.* **74**, 168–176 (2017).
47. Kopf, A. *et al.* The ocean sampling day consortium. *Gigascience* **4**, (2015).
48. Sunagawa, S. *et al.* Structure and function of the global ocean microbiome. *Science (80-.)*. **348**, (2015).
49. Knights, D. *et al.* Bayesian community-wide culture-independent microbial source tracking. *Nat. Methods* **8**, 761–763 (2011).
50. Spearman, C. The Proof and Measurement of Association between Two Things. *Am. J. Psychol.* **100**, 441–471 (1987).
51. Oren, A. The Family Methanobacteriaceae. in *The Prokaryotes: Other Major Lineages of Bacteria and The Archaea* (eds. Rosenberg, E., DeLong, E. F., Lory, S., Stackebrandt, E. & Thompson, F.) 165–193 (Springer Berlin Heidelberg, 2014). doi:10.1007/978-3-642-38954-2_411
52. Kroppenstedt, R. M. & Goodfellow, M. The family Thermomonosporaceae: Actinocorallia, actinomadura, spirillospora and thermomonospora. in *The Prokaryotes* (ed. Dworkin, M.) 682–724 (2006). doi:10.1007/0-387-30743-5_27
53. Normand, P., Benson, D. R., Berry, A. M. & Tisa, L. S. The Family Frankiaceae. in *The Prokaryotes: Actinobacteria* (eds. Rosenberg, E., DeLong, E. F., Lory, S., Stackebrandt, E. & Thompson, F.) 339–356 (Springer Berlin Heidelberg, 2014). doi:10.1007/978-3-642-30138-4_183
54. Franco, C. M. M. & Labeda, D. P. The Order Pseudonocardiales. in *The Prokaryotes: Actinobacteria* (eds. Rosenberg, E. et al.) 743–860 (Springer Berlin Heidelberg, 2014). doi:10.1007/978-3-642-30138-4_190
55. Albuquerque, L. & da Costa, M. S. The Families Conexibacteraceae, Patulibacteraceae and Solirubrobacteraceae. in *The Prokaryotes: Actinobacteria* (eds. Rosenberg, E., DeLong, E. F., Lory, S., Stackebrandt, E. & Thompson, F.) 185–200 (Springer Berlin Heidelberg, 2014). doi:10.1007/978-3-642-30138-4_200"

56. Phillips, R. W. *et al.* Kineococcus radiotolerans sp. nov., a radiation-resistant, gram-positive bacterium. *Int. J. Syst. Evol. Microbiol.* **52**, 933–938 (2002).
57. Tamura, T. & Suzuki, K.-I. The Suborder Kineosporiineae. in *The Prokaryotes: Actinobacteria* (eds. Rosenberg, E. *et al.*) 443–453 (Springer Berlin Heidelberg, 2014). doi:10.1007/978-3-642-30138-4_179"
58. Gorbushina, A. A. Life on the rocks. *Environ. Microbiol.* **9**, 1613–1631 (2007).
59. Hezbri, K. *et al.* Geodermatophilus pulveris sp. nov., a gamma-radiation-resistant actinobacterium isolated from the Sahara desert. *Int. J. Syst. Evol. Microbiol.* **66**, 3828–3834 (2016).
60. Normand, P. The Families Frankiaceae, Geodermatophilaceae, Acidothermaceae and Sporichthyaceae. in *The Prokaryotes: Volume 3: Archaea. Bacteria: Firmicutes, Actinomycetes* (eds. Dworkin, M. *et al.*) 669–681 (Springer New York, 2006). doi:10.1007/0-387-30743-5_26"
61. Lory, S. The Family Legionellaceae. in *The Prokaryotes: Gammaproteobacteria* (eds. Rosenberg, E., DeLong, E. F., Lory, S., Stackebrandt, E. & Thompson, F.) 387–389 (Springer Berlin Heidelberg, 2014). doi:10.1007/978-3-642-38922-1_370
62. Escalas, A. *et al.* Microbial functional diversity: From concepts to applications. *Ecology and Evolution* **9**, 12000–12016 (2019).
63. Fierer, N., Barberán, A. & Laughlin, D. C. Seeing the forest for the genes: Using metagenomics to infer the aggregated traits of microbial communities. *Front. Microbiol.* **5**, (2014).
64. Tignat-Perrier, R. *et al.* Microbial functional signature in the atmospheric boundary layer. *Biogeosciences* **17**, 6081–6095 (2020).
65. Khaled, A., Zhang, M., Amato, P., Delort, A.-M. & Ervens, B. Biodegradation by bacteria in clouds: An underestimated sink for some organics in the atmospheric multiphase system. *Atmos. Chem. Phys. Discuss.* **2020**, 1–32 (2020).
66. Garrison, V. H. *et al.* African and Asian Dust: From Desert Soils to Coral Reefs. *BioScience* **53**, 469–480 (2003).
67. Pedgley, D. E. Aerobiology: The Atmosphere as a Source and Sink for Microbes. in *Microbial Ecology of Leaves* (eds. Andrews, J. H. & Hirano, S. S.) 43–59 (Springer New York, 1991).
68. Tignat-Perrier, R. *et al.* Global airborne microbial communities controlled by surrounding landscapes and wind conditions. *Sci. Rep.* **9**, 14441 (2019).
69. Wehking, J. *et al.* Community composition and seasonal changes of archaea in coarse and fine air particulate matter. *Biogeosciences* **15**, 4205–4214 (2018).
70. Bowers, R. M. *et al.* Seasonal Variability in Bacterial and Fungal Diversity of the Near-Surface Atmosphere. *Environ. Sci. Technol.* **47**, 12097–12106 (2013).
71. Lang-Yona, N. *et al.* Airborne bacteria over oceans shed light on global biogeodiversity patterns. *under rev.*
72. Rahav, E. *et al.* The Impact of Atmospheric Dry Deposition Associated Microbes on the Southeastern Mediterranean Sea Surface Water following an Intense Dust Storm. *Front. Mar. Sci.* **3**, 127 (2016).

73. Krom, M. D. *et al.* Response of the Eastern Mediterranean Microbial Ecosystem to Dust and Dust Affected by Acid Processing in the Atmosphere. *Front. Mar. Sci.* **3**, 133 (2016).
74. Rahav, E. *et al.* Evaluating the Impact of Atmospheric Depositions on Springtime Dinitrogen Fixation in the Cretan Sea (Eastern Mediterranean) – A Mesocosm Approach. *Front. Mar. Sci.* **3**, 180 (2016).
75. Mesciöglu, E. *et al.* Dust-Associated Airborne Microbes Affect Primary and Bacterial Production Rates, and Eukaryotes Diversity, in the Northern Red Sea: A Mesocosm Approach. *Atmosphere (Basel)*. **10**, 358 (2019).
76. Paytan, A. *et al.* Toxicity of atmospheric aerosols on marine phytoplankton. *Proceedings of the National Academy of Sciences* **106**, 4601–4605 (2009).
77. Hazen, T. C. Handbook of Hydrocarbon and Lipid Microbiology. in *Handbook of Hydrocarbon and Lipid Microbiology* (ed. Timmis, K. N.) 2505–2514 (Springer Verlag, 2010). doi:10.1007/978-3-540-77587-4_185
78. Krumins, V., Mainelis, G., Kerkhof, L. J. & Fennell, D. E. Substrate-Dependent rRNA Production in an Airborne Bacterium. *Environ. Sci. Technol. Lett.* **1**, 376–381 (2014).
79. FDA. *2017 Summary Report on Antimicrobials Sold or Distributed for Use in Food-Producing Animals*. (2018).
80. Prospero, J. M. & Lamb, P. J. African Droughts and Dust Transport to the Caribbean: Climate Change Implications. *Science (80-)*. **302**, 1024 LP – 1027 (2003).
81. Brown, J. K. M. & Hovmöll, M. S. Aerial dispersal of pathogens on the global and continental scales and its impact on plant disease. *Science* **297**, 537–541 (2002).
82. Govi, G. Aerial diffusion of phytopathogenic fungi. *Aerobiologia (Bologna)*. **8**, 84–93 (1992).
83. Lang-Yona, N. *et al.* Annual distribution of allergenic fungal spores in atmospheric particulate matter in the Eastern Mediterranean; a comparative study between ergosterol and quantitative PCR analysis. *Atmos. Chem. Phys.* **12**, 2681–2690 (2012).
84. Kuparinen, A., Markkanen, T., Riikonen, H. & Vesala, T. Modeling air-mediated dispersal of spores, pollen and seeds in forested areas. *Ecol. Modell.* **208**, 177–188 (2007).
85. Rossi, V., Ponti, I., Marinelli, M., Giosuè, S. & Bugiani, R. Environmental Factors Influencing the Dispersal of *Venturia inaequalis* Ascospores in the Orchard Air. *J. Phytopathol.* **149**, 11–19 (2001).
86. Mondal, S. N., Gottwald, T. R. & Timmer, L. W. Environmental Factors Affecting the Release and Dispersal of Ascospores of *Mycosphaerella citri*. *Phytopathology®* **93**, 1031–1036 (2003).
87. Mims, S. A. & Mims, F. M. Fungal spores are transported long distances in smoke from biomass fires. *Atmos. Environ.* **38**, 651–655 (2004).
88. Kobziar, L. N. *et al.* Pyroaerobiology: the aerosolization and transport of viable microbial life by wildland fire. *Ecosphere* **9**, e02507 (2018).
89. Sinha, P., Jaeglé, L., Hobbs, P. V & Liang, Q. Transport of biomass burning emissions from southern Africa. *J. Geophys. Res. Atmos.* **109**, (2004).

90. Jaffe, D. *et al.* Long-range transport of Siberian biomass burning emissions and impact on surface ozone in western North America. *Geophys. Res. Lett.* **31**, (2004).
91. Lyu, Z., Shao, N., Akinyemi, T. & Whitman, W. B. Methanogenesis. *Curr. Biol.* **28**, R727–R732 (2018).
92. Misiukiewicz, A. *et al.* Review: Methanogens and methane production in the digestive systems of nonruminant farm animals. *Animal* 100060 (2020).
doi:<https://doi.org/10.1016/j.animal.2020.100060>
93. Semrau, J. D. Current knowledge of microbial community structures in landfills and its cover soils. *Appl. Microbiol. Biotechnol.* **89**, 961–969 (2011).
94. Zhao, R. *et al.* Antibiotic resistome in landfill leachate from different cities of China deciphered by metagenomic analysis. *Water Res.* **134**, 126–139 (2018).
95. Ramakrishnan, A. *et al.* Emerging contaminants in landfill leachate and their sustainable management. *Environ. Earth Sci.* **73**, 1357–1368 (2015).
96. Maki, T. *et al.* Aeolian Dispersal of Bacteria Associated With Desert Dust and Anthropogenic Particles Over Continental and Oceanic Surfaces. *J. Geophys. Res. Atmos.* **124**, 5579–5588 (2019).
97. Falkovich, A. H. Adsorption of organic compounds pertinent to urban environments onto mineral dust particles. *J. Geophys. Res.* **109**, D02208 (2004).
98. Falkovich, A. H. & Rudich, Y. Analysis of Semivolatile Organic Compounds in Atmospheric Aerosols by Direct Sample Introduction Thermal Desorption GC/MS. *Environ. Sci. Technol.* **35**, 2326–2333 (2001).
99. Archer, S. D. J. & Pointing, S. B. Anthropogenic impact on the atmospheric microbiome. *Nat. Microbiol.* **5**, 229–231 (2020).
100. Abatzoglou, J. T. & Williams, A. P. Impact of anthropogenic climate change on wildfire across western US forests. *Proc. Natl. Acad. Sci.* **113**, 11770–11775 (2016).
101. Liu, Y., Stanturf, J. & Goodrick, S. Trends in global wildfire potential in a changing climate. *For. Ecol. Manage.* **259**, 685–697 (2010).
102. Fankhauser, A. M. *et al.* Constraining the Impact of Bacteria on the Aqueous Atmospheric Chemistry of Small Organic Compounds. *ACS Earth Sp. Chem.* **3**, 1485–1491 (2019).

Figures

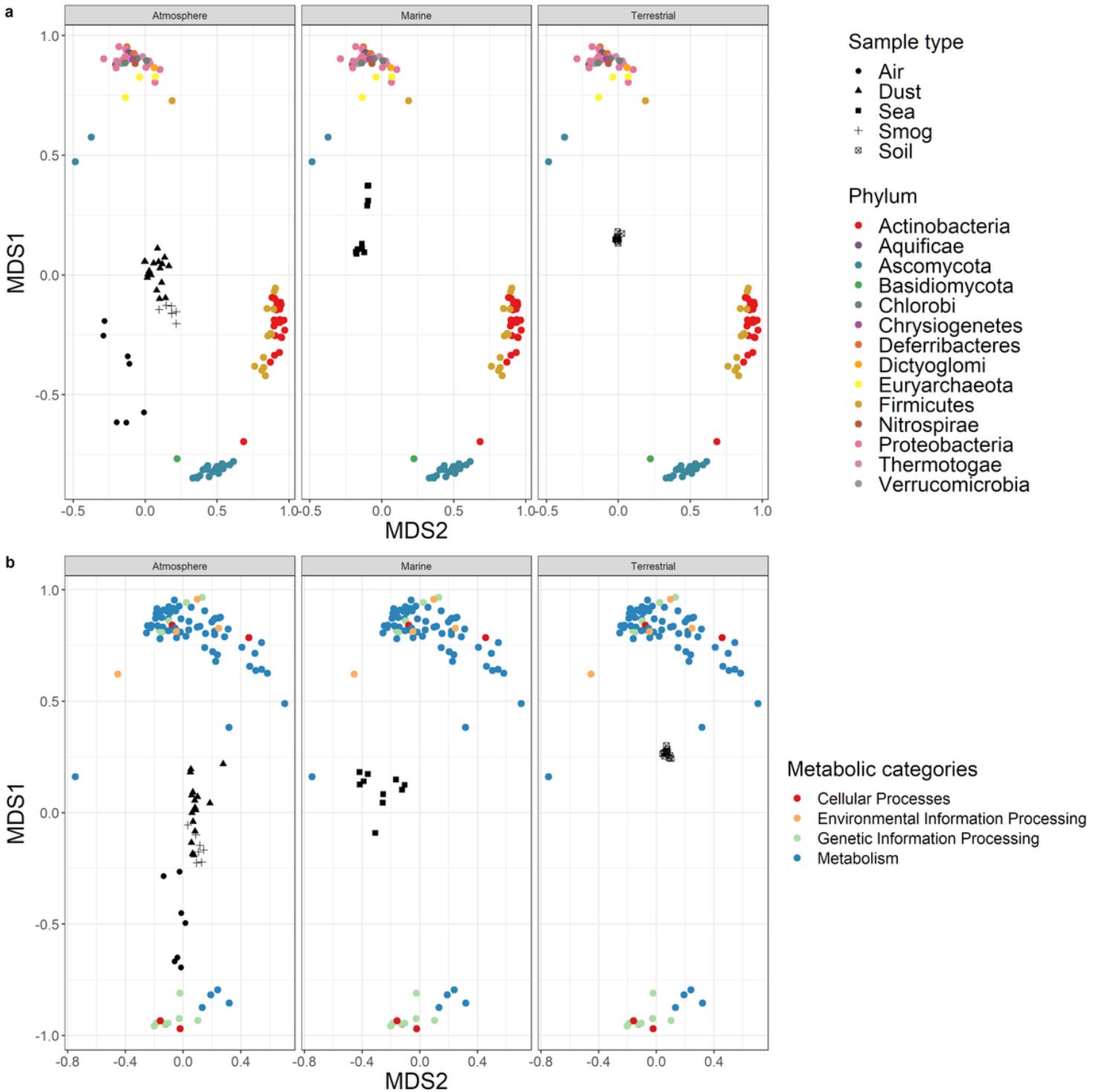


Figure 1

NMDS based on clr-transformed counts for taxa (a) and genes (b). Datasets are grouped according to sample types: Air: clouds and urban air; Dust: dust samples from Israel (this study) and from Saudi Arabia; Sea: samples from the Mediterranean and Red Seas; Smog: smog samples collected in Beijing; Soil: soil samples collected over a precipitation gradient in Israel. Figures are separated according to the environment they represent. Atmosphere includes all atmospheric samples (i.e., Dust, Air and Smog); Marine represents Sea samples and Terrestrial represents Soil samples. Metabolic categories were

annotated according to the KEGG database. Genera (a) and KEGG pathways (b) were correlated to the ordination axes (Spearman's correlation).

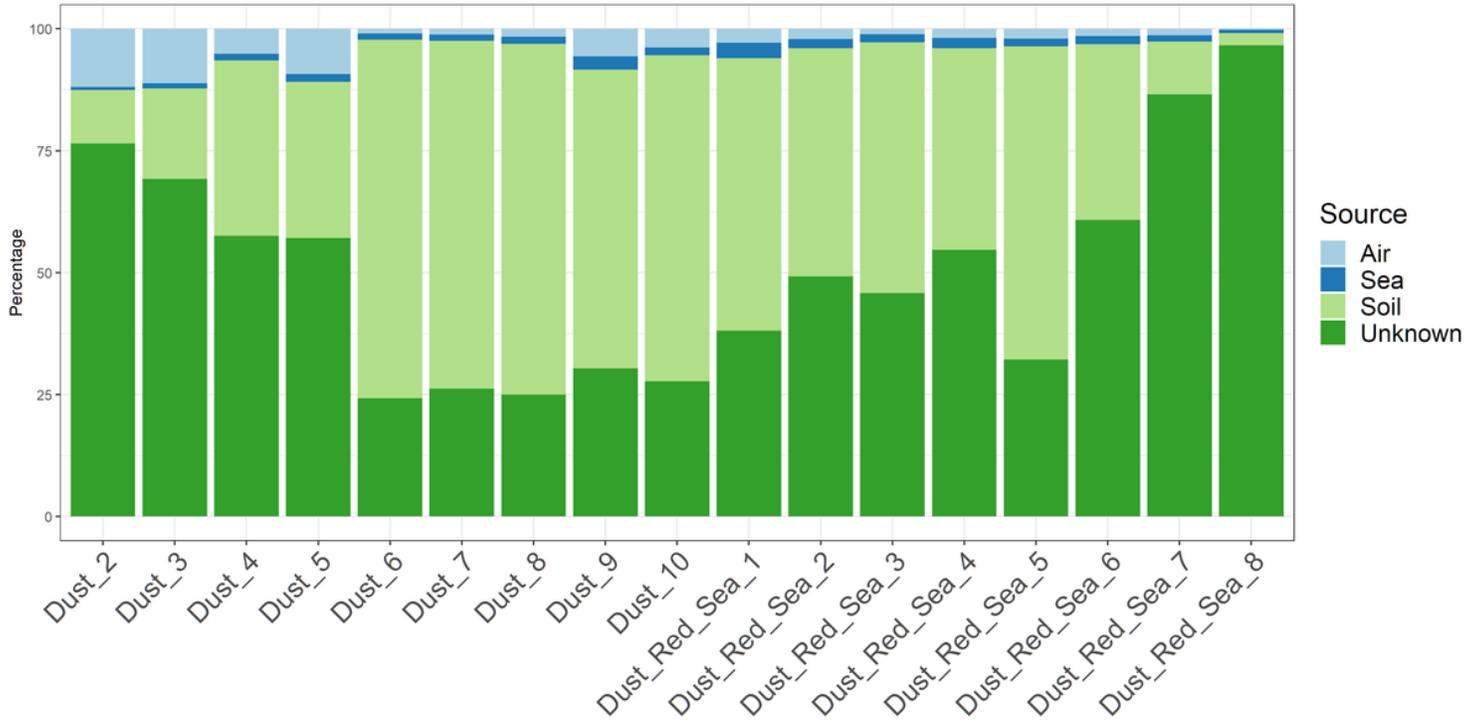


Figure 2

SourceTracker analysis of Dust samples. Sources were defined as Air (clouds and urban air), Sea (Mediterranean and Red Seas) and Soil (Israeli soils).

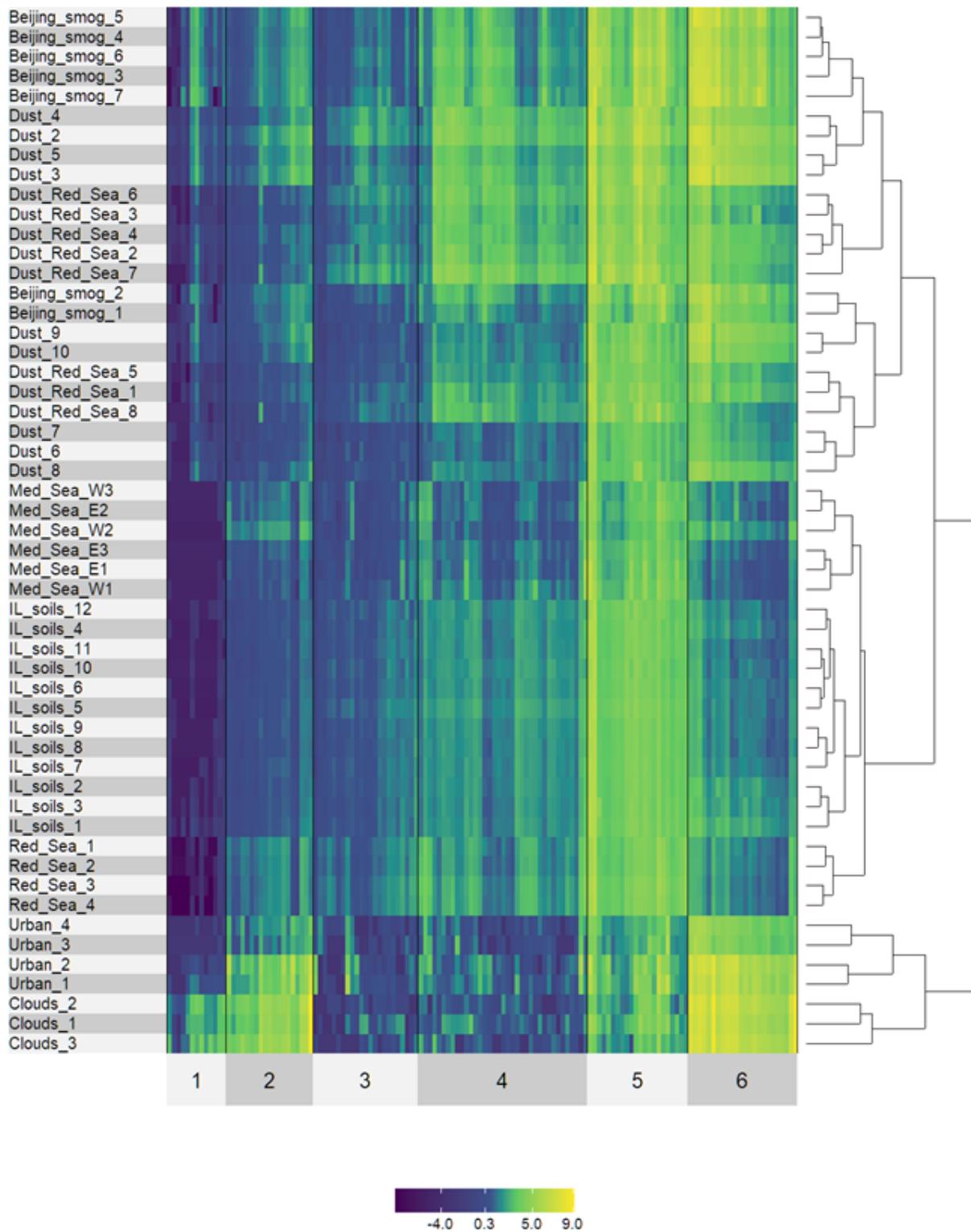


Figure 3

K-means clusters of genera that showed a significant negative correlation with PM10 concentrations in Israeli dust. The color scale represents clr-transformed counts.

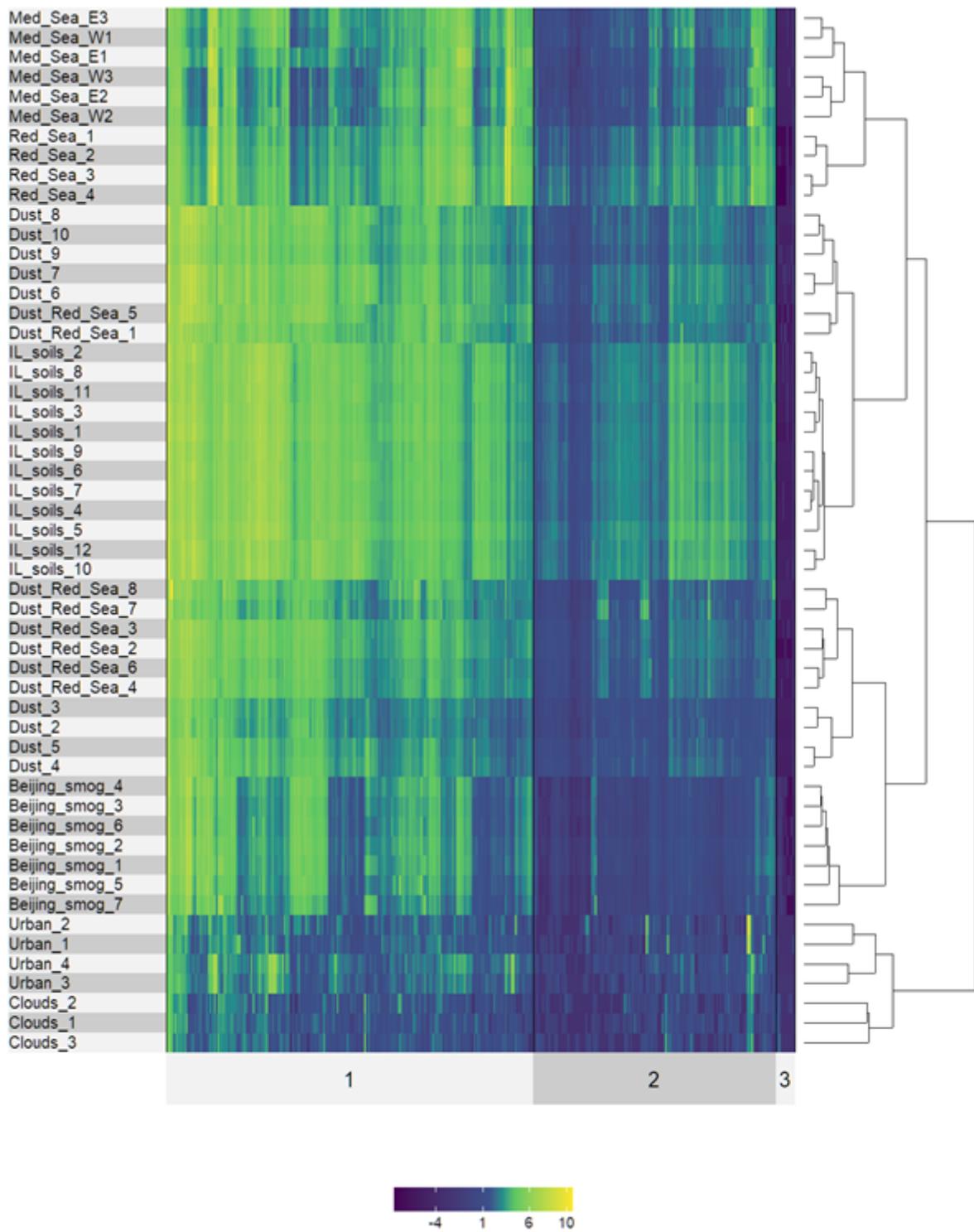


Figure 4

K-means clusters of genera that showed a significant positive correlation with PM10 concentrations in Israeli dust. The color scale represents cl_r-transformed counts.

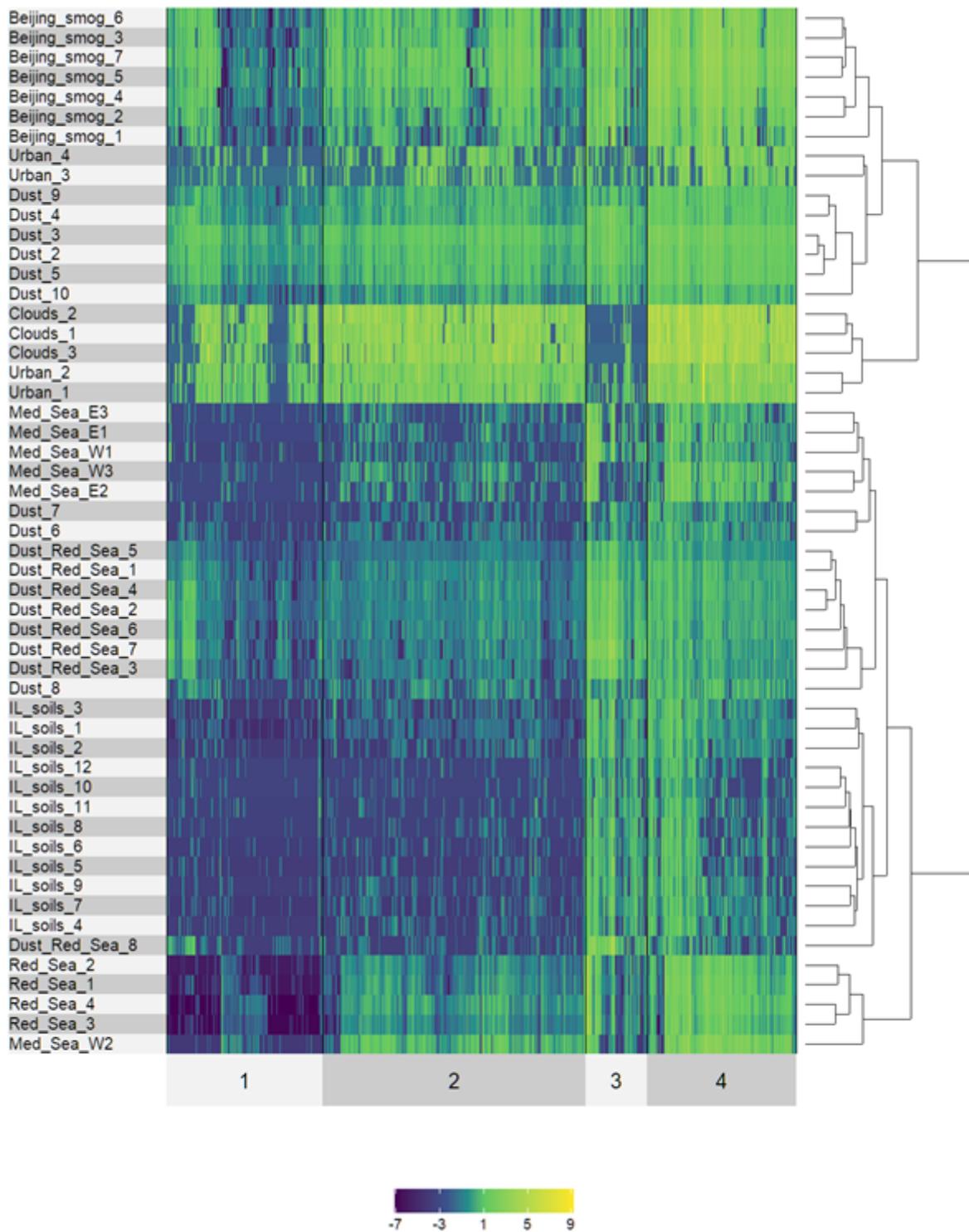


Figure 5

K-means clusters of genera that showed a significant negative correlation with PM10 concentrations in Israeli dust. The color scale represents clr-transformed counts.

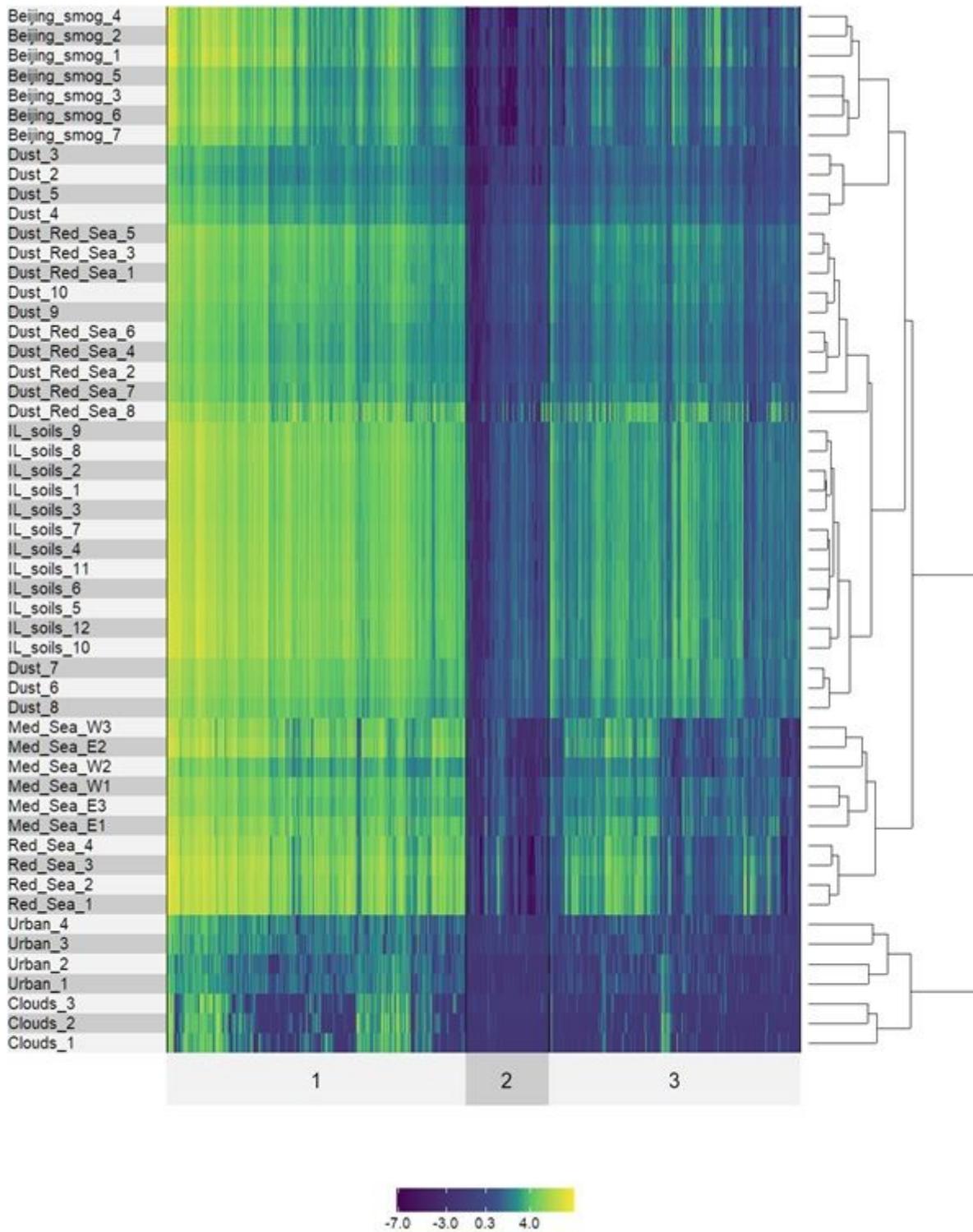


Figure 6

K-means clusters of genera that showed a significant positive correlation with PM10 concentrations in Israeli dust. The color scale represents clr-transformed counts.

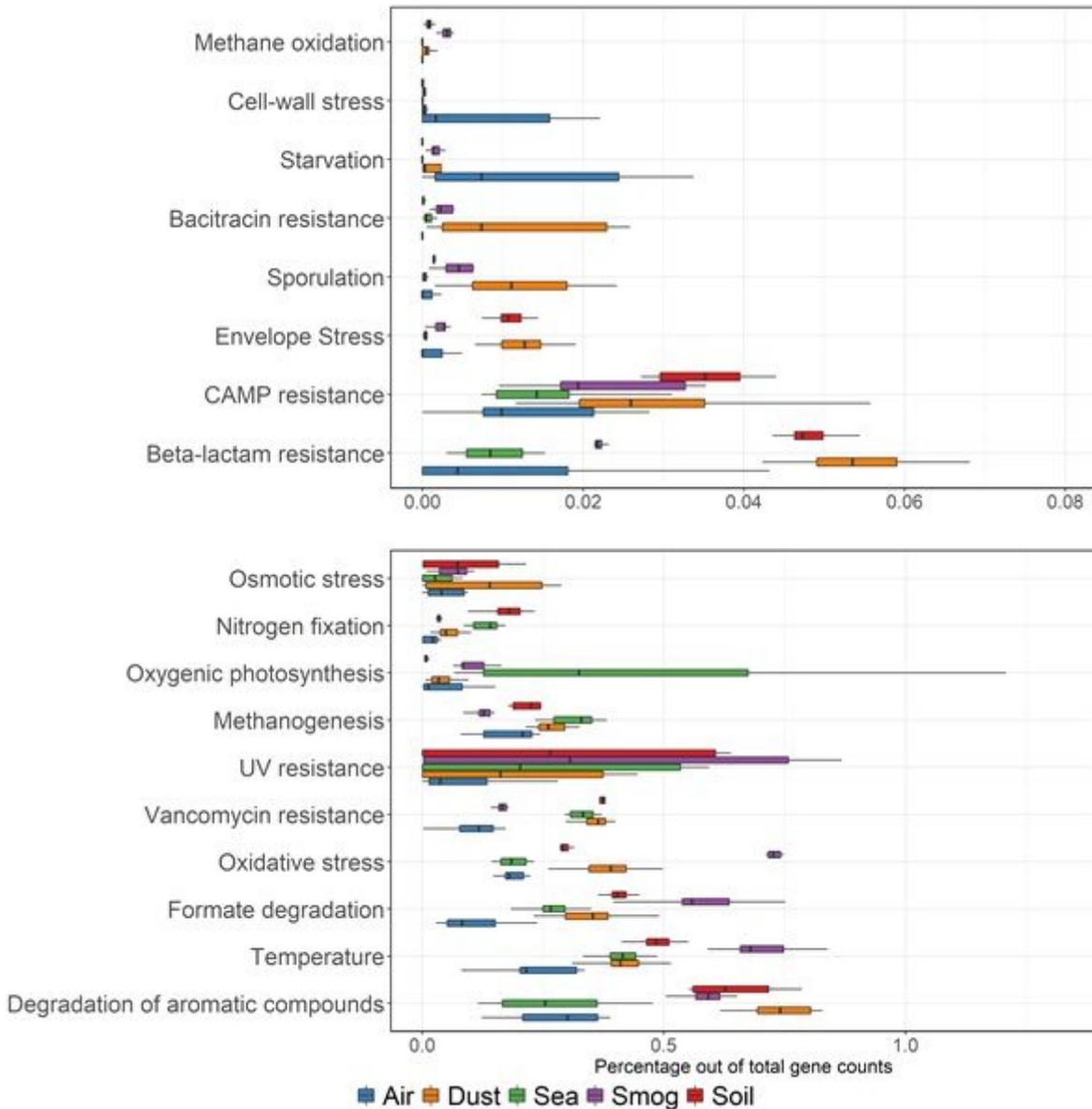


Figure 7

Relative abundances of selected functions, presented as percentages of total gene counts.

Supplementary Files

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- [TableS4a.xlsx](#)
- [TableS4b.xlsx](#)
- [TableS5.xlsx](#)

- [TableS6.xlsx](#)