

DOI: 10.5846/stxb201803260610

甄泉,王雅晴,洗超凡,郭卫俊,欧阳志云.沙尘暴对北京市空气细菌多样性特征的影响.生态学报,2019,39(2): - .

Zhen Q, Wang Y Q, Xian C F, Guo W J, Ouyang Z Y. Effects of sandstorms on the diversity characteristics of airborne bacteria in Beijing. Acta Ecologica Sinica, 2019, 39(2): - .

沙尘暴对北京市空气细菌多样性特征的影响

甄 泉^{1,2,3}, 王雅晴^{1,2}, 洗超凡^{1,2}, 郭卫俊⁴, 欧阳志云^{1,*}

1 中国科学院生态环境研究中心 城市与区域生态国家重点实验室, 北京 100085

2 中国科学院大学, 北京 100049

3 蚌埠医学院 公共卫生学院, 蚌埠 233030

4 浙江工商大学 环境科学与工程学院, 杭州 310018

摘要: 亚洲沙尘暴携带着各类污染物质,严重影响下风向地区大气过程及生态系统和相关人群的健康。然而,沙尘天气对中国华北地区空气细菌群落及多样性特征影响的研究较少。本文以北京市为例,系统研究了沙尘暴对城市空气细菌多样性特征的影响。2015年4月,通过定点采样连续收集了一次沙尘暴及其前后4天的空气颗粒物样本,DNA提取、PCR扩增后进行16S rRNA基因高通量测序。共获得169122条高质量序列,生物信息学分析表明,北京市空气细菌物种多样性较高,沙尘暴不能引起细菌OTU数目的增加,但沙尘暴天气下细菌群落Pielou、Shannon和Simpson指数显著增加。菌群分类分析发现,北京市空气细菌由35个细菌门构成,其中变形菌门(Proteobacteria)、放线菌门(Actinobacteria)、厚壁菌门(Firmicutes)和拟杆菌门(Bacteroidetes)相对丰度最高,分别占总序列的32.76%、28.09%、25.46%和6.32%;芽单胞菌门(Gemmatimonadetes)和酸杆菌门(Acidobacteria)次之,分别占序列总数的2.11%和1.81%,其他细菌门的相对丰度均低于1%。沙尘暴天气下,变形菌门(Proteobacteria)、拟杆菌门(Bacteroidetes)、芽单胞菌门(Gemmatimonadetes)和酸杆菌门(Acidobacteria)显著升高($P<0.05$),分别由31.67%、5.74%、1.82%、1.51%升高至41.46%、10.98%、4.48%和4.26%;而放线菌门(Actinobacteria)、厚壁菌门(Firmicutes)显著降低($P<0.05$),分别由28.84%、27.10%降低至22.13%和12.35%。冷杆菌属(*Psychrobacter*)、纤维单胞菌属(*Cellulomonas*)、不动杆菌属(*Acinetobacter*)、假单胞菌属(*Pseudomonas*)和梭菌属(*Clostridium*)可能含有人类条件病原菌,其在沙尘暴天气中相对丰度下降,但绝对丰度会大幅增加。沙尘暴能降低人体免疫力,因此致病菌潜在健康风险可能显著增强。通过聚类分析和主成分分析发现,沙尘暴日与非沙尘暴日空气细菌群落差异较大,而沙尘暴前后群落结构差异较小。沙尘暴前后空气细菌群落 α -多样性和 β 多样性均无显著差异,推测沙尘暴只能暂时影响空气细菌群落特征,不能显著改变其群落结构。本研究通过分析了沙尘暴侵袭下北京市空气细菌群落多样性特征及动态,为制定中国北方城市沙尘暴灾害预警措施和建立气传疾病的防控机制提供数据支持。

关键词: 沙尘暴;高通量测序;空气细菌;群落多样性

Effects of sandstorms on the diversity characteristics of airborne bacteria in Beijing

ZHEN Quan^{1,2,3}, WANG Yaqing^{1,2}, XIAN Chaofan^{1,2}, GUO Weijun⁴, OUYANG Zhiyun^{1,*}

1 State Key Laboratory of Urban and Regional Ecology, Research Center for Eco-Environmental Sciences, Chinese Academy of Sciences, Beijing 100085, China

2 University of Chinese Academy of Sciences, Beijing 100049, China

3 School of Public Health, Bengbu Medical College, Bengbu 233030, China

4 School of Environmental Science and Engineering, Zhejiang Gongshang University, Hangzhou 310018, China

基金项目: 国家自然科学基金重点项目(71533005);浙江省自然科学基金项目(LY17D050006)

收稿日期: 2018-03-26; **网络出版日期:** 2018-00-00

* 通讯作者 Corresponding author. E-mail: zyouyang@rcees.ac.cn

Abstract: Asian dust events usually transport various pollutants that considerably impact atmospheric processes, biological ecosystems, and human health in downwind areas. To date, the composition and biodiversity dynamics of airborne bacteria have rarely been investigated in urban regions during Asian sandstorms, especially in northern China. Taking Beijing as an example, this study systematically evaluated the effects of sandstorms on the diversity characteristics of airborne bacteria. Air samples were collected from the roof of buildings for nine consecutive days (April 11—19, 2015), covering a sandstorm day (April 15, 2015), as well as four days prior to and after it. After extracting DNA and performing PCR amplifying and 16S rRNA gene high-throughput sequencing, a total of 169,122 high-quality sequences were obtained. Bioinformatics analysis on these mass data showed a high richness of airborne bacteria in this city. Sandstorms could not result in increased OTU numbers, but could result in increased Pielou, Shannon, and Simpson indexes. Bacterial communities in the Beijing atmosphere consisted of 35 phyla. Among them, Proteobacteria, Actinobacteria, Firmicutes, and Bacteroidetes were the dominant phyla, which accounted for 32.76%, 28.09%, 25.46% and 6.32% of the total sequences, respectively; Gemmatimonadetes (2.11%) and Acidobacteria (1.81%) followed; whereas the relative abundance of the other 29 phyla were < 1%. During the sandstorm, the relative abundance of Proteobacteria, Bacteroidetes, Gemmatimonadetes, and Acidobacteria significantly increased from 31.67%, 5.74%, 1.82%, 1.51% to 41.46%, 10.98%, 4.48%, and 4.26%, respectively; whereas Actinobacteria and Firmicutes remarkably decreased from 28.84%, 27.10% to 22.13% and 12.35%, respectively. The genera *Psychrobacter*, *Cellulomonas*, *Acinetobacter*, *Pseudomonas*, and *Clostridium* may be opportunistic pathogenic bacteria that potentially threaten human health. During the sandstorm, the relative abundance of these genera decreased, but their absolute abundance increased considerably. Sandstorms potentially weaken human immune capabilities, and hence the potential risks caused by these pathogenic bacteria to human health would increase during sandstorms. Hierarchical cluster and principal component analyses on bacterial communities indicated large differences between the samples from sandstorm and non-sandstorm weather conditions, whereas differences between the samples collected before and after the sandstorm were not obvious. No significant differences were detected in α and β diversity in the comparisons of the air bacterial community collected before and after the sandstorm (t test and Adonis test, $P > 0.05$). Thus, sandstorms could only temporarily affect the characteristics of airborne bacterial communities, but not permanently change them. Together, this study analyzed the diverse characteristics and dynamics of airborne bacterial communities in Beijing under sandstorm conditions, and the results could provide data support for environmental disaster warnings, as well as the prevention and control of airborne diseases in northern China.

Key Words: sandstorm; high-throughput sequencing; airborne bacteria; community diversity

细菌是空气中微生物的主要组成部分,在地球的发展和演化中起着至关重要的作用^[1]。通过大气运动,空气中的细菌将遥远的生态系统连接起来,参与生态系统的物质循环和能量流动。它们还可以吸收和反射太阳辐射,影响大气系统能量收支与平衡^[1]。部分细菌可作为冰核参与云的形成及降雨降雪等大气过程^[2-3]。此外,空气中的病原细菌还可以传播疾病,引发组织感染,威胁人群健康^[4-7]。因此,评价这些生态环境、气候效应和人群健康危害需要了解空气细菌的群落特征及其动态变化。

沙尘天气是浮尘、扬沙和沙尘暴天气的统称,是一种由大风将地面沙尘吹(卷)起、或被高空气流带到下游地区而造成的一种大气混浊现象^[8]。在全球尺度上,沙尘天气每年转运的沙尘粒子可达 5—50 亿吨^[9]。其中,亚洲沙尘约贡献 20%^[7]。沙尘粒子携带丰富多样的细菌^[9-10],从沙尘源向下游地区扩散与传播,影响中国北方地区,韩国和日本,甚至能漂洋过海抵达数千公里以外的北美大陆和法国阿尔卑斯山脉^[9]。Cha^[11]研究了亚洲沙尘对首尔空气细菌的影响,发现沙尘日芽孢杆菌属(*Bacillus*)、节杆菌属(*Arthrobacter*)、游动微菌属(*Planomicrobium*)相对丰度增加,其群落结构较非沙尘日发生了较大改变。类似的,Cha^[12],Jeon^[13]等人在韩国的调查和 Hara^[14],Lee^[15],Maki^[16],Yamaguchi^[17]等人在日本调查均得到了沙尘能够影响空气细菌群落

的结果。Park^[18]比较了沙尘天气下北京与大阪空气细菌群落的差别,发现北京较大阪拥有数目更多,种类更丰富的空气细菌群落。以上研究表明,亚洲沙尘影响其下游地区空气细菌群落,距沙尘源越近空气细菌群落的变化程度越大。

相较于韩国、日本,我国北方地区较沙尘源更近。一些学者在戈壁沙漠^[19]、敦煌^[10, 20]等沙尘源区研究了沙尘日和非沙尘日空气细菌群落的差别,但在人口密集的华北地区,相关研究较少。亚洲沙尘对空气细菌的影响研究多基于浮尘和扬沙天气^[1, 18, 21],对污染强度更大,环境及健康危害也更大的沙尘暴天气研究较少。中国的首都北京,距沙尘源 500–2500 km,每年亚洲沙尘的沉降量约为 180 g/m³,是大阪的 3600–36000 倍^[22–23],沙尘气溶胶在大气气溶胶中占有较大的比重^[8, 24],影响着 2000 万以上常居人口的身体健康。本研究以北京市为例,采用 16S rRNA 高通量测序手段系统研究了沙尘暴前期、中期及后期空气细菌群落的变化,旨在为更深层次的理解沙尘暴影响空气细菌群落的方式及强度,对中国北方城市沙尘暴环境灾害的预警及相应政策法规修订也具有重要的参考价值。

1 材料与方法

1.1 样本采集

采样地点位于中国科学院生态环境研究中心某楼顶(40°0′31″N, 116°20′34″E), 2015 年 4 月 15 日北京市发生了自 2002 年以来最大强度的沙尘暴。

采用大流量颗粒物采样器(2031 型,青岛崂山应用技术研究所)收集了沙尘暴前期(Before sandstorm, 4 月 11–14 日,依次记做 B1–B4),沙尘暴中期(During sandstorm, 4 月 15 日,记做 DS)和沙尘暴后期(After sandstorm, 4 月 16–19 日,依次记做 A1–A4)的空气总悬浮颗粒物(Total Suspended Particulate, TSP)样本。采样时间为 10:00 至次日 9:00,以 1.05 m³/min 的流速连续采样 23 h。为避免细菌污染,玻璃纤维滤膜在采样前使用马弗炉 500℃煅烧 4 h,膜托在每天采样前使用 75%的酒精消毒。

1.2 DNA 提取

将滤膜剪成条状,置于 50 mL 无菌离心管中。使用灭菌 1×PBS 缓冲液震荡清洗 3 次,悬浊液经 0.2 μm PES 滤膜过滤后,剪碎并转移至 PowerBead 管。65℃水浴 PowerBead 管 10 min, Fastprep-24 快速核酸提取仪(MP Biomedicals, 美国)破碎细胞。余下步骤按照 PowerSoil DNA 提取试剂盒(MOBIO, 美国)说明书进行。

1.3 高通量测序

细菌 16S rRNA 基因 V4 高变区使用通用引物 515F/806R 扩增,引物在 5′-端添加特异性 barcode 序列修饰用于区分样本。PCR 反应程序如下:94℃预变性 30 sec;94℃变性 30 sec,50℃退火 30 sec,72℃延伸 45 sec,共 30 个循环;72℃延伸 5 min。PCR 产物送至测序公司,经纯化回收,等浓度混样构建文库,经 Qubit 定量和文库检测,在 HiSeq2000 测序平台实施双末端测序。

1.4 生物信息分析

初始序列拆分至各个样本后,FLASH^[25]拼接。使用 Qiime^[26]去除冗余序列、Usearch^[27]去除 singletons 和 chimeras。使用 Mothur^[28]将 fasta 序列与 Silva 参考序列对齐,预聚类降低序列中的噪音,并将所得到的高质量序列进行分类。去除不能注释及注释结果为真菌、古菌、线粒体和叶绿体的序列,根据序列 97%的相似度划分操作分类单元(Operational Taxonomic Units, OTU)^[29]。将 Mothur 生成的 OTU 表格转换成 biom 文件,在 Qiime 中进一步注释门、纲、目、科、属各个级别的分类信息。使用 R 语言 vegan 包,按照最少序列数对 OTU 表格进行重抽样。在每个样本含量相同的基础上,分析样本的 α-多样性,通过 OTU 数量、Shannon 指数、Simpson 指数、Pielou 指数体现。

1.5 统计分析

使用两独立样本 t 检验和单样本 t 检验对空气细菌 alpha 多样性指数进行比较;使用 hellinger 转化后的 OTU 矩阵进行非加权组平均法(unweighted pair-group method with arithmetic means, UPGMA)聚类分析和主成

分分析(Principal Component Analysis, PCA)分析;使用 adonis 检验分析细菌群落结构差异性;所有统计分析和绘图均在 R 语言中完成。

2 结果与讨论

2.1 沙尘暴对空气细菌群落 α -多样性的影响

高通量测序共获得 169122 条高质量的基因序列,每个样本获得 8611—37719 条序列。按照最小样本量,每个样本随机挑取 8611 条序列进行 α -多样性分析(表 1)。以 97% 序列相似度为 OTU 划分标准,共获得 4208 个 OTU,每个样本获得 1163—1337 个 OTU。沙尘暴前期,空气细菌 OTU 数目、Pielou、Shannon、Simpson 指数分别为 1258 ± 80 、 0.73 ± 0.04 、 5.17 ± 0.29 和 0.97 ± 0.01 ,与沙尘暴后期(上述 4 个指数依次为 1290 ± 22 、 0.74 ± 0.02 、 5.29 ± 0.13 和 0.98 ± 0.01)无显著差异($P > 0.05$)。沙尘暴日空气细菌含有 1291 个 OTU,与非沙尘暴日(1274 ± 53 个 OTU)无明显差别($P > 0.05$);而 Pielou、Shannon、Simpson 指数在沙尘暴日达到 0.77、5.50 和 0.99,显著高于非沙尘暴日的 0.73 ± 0.03 、 5.23 ± 0.22 和 0.97 ± 0.01 ($P < 0.05$)。

表 1 空气细菌群落的 α -多样性指数
Table 1 α -diversity index of airborne bacteria

指标 Index	前期 Before sandstorm				中期 During sandstorm		后期 After sandstorm			
	B1	B2	B3	B4	DS		A1	A2	A3	A4
OTU 数量(OTU number)	1308	1337	1222	1163	1291		1292	1279	1320	1269
Pielou 指数(Pielou index)	0.76	0.75	0.70	0.69	0.77		0.75	0.71	0.75	0.74
Shannon 指数(Shannon indx)	5.45	5.39	4.94	4.90	5.50		5.37	5.11	5.40	5.29
Simpson 指数(Simpson index)	0.98	0.98	0.96	0.96	0.99		0.98	0.97	0.98	0.98

Maki 等研究发现戈壁沙漠腹地沙尘日和非沙尘日空气细菌群落稀释曲线无明显高低^[19],沙尘天气没有显著增加空气细菌的物种多样性;而韩国首尔市的调查结果却显示沙尘日空气细菌 OTU 数目显著高于非沙尘日^[11, 13]。调查结果的差异可能是城市距沙源地距离及受沙尘天气影响程度不同所致。沙漠表层沙土是沙源地空气细菌的重要来源^[30],因此沙源地空气中可能含有比较全面的沙漠细菌类群,沙尘天气只是增加了空气细菌的总数,但没有增加细菌的种类;而首尔市距沙尘源较远,平时空气中细菌群落结构受沙尘影响程度较低,即使沙尘天气引入少量空气细菌,也容易在湿润的空气沉降,不能持久悬浮在空气中,因此沙尘天气引入沙源细菌容易导致 OTU 数量显著增加。沙尘暴天气下北京市空气细菌类群没有显著增加,其原因可能与戈壁等沙源地情况类似。北京市距沙尘源较近,本次沙尘暴前发生的多次沙尘天气^[31]已将沙尘细菌引入城市空气中,它们在干燥多风的空气中长期悬浮,沙尘粒子可能已经成为北京市空气颗粒物的重要组成部分。

与首尔、大阪等地^[13, 32-33]的研究结果一致,沙尘天气下北京空气细菌群落 Shannon 和 Simpson 指数显著升高($P < 0.05$)。深入分析空气细菌多样性指数升高原因发现,首尔、大阪等地沙尘日空气细菌 OTU 数目显著增加($P < 0.05$)均匀度(Pielou 指数反映)无明显变化($P > 0.05$);而北京市正好相反,沙尘暴日空气细菌 OTU 数目无明显变化($P > 0.05$)均匀度显著增加($P < 0.05$)。因此首尔、大阪空气细菌综合多样性指数的增加主要由物种多样性增加所致,而北京该指数的增加则更多受均匀度增加影响。北京沙尘暴空气中细菌群落具有更高的均匀性,表明相较于沙漠这种极端环境,空气环境对细菌的选择压力更大;而首尔、大阪等地沙尘日空气细菌群落 Pielou 指数无明显变化,可能是沙尘污染程度较低,空气的选择压力仍然起主导作用导致。

2.2 沙尘暴对空气细菌群落结构的影响

9 个空气细菌样本中发现 405 个共有 OTU,仅占 OTU 总数的 7.07%,但其所代表的序列却占据总序列的 85.32%。对各样本细菌群落分别进行门、属水平上的群落组成分析。门水平共发现 35 个细菌门,其中,变形菌门(Proteobacteria)、放线菌门(Actinobacteria)、厚壁菌门(Firmicutes)和拟杆菌门(Bacteroidetes)相对丰度最高(图 1),分别占总序列的 32.76%、28.09%、25.46%和 6.32%。芽单胞菌门(Gemmatimonadetes)和酸杆菌门

(*Acidobacteria*) 相对丰度次之,分别占序列总数的 2.11% 和 1.81%,其他细菌门的相对丰度均低于 1%。在属水平上(图 1),北京市空气细菌由 1120 个属构成,其中准确注释且相对丰度超过 1% 的优势菌属仅有 15 个,库克菌属(*Kocuria*)、马赛菌属(*Massilia*)、鞘脂单胞菌属(*Sphingomonas*)、嗜冷杆菌属(*Psychrobacter*) 和副球菌属(*Paracoccus*) 相对丰度最高,分别占总序列数的 5.06%、3.63%、3.13%、2.83% 和 2.31%;微红微菌属(*Rubellimicrobium*)、芽生球菌属(*Blastococcus*)、纤维单胞菌属(*Cellulomonas*) 等 10 属相对丰度在 1%—2% 之间。

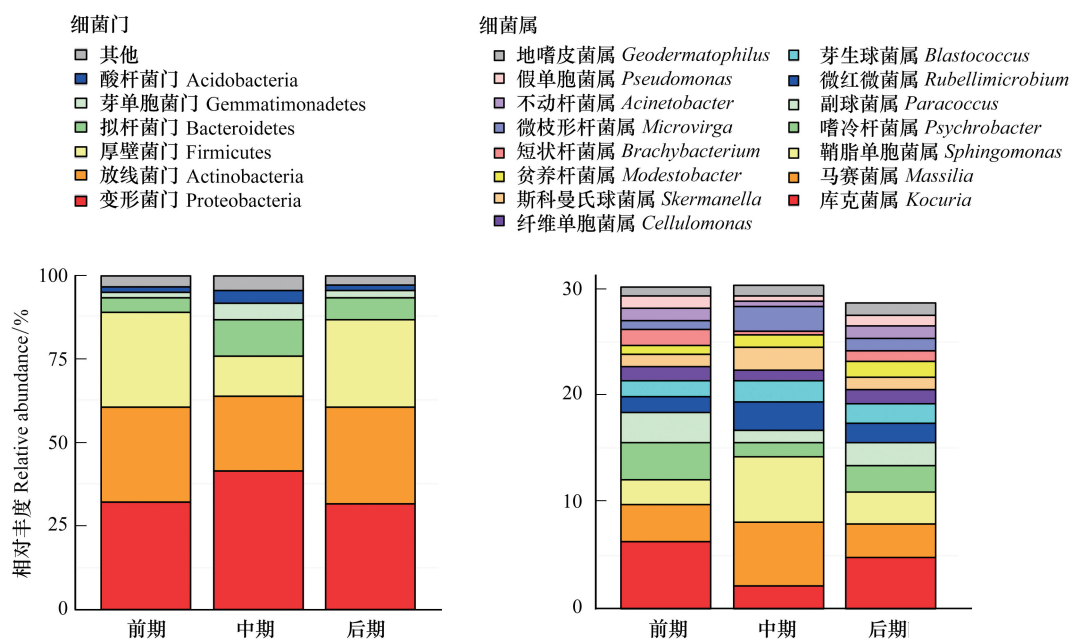


图 1 空气细菌群落结构(门/属级别)

Fig.1 Community structure of airborne bacteria at phylum and genus level

深入分析沙尘暴空气中优势菌群变化,门水平上(图 2)变形菌门(*Proteobacteria*)、拟杆菌门(*Bacteroidetes*)、芽单胞菌门(*Gemmatimonadetes*)和酸杆菌门(*Acidobacteria*)显著升高($P < 0.05$),分别由 31.67%、5.74%、1.82%、1.51% 升高至 41.46%、10.98%、4.48% 和 4.26%;而放线菌门(*Actinobacteria*)、厚壁菌门(*Firmicutes*)显著降低($P < 0.05$),分别由 28.84%、27.10% 降低至 22.13% 和 12.35%。属水平上(图 2),马赛菌属(*Massilia*)、鞘脂单胞菌属(*Sphingomonas*)、微红微菌属(*Rubellimicrobium*)、斯科曼氏菌属(*Skermanella*)、微枝形杆菌属(*Microvirga*) 相对丰度增加 0.98%—3.47% ($P < 0.05$);库克菌属(*Kocuria*)、嗜冷杆菌属(*Psychrobacter*)、副球菌属(*Paracoccus*)、纤维单胞菌属(*Cellulomonas*)、短杆菌属(*Brachybacterium*)、不动杆菌属(*Acinetobacter*) 和假单胞菌属(*Pseudomonas*) 相对丰度降低 0.21%—3.42% ($P < 0.05$);芽生球菌属(*Blastococcus*)、贫养杆菌属(*Modestobacter*)、地嗜皮菌属(*Geodermatophilus*) 在沙尘暴空气中无显著变化($P > 0.05$)。

共有 OTU、优势菌门(相对丰度 $\geq 1\%$) 代表绝大多数空气细菌序列,表明各样本共有类群,尤其是优势类群相对丰度的差异是导致细菌群落结构不同的主要原因,与 2.1 章节中沙尘暴日 Shannon、Simpson 指数增加主要受 Pielou 指数影响的结果一致。北京市空气细菌由变形菌门(*Proteobacteria*)、放线菌门(*Actinobacteria*)、厚壁菌门(*Firmicutes*) 和拟杆菌门(*Bacteroidetes*) 主导,这四个细菌门在亚洲沙尘源(如塔克拉玛干^[34]、戈壁沙漠^[19] 和敦煌^[10]) 及其下游地区(如太原^[21]、北京^[21]、首尔^[11-13, 21]、大阪^[35]) 的空气中同样占据主导地位。然而沙尘天气下,这些优势菌门在不同城市却表现出不一致特征。

本研究中,沙尘暴天气下,空气变形菌门(*Proteobacteria*)、拟杆菌门(*Bacteroidetes*) 相对丰度显著增加,放线菌门(*Actinobacteria*)、厚壁菌门(*Firmicutes*) 相对丰度显著降低。与戈壁沙漠空气细菌群落^[19] 相比, α -变形

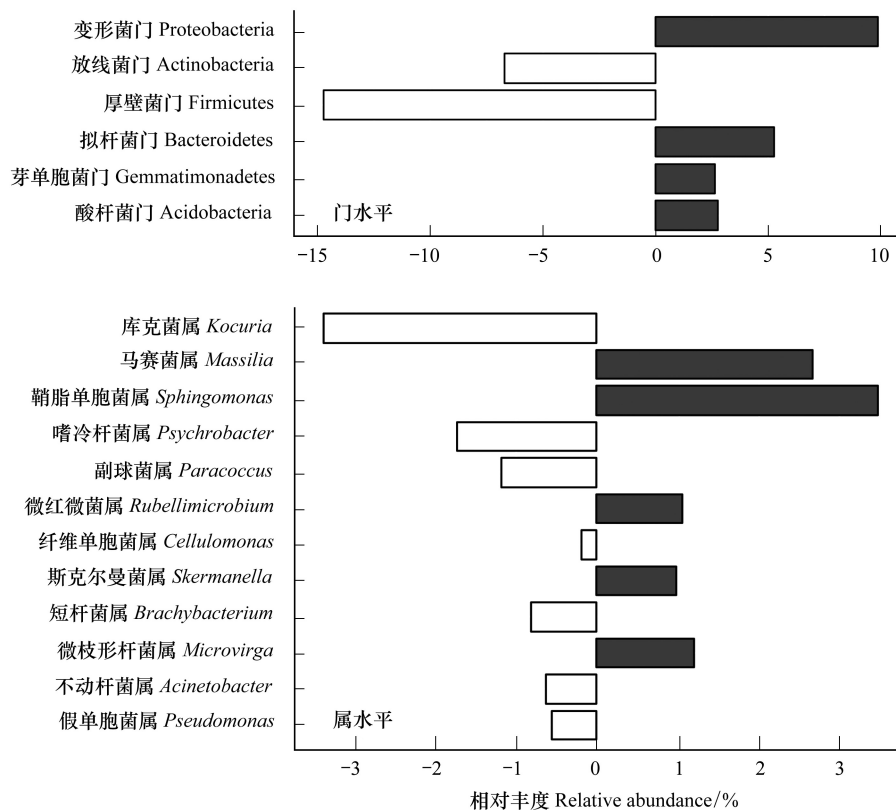


图2 沙尘暴中相对丰度显著变化的优势细菌类群

Fig.2 Abundant bacterial taxa which relative abundance significantly changed in Sandstorm

菌纲 (Alphaproteobacteria) 和厚壁菌门 (Firmicutes) 的变化趋势一致, 而拟杆菌门 (Bacteroidetes) 的变化趋势相反; 与首尔空气细菌群落^[12]相比, 变形菌门 (Proteobacteria)、放线菌门 (Actinobacteria) 的变化趋势相反。沙源地细菌群落特征的差异可能是研究结果不一致的首要原因。An 等调查证实亚洲沙尘两大沙源地塔克拉玛干沙漠和戈壁沙漠表层沙粒的细菌群落差异较大^[36], 格尔木市、库尔勒市沙源地变形菌门 (Proteobacteria)、拟杆菌门 (Bacteroidetes) 相对丰度远高于放线菌门 (Actinobacteria) 和厚壁菌门 (Firmicutes); 而敦煌和戈壁沙漠厚壁菌门 (Firmicutes) 相对丰度最高; Katra 等^[37]的调查也显示连续两次不同沙源的沙尘暴过程, 贝尔谢巴市空气细菌群落显著不同, 表现出沙尘源决定空气细菌群落的重要性。

属水平上非沙尘天气下库克菌属 (*Kocuria*), 马赛菌属 (*Massilia*), 冷杆菌属 (*Psychrobacter*), 鞘脂单胞菌属 (*Sphingomonas*), 副球菌属 (*Paracoccus*) 相对丰度最高 (>2%), Du 等人^[38-39]的调查也显示这些属在北京春季具有较高的丰度。沙尘暴天气下, 菌属的变化趋势取决于其在沙源细菌群落中的相对丰度。显著升高的优势菌属中, 马赛菌属 (*Massilia*)、鞘脂单胞菌属 (*Sphingomonas*)、微红微菌属 (*Rubellimicrobium*) 是戈壁、塔克拉玛干等亚洲沙漠的优势菌属, 也是亚洲沙尘事件的空气优势菌属^[11-12, 15-16, 21, 40]。斯克尔曼氏菌属 (*Skermanella*)^[41]和微枝形杆菌属 (*Microvirga*)^[12, 21]均从亚洲沙漠表层沙土中检测到其存在。

沙尘暴天气中相对丰度显著升高的优势属中, 基本不含人类病原菌, 而显著下降的优势属中, 冷杆菌属 (*Psychrobacter*)^[42], 纤维单胞菌属 (*Cellulomonas*)^[43], 不动杆菌属 (*Acinetobacter*)^[44], 假单胞菌属 (*Pseudomonas*)^[45]和梭菌属 (*Clostridium*)^[46]可能含有人类病原菌。Cao^[47]、Gao^[48]、Woo^[49]等人在空气中曾检测出鲍曼不动杆菌 (*Acinetobacter baumannii*)^[48-49]、铜绿假单胞菌 (*Pseudomonas aeruginosa*)^[48-49]、破伤风梭菌 (*Clostridium tetani*)^[48]、产气荚膜梭菌 (*Clostridium perfringens*)^[47]的存在, 它们可能导致肺炎、呼吸道感染、皮肤感染、菌血症, 甚至引发心内膜炎、脓胸、败血症等多种疾病^[50]。沙尘暴天气下, 空气细菌浓度 10^2 – 10^3 倍增^[13, 17, 35], 上述人类潜在病原菌相对丰度降低, 而绝对丰度却是大幅增加。加之沙尘暴降低了人体免疫

力^[9, 51], 机会致病菌的人群健康风险明显增强。

2.3 沙尘暴对空气细菌群落 β -多样性的影响

聚类分析(图 3)结果显示,沙尘暴发生时空气细菌群落发生显著改变,而沙尘暴前后期空气细菌群落比较相似;PCA 分析(图 4)得到一致结果,沙尘暴日空气细菌群落与非沙尘暴日相距较远,沙尘暴前后空气细菌群落更为接近。沙尘暴对空气细菌群落的显著影响主要体现在沙尘暴发生时。沙尘暴过后,沙尘粒子受重力作用自然沉降,其对空气细菌的主导地位也随之解除,空气细菌又恢复至原来的本地土壤、水体、植物等自然源为主导的状态^[52]。

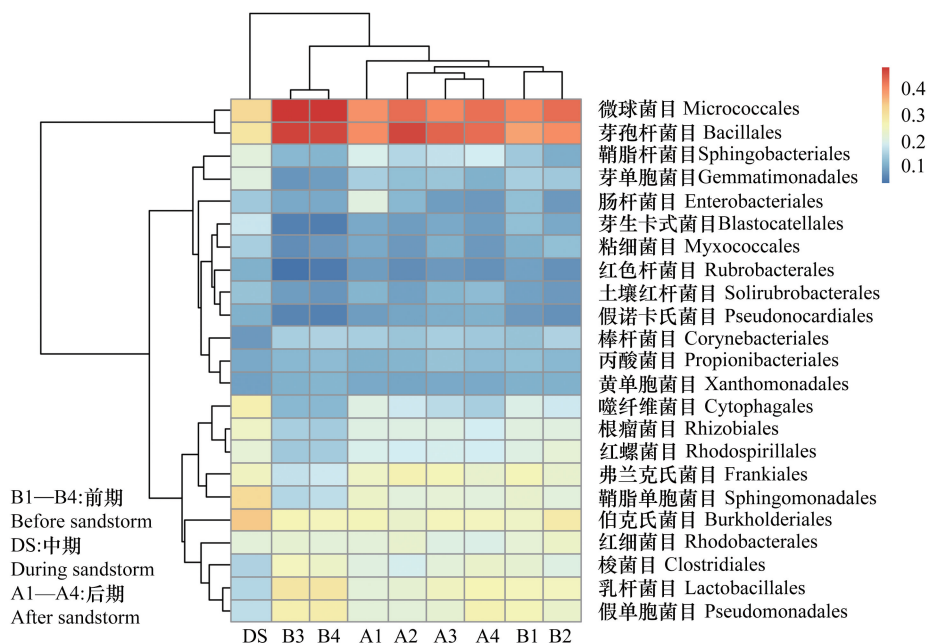


图 3 基于优势目的 UPGMA 聚类分析

Fig.3 Hierarchical cluster analysis of abundant orders using the UPGMA method

PCA 分析(图 4)显示,沙尘暴后期空气细菌群落位于沙尘暴日和沙尘暴前期之间,体现沙尘暴后期空气细菌群落结构向前期恢复的趋势。沙尘暴后期空气细菌群落较前期变异小,可能也受到沙尘暴的影响。沙尘暴前后期空气细菌群落没有显著差异(adonis 检验, $P > 0.05$),说明沙尘暴的对空气细菌群落的持续影响力较弱。日本学者的调查^[22, 35]发现,中等程度亚洲沙尘天气不能干扰大阪空气细菌群落结构。我们的研究进一步表明,即使是沙尘暴,也只能暂时影响空气细菌群落,不能引起其持久变化。

3 结论

(1)利用 16S rRNA 基因高通量测序技术,系统研究了沙尘暴对北京市空气细菌多样性特征的影响。结果表明,北京市空气细菌物种多样性较高,沙尘暴不能引起空气中细菌 OTU 数目的增加,但沙尘暴天气下空气细菌群落 Shannon、Simpson 指数的显著增加。

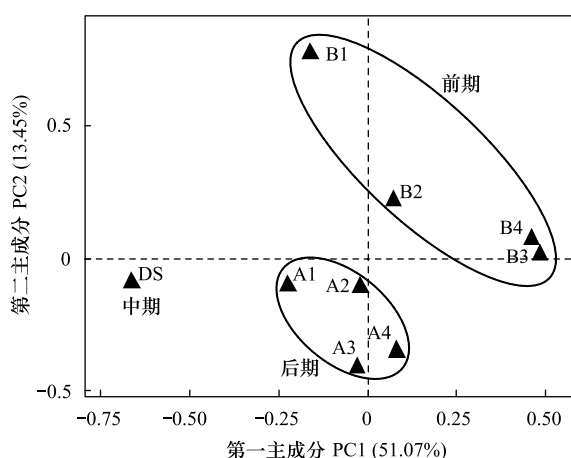


图 4 基于所有 OTU 的 PCA 分析

Fig.4 PCA analysis based on all OTUs

(2) 变形菌门 (Proteobacteria)、放线菌门 (Actinobacteria)、厚壁菌门 (Firmicutes) 和拟杆菌门 (Bacteroidetes) 是沙尘暴中期及前后期的主导细菌门, 沙尘暴天气变形菌门 (Proteobacteria)、拟杆菌门 (Bacteroidetes) 相对丰度显著升高, 放线菌门 (Actinobacteria)、厚壁菌门 (Firmicutes) 相对丰度显著降低。

(3) 冷杆菌属 (*Psychrobacter*)、纤维单胞菌属 (*Cellulomonas*)、不动杆菌属 (*Acinetobacter*)、假单胞菌属 (*Pseudomonas*) 和梭菌属 (*Clostridium*) 细菌在沙尘暴天气相对丰度下降, 但绝对丰度显著增加。上述的菌属可能含有人类条件病原菌, 且沙尘暴能降低人体免疫力, 因此沙尘暴天气的健康风险显著增强。

(4) 沙尘暴前后期空气细菌 OTU 数目、Pielou、Shannon、Simpson 指数, 群落结构均无显著差异 ($P < 0.05$), 表明沙尘暴后空气细菌迅速向原有群落结构恢复, 沙尘暴只能暂时影响空气中细菌群落特征。

参考文献 (References):

- [1] Yuan H L, Zhang D Z, Shi Y N, Li B Z, Yang J S, Yu X J, Chen N, Kakikawa M. Cell concentration, viability and culture composition of airborne bacteria during a dust event in Beijing. *Journal of Environmental Sciences*, 2017, 55: 33-40.
- [2] Christner B C, Cai R M, Morris C E, McCarter K S, Foreman C M, Skidmore M L, Montross S N, Sands D C. Geographic, seasonal, and precipitation chemistry influence on the abundance and activity of biological ice nucleators in rain and snow. *Proceedings of the National Academy of Sciences of the United States of America*, 2008, 105(48): 18854-18859.
- [3] Bowers R M, Lauber C L, Wiedinmyer C, Hamady M, Hallar A G, Fall R, Knight R, Fierer N. Characterization of airborne microbial communities at a high-elevation site and their potential to act as atmospheric ice nuclei. *Applied and Environmental Microbiology*, 2009, 75(15): 5121-5130.
- [4] Polymenakou P N. Atmosphere: a source of pathogenic or beneficial microbes? *Atmosphere*, 2012, 3(4): 87-102.
- [5] Liu H, Zhang X, Zhang H, Yao X W, Zhou M, Wang J Q, He Z F, Zhang H H, Lou L P, Mao W H, Zheng P, Hu B L. Effect of air pollution on the total bacteria and pathogenic bacteria in different sizes of particulate matter. *Environmental Pollution*, 2018, 233: 483-493.
- [6] Zhen Q, Deng Y, Wang Y Q, Wang X K, Zhang H X, Sun X, Ouyang Z Y. Meteorological factors had more impact on airborne bacterial communities than air pollutants. *Science of the Total Environment*, 2017, 601-602: 703-712.
- [7] Goudie A S. Desert dust and human health disorders. *Environment International*, 2014, 63: 101-113.
- [8] 韩晨, 祁建华, 谢锦测, 张文倩, 李先国, 张大海. 青岛近海春季沙尘天气可培养真菌及其潜在健康风险. *城市环境与城市生态*, 2015, 28(4): 18-23.
- [9] Griffin D W. Atmospheric movement of microorganisms in clouds of desert dust and implications for human health. *Clinical Microbiology Reviews*, 2007, 20(3): 459-477.
- [10] Puspitasari F, Maki T, Shi G Y, Bin C, Kobayashi F, Hasegawa H, Iwasaka Y. Phylogenetic analysis of bacterial species compositions in sand dunes and dust aerosol in an Asian dust source area, the Taklimakan Desert. *Air Quality, Atmosphere & Health*, 2015, 9(6): 631-644.
- [11] Cha S, Srinivasan S, Jang J H, Lee D, Lim S, Kim K S, Jheong W, Lee D W, Park E R, Chung H M, Choe J, Kim M K, Seo T. Metagenomic analysis of airborne bacterial community and diversity in Seoul, Korea, during December 2014, Asian dust event. *PLoS One*, 2017, 12(1): e0170693.
- [12] Cha S, Lee D, Jang J H, Lim S, Yang D, Seo T. Alterations in the airborne bacterial community during Asian dust events occurring between February and March 2015 in South Korea. *Scientific Reports*, 2016, 6: 37271.
- [13] Jeon E M, Kim H J, Jung K, Kim J H, Kim M Y, Kim Y P, Ka J O. Impact of Asian dust events on airborne bacterial community assessed by molecular analyses. *Atmospheric Environment*, 2011, 45(25): 4313-4321.
- [14] Hara K, Zhang D Z. Bacterial abundance and viability in long-range transported dust. *Atmospheric Environment*, 2012, 47: 20-25.
- [15] Lee S, Choi B, Yi S M, Ko G. Characterization of microbial community during Asian dust events in Korea. *Science of the Total Environment*, 2009, 407(20): 5308-5314.
- [16] Maki T, Puspitasari F, Hara K, Yamada M, Kobayashi F, Hasegawa H, Iwasaka Y. Variations in the structure of airborne bacterial communities in a downwind area during an Asian dust (Kosa) event. *Science of the Total Environment*, 2014, 488-489: 75-84.
- [17] Yamaguchi N, Ichijo T, Sakotani A, Baba T, Nasu M. Global dispersion of bacterial cells on Asian dust. *Scientific Reports*, 2012, 2: 525.
- [18] Park J, Li P F, Ichijo T, Nasu M, Yamaguchi N. Effects of Asian dust events on atmospheric bacterial communities at different distances downwind of the source region. *Journal of Environmental Sciences*, 2018, doi: 10.1016/j.jes.2017.12.019. (in press)
- [19] Maki T, Kurosaki Y, Onishi K, Lee K C, Pointing S B, Jugder D, Yamanaka N, Hasegawa H, Shinoda M. Variations in the structure of airborne bacterial communities in Tsogt-Ovoo of Gobi desert area during dust events. *Air Quality, Atmosphere & Health*, 2017, 10(3): 249-260.

- [20] Kakikawa M, Kobayashi F, Maki T, Yamada M, Higashi T, Chen B, Shi G Y, Hong C S, Tobo Y, Iwasaka Y. Dustborne microorganisms in the atmosphere over an Asian dust source region, Dunhuang. *Air Quality, Atmosphere & Health*, 2008, 1(4): 195-202.
- [21] An S, Sin H H, DuBow M S. Modification of atmospheric sand-associated bacterial communities during Asian sandstorms in China and South Korea. *Heredity*, 2015, 114(5): 460-467.
- [22] Park J, Ichijo T, Nasu M, Yamaguchi N. Investigation of bacterial effects of Asian dust events through comparison with seasonal variability in outdoor airborne bacterial community. *Scientific Reports*, 2016, 6: 35706.
- [23] Yamaguchi N, Baba T, Ichijo T, Himezawa Y, Enoki K, Saraya M, Li P F, Nasu M. Abundance and community structure of bacteria on Asian dust particles collected in Beijing, China, during the Asian dust season. *Biological & Pharmaceutical Bulletin*, 2016, 39(1): 68-77.
- [24] Wang Y, Zhuang G S, Tang A H, Zhang W J, Sun Y L, Wang Z F, An Z S. The evolution of chemical components of aerosols at five monitoring sites of China during dust storms. *Atmospheric Environment*, 2007, 41(5): 1091-1106.
- [25] Magoč T, Salzberg S L. FLASH: Fast length adjustment of short reads to improve genome assemblies. *Bioinformatics*, 2011, 27(21): 2957-2963.
- [26] Caporaso J G, Kuczynski J, Stombaugh J, Bittinger K, Bushman F D, Costello E K, Fierer N, Peña A G, Goodrich J K, Gordon J I, Huttley G A, Kelley S T, Knights D, Koenig J E, Ley R E, Lozupone C A, McDonald D, Muegge B D, Pirrung M, Reeder J, Sevinsky J R, Turnbaugh P J, Walters W A, Widmann J, Yatsunenko T, Zaneveld J, Knight R. QIIME allows analysis of high-throughput community sequencing data. *Nature Methods*, 2010, 7(5): 335-336.
- [27] Edgar R C. UPARSE: Highly accurate OTU sequences from microbial amplicon reads. *Nature Methods*, 2013, 10(10): 996-998.
- [28] Schloss P D, Westcott S L, Ryabin T, Hall J R, Hartmann M, Hollister E B, Lesniewski R A, Oakley B B, Parks D H, Robinson C J, Sahl J W, Stres B, Thallinger G G, Van Horn D J, Weber C F. Introducing mothur: Open-source, platform-independent, community-supported software for describing and comparing microbial communities. *Applied and Environmental Microbiology*, 2009, 75(23): 7537-7541.
- [29] Rognes T, Flouri T, Nichols B, Quince C, Mahé F. VSEARCH: A versatile open source tool for metagenomics. *PeerJ*, 2016, 4: e2584.
- [30] Smets W, Moretti S, Denys S, Lebeer S. Airborne bacteria in the atmosphere: Presence, purpose, and potential. *Atmospheric Environment*, 2016, 139: 214-221.
- [31] 赖芬芬, 孙军. 2015 年 3 月大气环流和天气分析. *气象*, 2015, 41(6): 786-792.
- [32] Mazar Y, Cytryn E, Erel Y, Rudich Y. Effect of dust storms on the atmospheric microbiome in the Eastern Mediterranean. *Environmental Science & Technology*, 2016, 50(8): 4194-4202.
- [33] Maki T, Hara K, Iwata A, Lee K C, Kawai K, Kai K, Kobayashi F, Pointing S B, Archer S, Hasegawa H, Iwasaka Y. Variations in airborne bacterial communities at high altitudes over the Noto Peninsula (Japan) in response to Asian dust events. *Atmospheric Chemistry and Physics*, 2017, 17(19): 11877-11897.
- [34] 王丹丹, 林青, 欧提库尔·玛合木提, 杨红梅, 娄恺. 塔克拉玛干沙尘暴源区空气细菌群落多样性. *微生物学报*, 2018, doi: 10.13343/j.cnki.wsxb.20170458. (in press)
- [35] Yamaguchi N, Park J, Kodama M, Ichijo T, Baba T, Nasu M. Changes in the airborne bacterial community in outdoor environments following asian dust events. *Microbes and Environments*, 2014, 29(1): 82-88.
- [36] An S, Couteau C, Luo F, Neveu J, DuBow M S. Bacterial diversity of surface sand samples from the Gobi and Taklamaken deserts. *Microbial Ecology*, 2013, 66(4): 850-860.
- [37] Katra I, Arotsker L, Krasnov H, Zaritsky A, Kushmaro A, Ben-Dov E. Richness and diversity in dust stormborne biomes at the southeast mediterranean. *Scientific Reports*, 2014, 4: 5265.
- [38] Du P R, Du R, Ren W S, Lu Z D, Fu P Q. Seasonal variation characteristic of inhalable microbial communities in PM_{2.5} in Beijing city, China. *Science of the total environment*, 2018, 610-611: 308-315.
- [39] Du P R, Du R, Ren W S, Lu Z D, Zhang Y, Fu P Q. Variations of bacteria and fungi in PM_{2.5} in Beijing, China. *Atmospheric Environment*, 2018, 172: 55-64.
- [40] Polymenakou P N, Mandalakis M, Stephanou E G, Tselepidis A. Particle size distribution of airborne microorganisms and pathogens during an intense African dust event in the eastern Mediterranean. *Environmental Health Perspectives*, 2007, 116(3): 292-296.
- [41] An H L, Zhang L, Tang Y L, Luo X S, Sun T J, Li Y W, Wang Y, Dai J, Fang C X. *Skermanella xinjiangensis* sp. nov., isolated from the desert of Xinjiang, China. *International Journal of Systematic and Evolutionary Microbiology*, 2009, 59(Pt 6): 1531-1534.
- [42] María O A J, Miguel S C J, Fabiola G A, Elizabeth G D, Araceli R C, Patricia A P, Claudia W A, Maribel G V, Gloria L Á, Jeanette G C A, Alberto D Q J, Alfonso M T, Ernesto R G J. Fatal *Psychrobacter* sp. infection in a pediatric patient with meningitis identified by metagenomic next-generation sequencing in cerebrospinal fluid. *Archives of Microbiology*, 2016, 198(2): 129-135.
- [43] Ohtaki H, Ohkusu K, Sawamura H, Ohta H, Inoue R, Iwasa J, Ito H, Murakami N, Ezaki T, Moriwaki H, Seishima M. First report of acute cholecystitis with sepsis caused by *Cellulomonas denverensis*. *Journal of Clinical Microbiology*, 2009, 47(10): 3391-3393.

- [44] Visca P, Seifert H, Towner K J. *Acinetobacter* infection-an emerging threat to human health. IUBMB Life, 2011, 63(12): 1048-1054.
- [45] Morrison Jr A J, Wenzel R P. Epidemiology of infections due to *Pseudomonas aeruginosa*. Reviews of Infectious Diseases, 1984, 6(S3): S627-S642.
- [46] Centers for Disease Control and Prevention (CDC). Severe *Clostridium difficile*-associated disease in populations previously at low risk-four states, 2005. MMWR. Morbidity and Mortality Weekly Report, 2005, 54(47): 1201-1205.
- [47] Cao C, Jiang W J, Wang B Y, Fang J H, Lang J D, Tian G, Jiang J K, Zhu T F. Inhalable microorganisms in Beijing's PM_{2.5} and PM₁₀ pollutants during a severe smog event. Environmental Science & Technology, 2014, 48(3): 1499-1507.
- [48] Gao J F, Fan X Y, Li H Y, Pan K L. Airborne bacterial communities of PM_{2.5} in Beijing-Tianjin-Hebei megalopolis, China as revealed by illumina MiSeq sequencing: a case study. Aerosol and Air Quality Research, 2017, 17(3): 788-798.
- [49] Woo A C, Brar M S, Chan Y, Lau M C Y, Leung F C C, Scott J A, Vrijmoed L L P, Zawar-Reza P, Pointing S B. Temporal variation in airborne microbial populations and microbially-derived allergens in a tropical urban landscape. Atmospheric Environment, 2013, 74: 291-300.
- [50] 李明远, 徐志凯. 医学微生物学(第三版). 北京: 人民卫生出版社, 2015: 176-211.
- [51] Honda A, Matsuda Y, Murayama R, Tsuji K, Nishikawa M, Koike E, Yoshida S, Ichinose T, Takano H. Effects of Asian sand dust particles on the respiratory and immune system. Journal of Applied Toxicology, 2014, 34(3): 250-257.
- [52] 周康辉, 方翀. 2015 年 4 月大气环流和天气分析. 气象, 2015, 41(7): 915-920.