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城市污水再生处理中微量有机污染物控制的关键难题与解决思路 王文龙,吴乾元,杜烨,黄南,陆韻,魏东斌,胡洪营







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冬青和女贞叶表面颗粒物微形态及叶际细菌群落结构

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摘要:植物叶表面颗粒物和微生物的结构动态对近地表环境和生态具有重要意义. 本研究以青岛市不同功能区冬青和女贞植 物的新鲜叶片为研究对象,利用环境扫描电镜观察叶表面颗粒物与微生物微形态特征及季节变化规律; 采用 Illumina 高通量 测序技术分析植物叶际细菌群落结构. 结果表明,冬青和女贞植物叶片颗粒物滞留量秋、冬季节高于春、夏两季,两种植物叶 片更易滞留 PM₁₀和 PM, , , 但冬青叶片的滞尘能力尤其是对粒径大于 10 μm 的颗粒物滞留效果高于女贞,城市主干道植物叶 片滯尘能力高于文教区和城市公园植物,受地面扬尘和汽车尾气影响较大;另植物叶际微生物组成存在季节差异,叶片易滯 留不同类型真菌、真菌孢子及菌丝体等,且两种植物因叶片革质更适宜菌丝体生长,此外女贞叶表皮还易镶嵌一种表面均匀 褶皱的真菌孢子,微生物丰富度城市公园 > 城市主干道 > 文教区,湿度对微生物的生长繁殖呈现积极影响;高通量测序下不 同季节的冬青和女贞植物叶际细菌群落差异显著,春季植物叶际细菌丰度最高,夏季最低,两种植物叶际细菌相对丰度最高 的优势菌门和优势菌纲为变形菌门(Proteobacteria)和 γ-变形菌纲(γ-Proteobacteria),且同一功能区两种植物共用一个核心微 生物群,另外受地面扬尘污染严重的城市主干道植物叶际细菌群落结构与其他两个功能区差异明显. 本研究结果证明城市不 同功能区典型绿化植物叶表面颗粒物与微生物群落结构之间存在显著相关性,为城市绿化建设提供参考.

关键词:叶表面颗粒物;微形态特征;叶际微生物;不同功能区;群落结构

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Micro-morphological Characteristics of Particles on Holly and Ligustrum Leaf Surfaces and Seasonal Changes in Bacterial Communities

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Abstract: The characteristics of particles and microorganisms on leaf surfaces have great significance for the near-surface environment and ecology. Here, fresh leaves of holly and Ligustrum were examined from different functional areas in Qingdao. Environmental scanning electron microscopy was used to observe particles and microorganisms on the leaf surfaces during different seasonal, and Illumina high-throughput sequencing was performed to analyze the phyllosphere bacterial community structure. The results showed that the retention of TSP on leaves in autumn and winter was higher than in spring and summer. The leaves of the two plants were more likely to retain PM₁₀ and PM_{2.5}; however, the dust retention capacity of holly leaves was higher than that of Ligustrum, especially for particle sizes greater than 10 µm. The numbers of particles on the leaf surfaces along an urban main road were higher than in two other locations, and were greatly affected by ground dust and automobile exhaust emissions. The community structure of phyllospheric microorganisms showed distinct seasonal variation, with different types of fungi, fungal spores, and mycelium observed on the leaf surfaces. Mycelium was more frequently detected on leathery leaves, and fungal spores with even folds were detected on the leaf epidermis of Ligustrum. The relative abundances of phyllospheric microorganisms were highest on leaves from an urban park, explained by a positive effect of humidity on growth. Significant differences in bacterial community abundance were observed between seasons. Specifically, bacterial abundance was highest in spring and lowest in summer. γ-Proteobacteria were the dominant bacteria, and the two plants shared a similar core microbial community. In addition, the phyllospheric bacterial community structure of leaves from urban arterial roads with ground dust pollution was significantly different from the leaves collected from other city areas. Our research results suggest a significant correlation between the leaf-surface particles and microbial community structure on representative plants in different areas of the city, which provides reference information for urban greening activities.

Key words: particulates on leaves; micromorphological characteristics; phyllosphere microorganism; different function areas; community structure

随着城市化的快速推进,颗粒物已经成为城市 的首要污染物^[1,2],空气中悬浮颗粒物(total suspended particle, TSP)聚集大量酸性氧化物、有害 重金属、真菌、细菌和病毒等[3],对环境和生物造 成危害. 生态系统中植物叶片作为滞留大气颗粒物 的主要器官[4],可以有效吸附阻挡粉尘颗粒.而叶 片滯留颗粒物能力受多种因素制约,叶片单位面积 大小及叶表面微结构对滞留 PM_{2.5}有直接影响^[5,6],

叶表面粗糙度也是滞尘能力的重要决定因素[7]:且 不同地点的植物对颗粒物的滞留效果差异显著[8], 连续暴露在汽车尾气排放地点的叶片颗粒物种类更 丰富[9]. 目前研究都集中在植物叶片微形态特

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征^[4,8,10],直接观察植物叶表面颗粒物微形态较少. 另外,受不同污染来源和气候条件等影响,不同城市植物叶表面颗粒物滞留差异明显^[11].且对植物叶表面颗粒物的研究主要集中在北京^[4,8,12]和南方城市^[13,14],青岛作为新一线城市,虽有较少研究针对青岛市部分植物叶片滞尘能力^[15,16],但依旧缺少植物叶表面颗粒物和微生物微形态的相关研究.

叶际微生物^[17]是叶片滯留颗粒物的特殊组成成分^[18],包括各种真菌、细菌和藻类等^[19].叶际微生物具有多种功能,部分叶际真菌可以促进植物生长^[9],影响全球的碳氮循环^[20];叶表面部分病原微生物互相作用可调节植物的抗病性^[21];且叶际微生物的某些成分会引发植物病害,使人和动物产生中毒、过敏和感染反应^[22].但目前研究主要集中在真菌^[23~25]和病原微生物^[21]的多样性变化,叶际微生物的微形态特征观察研究较少,而利用环境扫描电镜在低真空下对植物叶表面滞留微生物进行显微观察,可以反映自然状态下植物叶际微生物形貌与组成^[26].另有研究显示叶际细菌与空气中细菌存在一定差异^[27],且叶际细菌群落空间和时间差异性^[28]显著,本实验也对植物叶际细菌群落变化进行了研究.

本文以青岛市 3 个功能区的冬青和女贞植物为研究对象,采用 S-3500N 型环境扫描电镜在低真空下观察植物新鲜叶片滞留颗粒物和微生物的数量、粒径、类型及季节变化;同时叶际细菌通过Illumina 高通量测序^[29],对群落组成、相对丰度及优势菌差异等进行分析. 本研究全面了解了叶表面颗粒物和微生物分布规律和群落组成,以期为后续研究叶表面颗粒物和叶际微生物对植物、大气和生态环境的影响提供数据参考,对青岛市居民健康及旅游业的长远发展有着重要意义.

1 材料与方法

1.1 实验设计

1.1.1 研究区域概况

本研究地点选取青岛市青岛理工大学(文教

区)、山东路(城市主干道)和中山公园(城市公园).青岛理工大学位于青岛市市北区抚顺路,校内主要以学生活动为主;山东路是青岛市重要主干道,横跨两区,车流量、人流量都较大;中山公园位于青岛市市南区,三面环山,南向大海,植被景观特点突出,居民与旅客居多.具体取样地点见图 1.选取 3 个功能区距离主要道 5~10 m 处植物中下层无破损的叶片,采样高度平均为 1.5~1.7 m,同种植物每隔 3 m 取一次叶片,共做 5 次重复性实验.叶片受空气及人类活动影响明显,颗粒物种类数量丰富,且对人类健康有着直接影响,研究更具代表性.



图 1 植物样品采集地点分布示意

Fig. 1 Distribution of plant sample collection locations

1.1.2 选择样品概况

本实验样品选取冬青和女贞植物新鲜叶片. 两种植物属于常绿阔叶植物,广泛分布于青岛市各个功能区,滞尘能力较强,可以系统反映出叶际细菌群落变化. 具体植物及叶片性状描述见表 1.

1.2 样品采集期间气象要素

植物新鲜叶片采集时间为 2019 年的 4、7、10 月及 2020 年 1 月晴朗无风天气,若有雨推迟一周. 具体采样期间气象要素值见表2. 城市公园的相对

表 1 冬青和女贞植物及叶片性状描述

 Table 1
 Description of holly and ligustrum plants and leaf characters

 品种
 树型
 科属
 叶片大小
 叶片特征

 冬青
 常绿灌木
 寄生科
 中叶
 椭圆形,叶片革质,边缘呈锯齿状,无毛

 女贞
 常绿乔木
 木樨科
 中叶
 卵状椭圆形,叶片革质,边缘光滑,无毛

湿度高于文教区和城市主干道,而 PM_{2.5}、PM₁₀数量及污染物均低于其他地点;不同功能区空气中可吸入颗粒物及一次污染物夏、冬季节高于春、秋季节.春、夏季节主导风向为南风或偏南风,秋、冬季

节主导风向为西北风或北风.

1.3 样品处理及方法

1.3.1 叶片微观结构观察

用酒精擦拭过的镊子采集植物新鲜叶片放入无

表 2	2019 ~ 2020	年取样地点空气质量及污染物状况
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T 11 2	Air quality and			1.	1	c	2010 .	2020
Table 2	Air quality and	i pollutant	status at	sampling	locations	Irom	ZU19 to	2020

地点	日期(年-月-日)	AOI	质量等级	温度/℃	相对湿度/%	PM _{2.5}	PM_{10}	NO_2	SO_2
	2019-04-15	65	良	19	36	45	80	70	13
文教区	2019-07-20	101	轻度污染	30	58	76	90	64	9
入扒匹	2019-10-15	63	良	11	54	24	76	52	8
	2020-01-15	272	重度污染	6	48	224	195	114	47
	2019-04-15	65	良	20	37	50	85	72	18
城市主干道	2019-07-20	103	轻度污染	30	58	85	95	68	15
が山工1垣	2019-10-15	63	良	12	48	26	80	58	10
	2020-01-15	271	重度污染	6	50	236	305	118	49
	2019-04-15	55	良	15	68	12	60	17	3
城市公园	2019-07-20	100	轻度污染	31	68	75	83	14	3
がロス国	2019-10-15	55	良	13	56	10	61	15	9
	2020-01-15	268	重度污染	7	51	221	290	98	40

菌封口袋中封存,冰盒4℃保存,2h内送至实验室. 在距叶尖1/3近中脉处切取2cm×2cm大小的新鲜样品,叶片上表面朝上,用导电胶将样品直接粘于扫描电镜的样品台上,在低真空下采用RESOLUTION连续扫描模式,扫描电镜取9.6mm工作距离、电子束高压20.00kV,强度12.00,背散射电子BSE分辨率4.0nm,在500、1000和2000倍数对叶片表面进行观察并拍照.

1.3.2 叶际细菌 DNA 提取

5 次重复实验中采集适量叶片放在无菌水中用振荡器振荡 30 min 混合,用移液枪吸取无菌水反复冲洗叶面 10 min,收集含菌泥水,随后离心得到含叶际细菌的菌泥,提取细菌 DNA 的过程严格按照土壤 DNA 提取试剂盒的说明步骤进行. 提取后 DNA 样品纯度和浓度使用 NanoPhoto Meter 测定,纯度 A_{280} 值要求在 $1.8 \sim 2.0$ 之间. 将 DNA 样品置于-20%保存.

1.3.3 叶际细菌高通量测定

把 DNA 样品送至上海美吉生物医药科技有限公司进行 16S rRNA 高通量测序,细菌的通用引物序列为:338F(5'-ACTCCTACGGGAGGCAGCAG-3')和 806R(5'-GGACTACHVGGGTWTCTAAT-3'),扩增16S rDNA 的 V3-V4 区域,测序平台为 IIIumina MiSeq.

1.4 数据分析

将环境扫描电镜拍到的图像使用 Photoshop 软件进行色差对比处理,在 2000 倍图像下对图像中颗粒物进行数量统计. 用单因素分析比较植物叶片之间微形态结构的差异.

对 DNA 测序数据进行优化处理. 对优化序列提取非重复序列,便于降低分析中间过程冗余计算量. 去除没有重复的单序列,按照 97% 相似性对非重复序列(不含单序列)进行 OTUs(operational taxonomic

units)聚类,在聚类过程中去除嵌合体,得到 OTUs 的代表序列,选出与代表序列相似性在 97% 以上的序列.

通过单样本的 α 多样性分析反映微生物群落的丰富度和多样性,包括一系列统计学分析指数估计环境群落的物种丰度和多样性;群落 Bar 图可以直观呈现各样本优势物种的相对丰度(所占比重),通过分析不同样本群落组成可以反映样本间的差异和距离; Venn 图分析时选用相似水平为 97% 的OTUs 或其他分类学水平的样本表; PCoA 运用方差分解,将多组数据的差异反映在二维坐标图上,坐标轴取能够最大反映样品间差异的两个特征值; Bar 图使用 OriginPro 9.1 绘制, Venn 图和 PCoA 图利用 R 语言工具作图.

2 结果与分析

2.1 植物叶表面颗粒物与微生物微形态特征观察

利用环境扫描电镜对青岛市文教区、城市主干道和城市公园 4 个季节冬青和女贞植物叶表面颗粒物进行微形态观察,结果见图 2. 冬青树文教区春、夏季节叶片 TSP 低于秋、冬两季,春季叶片滞尘能力较差,表面纹理清晰,而叶片在秋、冬季节滞留 $D_p>10~\mu m$ 不规则颗粒物数量较多,造成植物叶表面纹理不可见; 另植物叶表面颗粒物之间易形成半透明菌丝体,分裂形成厚恒孢子或形成涡旋结构向外延伸,叶表面分布表面光滑饱满乳白色直径约 5~8 μm 的酵母菌及表面凹陷或分布均匀细小微孔的球状微生物等[图 2 (a)].

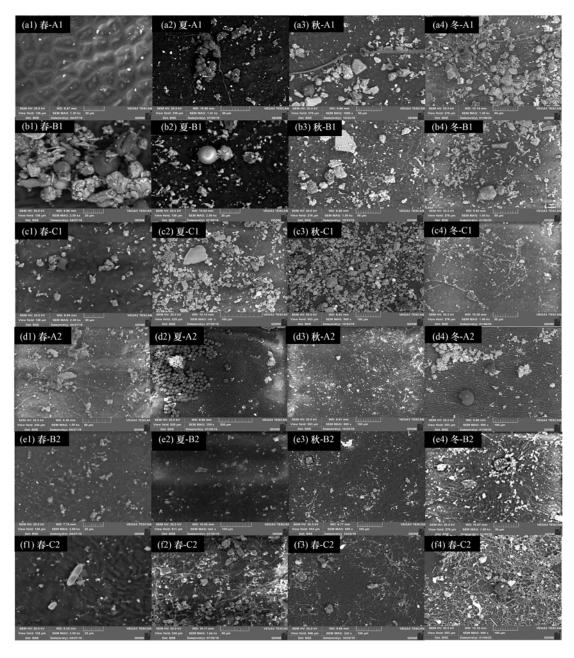
城市主干道冬青树叶片 TSP 总量密度较大,夏、秋和冬季节叶表面纹理不清晰,冬季叶表面颗粒物数量较多,且滞留了少量 $D_p > 50~\mu m$ 颗粒物;春季叶表面颗粒物周围附着基内菌丝,且出现大量具有1道萌发沟的花粉单粒及不同类型真菌,夏季

叶表面分布酵母菌和表面均匀孔隙的真菌,秋季出现霉菌分生孢子和酵母菌,冬季分布粒径约5~10μm 的粒状结晶颗粒物或为无机盐颗粒物[图2(b)].

冬青树中山公园春、夏季节叶表面 TSP 滞留量少于秋、冬两季,秋季叶表面纹理气孔不可见,冬季叶表面 PM_{2.5}和 PM₁₀数量较多;相比于其他功能区,叶际微生物种类数量较多,春、夏季节叶表面分布少量菌丝,夏季还滞留了椭圆形枝孢属分生孢子,秋、冬季节叶际微生物包括酵母菌、表面光滑或均匀凹陷的不同类型真菌及真菌孢子[图 2 (c)].

女贞作为常绿乔木植物,叶表面分布蜡质层.女贞树文教区更易滞留 PM_{10} 和 $PM_{2.5}$,分布均匀,秋、冬季节叶表面 TSP滞留量高于春、夏季节,冬季 D_p >10 μ m 颗粒物总量达到最高;对于叶际微生物,春、秋季节植物叶表皮镶嵌了均匀褶皱的真菌孢子,虽冬季滞留有不同类型真菌孢子,但因蜡质层较少,可见清晰纹理[图 2 (d)].

城市主干道女贞植物叶表面 TSP 滞留量较大, 与冬青相比, $D_p > 10 \mu m$ 颗粒物数量较少,秋季叶表面颗粒物总量略高于冬季; 叶际微生物与文教区相似,叶表皮镶嵌了真菌孢子,影响植物叶片纹理,秋



A1 表示文教区冬青, A2 表示文教区女贞, B1 表示城市主干道冬青, B2 表示城市主干道女贞, C1 表示城市公园冬青, C2 表示城市公园女贞, 下同

图 2 冬青和女贞植物叶表面微形态特征及季节变化

Fig. 2 Leaf surface micromorphological characteristics and seasonal changes of holly and ligustrum

季分布直径约 10~15 μm 的真菌的游动孢子,冬季叶片滞留了表面均匀分布小凸刺直径约 18 μm 的球状微生物及一些霉菌的分生孢子,且分布大量菌丝[图 2 (e)].

女贞树城市公园叶片易滞留 PM_{10} 和 $PM_{2.5}$,城市公园夏季颗粒物数量高于秋季,或与城市公园夏季人流量较大有关,冬季叶表面 TSP 及 $D_p > 10$ μm 颗粒物总量高于其他季节,表面纹理不可见;对于叶际微生物而言,叶表面在春季滞留直径约 20 μm 表面半透明杆状的某种藻类,春夏季节滞留不同类型球菌和真菌孢子,叶表面分布大量半透明菌丝,且叶表面同其他功能区相同,发现镶嵌在叶表皮的真菌孢子[图 2 (f)].

2.2 植物叶际细菌群落 α 多样性

对文教区、城市主干道和城市公园不同季节的 冬青和女贞植物叶际细菌进行高通量测序,按照最

小样本序列数进行抽平后,共得到356376条有效序 列. 样品稀释曲线达到平缓,表明本次测序数据量足 够. 通过 α 多样性分析可得到群落中物种的丰富度、 覆盖度和多样性等信息(表3). Shannon 指数越高、 Simpson 指数越低反映群落多样性(community diversity)越高, Chao 和 Ace 指数越高反映群落丰富 度(community richness)越高, Coverage 指数反映群落 覆盖度(community coverage)越高. 本研究中, 3 个功 能区冬青植物叶际细菌在不同季节 Shannon 和 Simpson 指数差异较小: 而 Chao 和 Ace 指数春季显 著高于其他季节,夏、秋和冬季节无显著差异,春季 细菌群落丰富度较高. 对于女贞植物叶际细菌来说, 夏季群落多样性要略低其他季节,但无显著性差异; 同时女贞叶表面微生物 Chao 和 Ace 指数夏季显著低 于其他季节,春、秋和冬季节叶际细菌群落丰富度较 高,且城市主干道与文教区、城市公园差异性显著.

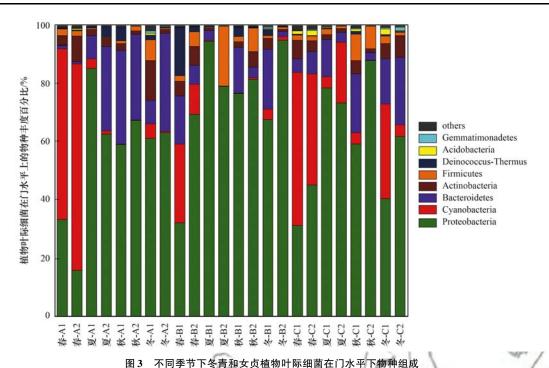
表 3 冬青和女贞植物叶际细菌不同季节内多样性指数

		Tabl	le 3 The o	α diversity	index of ba	cteria durir	ng different	seasons on	the leaves	s of holly ar	d ligustrur	n /	11
植物	指数		ブ	文教区	~ 1	^	城市	r主干道	18	7 1	切	市公园	() 1
但初	1日 安人	春季	夏季	秋季	冬季	春季	夏季	秋季	冬季	春季	夏季	秋季	冬季
	Shannon	2.51	2.79	2.79	2.79	2.25	3.50	3.14	3.28	3.55	2.96	4.78	4.39
1	Simpson	0.25	0.15	0.15	0.15	0.46	0.07	0.09	0.13	0.21	0.15	0.02	0.07
冬青	Ace	1 358.81	497.41	497.41	497.41	1 873.64	1237.24	644.75	873.95	1 906.71	557.28	1 753.62	1 991. 36
-	Chao	963.25	413.04	413.04	413.04	1 274. 41	892.02	481.75	661.39	1 540.29	458.39	1 383.25	1 471.97
	Coverage	0.98	0.99	0.99	0.99	0.97	0.99	0.99	0.99	0.97	0.99	0.97	0.97
1	Shannon	3.49	2.72	3.38	4.12	3.45	1.70	3.57	1.77	4.11	2.83	2.85	4.08
R	Simpson	0.07	0.14	0.10	0.06	0.09	0.32	0.07	0.33	0.09	0.11	0.12	0.06
女贞	Ace	765.23	213.71	1 059.12	1 492.83	976. 59	107.22	976.54	904.70	1 621.02	392.35	573.78	1 318.41
/	Chao	579.78	212.84	773.72	1 138. 23	725.62	90.60	788.09	726.75	1 328.04	329.14	413.79	949.33
	Coverage	0.99	1.00	0.99	0.98	0.99	1.00	0.99	0.99	0.98	1.00	0.99	0.98

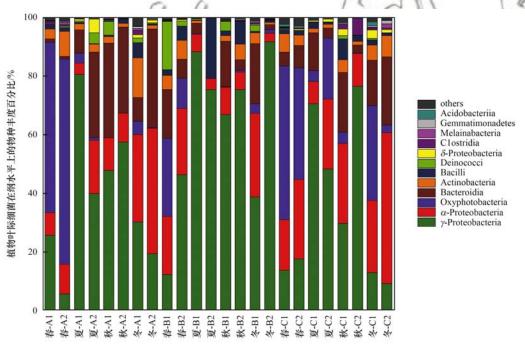
2.3 植物叶际细菌群落物种组成

冬青和女贞植物叶际细菌属于29门、63纲、 174 目、348 科、906 属和1 620种. 根据分类学注释 结果,分别在门和纲水平下进行样品物种丰度的统 计分析,将所有样本丰度占比少于0.01的物种归为 others. 在门水平上(图3), 共检测出8个菌门, 主要 为变形菌门 (Proteobacteria)、蓝藻门 (Cyanobacteria)、拟杆菌门(Bacteroidetes)、放线菌 门(Actinobacteria)、厚壁菌门(Firmicutes)、栖热链 球菌门(Deinococcus-Thermus)、酸杆菌门 (Acidobacteria)和芽单胞菌门(Gemmatimonadetes). Proteobacteria 属于不同季节冬青和女贞植物叶际细 菌的优势菌门,且在城市主干道植物叶际细菌中占 据绝对优势; Cyanobacteria 成为春季文教区和城市 公园的植物优势菌门,冬青和女贞植物叶际细菌分 别占比 58.7%、70.9% 和 52.6%、38.3%, 同时 Cyanobacteria 在冬季城市公园冬青叶际细菌中也成 为优势菌门; Bacteroidetes 在文教区和城市公园的 相对丰度要高于城市主干道植物,尤其在秋冬季节; 另夏季城市主干道植物叶际细菌菌门种类少于其他 季节,但无显著差异.

在纲水平(图 4),不同季节冬青和女贞植物共 检测出来 12 个菌纲,优势菌纲主要包括 γ-变形菌 纲 (γ-Proteobacteria)、 α-变 形 菌 纲 (α-Proteobacteria)、产氧光细菌纲(Oxyphotobacteria)、 拟杆菌纲(Bacteroidia)和放线菌纲(Actinobacteria). 其中 γ-Proteobacteria 在植物叶际细菌中相对丰度较高. 春季文教区和城市公园植物叶表面颗粒物优势菌纲为 Oxyphotobacteria, 冬青 和女贞分别占比58.2%、70.1%和52.4%、38.2%,而冬季植物叶际优势菌纲却为 α-Proteobacteria 和 Bacteroidia,植物叶际细菌优势菌纲会随着时间季节改变.相比于其他功能区,城市主干道植物叶际细菌在不同季节尤其是夏季的菌纲种类较少,γ-Proteobacteria 占绝对优势;另芽孢杆菌纲(Bacilli)仅在夏季女贞植物叶际细菌中占比较大,成为优势菌纲.此外,文教区和







不同季节下冬青和女贞植物叶际细菌在纲水平下物种组成

Fig. 4 Abundance of bacteria at the class level during different seasons on the leaves of holly and ligustrum

城市公园相同季节植物叶际菌纲丰度差异性较小, 但与城市主干道差异性显著.

植物叶际细菌群落 Venn 图分析

Venn 图可用于统计多组中所共有和独有的物 种数目,可以比较直观地展现不同环境样本中物种 组成相似性及重叠情况. 在 OTUs 水平上,对不同 季节冬青和女贞植物叶际细菌进行 Venn 图分析 (图5),发现4个季节植物叶际细菌共7709个 OTUs,叶际细菌 OTUs 总数春季 > 冬季 > 秋季 > 夏

季,夏季数量最低,共895个 OTUs. 其中 4 个季节 植物共有 OTUs 共 590 个,分别占 23.9%、65.9%、 30.4% 和 24.1%, 夏季植物叶际细菌共有 OTUs 占 绝大部分. 每个季节都与其他季节存在共同 OTUs,同时具有特有 OTUs. 可见,每个季节植物既 有与其他季节相同的叶际细菌群落,也存在自己 特有叶际细菌.

2.5 植物叶际细菌 PCoA 分析

PCoA(principal coordinates analysis)分析,即主



图 5 不同季节植物叶际细菌 OTUs 水平下韦恩图

Fig. 5 Venn diagram of OTUs of plant leaf bacteria during different seasons

坐标分析,是一种非约束性的数据降维分析方法,可 用来研究样本群落组成的相似性或差异性. 两样本 点越接近,表明两样本物种组成越相似.对3个功能 区不同季节冬青和女贞植物叶际细菌在 OTUs 水平 上进行 PCoA 分析(图 6). 可见第一主成分贡献率 为 20.8%,第二主成分的贡献率为 15.87%. 不同季 节和不同功能区的两种植物叶际细菌群落组成都存 在差异性,且春季文教区和城市公园植物叶际细菌 组成与其他季节差异明显. 另春、秋两季不同功能 区两种植物之间叶际细菌组成相似度较高,但是春 季植物叶际细菌在第一主成分上差异性明显,而秋 季叶际细菌在第二主成分上差异性较大: 夏、冬季 节同一功能区两种植物之间叶际细菌差异性明显, 且均是第二主成分差异性高于第一主成分. 可见造 成叶际细菌群落差异的第一主成分或为季节因素和 不同功能区,第二成分或为不同类型植物和季节 因素.

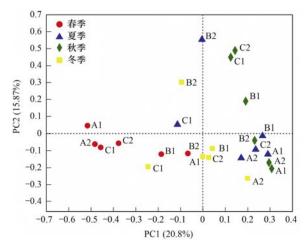


图 6 不同季节植物叶际细菌在 OTUs 水平下的 主坐标分析分析

Fig. 6 PCoA analysis of plant leaf bacteria OTUs during different seasons

3 讨论

3.1 植物叶表面颗粒物滞尘能力对比

不同季节植物叶片 TSP 滞留量存在显著性差 异. 本研究中,冬青和女贞植物叶片 TSP 滞留量秋、 冬两季高于春、夏季节,在冬季达到最高,这不同于 张维康等[30]研究发现的北京路边阔叶植物叶片在 夏季滞尘能力最高. 两个城市空气污染物来源、气 候和温度等都相差较大,空气中颗粒物数量种类也 存在明显差异,致使叶片滞留颗粒物也存在季节差 异. 青岛市春季空气质量良好,污染物浓度较低,空 气中颗粒物相比于其他季节较少,植物叶片滞留 TSP 较少;而秋、冬两季植物叶表面 TSP 总量密度 较大,不仅基于春、夏季节叶表面滞留颗粒物的累 积,还因冬季燃料燃烧导致空气颗粒物污染严重,继 而沉降到植物叶表面. 同时叶表面颗粒物的滞留差 异直接影响叶片微形态结构,与鲁绍伟等[31]的研究 结果相同. 滞留在叶表面的颗粒物能够让叶片微观 结构发生变化[32],春季植物叶片 TSP 滞留量较少, 叶表面纹理清晰,而叶片滞留颗粒物较多的季节,几 乎看不到植物叶表面纹理,同时女贞树叶表面分布 有蜡质层,叶表面粗糙,同样会造成纹理气孔不 可见.

冬青和女贞叶片滞留 $PM_{2.5}$ 和 PM_{10} 能力高于 D_p > 10 μ m 颗粒物,与多种园林植物叶片滞留颗粒物特征相同 $[^{33}^{-35}]$. 另本研究显示女贞植物叶片 TSP和 D_p > 10 μ m 颗粒物滞留量低于冬青植物,高国军等 $[^{36}]$ 的研究也表明了乔木植物叶片单位面积 TSP小于灌木植物,不同类型植物对颗粒物的滞留具有选择作用 $[^{33}]$,女贞作为乔木植物树干较高,叶片受风力因素较大,叶表面 D_p > 10 μ m 颗粒物容易被吹散;而冬青作为小型灌木,叶表面易吸附地面扬尘和汽车排放的污染物,滞尘能力较大且受到人类活动影响明显.

城市主干道植物叶表面 TSP 总量高于文教区和城市公园.城市主干道一次污染物要高于其他功能区,致使植物叶片更易吸附滞留颗粒物,文献[9,37]也证明了该结论.不同环境地点对植物吸附颗粒物的数量有明显影响^[38],文教区、城市主干道和城市公园分别属于3个不同城市功能区,叶表面颗粒物分别来源于不同的生产生活活动.城市主干道商业活动较多,人流车流量较大,近地面空气中颗粒物和污染物多且复杂,植物距离污染源较近,增加了叶表面与颗粒物的接触面积,使得叶片滞尘能力提高,而文教区和城市公园污染较轻,空气中颗粒物数量较少,叶表面的滞尘能力也相对下降.

3.2 植物叶表面微生物微形态特征分析

冬青和女贞植物叶片在低真空下可以观察到不同类型真菌、真菌孢子和菌丝体等,但不同季节叶际微生物组成却差异明显.其中秋、冬季节叶表面更易出现菌丝体和真菌孢子,这与王琳^[39]的研究发现秋、冬季节空气气溶胶中更易发现真菌孢子相似,真菌更适宜低温干燥环境,且叶际微生物与空气中微生物具有较高相似性.

冬青和女贞植物球状微生物较多,酵母菌和孢子是叶表面最活跃的微生物 $[^{40}]$.有研究表明大气颗粒物中球菌约占 $66\%^{[41]}$,且球状微生物比表面积大,受重力或其他机制影响更易被滞留在叶表面.叶表面分生孢子易滞留霉菌分生孢子和枝孢属分生孢子等,且两种植物叶表面易形成菌丝体并分裂形成厚恒孢子,冬青树叶表面菌丝容易缠绕在 $D_p > 10$ μ m 颗粒物上,女贞叶表面蜡质层更有利于菌丝体生长繁殖.另女贞叶表皮镶嵌一种均匀褶皱的真菌孢子,对叶表面纹理造成影响.

对不同功能区叶际微生物分析可发现冬青和女贞植物叶际微生物丰度城市公园>城市主干道>文教区.城市公园因湿度较大,更适应微生物的生长繁殖,与郭徐鹏^[42]的研究结果相同;同时城市公园更靠近海洋,受海洋季风影响较大,相比于其他两个地点植物叶表面更容易发现藻类.可见,叶片滞留颗粒物和微生物的分布规律有一定差异性,微生物属于生命颗粒物,除了受外界因素影响,还会选择适宜环境生长繁殖.

3.3 植物叶际细菌群落多样性和丰富度分析

3个功能区不同季节冬青和女贞植物叶际细菌群落多样性无显著差异. Laforest-Lapointe 等[43]的研究表明,叶际细菌群落组成变化地点和时间因素仅占11%和1%,即使在较长地理距离范围内,相同植物种内细菌群落分化也较小[44],同种植物叶际细菌群落多样性受季节和地点影响较小;另外,相同季节和功能区的冬青和女贞两种植物叶际细菌多样性相似度较高,Vokou等[27]对多种植物进行研究也发现了同地区常绿阔叶植物叶际细菌多样性相似度较高.而对于植物叶际细菌丰度而言,春季叶际细菌群落丰富度显著低于其他季节,而夏季植物叶际细菌群落丰富度显著低于其他季节,与之前研究结果一致[45-47];植物叶际细菌丰富度在空间[48]和时间[49]是不同的,不同植物即使在同一功能区群落丰度也存在较大差距.

冬青和女贞植物叶际细菌的优势菌门都为 Proteobacteria,这与前人研究都进一步证明了 Proteobacteria 在植物叶际细菌中的优势地位^[50~52], 同样也是不同季节植物叶际细菌之间存在共享 OTUs 的特征表现. 而在纲水平上, 冬青和女贞植物 叶际细菌优势菌纲为 γ-Proteobacteria, 与枸杞^[53]叶 际细菌优势菌纲相同,但不同于青杨[42]和大花蕙 兰[20],植物叶际细菌对不同宿主植物及环境有一定 适应性和选择性. 研究还发现同一功能区两种植物 的优势菌门和优势菌纲相似, Shade 等[54]的研究表 明同一地点植物共享一个核心微生物群,受到相同 的气候和其他大环境条件的影响[55],但通过各种媒 介(即空气、雨水和土壤等)的扩散到其他植物叶片 上[56],不同类型植物叶片滞留叶际细菌的能力又存 在差异,导致同一功能区不同种植物虽核心菌群相 同,但菌群丰度却存在明显差异.另外,污染严重的 城市主干道与其他功能区相比植物叶际细菌群落组 成和丰度存在显著差异,不同污染程度的植物叶际 微生物群落存在差异[57,58]. 城市主干道植物周围颗 粒污染物来源复杂,且受到大气扩散、沉积和叶片 表面特征等影响,导致植物叶际细菌群落与污染较 轻的功能区差异性显著. 可见,季节、功能区和植物 类型都会影响植物叶际细菌群落.

4 结论

- (1)冬青和女贞叶片 TSP 滞留能力秋、冬季节高于春、夏两季,在冬季达到最高. 两种植物滞留 PM_{10} 和 $PM_{2.5}$ 能力高于滞留 $D_p > 10$ μ m 颗粒物,另冬青叶表面 TSP 和 $D_p > 10$ μ m 颗粒物数量密度高于女贞植物. 且污染严重的城市主干道植物叶片滞尘量高于文教区和城市公园. 叶表面颗粒物滞留量直接影响叶片微形态结构,颗粒物滞留较多将导致叶片纹理不清晰.
- (2)冬青和女贞植物叶际微生物群落结构存在季节差异.且植物叶片在低真空下可以观察到不同类型真菌、真菌孢子和菌丝体等,球状微生物占比较大,包括酵母菌、表面饱满或有褶皱真菌等;孢子包括霉菌分生孢子、枝孢属分生孢子和厚恒孢子等,且真菌孢子更适宜低温干燥环境.叶表面微生物丰富度城市公园>城市主干道>文教区,城市公园因湿度较大,更适宜微生物的生长繁殖,且更易发现藻类.叶际微生物分布规律与叶片滞留颗粒物存在差异,微生物属于生命颗粒物,不仅受外界条件影响,还会选择适宜环境滞留.
- (3)不同季节冬青和女贞植物叶际细菌群落多样性无显著性差异,但细菌群落丰富度差异性明显,且叶际优势菌存在季节动态变化. 两种植物叶际细菌的 优势 菌门为 Proteobacteria;在 纲水 平, γ -Proteobacteria、 α -Proteobacteria 和 Oxyphotobacteria

在植物叶际细菌中相对丰度较高.同一功能区冬青和女贞植物叶际细菌多样性相似,而对于不同功能区而言,城市主干道叶际细菌群落结构与其他功能区差异显著,不同程度的污染环境对植物叶际细菌的滞留影响明显.

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《环境科学》多项引证指标名列前茅

2020年12月29日,中国科学技术信息研究所在中国科技论文统计结果发布会上公布了2019年度中国科技论文统计结果. 统计结果显示《环境科学》2019年度总被引频次12057,影响因子2.256,多项引证指标位居环境科学技术及资源科学技术类科技期刊前列.

HUANJING KEXUE

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