

目次

河北南部城市臭氧和 VOCs 的污染特征及传输贡献	赵江伟, 聂赛赛, 于玉洁, 王帅, 崔建升, 王玮, 任晓伟, 朱烁 (4775)
基于卫星和地面监测的河西走廊 O ₃ 浓度时空分布及潜在源区分析	李锦超, 曹春, 方锋, 唐千惠, 梁朕月 (4785)
海南岛臭氧污染时空变化及敏感性特征	符传博, 丹利, 佟金鹤, 徐文帅 (4799)
华东地区对流层 O ₃ 和前体物 HCHO 及 NO ₂ 的时空特征	王晓雯, 刘昊霞, 王扬, 宋宜凯 (4809)
2006~2020 年广东省大气甲醛排放演变特征	明桂英, 朱曼妮, 沙青娥, 张雪驰, 饶思杰, 陈诚, 刘慧琳, 郑君瑜 (4819)
淄博市涂装行业 VOCs 排放水平及减排潜力	黄玥润, 杨文, 王秀艳, 王雨燕, 程颖, 王帅 (4832)
西安市城市降尘和土壤尘 PM ₁₀ 和 PM _{2.5} 中碳组分特征	沈利娟, 王红磊, 孙杰娟, 刘诗云, 刘焕武, 赵天良 (4843)
长江流域径流模拟及其对极端降雨的响应	高爽, 凌超普, 汤水荣, 王心亮, 王慧勇, 孟磊, 颜晓元 (4853)
黄河流域内蒙古段水化学同位素特征及水体转化关系	裴森森, 段利民, 苗平, 潘浩, 崔彩琪, 张波, 籍健勋, 罗艳云, 刘廷玺 (4863)
古堆泉域岩溶地下水水化学特征及成因	唐春雷, 申豪勇, 赵春红, 王志恒, 谢浩, 赵一, 梁永平 (4874)
华北平原典型城市(石家庄)地下水重金属污染源解析与健康风险评价	陈慧, 赵鑫宇, 常帅, 宋圆梦, 卢梦淇, 赵波, 陈昊达, 高赛, 王琳静, 崔建升, 张璐璐 (4884)
湖南东江湖表层沉积物重金属的空间分布、形态及生态风险	张同亮, 衣丽霞, 黎常成, 袁首枢, 豆奕轩, 田胜艳, 林尚璇 (4896)
太湖水华前表层水 CDOM 的光谱特征与来源解析	王永强, 卢少勇, 黄蔚, 韩镇阳, 国晓春 (4906)
白洋淀典型水域 COD 的组成及各组分贡献	李琦, 张超, 张文强, 温胜芳, 单保庆, 季恺悦 (4915)
白洋淀典型抗生素的源解析及其特定源风险评估	宋圆梦, 赵波, 卢梦淇, 赵鑫宇, 陈慧, 陈昊达, 高赛, 王琳静, 崔建升, 张璐璐 (4927)
雅鲁藏布江中游河流生态系统健康状态对水环境因子的响应	李晓东, 杨清, 刘惠秋, 巢欣, 杨胜烟, 巴桑 (4941)
鄱阳湖典型河湖交汇区浮游植物现状分析	于新平, 陈宇炜, 刘金福, 王俊颖, 徐光锋, 邹浩月, 陈楠, 夏雨 (4954)
不同黑臭程度下城市河道浮游植物群落结构、多样性和功能群	张琪琪, 曾劼, 尹卓, 冯杰, 刘静, 修宇鑫, 刘国, 许春阳 (4965)
反硝化脱氮对太湖蓝藻水华态势的影响	李昌杰, 许海, 詹旭, 张铮惠, 朱梦圆, 邹伟, 肖曼, 倪子怡, 朱慧 (4977)
密云水库入库河流微生物群落演替对氮素形态转化的影响	辛苑, 张耀方, 李添雨, 叶芝茜, 申佩弘, 魏源送, 高超龙, 舒舒兴, 张俊亚 (4985)
城市尾水氮代谢过程中芽孢杆菌对微藻作用机制	赵志瑞, 马超, 颜嘉晨, 李书缘, 李晴, 胡紫如, 呼庆, 刘硕, 万敬敏 (4996)
高效异养硝化-好氧反硝化菌 <i>Glutamicibacter</i> sp. WS1 低温下对多种氮源的脱氮特性及氮代谢机制	魏淑惠, 罗晓, 吕鹏翼, 马文凯, 苏金卫, 李伟, 崔建升 (5006)
大环内酯类抗生素在饮用水处理过程中的污染特征及其氯化反应机制	岑霞, 程思宇, 石宗民, 谢卓鸿, 张凌菱, 杨滨, 应光国 (5017)
不同堆肥工艺处理的城市污水污泥对滨海湿地土壤中养分释放特征和潜力的影响	贾培寅, 王馨, 花玉婷, 姜志翔 (5025)
淮河流域安徽段水体和沉积物微塑料赋存特征及风险评估	张海强, 高良敏, 葛娟, 赵兴兰, 张振, 慕明, 邱宇辉 (5036)
黄河三角洲湿地表层沉积物中微塑料的分布、来源和风险评估	耿娜, 赵广明, 张大海, 袁黎明, 李先国 (5046)
固原市农田土壤微塑料的分布特征及风险评估	马贵, 丁家富, 周悦, 周炎炎, 廖彩云, 海金涛, 王欢 (5055)
鄱阳湖湿地淹水与非淹水状态下微塑料表面细菌群落分布特征	赵俊凯, 陈旭, 胡婷婷, 廖轶颖, 邹龙, 简敏菲, 刘淑丽 (5063)
聚苯乙烯微塑料对铜绿假单胞菌生物膜形成和结构变化的影响	陶辉, 于多, 杨兰, 陈泽扬, 周灵沁, 罗雲鑫 (5071)
聚苯乙烯微塑料对小白菜生长、生理生化及冠层温度特性的影响	郭冰林, 丰晨晨, 陈悦, 林迪, 李岚涛 (5080)
聚乙烯与磺胺二甲嘧啶复合胁迫对大豆种子萌发及幼苗生长生理特征的影响	赵肖琼, 张恒慧, 赵润枝, 张新梅 (5092)
离子类型、强度和铁氧化物影响下微塑料的迁移行为及模型计算	张然, 于可飞, 黄磊, 陈雅丽, 马杰, 翁莉萍, 李永涛 (5102)
土地利用和气候变化对农牧交错带生态系统服务的影响	徐文彬, 饶良懿 (5114)
有机碳流失对土壤侵蚀的响应及其驱动因素: 基于 Meta 分析	刘小岚, 黄金权, 齐瑜洁, 李威闻, 刘纪根, 陈燕飞, 高绣纺 (5125)
平衡施肥与秸秆覆盖对紫色土坡耕地养分及其化学计量的影响	张高宁, 徐绮雯, 何丙辉, 李天阳, 冯梦蝶, 梁珂 (5135)
稀土-重金属共污染土壤中真菌群落结构特征及主导影响因素	罗颖, 李敬伟, 袁浩, 包智华 (5145)
周年轮作休耕对土壤 AMF 群落和团聚体稳定性的影响	鲁泽让, 夏梓泰, 芦美, 赵吉霞, 李永梅, 王自林, 范茂攀 (5154)
华北平原典型城市土壤微生物群落时空变化规律及其驱动因子	赵鑫宇, 陈慧, 常帅, 宋圆梦, 赵波, 卢梦淇, 崔建升, 张璐璐 (5164)
高量秸秆还田配施芽孢杆菌对沙化土壤细菌群落及肥力的影响	聂扬眉, 步连燕, 陈文峰, 安德荣, 韦革宏, 王红雷 (5176)
基于大田试验的铅镉复合污染土壤中甜糯玉米低积累特性	唐乐斌, 李龙, 宋波, 董心月, 韦美溜 (5186)
不同阻控措施对生菜中镉铅累积及品质的影响	周洪印, 李嘉琦, 包立, 夏运生, 王晟, 吴龙华, 张乃明 (5196)
纳米氧化铜对镉胁迫下小油菜生理生化和重金属累积的影响	王诗琪, 孙约兵, 黄青青, 徐应明, 董如苗, 孟庆尧 (5204)
不同污染区巨菌草生物炭内源污染物分布及其生物毒性	刘莉雅, 崔红标, 刘笑生, 张雪, 董婷婷, 章腾, 周静 (5214)
CuFeO ₂ 改性生物炭对四环素的吸附特性	刘国成, 张新旺, 信帅帅, 王倩文, 阎清华, 周成智, 辛言君 (5222)
基于 Meta 分析的全氟化合物对鱼类生态毒性效应	陆宏, 周锦阳, 杨帆, 王蓓莉, 程治文, 申哲民, 袁涛 (5231)
基于 PMF 模型的县域尺度土壤重金属来源分析及风险评估	郑永立, 温汉辉, 蔡立梅, 罗杰, 汤端阳, 武妙, 李慧, 李鼎 (5242)
基于 PMF-PCA/APCS 与 PERI 的菏泽油田牡丹种植区表层土壤重金属潜在来源识别及生态风险评估	赵庆令, 李清彩, 安茂国, 于林松, 万鑫, 曹付恒, 韩文撑, 陈娟, 王天鸽 (5253)
重庆市煤矸山周边农产品镉健康风险评估及土壤环境基准值推导	马杰, 余泽蕾, 王胜蓝, 邓力, 孙静, 刘萍, 徐敏 (5264)
安徽典型硫铁矿集中开采区土壤重金属污染特征及来源解析	贾晗, 刘军省, 王晓光, 鞠林雪, 何鑫, 周建伟, 张羲 (5275)
湘东北典型河源区土壤重金属分布特征、来源解析及潜在生态风险评估	杨振宇, 廖超林, 邹炎, 谢伍晋, 陈晓威, 张驭飞 (5288)
不同含量外源镉在红壤中的变化特征	周子阳, 庞瑞, 宋波 (5299)
CO ₂ 泡沫混凝土碳封存潜力分析	张源, 他旭鹏, 覃述兵, 郝佑民 (5308)
综合环境社会经济指标的优先污染物筛选方法研究: 以电子废物拆解为例	陈源, 蔡震, 李金惠 (5316)
《环境科学》征订启事(4808) 《环境科学》征稿简则(4831) 信息(4842, 5124, 5153)	

华北平原典型城市土壤微生物群落时空变化规律及其驱动因子

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摘要:微生物群落结构和功能受多种环境因素的共同影响,为阐明典型城市土壤中微生物群落的时空变化规律及其主要影响因素,亟需开展典型城市土壤中微生物群落时空动态变化特征研究。鉴于此,选取华北平原典型城市——石家庄市为研究区,共设置12个采样点,分别于2021年6月(夏季)和9月(秋季)采集表层土壤,并利用16S rRNA高通量测序技术对土壤中微生物群落结构及功能进行研究,探究其时空变化规律;同时运用Pearson相关性分析建立微生物群落与环境因子间的相关性,识别微生物群落时空变化的主要驱动因子。结果表明:①石家庄市表层土壤中主要优势菌门为放线菌(Actinobacteriota)和变形菌(Proteobacteria);就季节变化而言,在门水平上,Actinobacteriota和Proteobacteria相对丰度均呈降低趋势;在属水平上,夏季主要优势菌属为节杆菌属(*Arthrobacter*)和未知分类菌属,秋季主要优势菌属为*Arthrobacter*和*Candidatus_Nitrocosmicus*,且优势菌属呈显著季节差异($P < 0.05$);②就季节变化而言,Simpson、Ace和Chao指数均值呈升高趋势,而OTU均值呈降低趋势;就空间差异而言,Shannon指数和Simpson指数呈显著空间差异($P < 0.01$ 和 $P < 0.05$);③各类功能基因无显著季节和空间差异;其中能源生产和转换类功能基因相对丰度最高,在夏季和秋季的相对丰度分别为24.06%~24.84%和24.63%~25.98%;④微生物群落结构组成、多样性指数和功能基因与喹诺酮类抗生素(QNs)、总磷(TP)和硝氮(NO_3^- -N)呈显著相关,且与QNs显著相关性最强($|r| > 0.900$);这表明在石家庄市表层土壤中抗生素成为影响微生物群落变化的主要驱动因素。因此,为保障城市土壤中微生物群落结构和功能稳定,应进一步加强土壤中抗生素污染的综合管控。

关键词:土壤;微生物群落;多样性指数;功能基因;时空分布;驱动因素

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Spatial-temporal Changes and Driving Factors of Soil Microbial Communities in a Typical City of North China Plain

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Abstract: The structure and function of microbial communities are affected by several environmental factors. To clarify the spatial-temporal changes and main influencing factors of soil microbial communities in a typical pharmaceutical city, it is urgent to study the spatial-temporal changes in microbial communities in soils for typical cities. Shijiazhuang City was selected as the study area, and 12 sampling sites were selected. The topsoil was collected in June (summer) and September (autumn) of 2021. The 16S rRNA high-throughput sequencing technology was used to study the structure and function of microbial communities in the soil and explore their spatial-temporal changes. Concurrently, Pearson correlation analysis was applied to establish the correlation between the microbial community and environmental factors, and identify the main driving factors of temporal and spatial changes in the microbial community. The results showed that: ① Actinobacteriota and Proteobacteria were the main dominant bacteria in the surface soil of Shijiazhuang City; at the phylum level, the relative abundance of Actinobacteria and Proteobacteria decreased from summer to autumn; at the genus level, the dominant genera were *Arthrobacter* and unknown genera in summer and *Arthrobacter* and *Candidatus_Nitrocosmicus* in autumn, which showed significant seasonal differences ($P < 0.05$). ② For seasonal variation, the mean values of the Simpson, Ace, and Chao indices increased, whereas the mean values of OTU decreased; for spatial variation, the Shannon and Simpson indices showed significant spatial difference ($P < 0.01$ and $P < 0.05$). ③ There were no significant spatial-temporal differences in various functional genes; thereinto, the relative abundances of energy production and transformation functional genes were the highest (24.06%-24.84% in summer and 24.63%-25.98% in autumn, respectively). ④ The compositions of microbial community, diversity index, and functional genes were significantly correlated with quinolone antibiotics (QNs), total phosphorus (TP), and nitrate nitrogen (NO_3^- -N), most significantly correlated with QNs ($|r| > 0.900$), which indicated that antibiotics were the main driving factor of soil microbial communities. Therefore, to ensure the stability of microbial community structure and function in urban soil, the comprehensive management and control of antibiotic pollution in soil should be further strengthened.

Key words: soil; microbial community; diversity index; functional genes; temporal and spatial distribution; driving factors

微生物群落(包括细菌、古菌和真菌)作为驱动生物地球化学循环和维持生态系统功能的基础^[1,2],在土壤生态系统中发挥着重要作用。土壤微生物群落易受环境变化的影响,其结构组成、多样性和功能在不同环境条件下存在显著差异^[3~5]。季节变化作为微生物群落变化的重要驱动因素,其可

通过影响植被类型、生物量、水分和温度进而改变

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微生物群落的结构和功能^[6~10]. 其中温度变化可改变土壤中微生物群落的生物活性和结构组成^[11]. 而土壤水分是影响微生物群落多样性和功能时空变化的关键因素^[12]. 当土壤被雨水淹没时,会加剧土壤中微生物群落的生物转化机制^[13]; 洪涝变化会显著改变湿地土壤中微生物群落多样性和组成,进而影响土壤生物地球化学循环^[14]; 降水量的增加会显著增加高寒草地土壤中真菌群落的 α 多样性^[15]. 此外,降雨量的变化可导致表层土壤理化性质(如氮和磷等营养元素含量)发生改变^[16],而土壤理化性质与土壤微生物群落结构密切相关^[17]. 华北平原的雨季集中在 7~8 月^[18],研究雨季前后(6 月和 9 月)土壤微生物群落的空间分布特征,对于了解不同水文时期土壤微生物群落的时空变化规律具有重要意义.

整体而言,微生物群落的结构和功能受多种环境因子的共同影响,且不同季节的主要环境影响因素存在差异^[19],如: pH 和总氮(TN)与中亚热带毛竹林土壤中微生物群落结构显著相关($P < 0.05$)^[20]; TN 和总碳(TC)是驱动尾矿坝面土壤中微生物群落结构动态变化的主要环境因子^[21]; pH、TC 和多酚氧化酶是戴云山夏季土壤微生物群落变化的主要驱动因子,而有效磷、全钾和 TC 是戴云山冬季土壤微生物群落变化的主要驱动因子^[10]. 然而,此前研究多集中于土壤 pH、电导率(EC)、TN、TC、可溶性有机碳含量和有效磷含量等理化因子对土壤微生物群落的影响,而较少关注土壤中各形态氮和土壤粒径的影响. Li 等^[22]研究表明,亚硝氮(NO_2^- -N)是解释湿地土壤中门水平微生物群落结构变化和组成的主要因素之一; 赵河^[23]和王华林^[24]研究发现,微生物群落结构和多样性与氨氮(NH_4^+ -N)、硝氮(NO_3^- -N)和总磷(TP)显著相关,土壤粒径也会影响微生物群落分布. 因此,本研究拟选择 NH_4^+ -N、 NO_3^- -N、 NO_2^- -N、土壤粒径和 TP 作为环境影响因子. 此外,由于微生物群落对环境变化敏感,抗生素等污染物也会影响微生物群落的结构和功能^[25],如:低含量四环素和土霉素能显著降低土壤中细菌和真菌数量^[26]; 恩诺沙星能降低微生物群落的活性^[27]等. 有研究表明,喹诺酮类(QNs)抗生素在京津冀地区土壤中的污染最为严重^[28]. 由于 QNs 吸附能力较强,在土壤中不断蓄积,最终会影响土壤微生物群落的多样性及功能. 因此,为保障土壤生态系统的稳定,亟需进一步探究 QNs 污染土壤中微生物群落结构及功能变化的主要驱动因素.

石家庄市作为华北平原的典型城市,制药企业发达. 其中华北制药集团的抗生素原料药总产量占

全国总产量的 15% 左右. 石家庄市表层土壤中存在抗生素污染,且已证实会对土壤微生物群落的多样性及功能产生影响^[29]. 但目前对石家庄市土壤中微生物群落的时空变化特征及其驱动因子的研究仍未见报道. 因此,本研究分别于 2021 年 6 月和 9 月采集了 12 个代表性表层土壤样品,利用 16S rRNA 高通量测序技术对土壤中微生物群落组成、功能和多样性进行研究,明晰其季节动态变化特征,同时建立微生物群落与多种环境因子(QNs 和土壤理化性质)的相关性,识别长期抗生素污染土壤中微生物群落时空变化的主要环境驱动因子,以期为土壤生态系统的恢复与稳定提供理论支撑.

1 材料与方法

1.1 研究区域概况与样品采集

石家庄市有“华北药都”之称,位于河北省中南部(东经 $113^{\circ}30'$ ~ $115^{\circ}20'$,北纬 $37^{\circ}27'$ ~ $38^{\circ}47'$ 之间),属温带季风气候,季节性变化显著. 按地貌类型和土地利用可分为西部山区、中部山麓平原建设用地和东部平原农业用地. 其中中部建设用地制药企业发达,分布有三大产业园区^[30]. 2021 年《经济日报》称,截至 2020 年底,石家庄市医药工业规模位居全市主导产业前列,创造了占全市工业 36.5% 的利润.

根据石家庄市地形,并结合其空间方位,分别在其东部(S1)、北部(S2)、中部(S3)和南部(S4)各均匀布设 3 个采样点,共 12 个采样点,依次为 S1(木邱、木连城、寺头)、S2(协神、正莫、连家庄)、S3(方台、宋营、固营)和 S4(南清河、北王里、高邑城关),采样点分布见图 1. 其中 S1、S2 和 S4 区域主要为农业用地,而 S3 区域主要为建设用地. 用干净的铁铲采集 0~25 cm 的表层土壤,使用聚乙烯袋进行密封装袋标记,放入恒温箱低温保存,运送回实验室后放入 -20°C 冰箱进行冷冻保存直至分析.

1.2 DNA 提取与高通量测序

将新鲜土壤样品过 40 目筛,取适量过筛后的土壤转移至聚乙烯离心管内,标记后进行密封低温保存,并送至测序公司进行高通量测序. 使用 A. E. Z. N. A. TM 土壤 DNA 试剂盒进行土壤样品 DNA 提取. 采用引物 341F ($5'$ -CCTACGGGNGGCWGCAG- $3'$) 和 806R ($5'$ -GGACTACHVGGGTWTCTAAT- $3'$) 进行 PCR 扩增. 扩增产物经切胶回收后,建立基因文库并进行定量和 DNA 序列质控,然后利用 Illumina MiSeq 平台进行 16S rRNA 高通量测序. 在 95% 相似水平下进行 OTU 聚类分析. 本研究的高通量测序由上海美吉生物有限公司完成.

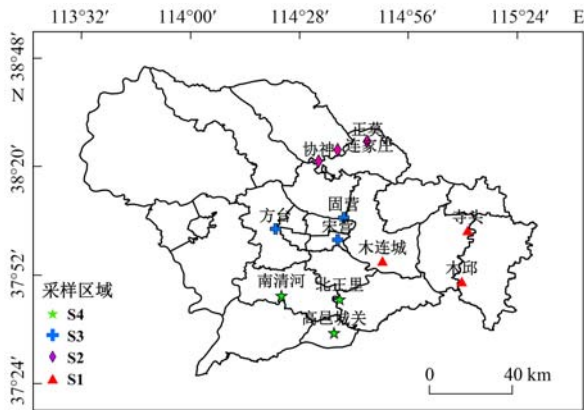


图1 采样点分布示意

Fig. 1 Distribution of sampling sites

1.3 环境因子分析

称取 40 g 土壤样品,风干粉碎后过 40 目筛,混合均匀,使用 LE400-05(USA) 粒径分析仪测定土壤粒径.将剩余土壤样品分为两份,一份用于 TP 测定,另一份用于 $\text{NH}_4^+\text{-N}$ 、 $\text{NO}_2^-\text{-N}$ 和 $\text{NO}_3^-\text{-N}$ 含量的测定.将测定 TP 所需土壤进行自然风干、粉碎和混匀,混匀后的样品分为两份,一份根据 HJ 613-2011 标准测定土壤样品(风干土壤)的干物质含量,另一份过 100 目筛,然后根据 HJ 632-2011 标准测定 TP 含量.将测定氮含量所需土样去除杂物、混匀并过 100 目筛,将过筛后的样品分为两份,其中一份根据 HJ 613-2011 标准测定土壤样品(新鲜土壤)的干物质含量,另一份称取 40 g 土壤样品,根据 HJ 634-2011 标准测定土壤中 $\text{NH}_4^+\text{-N}$ 、 $\text{NO}_2^-\text{-N}$ 和 $\text{NO}_3^-\text{-N}$ 含量.土壤中 QNs 的测定方法参照文献[31].

1.4 数据统计与分析

采用 SPSS 25.0 和 Excel 软件进行土壤理化因子的数据处理与统计,采用单因素方差分析进行差异显著性检验,采用 Pearson 法进行相关性分析.利用 Origin 2018 软件绘制理化因子、 α 多样性指数和 COG 功能基因柱状分布.通过美吉生信云分析平台进行微生物群落物种组成分析、 α 多样性分析、PICRUSt 功能预测、VIF 方差膨胀因子筛选、相关性分析和 Kruskal-Wallis 秩和检验.使用 ArcGIS 软件绘制 QNs 含量空间分布.

2 结果与分析

2.1 微生物群落时空分异特征

就微生物群落物种组成而言,6 月检测出 39 门 1 028 属,共计 2 171 种;9 月共检出 51 门 961 属,共计 1 953 种.从 6~9 月,土壤中主要菌门种类并无显著变化,放线菌(Actinobacteriota)和变形菌(Proteobacteria)为主要优势菌门,相对丰度均呈降

低趋势.在属水平上,5 种相对丰度较高的菌属由某未知分类菌属、节杆菌属(*Arthrobacter*)、鞘氨醇单胞菌属(*Sphingomonas*)、*norank_f_JG30-KF-CM45* 和类诺卡氏菌属(*Nocardioides*)变为节杆菌属、*Candidatus_Nitrocosmicus*、*Nitrososphaeraceae*、芽孢杆菌属(*Bacillus*)和鞘氨醇单胞菌属[图 2(e)和图 2(g)].其中,夏季(6 月)土壤中的主要优势菌属为未知分类菌属和节杆菌属,相对丰度分别为 3.66% (S2)~6.01% (S4)和 2.45% (S1)~7.65% (S3);而秋季(9 月)土壤中主要优势菌属为节杆菌属和 *Candidatus_Nitrocosmicus*,相对丰度分别为 3.71% (S2)~7.41% (S3)和 2.85% (S3)~7.99% (S4).根据 Venn 图和 Kruskal-Wallis 秩和检验,属水平微生物群落组成空间差异显著($P < 0.05$),而门水平微生物群落组成并无显著空间差异.

2.2 多样性指数和 OTU 的时空分异特征

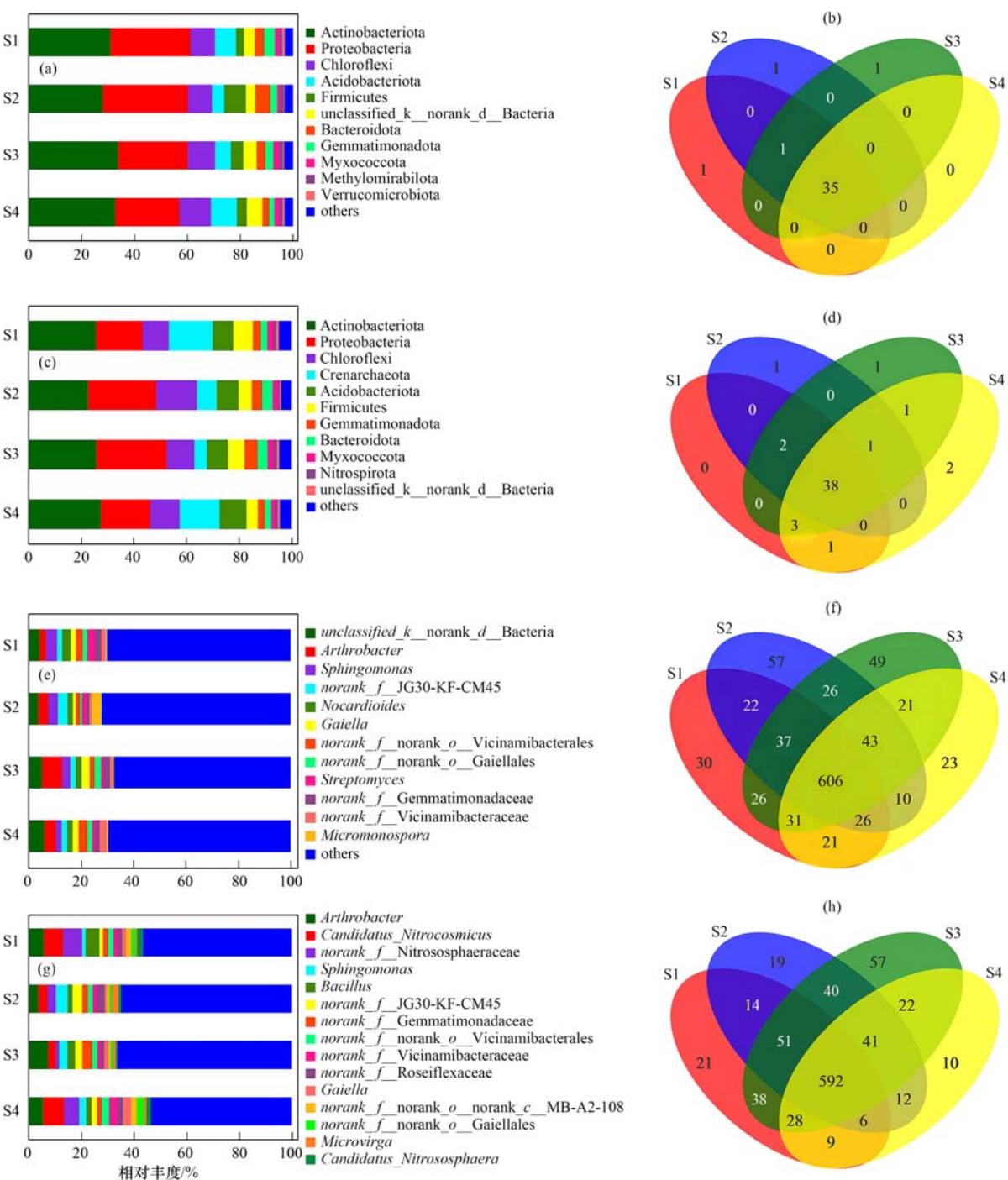
6 月和 9 月的 Coverage 指数平均值均高于 0.98,表明测序结果能够较为真实地反映样本中的微生物情况.OTU 聚类 and α 多样性分析结果表明(图 3),从 6~9 月,Simpson、Ace 和 Chao 指数平均值分别由 0.008、3 227 和 3 236 升高为 0.011、3 331 和 3 332,而 OTU 平均值由 2 720 降低为 2 589,表明土壤中微生物群落多样性呈降低趋势,而丰度呈升高趋势.单因素方差分析结果表明,6 月 Shannon、Simpson、Ace 和 Chao 指数空间差异显著($P < 0.05$),9 月 Shannon 指数和 Simpson 指数空间差异显著($P < 0.01$ 和 $P < 0.05$).

2.3 COG 功能基因时空分异特征

COG 功能基因相对丰度如图 4 所示.从 6~9 月,8 种 COG 功能基因相对丰度并无显著变化.其中能源生产和转换类功能基因相对丰度最高,其次为翻译、核糖体结构和生物发生类、无机离子转运和代谢类以及信号转导机制类功能基因,相对丰度分别由: 24.06%~24.84%、17.14%~17.64%、18.26%~19.53% 和 18.46%~19.72% 变为 24.63%~25.98%、17.80%~20.18%、17.88%~18.45% 和 16.79%~18.84%.且单因素方差分析结果表明,6 月和 9 月各类 COG 功能基因丰度并无显著空间差异.

2.4 土壤理化因子时空分异特征

土壤理化因子分析结果表明(图 5),从 6~9 月,土壤中黏粒的占比平均值显著降低(9.94% 和 4.45%),而砂粒的占比平均值显著升高(8.31% 和 16.82%); $\omega(\text{TP})$ 、 $\omega(\text{NH}_4^+\text{-N})$ 、 $\omega(\text{NO}_3^-\text{-N})$ 和 $\omega(\text{NO}_2^-\text{-N})$ 平均值分别由 836.4、16.79、13.26 和



(a) 和 (c) 分别为 6 月和 9 月土壤中门水平微生物群落组成；(b) 和 (d) 分别为 6 月和 9 月土壤中门水平物种组成 Venn 图；(e) 和 (g) 分别为 6 月和 9 月土壤中属水平微生物群落组成；(f) 和 (h) 分别为 6 月和 9 月土壤中属水平物种组成 Venn 图

图 2 土壤中微生物群落时空分布

Fig. 2 Temporal and spatial distribution of microbial communities in soil

0.6327 mg·kg⁻¹ 升高为 964.5、24.45、78.57 和 1.113 mg·kg⁻¹，其中 NO₃⁻-N 含量平均值显著升高。且单因素方差分析结果表明，各理化因子并无显著空间差异。

2.5 土壤中 QNs 时空分异特征

对土壤中 QNs 分析结果表明 (图 6)，从 6 ~ 9 月，ω (总 QNs) 平均值由 41.53 μg·kg⁻¹ 升高为 92.39 μg·kg⁻¹，整体呈 S3 > S4 > S1 > S2 的空间分

布特征。其中 OFL、ENO、NOR、CIP、ENR、OXO 和 FLU 的检出率均高于 50%。除 ENO 外，ω (OFL)、ω (NOR)、ω (CIP)、ω (ENR)、ω (OXO) 和 ω (FLU) 平均值均呈升高趋势，分别由 1.36、15.86、13.72、5.11、0.06 和 0.35 μg·kg⁻¹ 升高为 3.45、72.49、31.94、6.15、0.38 和 6.48 μg·kg⁻¹。且单因素方差分析结果表明，NOR 含量和总 QNs 含量呈显著空间差异 (P < 0.05)。

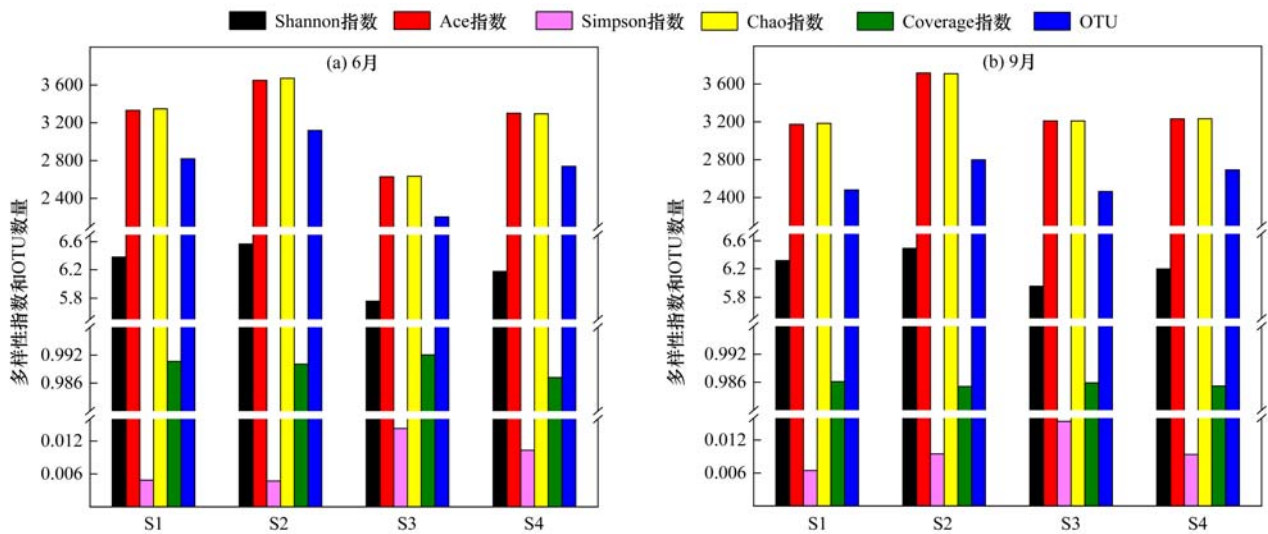


图3 多样性指数和 OTU 数量的时空分布

Fig. 3 Spatial and temporal distribution of diversity index and OTU number

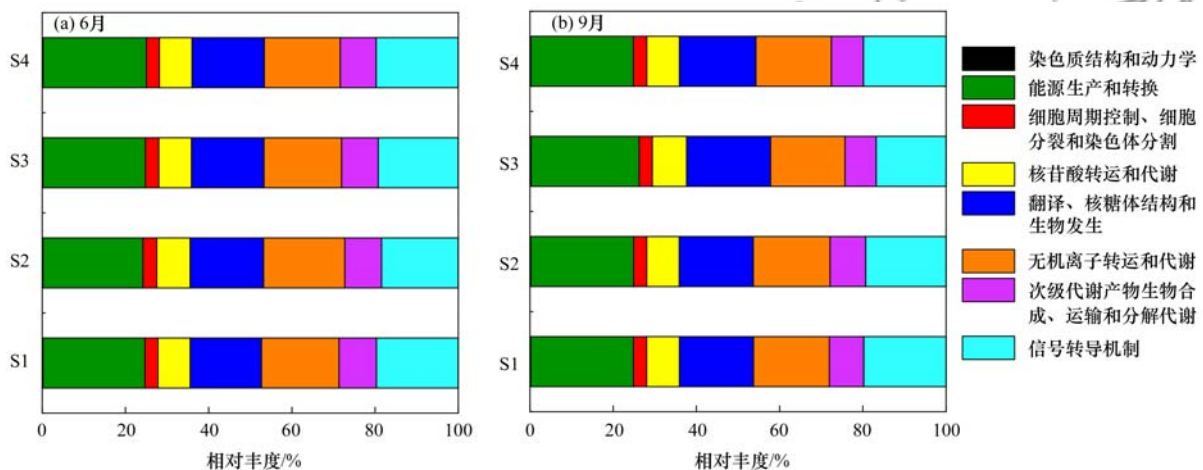


图4 COG 功能基因相对丰度

Fig. 4 Relative abundance of COG functional genes

2.6 微生物群落与环境因子的相关性

将6月和9月各QNs和理化因子分别进行VIF方差膨胀系数筛选,以去除共线性较高的环境因子,筛选后的环境因子与多样性指数和COG功能基因进行Pearson相关性分析(图7).结果表明,QNs与多样性指数和功能基因存在广泛显著相关性[图7(a)和图7(b)],而理化因子与多样性指数和功能基因相关性并不显著[图7(c)和图7(d)].从6~9月,理化因子与多样性指数和功能基因相关性显著增强.夏季(6月)微生物群落多样性和COG功能基因的主要驱动因子为NOR、CIP、ENR和总QNs,而秋季(9月)微生物群落多样性和COG功能基因的主要驱动因子为ENO、NOR、CIP、OXO、FLU、总QNs、TP和 NO_2^- -N.

将进行VIF方差膨胀系数筛选后的环境因子与微生物群落进行Pearson相关性分析,结果表明(图8),QNs和土壤理化因子与门水平和属水平微生物

群落存在显著相关性.其中ENR和CIP与*Gaiella*、*Gemmatimonadaceae*、*Myxococcota*和*Nitrospirota*显著正相关($P < 0.05$),NOR和总QNs与*Arthrobacter*显著正相关($P < 0.01$);TP和 NO_3^- -N与*Streptomyces*显著正相关($P < 0.01$),与*Bacillus*显著负相关($P < 0.05$ 和 $P < 0.01$); NO_2^- -N与*Candidatus Nitrocosmicus*显著正相关($P < 0.05$).总体上,TP、 NO_3^- -N、CIP和ENR是驱动夏季(6月)微生物群落组成变化的主要环境因子,CIP、ENR和NOR是驱动秋季(9月)微生物群落组成变化的主要环境因子.

3 讨论

3.1 土壤中微生物群落的时空变化规律

因不同地区土壤的土地利用类型、气候及地形条件存在差异,导致微生物群落的组成和多样性呈现空间差异^[32-35].在本研究中,石家庄市不同区域

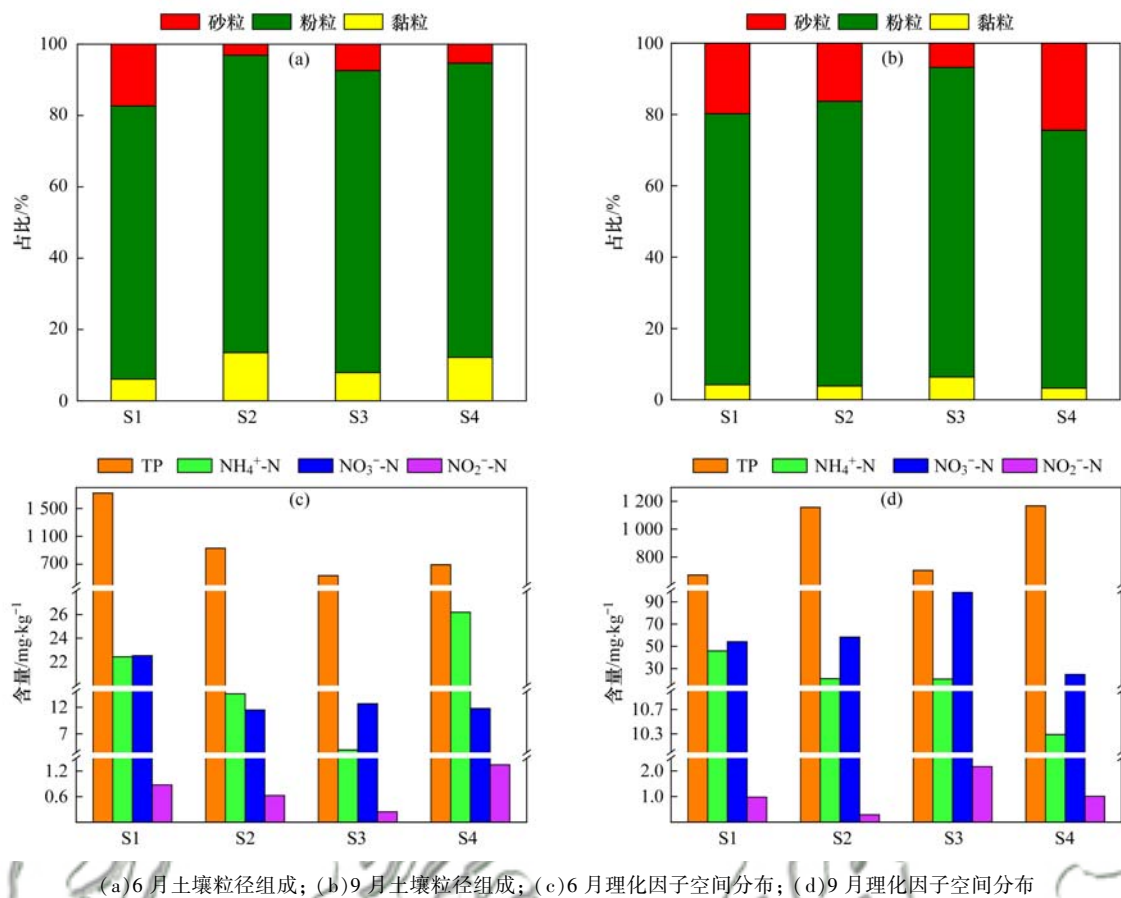


图 5 土壤理化因子时空分布
Fig. 5 Spatial and temporal distribution of soil physical and chemical factors

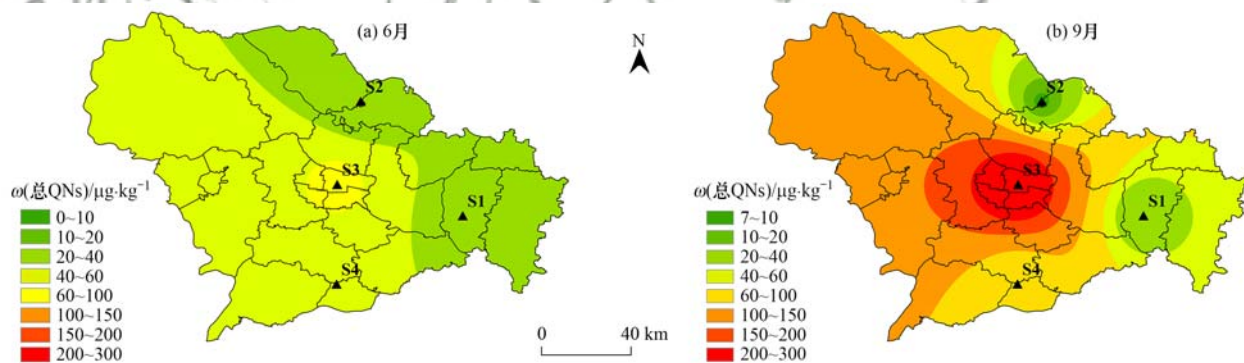
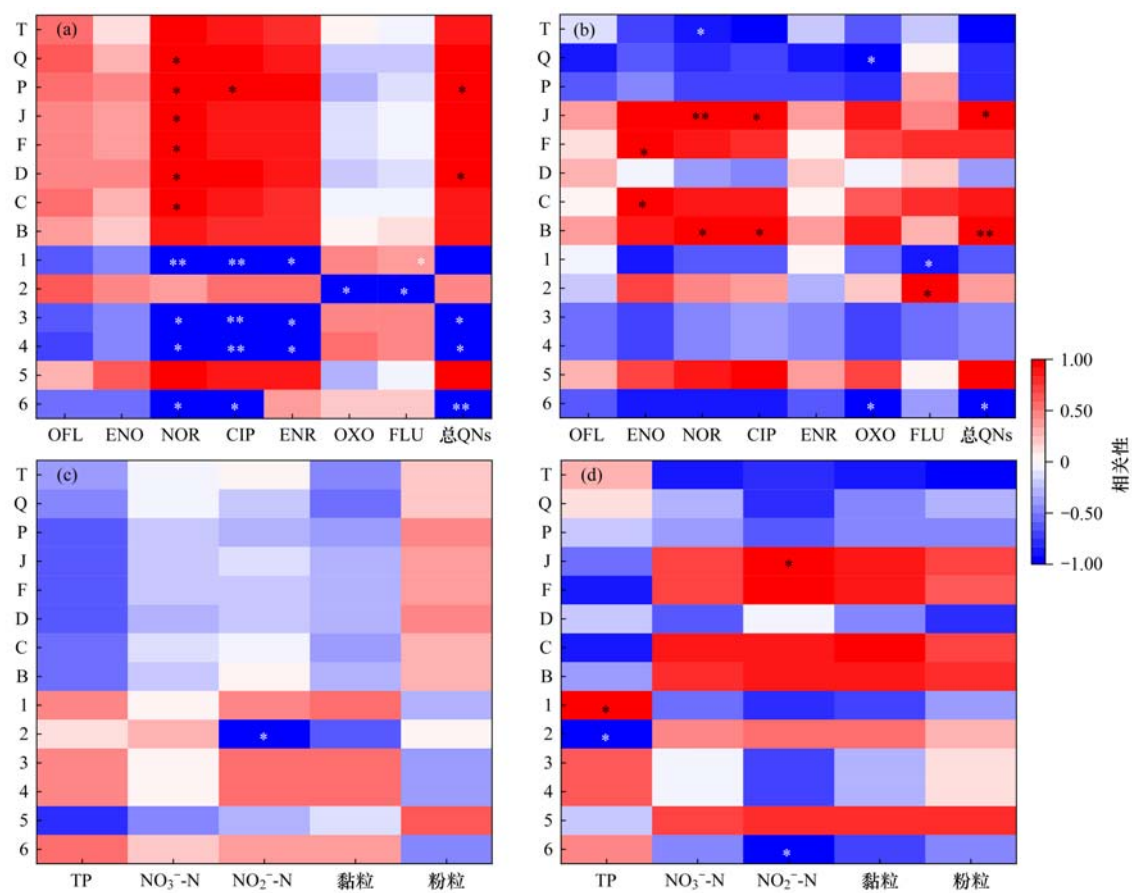


图 6 土壤中总 QNs 含量的时空分布
Fig. 6 Spatial and temporal distribution of total QNs content in soil

优势菌属的空间差异显著,而优势菌门组成并无显著空间差异,这与此前研究的结果一致^[29].本研究土壤中优势菌门主要包括变形菌、放线菌、酸杆菌、绿弯菌和厚壁菌等,与我国(西)北部草原、西部高原草甸、东部城市农田和南部林地土壤中优势菌门组成高度相似(见表1)^[36~41],因此土壤中优势菌门组成受环境条件的影响较小.其中变形菌门为丰度最高的优势菌门,其次为放线菌门,这与高雪峰等^[42]和刘洋等^[43]研究的结果一致.优势种对外界环境条件(如污染物)具有一定适应性,可利用特定的降解酶经多种代谢途径满足自身生长和发展需

求^[44].其中放线菌大量存在于土壤表层,可利用水解酶实现对植物细胞壁的降解^[45];厚壁菌作为石家庄表层土壤中优势菌门之一,包括杆菌和梭菌等,其中杆菌可有效分解土壤中的碳基污染物^[46,47].

就季节变化而言,从6~9月,本研究土壤中的厚壁菌相对丰度和 QNs 含量显著升高,表明厚壁菌的相对丰度变化可能与表层土壤中 QNs 等碳基污染物含量有关.此外,从6~9月,微生物群落的 α 多样性和优势物种丰度整体呈降低趋势,且主要优势菌属组成发生显著变化,这与 Chen 等^[48]研究的结果一致,这可能与汛期前后降雨量和气温的变化有



(a)和(b)分别为6月和9月QNs与多样性指数、OTU和功能基因相关性；(c)和(d)分别为6月和9月理化因子与多样性指数、OTU和功能基因相关性；B、C、D、F、J、P、Q和T分别为染色质结构和动力学类、能源生产和转换类、细胞周期控制、细胞分裂和染色体分割类、核苷酸转运和代谢类、翻译、核糖体结构和生物发生类、无机离子转运和代谢类、次级代谢产物生物合成、运输和分解代谢类及信号转导机制类功能基因；1、2、3、4、5和6分别为OTU、Coverage指数、Chao指数、Ace指数、Simpson指数和Shannon指数；*表示 $P < 0.05$ ，**表示 $P < 0.01$

图7 环境因子与多样性指数、OTU和功能基因相关性

Fig. 7 Correlation between environmental factors and diversity index, OTU, and functional genes

表1 我国不同地区土壤中优势菌门组成

Table 1 Composition of dominant bacteria in soils of different regions in China

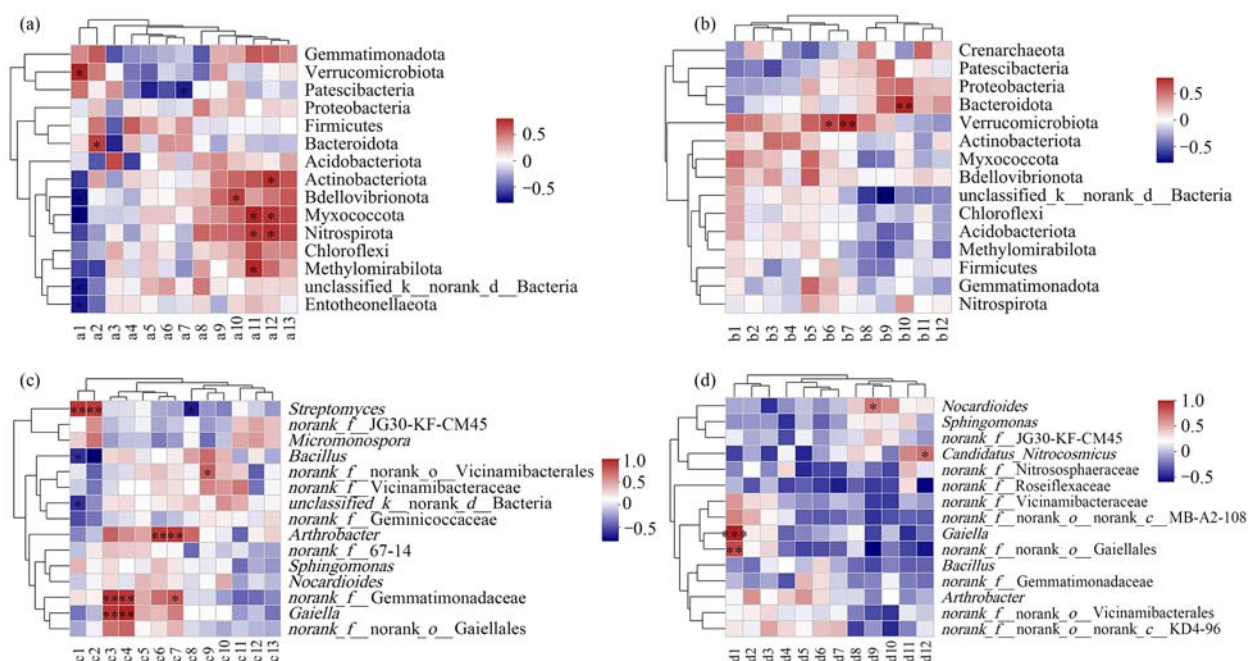
地点	土地利用类型	优势菌门	文献
陕西咸阳	试验农田	变形菌、放线菌、芽单胞菌、酸杆菌和绿弯菌	[36]
云南昆明	林地	变形菌、放线菌、芽单胞菌、酸杆菌和绿弯菌	[37]
内蒙和新疆	草原	变形菌、放线菌、酸杆菌和绿弯菌	[38]
湖北黄石	尾矿池	变形菌、放线菌、酸杆菌、绿弯菌和厚壁菌	[39]
山东青州	农田	变形菌、放线菌、芽单胞菌和酸杆菌	[40]
青藏高原	草甸和草原	变形菌、放线菌、酸杆菌和拟杆菌	[41]
河北石家庄	农田、林地、草地和荒地等	变形菌、放线菌、酸杆菌、绿弯菌和厚壁菌	本研究

关. 温度升高和降水量的增加,有助于提高微生物群落的活性和生产力^[49,50]. 9月汛期结束,降水量和土壤表层温度较6月显著降低,改变了土壤中微生物群落组成和酶活性^[50,51],抑制了微生物群落的生长,导致微生物群落 α 多样性和丰度降低.

3.2 土壤中微生物群落的主要环境驱动因子

就微生物群落组成而言,在夏季,微生物群落组成变化的主要驱动因子为TP、 NO_3^- -N、CIP和ENR;而在秋季,微生物群落组成变化的主要驱动

因子为CIP、ENR和NOR. 其中TP和 NO_3^- -N对微生物群落主要表现为抑制作用,这之前研究的结果不同^[10,52],这可能是由于石家庄市土壤中氮、磷含量处于饱和状态,TP和 NO_3^- -N含量的升高导致土壤pH降低,从而降低微生物的碳利用效率,抑制微生物群落的生长^[53]. Bloem等^[54]和Riaz等^[55]研究也证实,抗生素会影响微生物群落结构. 秋季降雨量和温度降低,导致抗生素等污染物的垂向迁移减弱和吸附能力增强,不断在表层土壤中吸附蓄积,最终导



(a) 6 月环境因子与门水平微生物群落相关性; (b) 9 月环境因子与门水平微生物群落相关性; (c) 6 月环境因子与属水平微生物群落相关性; (d) 9 月环境因子与属水平微生物群落相关性; a1. TP, a2. NO_3^- -N, a3. OXO, a4. 黏粒, a5. 粉粒, a6. NO_2^- -N, a7. ENO, a8. FLU, a9. NOR, a10. OFL, a11. ENR, a12. CIP, a13. QNs; b1. OXO, b2. FLU, b3. ENO, b4. ENR, b5. 粉粒, b6. 黏粒, b7. NOR, b8. NO_2^- -N, b9. NO_3^- -N, b10. CIP, b11. TP, b12. OFL; c1. TP, c2. NO_3^- -N, c3. CIP, c4. ENR, c5. OFL, c6. NOR, c7. QNs, c8. 粉粒, c9. OXO, c10. FLU, c11. NO_2^- -N, c12. 黏粒, c13. ENO; d1. ENR, d2. ENO, d3. OXO, d4. FLU, d5. 粉粒, d6. 黏粒, d7. NOR, d8. OFL, d9. NO_3^- -N, d10. CIP, d11. TP, d12. NO_2^- -N; * 表示 $P < 0.05$, ** 表示 $P < 0.01$, *** 表示 $P < 0.001$; 色柱对应数值为 Pearson 相关性系数, 蓝色对应相关性系数为负值, 表示为负相关, 红色对应相关性系数为正值, 表示为正相关

图 8 土壤环境因子与微生物群落组成相关性

Fig. 8 Correlation between soil physical and chemical factors and microbial community composition

致抗生素含量升高^[56~58]. 本研究土壤中 CIP 和 ENR 对微生物群落主要表现为促进作用, 这表明在特定含量范围内, 随着抗生素含量的升高, 会刺激具有相应抗性(耐受性)的优势种丰度的增加, 如: Li 等^[59]研究发现, 亚硝基螺旋菌群某一支菌属丰度随着磺胺嘧啶含量的升高($0 \sim 20 \text{ mg} \cdot \text{kg}^{-1}$) 逐渐增加. 由夏季进入秋季后, 土壤中 TP 和 NO_3^- -N 含量升高, 但与微生物群落的相关性减弱, 可能是氮、磷含量的升高使得土壤中优势种转变为对养分需求低的微生物类群^[60].

就微生物群落多样性及功能而言, 在夏季, 微生物群落多样性及功能变化的主要驱动因子为 NOR、CIP 和 ENR; 而在秋季, 微生物群落多样性及功能变化的主要驱动因子为 NOR、CIP、ENO、OXO、FLU、TP 和 NO_2^- -N. 其中 QNs 与微生物群落多样性指数呈显著负相关, 表明 QNs 会降低土壤中微生物群落多样性, 这与已有研究的结果一致^[61~63], 如: NOR 处理 28d 后可显著降低微生物群落的 Shannon 指数^[64]; 高浓度的 QNs 残留能显著降低土壤微生物群落多样性^[65]. 此外, 微生物群落的功能也被证实与抗生素等污染物显著相关, 如: OFL 等抗生素能

增加沉积物中微生物群落的反硝化功能基因的丰度^[66], 同时 OFL 可降低微生物群落的固氮能力、光合能力和代谢能力^[67]. 在本研究中, QNs 对微生物群落功能基因的影响呈现季节性动态变化, 可能与功能基因对抗生素等污染胁迫的降解反应及毒性响应差异有关^[68]. NOR 和 CIP 作为石家庄市表层土壤中检出含量最高的 QNs, 在夏季, 其与各类代谢类和信号转导机制类功能基因正相关, 说明其可作为信号分子刺激微生物群落进行繁殖代谢活动; 而在秋季, 与信号转导机制类功能基因负相关, 这可能是由于秋季表层土壤中抗生素含量升高, 抑制了微生物群落信号转导功能, 降低了微生物群落的抗干扰能力^[69,70]. 总而言之, 相较于 TP、 NO_3^- -N、 NO_2^- -N、 NH_4^+ -N 和土壤粒径等理化因子而言, QNs 是驱动石家庄市表层土壤中微生物群落结构及功能变化的主要环境因子.

不同地区土壤中微生物群落的环境驱动因子不同. 在自然环境条件下, 除季节更替导致的温度和水分变化产生的影响外, 土壤微生物群落主要受土壤理化因子的影响. 如: Wang 等^[71]对高原湖岸土壤的研究表明, OTU 水平微生物群落组成的差异主要由

盐度、TOC 和 TN 决定; Li 等^[22]对黄河三角洲湿地土壤的研究表明,TK、Mn⁴⁺、NO₂⁻-N 和 Na 是土壤中微生物群落结构和组成变化的主要驱动因子. 而石家庄市作为典型制药城市,在本研究中表层土壤中的微生物群落主要受 QNs 等抗生素的影响; Zhang 等^[17]研究发现,在重金属污染土壤中,Cr 和 Cd 是驱动微生物群落结构变化的环境因子. 这表明当土壤环境受到污染时,污染物可能成为土壤微生物群落变化的主要驱动因子.

4 结论

(1)就微生物群落组成季节变化而言,土壤中优势菌属种类发生改变;就空间分异而言,优势菌属存在显著空间差异.

(2)就多样性指数和功能基因季节变化而言,微生物群落多样性呈降低趋势,而丰度呈升高趋势;就空间分异而言,多样性指数呈显著空间差异.

(3)就环境因子季节变化而言,表层土壤中黏粒占比呈降低趋势,而砂粒占比、NO₃⁻-N 含量和 QNs 含量呈升高趋势;就空间分异而言,QNs 呈显著空间差异.

(4)就微生物群落结构变化的驱动因子而言,夏季为 TP、NO₃⁻-N、CIP 和 ENR,而秋季为 CIP、ENR 和 NOR;就微生物群落多样性及功能变化的驱动因子而言,夏季为 NOR、CIP 和 ENR,而秋季为 NOR、CIP、ENO、OXO、FLU、TP 和 NO₂⁻-N.

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环境科学

CONTENTS

Pollution Characteristics and Transport contributions of Ambient Ozone and Volatile Organic Compounds in Southern Hebei Cities	ZHAO Jiang-wei, NIE Sai-sai, YU Yu-jie, <i>et al.</i> (4775)
Temporal and Spatial Distributions of O ₃ Concentration and Potential Source Area Analysis of Hexi Corridor Based on Satellite and Ground Monitoring	LI Jin-chao, CAO Chun, FANG Feng, <i>et al.</i> (4785)
Spatial and Temporal Variations in Ozone Pollution and Sensitivity Characteristics in Hainan Island	FU Chuan-bo, DAN Li, TONG Jin-he, <i>et al.</i> (4799)
Temporal and Spatial Characteristics of Troposphere O ₃ and Precursors HCHO and NO ₂ in East China	WANG Xiao-wen, LIU Min-xia, WANG Yang, <i>et al.</i> (4809)
Evolution Characteristics of Atmospheric Formaldehyde Emissions in Guangdong Province from 2006 to 2020	MING Gui-ying, ZHU Man-ni, SHA Qing-e, <i>et al.</i> (4819)
VOCs Emission Level and Emission Reduction Potential of Coating Industry in Zibo	HUANG Yue-run, YANG Wen, WANG Xiu-yan, <i>et al.</i> (4832)
Pollution Characteristics of Carbonaceous Components in PM ₁₀ and PM _{2.5} of Road Dust Fall and Soil Dust in Xi'an	SHEN Li-juan, WANG Hong-lei, SUN Jie-juan, <i>et al.</i> (4843)
Runoff Simulation and Its Response to Extreme Precipitation in the Yangtze River Basin	GAO Shuang, TI Chao-pu, TANG Shui-rong, <i>et al.</i> (4853)
Water Chemical Isotope Characteristics and Water Transformation Relationship in Mongolian Section of the Yellow River Basin	PEI Sen-sen, DUAN Li-min, MIAO Ping, <i>et al.</i> (4863)
Hydrochemical Characteristics and Formation Causes of Ground Karst Water Systems in Gudui Spring Catchment	TANG Chun-lei, SHEN Hao-yong, ZHAO Chun-hong, <i>et al.</i> (4874)
Source Analysis and Health Risk Assessment of Heavy Metals in the Groundwater of Shijiazhuang, a Typical City in North China Plain	CHEN Hui, ZHAO Xin-yu, CHANG Shuai, <i>et al.</i> (4884)
Spatial Distribution, Speciation, and Ecological Risk Assessment of Heavy Metals in Surface Sediments of Dongjiang Lake, Hunan Province	ZHANG Tong-liang, YI Li-xia, LI Chang-cheng, <i>et al.</i> (4896)
Spectral Characteristics and Source Analysis of Chromophoric Dissolved Organic Matter in Surface Water of Taihu Lake Before Cyanobacterial Blooming	WANG Yong-qiang, LU Shao-yong, HUANG Wei, <i>et al.</i> (4906)
Chemical Oxygen Demand(COD) Composition and Contribution in Typical Waters of Baiyangdian Lake	LI Qi, ZHANG Chao, ZHANG Wen-qiang, <i>et al.</i> (4915)
Source Apportionment and Source-specific Risk of Typical Antibiotics in Baiyangdian Lake	SONG Yuan-meng, ZHAO Bo, LU Meng-qi, <i>et al.</i> (4927)
Response of River Ecosystem Health Status to Water Environmental Factors in the Middle Reaches of Yarlung Zangbo River	LI Xiao-dong, YANG Qing, LIU Hui-qi, <i>et al.</i> (4941)
Analysis on the Current Situation of Phytoplankton in the Typical River- Lake Ecotone of Lake Poyang	YU Xin-ping, CHEN Yu-wei, LIU Jin-fu, <i>et al.</i> (4954)
Phytoplankton Community Structure, Diversity, and Functional Groups in Urban River Under Different Black and Odorous Levels	ZHANG Qi-qi, ZENG Jie, YIN Zhuo, <i>et al.</i> (4965)
Influence of Denitrification on Cyanobacterial Blooms Trends in Lake Taihu, China	LI Chang-jie, XU Hai, ZHAN Xu, <i>et al.</i> (4977)
Effects of Nitrogen Speciation Transformation on Microbial Community Succession in Input Rivers of Miyun Reservoir	XIN Yuan, ZHANG Yao-fang, LI Tian-yu, <i>et al.</i> (4985)
Action Mechanism of <i>Bacillus</i> on Microalgae During Nitrogen Metabolism in Urban Tailwater	ZHAO Zhi-ni, MA Chao, YAN Jia-chen, <i>et al.</i> (4996)
Nitrogen Removal Characteristics and Metabolism Mechanism of High-Efficiency Cold-Tolerant Heterotrophic Nitrification-Aerobic Denitrification Bacterium <i>Glutamicibacter</i> sp. WSI for Various Nitrogen Sources at Low Temperature	WEI Bo-hui, LUO Xiao, LÜ Peng-yi, <i>et al.</i> (5006)
Pollution Characteristics of Macrolide Antibiotics During Drinking Water Treatment and Their Chlorination Reaction Mechanism	CEN Xia, CHENG Si-yu, SHI Zong-min, <i>et al.</i> (5017)
Effects of Different Biochar and Effective Microorganism Agent Improvement Approaches on the Nutrient Release Characteristics and Potential of Compost	JIA Pei-yin, WANG Xin, HUA Yu-ting, <i>et al.</i> (5025)
Occurrence Characteristics and Risk Assessment of Microplastics in Water and Sediments of Anhui Section of Huaihe River Basin	ZHANG Hai-qiang, GAO Liang-min, GE Juan, <i>et al.</i> (5036)
Distribution, Sources, and Risk Assessment of Microplastics in Surface Sediments of Yellow River Delta Wetland	GENG Na, ZHAO Guang-ming, ZHANG Da-hai, <i>et al.</i> (5046)
Distribution Characteristics and Risk Assessment of Microplastics in Farmland Soil in Guyuan	MA Gui, DING Jia-fu, ZHOU Yue, <i>et al.</i> (5055)
Distribution Characteristics of Microplastic Surface Bacterial Communities Under Flooded and Non-flooded Conditions in Nanjishan Wetland of Poyang Lake	ZHAO Jun-kai, CHEN Xu, HU Ting-ting, <i>et al.</i> (5063)
Influence of Polystyrene Microplastics on the Formation and Structural Change of <i>Pseudomonas aeruginosa</i> Biofilm	TAO Hui, YU Duo, YANG Lan, <i>et al.</i> (5071)
Effects of Polystyrene Microplastics on Growth, Physiology, Biochemistry, and Canopy Temperature Characteristics of Chinese Cabbage Pakchoi (<i>Brassica chinensis</i> L.)	GUO Bing-lin, FENG Chen-chen, CHEN Yue, <i>et al.</i> (5080)
Effects of Combined Stress of Polyethylene and Sulfamethazine on Seed Germination, Seedling Growth, and Physiological Characteristics of Soybean	ZHAO Xiao-qiong, ZHANG Heng-hui, ZHAO Run-zhu, <i>et al.</i> (5092)
Transport and Model Calculation of Microplastics Under the Influence of Ionic Type, Strength, and Iron Oxide	ZHANG Ran, YU Ke-fei, HUANG Lei, <i>et al.</i> (5102)
Impacts of Land Use and Climate Change on Ecosystem Services in Agro-pastoral Ecotone	XU Wen-bin, RAO Liang-yi (5114)
Response of Organic Carbon Loss to Soil Erosion and Its Drivers: A Meta-analysis	LIU Xiao-lan, HUANG Jin-quan, QI Yu-jie, <i>et al.</i> (5125)
Effects of Balanced Fertilization and Straw Mulching on Soil Nutrients and Stoichiometry in Purple Soil Slope	ZHANG Gao-ming, XU Qi-wen, HE Bing-hui, <i>et al.</i> (5135)
Characteristics and Dominant Influencing Factors of the Fungal Community Structure in Soils Co-contaminated with Rare Earth Elements and Heavy Metals	LUO Ying, LI Jing-wei, YUAN Hao, <i>et al.</i> (5145)
Effects of Annual Crop Rotation and Fallow on Soil AMF Community and Aggregate Stability	LU Ze-rang, XIA Zi-tai, LU Mei, <i>et al.</i> (5154)
Spatial-temporal Changes and Driving Factors of Soil Microbial Communities in a Typical City of North China Plain	ZHAO Xin-yu, CHEN Hui, CHANG Shuai, <i>et al.</i> (5164)
Effect of High-volume Straw Returning and Applying <i>Bacillus</i> on Bacterial Community and Fertility of Desertification Soil	NIE Yang-mei, BU Lian-yan, CHEN Wen-feng, <i>et al.</i> (5176)
Low Accumulation Characteristics of Sweet-waxy Maize in Pb and Cd Complex Contaminated Soils Based on Field Trials	TANG Le-bin, LI Long, SONG Bo, <i>et al.</i> (5186)
Effects of Different Control Measures on Cadmium and Lead Accumulation and Quality in Lettuce	ZHOU Hong-yin, LI Jia-qi, BAO Li, <i>et al.</i> (5196)
Effects of Nano-copper Oxide on Physiobiochemical Properties of <i>Brassica chinensis</i> L. and Its Heavy Metal Accumulation Under Cadmium Stress	WANG Shi-qi, SUN Yue-bing, HUANG Qing-qing, <i>et al.</i> (5204)
Distribution and Biototoxicity of Endogenous Pollutants in <i>Pennisetum</i> sp. Biochar from Different Polluted Areas	LIU Li-ya, CUI Hong-biao, LIU Xiao-sheng, <i>et al.</i> (5214)
Adsorption Characteristics of Tetracycline by CuFeO ₂ -modified Biochar	LIU Guo-cheng, ZHANG Xin-wang, XIN Shuai-shuai, <i>et al.</i> (5222)
Ecological Toxic Effect of Perfluorinated Compounds on Fish Based on Meta-analysis	LU Hong, ZHOU Jin-yang, YANG Fan, <i>et al.</i> (5231)
Source Analysis and Risk Assessment of Heavy Metals in Soil of County Scale Based on PMF Model	ZHENG Yong-li, WEN Han-hui, CAI Li-mei, <i>et al.</i> (5242)
Potential Source Identification and Ecological Risk Assessment of Heavy Metals in Surface Soil of Heze Oil Peony Planting Area Based on PMF-PCA/APCS and PERI	ZHAO Qing-ling, LI Qing-cai, AN Mao-guo, <i>et al.</i> (5253)
Health Risk Assessment and Environmental Benchmark of Cadmium in Farmland Soils around the Gangue Heap of Coal Mine, Chongqing	MA Jie, SHE Ze-lei, WANG Sheng-lan, <i>et al.</i> (5264)
Pollution Characteristics and Sources of Heavy Metals in Soil of a Typical Pyrite Concentrated Mining Area in Anhui Province	JIA Han, LIU Jun-xing, WANG Xiao-guang, <i>et al.</i> (5275)
Distribution Characteristics, Source Analysis and Potential Ecological Risk Assessment of Soil Heavy Metals in Typical River Source Areas of Northeastern Hunan Province	YANG Zhen-yu, LIAO Chao-lin, ZOU Yan, <i>et al.</i> (5288)
Variation Characteristics of Exogenous Cadmium with Different Contents in Red Soil	ZHOU Zi-yang, PANG Rui, SONG Bo (5299)
Analysis of Carbon Storage Potential of CO ₂ Foamed Concrete	ZHANG Yuan, TA Xu-peng, QIN Shu-bing, <i>et al.</i> (5308)
Research on the Screening Method of Priority Pollutants with Integrated Environmental Socio-economic Indicators: Example of E-waste Dismantling	CHEN Yuan, CAI Zhen, LI Jin-hui (5316)