



专论与综述

污水处理过程中微生物群落多样性及其对环境因子响应的研究进展

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摘要: 污水生物处理系统的性能和稳定性与微生物群落结构和动态密切相关。通过深入了解活性污泥中微生物群落结构及其影响因素,有助于提高污水厂污染物的去除效果。在不同污水活性污泥处理系统中细菌群落主要以变形菌、绿弯菌、放线菌、厚壁菌和拟杆菌为功能菌群;活性污泥中寄居的大多数真菌来自于子囊菌门,还有少量担子菌门;古菌以产甲烷菌为主;而病毒中分布最广的噬菌体和致病性病毒是最主要的关注点。本文通过对相关文献分析及总结,综述了进水组成、不同处理工艺、参数(理化参数和运行参数)、地理位置和气候条件等环境因子对活性污泥中细菌、真菌、古菌以及病毒群落组成的影响,尽可能全面地介绍污水厂微生物群落多样性及其对环境因子的响应。同时,对未来研究方向进行探讨,以期能够为活性污泥中功能微生物的应用及调控提供理论和应用基础。

关键词: 污水生物处理, 微生物群落, 环境因子, 功能微生物

Microbial community diversity and its response to environmental factors during sewage treatment

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Abstract: The performance and stability of the biological wastewater treatment system are closely related to the microbial community structure and dynamics. An in-depth understanding of the microbial community structure in activated sludge and its influencing factors can contribute to improve pollutants removal in the treatment process. In different sewage wastewater treatment systems, the distribution of

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bacterial communities mainly consists of *Proteobacteria*, *Campylobacter*, *Actinomycetes*, *Pachythrmycetes* and *Bacteroides*. Most of the fungi living in activated sludge belong to *Ascomycota* and a small amount of *Basidiomycota*. Meanwhile, the most widely distributed bacteriophages and pathogenic viruses in the virus are the major concerns. By analyzing and summarizing relevant literatures, this paper reviews the environmental factors such as influent composition, different treatment processes, parameters (physicochemical parameters and operating parameters), geographical location and climatic conditions on bacteria, fungi, archaea and virus in activated sludge, and introduces the diversity of microbial communities in sewage plants and their responses to environmental factors as comprehensively as possible. At the same time, the future research direction is discussed in order to provide theoretical and application basis for regulation of functional microorganisms in activated sludge.

Keywords: Sewage biological treatment, Microbial community, Environmental factors, Functional microorganism

微生物是地球上生物多样性最为丰富的群体^[1], 在碳(C)、氮(N)、硫(S)、磷(P)等生物地球化学循环中扮演重要角色。随着宏基因组测序技术的不断发展^[2], 微生物的多样性和分布正被广泛地研究^[3-4]。微生物在污水处理厂(wastewater treatment plants, WWTPs)污水净化中扮演重要角色, 为公众和环境健康带来益处。在众多工业废水和生活污水处理工艺中, 活性污泥是一个高度复杂的微生物群落^[5], 主要是由原核生物、真核生物和噬菌体等形成的一个复杂生态网络^[6], 能够降解各种有机污染物^[7-10]。因此, 通过对污水处理厂微生物群落生态的全面了解, 可以揭示在污水处理厂运行过程中各因素对微生物群落的影响, 对后期污水处理系统运行的优化具有指导作用。

本文将重点分析污水处理厂活性污泥中细菌、真菌、古菌以及病毒的多样性及其影响因素, 为污水处理条件优化以及提高污水厂运行效率提供理论基础。

1 污水处理中微生物群落结构及多样性

采用高通量测序技术研究污水处理厂中的微生物群落多样性和组成, 显著增强了对污水处理厂微生物群落的了解。Zhang 等^[11]采用 Illumina MiSeq 高通量测序技术, 对滁州市城市污水处理系统中 5 个活性污泥样品(预缺氧池、缺氧池、

好氧池、配水井和污泥脱水机站)中微生物群落结构进行了研究, 结果表明在所有活性污泥样品中, 以变形菌(22.92%–34.42%)、绿弯菌(17.63%–31.81%)、放线菌(6.46%–14.64%)、酸杆菌(3.64%–7.45%)、拟杆菌(2.98%–9.53%)和厚壁菌(2.43%–2.97%)为优势菌。Osunmakinde 等^[12]对南非豪登省 3 个污水处理厂的细菌群落结构和病原菌的分布进行了研究, 结果发现变形菌、放线菌和厚壁菌是主要的优势菌, 而玫瑰单胞菌属和肠杆菌属是优势病原菌属。采用升流式厌氧污泥床反应器处理马铃薯淀粉加工废水, 研究污泥颗粒的微生物多样性, 结果发现功能性种群丰度最高的是绿弯菌(28.91%), 其次是广古菌(22.13%)、厚壁菌(16.70%)、变形菌(16.25%)和拟杆菌(7.73%)^[13]。因此, 在不同污、废水活性污泥处理系统中细菌群落分布主要以变形菌、绿弯菌、放线菌、厚壁菌和拟杆菌为功能菌群。

冬季容易引起活性污泥膨胀, 污泥膨胀会对微生物群落造成一定影响。Yang 等^[14]研究了 3 个不同规模污水厂在污泥膨胀与非污泥膨胀过程中的细菌和病毒, 发现污泥膨胀影响了细菌和噬菌体的丰度, 而且细菌和病毒的多样性降低。通过对污水厂细菌群落组成及多样性分析, 以确定引起活性污泥膨胀的细菌种类。Jin 等^[15]采用培养和 16S rRNA 基因克隆文库分析相结合的方法, 对 A²/O 工艺污水处理厂中膨胀污泥的细菌群落进行

了研究, 结果发现 γ -变形菌是该菌群的优势菌, 其次是厚壁菌门、拟杆菌门、 β -变形菌门和 α -变形菌门。Xu 等^[16]在新疆维吾尔自治区北部的不同污水处理厂收集活性污泥样本, 测序结果表明膨胀性污泥中腐螺旋菌属(*Saprospiraceae*)、黄杆菌属(*Flavobacterium*)和四球虫属(*Tetrasphaera*)的相对丰度分别为 12.0%、8.3%和 5.2%, 均高于正常样品。

随着下一代测序(next-generation sequencing, NGS)等先进分子技术的出现, 真菌群落的丰度、生态功能和相互作用等问题越来越受到重视。Assress 等^[17]利用高通量测序技术研究了南非豪登省 3 个不同污水处理厂的进水和出水样品中真菌的生物多样性, 结果表明污水处理厂中真菌群落可分为 6 个门、31 个纲、361 个属; 分类学鉴定结果表明担子菌门(*Basidiomycota*)和子囊菌门(*Ascomycota*)是最主要的两个门, 分别占 48.38%和 38.36%。这之前对污水处理厂活性污泥中真菌群落的研究^[18]相一致。秦文韬等^[19]利用高通量测序技术对我国南北区域城市污水处理系统内的真菌群落进行了研究, 结果发现南北真菌群落结构存在显著差异, 南方主要以粪壳菌纲(*Sordariomycetes*)和球囊菌纲(*Glomeromycetes*)为优势菌纲, 以 *Ophiocordyceps* 和链格孢属(*Alternaria*)为优势菌属, 而北方则主要以 *Tremellomyces* 和酵母纲(*Saccharomycetes*)为优势菌纲, 以丝孢酵母属(*Trichosporon*)和酵母属(*Saccharomyces*)为优势菌属, 其中 *Trichosporon* 和 *Sordariomycetes* 是常见的病原菌。

病毒在污水处理系统中也起着重要作用。对于可以侵染和杀死细菌的病毒, 噬菌体在调控活性污泥中细菌群落的结构和功能方面可能发挥着重要作用^[20]。有研究表明, 活性污泥中病毒丰度变化与细菌总数、氨氧化细菌(ammonia oxidising bacteria, AOB)丰度、群落组成、化学需氧量(chemical oxygen demand, COD)和氨氮排放浓度以及系统功能有显著的相关性, 这表明病

毒在活性污泥体系中可能是控制细菌数量、群落结构和功能稳定性的关键因素之一^[21]。然而关于医院废水系统中抗生素抗性基因(antibiotic resistance genes, ARGs)、病毒及其相关细菌之间的联系知之甚少。Petrovich 等^[22]使用宏基因组学, 对以色列一座医院废水处理系统中的 ARGs、dsDNA 病毒和细菌的组成进行了研究, 结果在该系统中没有发现病毒序列和 ARGs 之间的强相关性, 说明噬菌体可能不是该系统中 ARGs 转移的重要载体。另外有研究表明, 从污水中分离出的 EcoM017 噬菌体可以减少细菌附着并裂解大肠杆菌相关的生物膜细胞, 以降低生物污垢形成的可能性^[23]。

2 污水处理中微生物群落对环境因子的响应

2.1 进水组成

2.1.1 污水性质

由于城市工业废水与生活污水组成成分以及污染程度不同, 使得污水处理系统的运行方式和条件也有所不同。污水处理中微生物群落扮演着重要的角色, 而不同的进水水质会对活性污泥菌群组成和生物多样性产生影响^[24](表 1)。因此, 在不同性质污水处理过程中, 对微生物群落影响的研究很有必要。在污水处理中, 活性污泥微生物群落结构与进水类型有关, 硝化螺旋菌纲(*Nitrospira*)、暖绳菌科(*Caldilineaceae*)和厌氧绳菌纲(*Anaerolineaceae*)与生活污水处理系统高度相关, 而陶厄氏菌属(*Thauera*)是工业污水处理系统中最丰富的芳烃降解菌^[25]。在好氧处理中, 好氧颗粒污泥(aerobic granular sludge, AGS)在结构上由颗粒和絮凝体组成, 颗粒中细菌和古菌较多, 絮凝体中真菌较多, 而工业和生活污水混合的废水特性在保持 AGS 中微生物的丰富度和均匀性方面发挥了重要作用^[26]。纺织工业废水通常含有高浓度的染料、染色添加剂和各种化学品, 具有高盐浓度, 使纺织废水难以处理。在处理纺织

表 1 进水组成对微生物群落的影响

Table 1 Influence of influent composition on microbial community

进水组成 Influent composition	主要结论 Main conclusions	参考文献 References
污水性质 Sewage properties	不同进水水质对活性污泥菌群组成和多样性产生影响 Different influent water quality affects the composition and diversity of activated sludge flora 活性污泥微生物群落结构与进水类型有关, <i>Nitrospira</i> 、 <i>Caldilineaceae</i> 和 <i>Anaerolineaceae</i> 与生活污水处理系统高度相关, 而 <i>Thauera</i> 是工业污水处理系统中最丰富的芳烃降解菌 The microbial community structure of activated sludge is related to the type of influent. <i>Nitrospira</i> , <i>Caldilineaceae</i> and <i>Anaerolineaceae</i> are highly related to domestic sewage treatment systems, and <i>Thauera</i> is the most abundant aromatic hydrocarbon degrading bacteria in industrial sewage treatment systems 工业和生活污水混合的废水特性在保持 AGS 中微生物的丰富度和均匀性方面发挥了重要作用 The wastewater characteristics of industrial and domestic sewage mixture play an important role in maintaining the richness and uniformity of microorganisms in AGS 高盐度、高有机负荷是驱动纺织废水处理厂微生物群落组成的重要潜在变量 High salinity and high organic load are important potential variables that drive the composition of microbial communities in textile wastewater treatment plants AOA 丰度在中盐度时降低, 在高盐度时增加, 而 AOB 丰度则呈现相反的趋势 AOA abundance decreases at medium salinity and increases at high salinity, while AOB abundance presents the opposite trend 废水的可生化性决定了大型污水处理厂的微生物群落聚集机制 The biodegradability of wastewater determines the microbial community aggregation mechanism of large sewage treatment plants 进水来源相似的污水处理厂微生物群落具有聚集的趋势 Microbial communities of sewage treatment plants with similar influent sources have a tendency to aggregate	[24] [25] [26] [27] [28] [29] [30-31]
污水成分 Sewage composition	铜等废水中常见的重金属会改变硝化菌的群落结构, 降低硝化菌的活性 Heavy metals commonly found in copper and other wastewater will change the community structure of nitrifying bacteria and reduce the activity of nitrifying bacteria 在中温厌氧消化稳定的样品中, 铬、铜、镍、铅、锌 5 种元素和六溴环十二烷、三溴二苯醚与细菌群落结构显著相关 In the samples with stable anaerobic digestion at moderate temperature, chromium, copper, nickel, lead, zinc and hexabromocyclododecane and tribromodiphenyl ether were significantly related to the bacterial community structure 铬胁迫抑制了微生物的大部分代谢途径和功能基因群落, 铬浓度的增加改变了 EPS 的组成及功能组分, 而且降低了大多数细菌的丰度 Chromium stress inhibits most of the microbial metabolic pathways and functional gene communities. Increasing chromium concentration changes the composition and functional components of EPS and reduces the abundance of most bacteria 镉浓度变化对微生物群落结构及多样性有显著影响, 且参与氮去除的亚硝化细菌、硝化螺旋菌、嗜酸杆菌的相对丰度随镉浓度的增加而显著降低 Changes in cadmium concentration have a significant impact on the microbial community structure and diversity, and the relative abundance of nitrosating bacteria, nitrospiral bacteria, and acidophilus involved in nitrogen removal decreases significantly with increasing concentration 长期重金属(Cd、As、Pb 和 Zn)污染对微生物群落组成有显著影响 Long-term heavy metal (Cd, As, Pb, and Zn) pollution has a significant effect on microbial community composition 在 A ² /O 工艺中随着 K ⁺ 浓度的增加, 缺氧区 <i>Candidatus-Competibacter</i> 、 <i>Acinetobacter</i> 和 <i>Azoarcus</i> 的丰度下降, 这可能导致反硝化除磷能力下降 With the increase of K ⁺ concentration in the A ² /O process, the abundance of <i>Candidatus-Competibacter</i> , <i>Acinetobacter</i> and <i>Azoarcus</i> in the anoxic zone decreases, which may lead to a decrease in denitrifying phosphorus removal capacity	[32] [33] [34-35] [36] [37] [38]

工业废水中,高盐度、高有机负荷是驱动纺织废水处理厂微生物群落变化的重要因素^[27],活性污泥中厌氧氨氧化古菌(ammonia oxidising archaea, AOA)和厌氧氨氧化细菌(AOB)群落对不同盐度的胁迫反应不同,AOA 丰度在中等盐度时降低、在高盐度时增加,而 AOB 丰度则呈现相反的趋势^[28],由于氨氧化菌分离和纯培养比较困难,所以其在硝化过程中的作用和效率有待研究。另外,有研究表明废水的可生化性决定了大型污水处理厂的微生物群落聚集^[29],进水来源相似的污水处理厂微生物群落具有聚集的趋势^[30-31]。然而,目前还缺少污水处理厂微生物群落聚集机制的相关研究。

2.1.2 进水成分

生活污水与工业废水组成中,最主要的差异在于工业废水中含有大量重金属等非常规污染物,这对污水处理带来一定困难。因此,研究重金属对污水处理微生物群落的影响显得尤为重要。活性污泥系统中微生物群落多样性和组成对废水处理效率至关重要,废水中常见的重金属铜会改变硝化菌的群落结构,降低硝化菌的活性^[32]。Stiborova 等^[33]研究了 6 种废水污泥中细菌群落结构和多样性,结果发现,在中温厌氧消化稳定的样品中,铬、铜、镍、铅、锌 5 种元素和六溴环十二烷、三溴二苯醚与细菌群落结构显著相关,细菌群落结构 85% 的变化可以归因于这些污染物。探索持久性铬处理对污水处理系统微生物群落和功能影响的研究,结果表明,铬胁迫抑制了微生物的大部分代谢途径和功能微生物群落,同时铬浓度的增加改变了胞外聚合物(extracellular polymeric substances, EPS)的组成及功能组分,而且降低了大多数细菌的丰度^[34-35]。另外,镉可能改变活性污泥系统中的微生物群落结构,有研究发现 10 mg/L 的镉对 COD 的去除有不利影响,降低了微生物群落多样性,改变了整个微生物群落结构;参与氮去除的亚硝化细菌、硝化螺旋菌、嗜酸杆菌的相对丰度随镉浓度的增加而显著降低^[36]。总之,长期重金属(Cd、As、Pb 和 Zn)

污染对微生物群落组成有显著影响^[37]。在 A²/O 工艺中随着 K⁺浓度的增加,缺氧区 *Candidatus-Competibacter*、不动杆菌属(*Acinetobacter*)和固氮弧菌属(*Azoarcus*)的丰度下降,这可能导致反硝化除磷能力下降,然而某些厚壁菌属的丰度有所增加,这与微生物耐盐能力的增强相一致^[38]。

2.2 不同处理工艺

采用不同污水处理工艺的微生物群落存在显著差异(表 2)。孔晓等^[39]对农村污水膜生物反应器系统中微生物群落进行了研究,发现在不同处理工艺阶段主要细菌类群及丰度明显不同,调节池的物种丰度最高。Islam 等^[40]对某污水处理厂中好氧活性污泥池、回用污泥和厌氧消化池中 AOA、AOB 及其 *amoA* 基因的丰度进行了测定,结果发现 AOA-*amoA* 基因在硝化池中比在活性污泥池中更活跃。Ouyang 等^[41]采用高通量测序技术研究了 3 种不同制药废水处理系统曝气池中污泥样品的微生物群落结构,结果表明,不同的处理工艺对细菌群落结构有很大的影响。房平等^[42]对比了某污水厂 A²/O 工艺和 A²/O-MBR 工艺产生的剩余污泥在微波预处理-厌氧消化过程中的古菌群落变化,结果两种剩余污泥的古菌群落结构差异较大,A²/O-MBR 污泥中甲烷丝菌属和甲烷八叠球菌属丰度分别比 A²/O 污泥多 3.68% 和 19.73%,污泥中有机组分不同是引起古菌群落结构变化的重要影响因素。Qin 等^[43]研究了 3 个污水处理厂不同工艺(A²/O、DE 氧化沟和卡鲁塞尔氧化沟)活性污泥中细菌和古菌群落结构及多样性,结果发现它们的群落组成存在显著差异,A²/O 中细菌相对丰富,主要为地发菌属(*Geothrix*)、甲烷螺菌属(*Methanospirillum*)、*Allochro-matium*、*Fimbriimonas* 和贪噬菌属(*Variovorax*); DE 氧化沟中主要为 *Candidatus-Accumulibacter*、不动细菌属(*Acinetobacter*)、军团菌属(*Legionella*)、菌胶团(*Zoogloea*)和硝化螺菌属(*Nitrospira*); 而卡鲁塞尔氧化沟中主要为出芽菌属(*Gemmata*)、盐水杆菌(*Salinibacterium*)、*Methylibium*、浮游霉状菌属

表 2 不同处理工艺的微生物群落差异

Table 2 Differences in microbial communities in different treatment processes

污水处理工艺	主要结论	参考文献
Sewage treatment process	Main conclusions	References
膜生物反应器	农村污水膜生物反应系统中, 不同处理工艺阶段主要细菌类群及丰度	[39]
Membrane bioreactor	明显不同, 调节池中的物种丰度最高 In the rural wastewater membrane biological reaction system, the main bacterial groups and abundances in different treatment process stages are obviously different, and the species abundance in the regulating tank is the highest	
好氧活性污泥/回用污泥/厌氧硝化污泥	AOA- <i>amoA</i> 基因在硝化池中比在活性污泥池中更活跃	[40]
Aerobic activated sludge/Reused sludge/Anaerobic nitrification sludge	AOA- <i>amoA</i> gene was more active in the digesters than in the activated sludge tanks	
曝气池 Aeration tank	不同的处理工艺和废水质量对细菌群落结构有很大的影响	[41]
	Different treatment processes and wastewater quality have a great influence on the bacterial community structure	
厌氧-缺氧-好氧/厌氧-缺氧-好氧-膜生物反应器	两种剩余污泥的古菌群落结构差异较大, A ² /O-MBR 污泥中甲烷丝菌属和甲烷八叠球菌属丰度分别比 A ² /O 污泥多 3.68%和 19.73%	[42]
Anaerobic-anoxic-aerobic/Anaerobic-anoxic-aerobic-membrane	The archaeal community structure of the two remaining sludges is quite different, and the abundance of methanothrix and methanosarcina in A ² /O-MBR sludge is 3.68% and 19.73% more than that of A ² /O sludge, respectively	
厌氧-缺氧-好氧/DE 氧化沟/卡鲁塞尔氧化沟	细菌和古菌群落及其活性污泥的功能, 它们的群落组成有很大的差异	[43]
Anaerobic-anoxic-aerobic/DE oxidation ditch/Carrousel oxidation ditch	The function of bacterial and archaeal communities and their activated sludge, their community composition is very different	
序批式活性污泥-磁场	活性污泥中细菌群落结构变化与污水处理效率存在一定相关性, 外加	[44]
Sequencing batch reactor activated sludge-magnetic field	磁场通过改变微生物群落结构影响影响污水处理效果 There is a certain correlation between the change of bacterial community structure in the activated sludge and the efficiency of sewage treatment, and the effect of the external magnetic field on the sewage treatment effect by changing the microbial community structure	
芬顿-序批式活性污泥	芬顿氧化处理后的钻井废水可生化性得到提高, 处理后的钻井废水与	[45]
Fenton-sequencing batch reactor activated sludge	模拟废水的优势菌群存在差异 The biodegradability of the drilling wastewater after Fenton oxidation treatment is improved, and there are differences in the predominant flora of the treated drilling wastewater and simulated wastewater	

(*Planctomyces*)和溶杆菌属(*Lysobacter*); 甲烷菌是古细菌的优势菌, 其相对丰度为 A²/O>DE 氧化沟>卡鲁塞尔氧化沟。由此可见, 不同的污水处理工艺对活性污泥微生物群落结构及多样性有重要影响。

耿淑英等^[44]采用 MiSeq 高通量测序技术解析了磁场条件下活性污泥微生物群落多样性变化, 结果表明, 在中等磁感应强度(B=7×10⁻² T)时, 活性污泥微生物丰度及多样性最高。活性污泥中细菌群落结构变化与污水处理效率存在一定相关性, 外加磁场通过改变微生物群落结构影响污水

处理效果。Zhang 等^[45]采用 Fenton-SBR 工艺对含有聚丙烯酰胺(HPAM)的钻井废水进行处理, 结果表明, 芬顿氧化处理后的钻井废水可生化性得到提高, 处理后的钻井废水与模拟废水的优势菌群存在差异。因此, Fenton-SBR 工艺在处理钻井废水方面具有潜在的应用前景。

污水处理厂中含有高密度和多样性的病毒, Petrovich 等^[46]对两个不同处理工艺的污水处理厂污水和生物量样本中的 dsDNA 病毒和细菌群落组成及多样性进行了研究, 结果发现两个污水处理厂存在相似的病毒序列家族, 但相对丰度不同;

此外,病毒宿主的细菌群落结构与整个细菌群落有显著差异,而对于污水处理厂病毒组功能及其与环境因子间的关系需要进一步的研究。

2.3 参数

已有研究表明,温度、pH、溶氧量、底物初始浓度和氮浓度等参数都会影响微生物的生长繁殖^[47]。

2.3.1 理化参数

理化参数是影响污水处理微生物群落结构的重要环境因子^[48](表3)。反硝化过程是污水处理厂生物脱氮的关键。通过研究不同处理工艺和进水特性的污水处理厂微生物群落变化,结果发现五日生化需氧量(five-day biochemical oxygen demand, BOD₅)是影响微生物群落结构最重要的环境变量^[30]。Zhang 等^[11]采用高通量测序技术对滁州市城市污水处理系统中活性污泥的微生物群落结构进行了研究,冗余分析表明 pH、总磷(total phosphorus, TP)和 COD 是影响细菌群落分布的重要环境因子。Zhang 等^[49]采用 Illumina 测序法测定了 18 个地理分布的 WWTPs 活性污泥真菌群落,冗余分析表明,温度、TP、pH 和氨氮对真菌群落有显著影响。Xu 等^[50]研究了常规污水处理卡鲁塞尔氧化沟系统厌氧区、缺氧区和缺氧生物区微生物群落多样性,结果表明水温、进水氨氮、进水 COD 和出水 COD 等环境变量与微生物群落显著相关($P<0.05$)。Gao 等^[51]对上海市四座城市污水处理厂活性污泥的微生物群落组成及多样性进行了研究,变形菌是最主要的发育类群,其次是拟杆菌门和厚壁菌门,而且进水 COD 和 pH 对微生物群落组成的影响最大。在活性污泥系统中,缺氧、好氧和厌氧条件的变化有利于促进不同微生物的生长。在上流式厌氧曝气生物滤池(up-flow anaerobic filter-biological aerated filter, UAF-BAF)中,反硝化菌的丰度随化学需氧量氮比(COD/N)值的降低而降低^[52]。另外,有研究发现在反硝化 MBBR 中,随着 NO₃⁻-N 和总氮(total nitrogen, TN)浓度增加,填料生物膜和底泥中各脱氮基因拷贝数增大;

nirK、*nirS* 和 Anammox 等基因拷贝数也随 NO₃⁻-N 浓度的增加而增大^[53]。

在上流式厌氧污泥床(upflow anaerobic sludge blanket, UASB)反应器中, pH 胁迫对微生物群落有显著影响,随着 pH 值降低,优势产氢产甲烷菌主要由甲烷杆菌属(*Methanobacterium*)和甲烷螺菌属(*Methanospirillum*)组成^[54]。对于南方和北方不同污水处理厂活性污泥中存在类硝基还原菌的古细菌,研究表明挥发性固体和 pH 是硝基还原菌类古细菌群落结构的最重要影响因子($P<0.05$),而且挥发性固体与硝基还原菌类古细菌多样性呈正相关^[55]。另外,有研究发现,环境中的极端温度和 pH 值会影响噬菌体的活性,而细菌孢子可以作为噬菌体基因组的保护壳,抵御环境因子对噬菌体带来的压力^[56]。

2.3.2 运行参数

曝气池是污水厂处理污水的一个重要工艺,池内提供一定的污泥停留时间,满足好氧微生物所需的氧量以及使污水与活性污泥充分接触的混合条件,使微生物可以最大效益地进行有氧呼吸,从而去除污水中的 COD。有研究发现污泥龄对曝气池微生物群落的多样性、组成和共生模式有显著影响,在污泥龄为 10–20 d 时,曝气池中的微生物群落具有最高的生物多样性、最低的随机过程影响、更稳定的分子生态网络结构、最低的污泥膨胀风险和更高的脱氮潜力^[57]。另外,较长的污泥龄促进了细菌的稳定硝化作用,而较短的污泥龄促进了更高的真核生物多样性,增加了功能稳定性^[58]。随着水力停留时间和固体停留时间延长以及活性污泥中硝酸盐浓度的升高,微丝菌属(*Candidatus Microthrix*)的 16S rRNA 和 rDNA 的拷贝数均增加,而其丰度与污水处理厂有机物和总氮的去除率呈显著正相关^[59]。在曝气池中可以通过控制曝气程度来调节溶解氧(dissolved oxygen, DO),进而影响细菌群落丰度^[60],在高溶解氧浓度下微生物多样性降低^[61]。

表 3 参数对微生物群落的影响

Table 3 Effects of parameters on microbial communities

参数	主要结论	参考文献
Parameters	Main conclusions	References
理化参数	BOD ₅ 是影响微生物群落结构最重要的环境变量	[30]
Physical and chemical parameters	BOD ₅ is the most important environmental variable affecting microbial community structure	[11]
	pH、TP 和 COD 是影响细菌群落分布的重要环境因子	
	pH, TP and COD are important environmental factors that affect the distribution of bacterial communities	[49]
	温度、TP、氨氮和 pH 对真菌群落有显著影响	
	Temperature, TP, ammonia nitrogen and pH have significant effects on the fungal community	[50]
	水温、进水氨氮、进水 COD 和出水 COD 等环境变量与微生物群落显著相关($P<0.05$)	
	Environmental variables such as water temperature, influent ammonia nitrogen, influent COD, and effluent COD are significantly correlated with microbial communities ($P<0.05$)	[51]
	进水 COD 和 pH 对微生物群落组成的影响最大	
	Influent COD and pH have the greatest impact on microbial community composition	[52]
	在上流式厌氧曝气生物滤池(UAF-BAF)中, 反硝化菌、与氮代谢有关的基因和胞外聚合物(EPS)丰度随 COD/N 比值的降低而降低	
	In the upflow anaerobic aerated biological filter (UAF-BAF), the abundance of denitrifying bacteria, genes related to nitrogen metabolism, and extracellular polymer (EPS) decrease as the COD/N ratio decreases	[53]
	在反硝化 MBBR 中, 随着 NO ₃ ⁻ -N 和 TN 浓度增加, 填料生物膜和底泥中各脱氮基因拷贝数增大; <i>nirK</i> 、 <i>nirS</i> 和 <i>Anammox</i> 等基因拷贝数也随 NO ₃ ⁻ -N 浓度的增加而增大	
运行参数	In the denitrifying MBBR, as the NO ₃ ⁻ -N and TN concentrations increase, the copy number of each denitrification gene in the filler biofilm and sediment increases; the copy numbers of genes such as <i>nirK</i> , <i>nirS</i> , and <i>Anammox</i> also increase with the increase of NO ₃ ⁻ -N concentration	[54]
	在上流式厌氧反应器(UASB)中, pH 胁迫对微生物群落有显著影响	
	In the up flow anaerobic sludge blanket (UASB) reactor, pH stress has a significant effect on the microbial community	[55]
	挥发性固体和 pH 是硝基还原菌类古细菌群落结构的最重要影响因子($P<0.05$), 而且挥发性固体与硝基还原菌类古细菌多样性呈正相关	
	Volatile solids and pH are the most important factors affecting the community structure of nitro-reducing bacteria archaea ($P<0.05$), and volatile solids are positively correlated with the diversity of nitro-reducing bacteria archaea	[56]
	环境中的极端温度和 pH 值会影响噬菌体的活性, 而细菌孢子可以作为噬菌体基因组的保护壳, 抵御环境因子对噬菌体带来的压力	
	Extreme temperature and pH in the environment will affect the activity of the bacteriophage, and bacterial spores can be used as a protective shell of the bacteriophage genome to resist the pressure of environmental factors on the bacteriophage	[57]
	在污泥龄为 10–20 d 时, 曝气池中的微生物群落具有最高的生物多样性、最低的随机过程影响、更稳定的分子生态网络结构、最低的丝状污泥膨胀风险和更高的脱氮潜力	
	When the sludge age is 10–20 days, the microbial community in the aeration tank has the highest biodiversity, the lowest random process impact, a more stable molecular ecological network structure, the lowest filamentous sludge swelling risk and higher denitrification potential	[58]
	较长的污泥龄促进了稳定的细菌硝化作用, 而较短的污泥龄促进了更高的真核生物多样性, 增加了功能稳定性	
	Longer sludge age promotes stable bacterial nitrification, while shorter sludge age promotes higher eukaryotic biodiversity and increases functional stability	[59]
	随着水力停留时间和固体停留时间延长以及活性污泥中硝酸盐浓度的升高, <i>Candidatus Microthrix</i> 的 16S rRNA 和 rDNA 的拷贝数均增加, 而 <i>Candidatus Microthrix</i> 的丰度与污水处理厂有机物和总氮的去除率呈显著正相关	
	With the extension of hydraulic retention time and solid retention time and the increase of nitrate concentration in activated sludge, the copy number of 16S rRNA and rDNA of <i>Candidatus Microthrix</i> increased, while the abundance of <i>Candidatus Microthrix</i> and the organic matter and total nitrogen of sewage treatment plant removal rate was significantly positively correlated	

(待续)

(续表 3)

溶解氧影响细菌群落丰度, 在高溶解氧浓度下微生物多样性降低	[60-61]
Dissolved oxygen affects the abundance of bacterial communities, and microbial diversity decreases at high dissolved oxygen concentrations	
DO 和 C/N 比值是影响群落结构变化的两个最主要因素	[18,62]
DO and C/N ratios are the two most important factors that affect the change of community structure	
高 C/N 比废水中细菌和古菌的多样性以及 TN 和 TP 的去除率较高, 其中变形菌、酸杆菌和拟杆菌是最丰富的种类。	[63]
In the wastewater with high C:N ratios, bacterial and archaeal diversities and TN and TP removal efficiencies were generally higher, with <i>Proteobacteria</i> , <i>Acidobacteria</i> , and <i>Bacteroides</i> being the most abundant phyla	
温度和曝气量对厌氧消化过程中肠球菌的丰度有显著影响	[64]
Temperature and aeration have a significant effect on the abundance of enterococci during anaerobic digestion	
硝化池温度会对微生物群落结构产生影响, 古菌群落结构与产生的挥发性脂肪酸浓度密切相关	[65]
The temperature of the nitrification tank will affect the microbial community structure, and the archaeal community structure is closely related to the concentration of volatile fatty acids produced	
温度由 35 °C 降低到 25 °C 时, 厌氧氨氧化细菌和反硝化菌受到青睐, 而厌氧阴囊菌和梭状芽胞菌则受到负面影响	[66]
When the temperature is lowered from 35 °C to 25 °C, anaerobic ammonia-oxidizing bacteria and denitrifying bacteria are favored, while <i>Anaerolineales</i> and <i>Clostridiales</i> are negatively affected	

在污水处理厂中, 真菌群落存在相似的生物多样性, 但群落结构不同, 而 DO 和碳氮比(C/N) 值是影响群落结构变化的两个最主要因素^[18,62]。Gu 等^[63]利用高通量 16S rRNA 基因扩增子测序技术分析了不同 C/N 比下畜禽养殖废水处理反应器中的微生物群落, 结果发现不同的 C/N 比值对畜禽养殖废水中细菌和古菌群落结构、共生网络具有重要影响; 在高 C/N 比废水中, 细菌和古菌的多样性以及总氮和总磷的去除效率较高, 其中变形菌、酸杆菌和拟杆菌是最丰富的菌群。

为探讨城市污泥高温好氧消化预处理强化污泥厌氧消化对肠道病原菌肠球菌的影响, 房平等^[64]研究了 3 组不同曝气量的高温好氧预处理效果, 在高温好氧预处理过程中, 由于反应器温度较高以及曝气量较大, 体系内处于微好氧状态, 肠球菌生长环境较差, 有少量的肠球菌不能适应外部环境而死亡, 在后续的厌氧消化过程的水解阶段, 反应器内产生大量的挥发性有机酸, 对肠球菌有毒害作用, 所以肠球菌的数量在厌氧消化过程中大量减少。另外, Kor-Bicakci 等^[65]研究了微波预处理和消化池温度对微生物群落结构的影响, 结果发现微生物群落结构主要受消化池温度影响, 古菌群落结构与产生的挥发性脂肪酸浓度密切相关, 在低挥发性脂肪酸浓度的中温消化池中, 甲烷八叠球菌属是最丰富的

产甲烷菌, 而在高挥发性脂肪酸浓度的中温消化池中, 氢营养型产甲烷菌占优势。

de Almeida Fernandes 等^[66]评价了温度对厌氧氨氧化序批式反应器脱氮性能和微生物多样性的影响, 结果表明, 当温度从 35 °C 降低到 25 °C 时, 氨氧化细菌和反硝化菌受到青睐, 而厌氧绳菌目(*Anaerolineales*)和梭菌目(*Clostridiales*)则受到负面影响。以上运行参数大多是在实验室反应器中调试运行, 而这些过程的实际应用有待进一步的研究。

2.4 地理位置

不同污水处理厂微生物群落组成存在差异, 但是其存在核心微生物群落(表 4)。Wu 等^[67]通过系统的全球取样, 分析了来自 6 大洲 23 个国家 269 个污水处理厂的约 1 200 个活性污泥样本的 16S rRNA 基因序列, 分析结果表明, 全球活性污泥细菌群落中存在核心细菌群落, 活性污泥细菌群落没有明显的纬度梯度。

真菌是活性污泥污水处理厂各种功能的重要组成部分, 对于真菌群落的地理特征已有报道。有研究表明, 活性污泥真菌群落组成在不同污水处理厂之间存在差异, 氧化沟和厌氧-缺氧-好氧(A²O)系统之间也存在差异^[49]。另外, 不同污水处理厂间真菌群落结构和海拔高度表现出显著的距离衰减关系^[18]。

表 4 不同地理位置污水处理厂的微生物群落差异

地理位置	主要结论	参考文献
Geographic location	Main conclusions	References
地理分布	活性污泥真菌群落组成在不同污水处理厂之间存在差异, 氧化沟和厌氧/缺氧/好氧(A ² /O)系统	[49]
Geographical distribution	之间也存在差异 Activated sludge fungal community composition differs between different sewage treatment plants, as well as between oxidation ditch and anaerobic-anoxic-aerobic (A ² /O) system 全球活性污泥系统中存在核心细菌群落, 活性污泥细菌群落没有明显的纬度梯度	[67]
海拔高度	There is a core bacterial community in the global activated sludge system, and there is no obvious latitude gradient in the activated sludge bacterial community 3 个厌氧反应器中, 微生物群落结构与海拔高度之间没有显著的相关性	[68]
Altitude	In the three anaerobic reactors, there was no significant correlation between microbial community structure and altitude 污水处理厂的古生物群落以甲烷菌为主(84.6%), 高海拔地区的古生物群落丰富度与环境变量的相关性大于低海拔地区 Paleontological communities in sewage treatment plants are dominated by methane bacteria (84.6%), and the correlation between the richness of paleontological communities at high altitudes and environmental variables is greater than at low altitudes	[69]

污水处理厂所在的海拔高度不同,微生物群落可能存在差异。在对赤霉素(gibberellin, GA)废水的处理中, Ouyang 等^[68]采用 16S rRNA 基因测序检测内循环(internal circulation, IC)和 2 个 UASB 反应器内的微生物群落,并研究采样海拔对微生物群落的影响, Mantel 分析表明在 3 个厌氧反应器中,微生物群落结构与海拔高度之间没有显著的相关性。Niu 等^[69]对我国海拔 3 660 m 的 20 个活性污泥污水处理厂的古细菌群落进行了调查,结果表明污水处理厂的古生物群落以甲烷菌为主(84.6%),高海拔地区的古生物群落丰富度与环境变量的相关性大于低海拔地区。综上所述,不同地理位置污水处理厂存在核心微生物群落,核心物种的分离培养对于提高污水处理效率至关重要,而活性污泥中微生物群落复杂度高,因此核心物种的分离培养工作仍然是一大挑战,值得关注。

2.5 气候条件

2.5.1 季节变化

活性污泥微生物群落在污水处理过程中起着关键作用,而微生物群落受气候条件的影响(表 5)。活性污泥的活性受季节性温度变化的影响,但在一定温度范围内,微生物群落结构中优势菌属的演替在一定程度上有利于维持活性污泥

在温度变化下的功能稳定性^[80]。通过 16S rRNA 基因高通量测序, Liu 等^[70]揭示了广州市 8 个污水处理厂活性污泥微生物群落的显著季节变异性,结果表明不同季节的微生物群落有很大的差异($R=0.72$, $P=0.001$),而且在 $P=0.01$ 的水平上,季节内微生物群落相似性显著高于季节间相似性。Zhang 等^[71]从 4 个规模相同的污水处理厂每个生物处理单元采集了季节性样本,结果发现季节变化对活性污泥菌群的影响大于不同污水处理系统的变化,相对而言,季节变化对细菌群落有较强的影响。Wei 等^[72]研究了我国 5 个污水处理厂活性污泥中细菌和真菌分别在夏季、冬季的分布及多样性,与细菌相比,真菌丰度随季节变化较大,而细菌的多样性随季节的变化差异较真菌显著。Liu 等^[73]利用高通量测序技术,研究了不同污染物浓度和温度等季节变化对污水处理厂厌氧/缺氧/好氧(A²/O)系统微生物群落结构的影响,结果表明,季节变化会改变微生物群落结构,但不足以改变功能基因。对于厌氧污水处理系统中微生物群落结构变化,有研究表明,在 UASB 反应器中,细菌和真菌群落结构的变化不受季节性的影响^[74]。水平潜流人工湿地(horizontal subsurface flow, HSSF)中主要利用细菌和古菌处理废水中的

表 5 气候条件对污水处理微生物群落的影响

Table 5 Impact of climatic conditions on microbial communities in wastewater treatment

气候条件 Climatic conditions	主要结论 Main conclusions	参考文献 References
季节变化 Seasonal changes	不同季节的微生物群落有很大的差异, 且在 $P=0.01$ 的水平上, 季节内微生物群落相似性显著高于季节间 [70] 相似性 The microbial community varies greatly in different seasons, and at the level of $P=0.01$, the similarity of the microbial community within the season is significantly higher than that between the season 季节变化对细菌群落有较强的影响 [71] Seasonal changes have a strong influence on bacterial communities 真菌丰度随季节变化较大, 而细菌的多样性随季节的变化差异较真菌显著 [72] The abundance of fungi varies greatly with seasons, and the diversity of bacteria varies with seasons 季节变化会改变微生物群落结构, 但不足以改变功能基因 [73] Seasonal changes will change microbial community structure, but not enough to change functional genes 在上流式厌氧污泥床(UASB)反应器中, 原生菌和真菌群落结构的变化不受季节性的影响 [74] In the up flow anaerobic sludge blanket (UASB) reactor, the changes in the structure of protozoan and fungal communities are not affected by the seasonality 水平潜流人工湿地(HSSF)中主要利用细菌和古菌处理废水中的有机物, 而古菌群落结构不受季节的影响 [75] In horizontal subsurface flow constructed wetlands (HSSF), bacteria and archaea are mainly used to treat organic matter in wastewater, while archaeal community structure is not affected by seasons 污泥中的病毒也受季节变化的影响, 病毒在冬季可以存活很长时间, 尤其是在污泥的深层处 [76] Viruses in sludge are also affected by seasonal changes. Viruses can survive for a long time in winter, especially deep in the sludge 气候温度是影响污水处理厂微生物群落多样性和结构的重要参数 [77-78] Climate temperature is an important parameter that affects the diversity and structure of microbial communities in sewage treatment plants	
降雨变化 Change in rainfall	雨季和旱季两个时间点的微生物多样性基本稳定 The microbial diversity at the two time points of rainy season and dry season is basically stable	[79]

有机物, 研究发现细菌主要由厚壁菌(42%)、变形菌(33%)和拟杆菌(25%)组成, 古菌主要以甲烷八叠球菌目为主, 而且该古菌群落结构不受季节的影响^[75]。然而, 亚硝基还原菌类古菌的丰度存在季节性变化, 秋冬季丰度高于春夏季^[55]。另外, 污泥中的病毒也受季节变化的影响, 病毒在冬季可以存活很长时间, 尤其是在污泥的深层处^[76]。

有研究发现, 温度是影响污水处理厂微生物群落多样性和结构的重要参数^[77]。对芬兰极地北极圈地区污水处理厂微生物群落的结构分析发现, 在生物反应器中, 真菌比细菌和古细菌竞争更激烈, 北极圈温度是影响污水处理厂微生物的主要因素^[78]。

2.5.2 降雨变化

众所周知, 降雨是影响微生物系统发育和功

能结构的因素之一。Bedoya 等^[79]使用 NGS 技术对哥伦比亚的一个污水处理厂中微生物多样性进行了研究, 结果表明雨季和旱季两个时间点的微生物多样性基本稳定; 尽管优势菌种的微生物丰度发生了变化, 但对厌氧消化器的性能没有影响。

2.6 其他因素

Wang 等^[81]通过比较有(A²/O-B)和无(A²/O-C)有益微生物的厌氧-缺氧-好氧过程的微生物生物量和群落组成变化, 结果表明接种有益微生物可增加微生物在去除有机物质、营养物(即动物胶菌属、脱氮单胞菌属、硝化螺旋菌属和亚硝化单胞菌属)和减少剩余污泥(即变形菌属和拟杆菌属)方面的比例。有研究表明, 某些真菌辅助废水处理工艺的失败可能是由于本土真菌而非细菌的竞争造成

的^[82]。然而对于厌氧处理, 有机底物种类是显著影响微生物群落最主要的因素之一^[83]。在主流废水中存在不同和潜在活性的厌氧氨氧化细菌, 而特定挥发性脂肪酸可能影响高 COD 废水中厌氧氨氧化细菌的多样性和丰富度^[84]。N-酰基-L-高丝氨酸内酯(N-acyl-L-homoserine lactones, AHLs)可以调节活性污泥微生物群落的 AOA 和 AOB 群落组成以及氨氧化活性^[85]。污水处理厂微生物群落的多样性与药物生物转化间存在显著的正相关, 脱氢酶、酰胺酶和单加氧酶等代谢基因与药物生物转化密切相关^[86]。除此之外, 微生物滞留期(microbial residence time, MRT)也是影响污水处理中活性污泥微生物群落的一个重要因素^[87]。

3 结论与展望

在污水处理系统中, 活性污泥是一个由多种细菌、真菌、古菌、原生动物和病毒组成的高度复杂的微生物生态系统。活性污泥中最主要的细菌包括变形菌门(*Proteobacteria*)、拟杆菌门(*Bacteroidetes*)、放线菌门(*Actinobacteria*)和后壁菌门(*Firmicutes*), 最主要的古菌是广古菌门(*Euryarchaeota*), 活性污泥中寄居的大多数真菌来自于子囊菌门, 还有少量担子菌门^[20]。含有高度复杂微生物群落的活性污泥系统, 在污水处理厂污染物去除中起至关重要的作用。了解活性污泥中微生物群落的多样性和结构组成是解决基本生态问题和污水处理工程的关键。通过高通量测序等技术进行的微生物生态学研究进展迅速, 极大地促进了研究者对污水厂活性污泥微生物群落组成及多样性的了解, 揭示了活性污泥中微生物群落对环境因子的响应。

未来污水处理厂活性污泥系统中微生物的研究趋势为:

(1) 建立数学模型, 以深入理解微生物群落的功能以及环境因子对其调控的规律。近年来有研究发现活性污泥中微生物群落受环境因子调控, 这对污水处理厂污染物去除效率造成直接影

响。然而, 目前关于环境因子对活性污泥中微生物群落调控规律以及活性污泥中微生物群落的功能尚未知晓。房平等^[88]利用 ASM1 模型在污水生物添加强化脱氮等方面已有研究, 期望未来能开发数学模型, 以便于利用活性污泥中微生物群落信息了解环境因子对活性污泥中微生物群落的调控规律, 从而预测污水厂的性能, 对于提高活性污泥中微生物生态学和环境工程的知识至关重要。

(2) 污水厂活性污泥中核心物种的分离鉴定。尽管污水厂中微生物群落受环境因子影响, 活性污泥微生物群落具有多样性, 但其具有与活性污泥性能密切相关的核心微生物群落。然而, 活性污泥中微生物数量庞大、复杂度高, 所以活性污泥中微生物物种尤其是一些核心物种的分离培养难度大。目前分离培养的微生物比例很小, 还有大量的微生物物种没能成功分离培养, 因此对污水厂活性污泥中核心物种的分离培养仍然是一大难题。Jin 等^[89-91]从污水处理厂活性污泥中分离了 3 株细菌分类学新种, 其中 *Gordonia phthalatica* sp. QH-11^T 具有很强的有机污染物降解能力, 在有机污染物修复方面具有重要的应用潜力, 然而大量的细菌类群仍然尚未分离。近期中国科学院科研人员发现了一类广泛存在于城市污水处理系统中的新型微生物——中科微菌科, 其是污水厂活性污泥核心菌种之一, 该菌株在污水处理厂中除了有效分解各种有机污染物外, 还能够减少硝酸盐和磷的积累, 脱氮除磷效果非常显著, 提高了污水处理效率^[92]。下一步他们将利用高通量测序的研究手段及结合国内外相关的文献调研, 针对一些核心物种开展更多的分离鉴定工作, 能够为功能微生物在污水处理厂的应用奠定理论及应用基础。

(3) 需加强污水厂微生物群落中病毒的功能及其与环境因子间相关关系方面的研究。污水处理系统中病毒是活性污泥中的组成部分, 其中分

布最广的是噬菌体和致病性病毒, 这些病毒能显著影响水生系统中的微生物群落^[46]。近年来, 污水处理厂中病毒检测研究比较广泛^[93-96], 而关于活性污泥中病毒组功能的相关研究较少, 后期需要加强环境因子对污水处理厂中病毒组的影响及其功能方面的研究, 为污水处理厂中病毒风险防控及提高污水净化效率提供理论基础。

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