

堆肥降解抗生素及抗性基因研究进展

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摘要: 抗生素残留污染问题受到广泛关注并引起高度重视。堆肥能够有效降解堆肥体系中的抗生素, 是一种绿色环保的固体有机废弃物处置方法。本文阐述了国内外固体废弃物堆肥降解抗生素研究进展, 总结了抗生素在环境中的降解途径、影响抗生素在堆肥中降解的因素和抗生素在环境中的降解过程, 并对未来的发展趋势进行展望。

关键词: 堆肥; 抗生素; 微生物; 调理剂

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Research Progress in Antibiotic Degradation and Resistance Genes During Composting

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Abstract: The problem of antibiotic residue pollution has attracted widespread attention and high priority from researchers. Composting is an environmentally friendly method for solid organic waste disposal, which can effectively degrade antibiotics in the composting system and thus serves as a green disposal technology. This paper reviews the research progress in antibiotic degradation during solid waste composting at home and abroad, summarizes the degradation pathways of antibiotics in the environment, factors influencing antibiotic degradation during composting, and the degradation processes of antibiotics in environmental systems. Finally, future development trends in this field are prospected.

Key words: Composting; Antibiotic; Microorganism; Conditioning agent

全球每年生产抗生素约 10 万 t, 其中三分之二应用于畜牧业^[1]。中国的抗生素年使用量远远高于其他国家^[2]。许多兽药抗生素在动物体内会经历不完全的吸收和代谢, 导致很大一部分给药剂量作为活性化合物被排出^[3]。畜牧业产生的动物粪便没有得到充分的预处理, 常常直接作为生粪肥施用到农田中, 并进一步扩散到地表水和地下水中^[4], 产生严重的生态影响^[5]。四环素类抗生素(Tetracyclines, TCs)包括金霉素(Chlortetracycline, CTC)、土霉素(Oxytetracycline, OTC)、四环素(Tetracycline, TC)及其半合成衍生物如甲烯土霉素、强力霉素等, 是由放线

菌产生的广谱药物, 在兽医领域应用广泛, 是当前最常用的一类药物。在农业不断发展的过程中, 我国每年产生的农业废弃物达 40 多亿 t^[6], 其中禽畜粪便占 65%, 秸秆占 17.5%^[7], 这些废弃物随意放置容易滋生细菌, 产生污染, 增加人畜共患病传播的可能性^[8], 直接应用于土壤会将抗生素有毒代谢物、重金属、致病细菌和多种无机盐引入土壤, 影响植物发育和土壤健康^[9-11]。本文系统地综述堆肥过程中不同环境条件对抗生素和抗性基因的影响, 以为抗生素降解提供理论参考。

1 堆肥对抗生素及抗性基因的影响

堆肥是一种出现较早的将农业废弃物通过生物转化进行处理的技术, 畜禽粪便和秸秆等农业废弃物的生物化学降解过程中, 关键在于优化其碳氮平衡, 同时精确管理含水率、温度及 pH 值等环境参数。将农业废弃物中可降解的物质转化为稳定的腐殖肥料, 形成一种类似腐殖质土壤的物

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质,可用作肥料或土壤改良剂^[12]。四环素类抗生素容易被微生物分解利用^[13-14],高温好氧堆肥是去除四环素的一种有效途径,使堆肥残渣可以更安全地用于农田^[15-16]。LU等^[17]使用猪粪和水葫芦堆肥,堆肥结束后测定TCs浓度小于初始浓度的0.5%。WANG等^[18]也通过堆肥使猪粪中的OTC去除89.9%。

堆肥是一种将废弃物转化为有机肥料的有效方法,抗性基因(ARGs)的存在会降低抗生素的治疗效果,而高温堆肥可以显著提高ARGs的去除率^[19],这一过程取决于堆肥的温度^[20-21]。如果堆肥的湿度和碳氮比(C/N)合适,温度会随时间呈指数上升,大部分ARGs在高温阶段被去除^[22]。堆肥过程中ARGs的去除受到多种环境因素的影响。pH值可能改变堆肥基质的化学特性,改变DNA结构,导致ARGs与基质表面的结合能力不同^[23-24]。水分也会影响ARGs的分布^[25]。一般来说,在高温阶段,水分和C/N主要影响ARGs的分布;在中温阶段,温度主要影响ARGs的分布;在成熟阶段,pH值起主要作用。因此,堆肥的不同时期对ARGs剖面的变化具有重要的影响^[19]。

2 影响堆肥降解抗生素效果的条件

2.1 堆肥原料基本性质

堆肥原料的pH值、C/N、含水率和抗生素初始浓度等性质对抗生素的降解效果有一定的影响。学者针对堆肥起始条件展开研究,以提高堆肥对材料中抗生素的去除效率。WU等^[26]改变鸡粪和甘蔗渣堆肥的时间、抗生素种类、蔗渣百分比和初始抗生素浓度,对这些条件进行人工神经网络分析。结果表明,时间、抗生素种类、蔗渣百分比和初始抗生素浓度对TCs降解的相对重要性分别为80.43%、7.95%、6.43%和5.17%。原料混合比例不同,直接影响堆体的C/N和含水率,C/N和含水率是高温阶段最重要的环境因子,抗生素的降解与C/N和水分呈正相关。研究证实,在堆肥过程中,微生物种群也会随着C/N的增加和一定范围内的含水量的增加而增加^[27]。由于C和N是参与堆肥过程的微生物细胞生长所需的两种主要营养素,因此成功的堆肥需要一个最佳的初始C/N^[28-30]。一般来说,初始C/N在20~50是最好的^[31]。RAVINDRAN等^[32]在研究鸡粪与蚯蚓联合堆肥时发现,C/N为40的蚯蚓堆肥质量最好。因此,C/N和水分也直接或间接导致抗性基因的变异^[31]。

2.2 温度

高温条件下,大多数抗生素性质不稳定。时

红蕾、SHI等^[33-34]以人粪便和锯末为试验材料,对比不同温度条件下的好氧堆肥试验对抗生素的去除效果。结果表明,四环素、金霉素、磺胺嘧啶和环丙沙星在高温堆肥中的去除率都达到90%以上,对比而言,金霉素及四环素类物质的降解速率受堆肥温度的影响程度高于磺胺嘧啶与环丙沙星。在高温阶段四环素类抗生素降解速率较快,高温也是影响猪粪堆肥过程中抗生素降解的一个主要因素^[19]。

2.3 外源添加剂

微生物接种剂、膨胀剂(疏松剂)和特定功能调理剂等物质被添加到堆肥物料中,能够进一步加快堆肥进程,提高堆肥产品的品质^[35]。

2.3.1 微生物接种剂

在加快堆肥腐熟过程中,多种微生物菌群的协同作用效果要远高于单一菌群的作用^[36]。因此,为了提高堆肥体系中微生物菌群的数量,加速堆肥反应过程,人工接种微生物的方法被广泛研究。接种木质素降解菌会影响中温阶段的细菌群落,可以降低ARGs的传播风险,提高堆肥产品的安全性,提高堆肥质量^[37]。接种白腐真菌不仅可以减少铵态氮的损失,也会加速堆肥中四环素降解,同时不会影响堆肥产品腐熟度^[38]。添加0.5%的枯草芽孢杆菌可以通过影响堆肥的pH值来抑制ARGs的传播^[39]。孟应宏等^[40]在堆肥成品中成功筛选出一株能够高效降解土霉素的菌株(T₄菌),添加T₄菌后,土霉素去除率升高。仇天雷等^[41]也证实,添加菌剂对提高鸡粪堆肥高温期温度具有明显的促进作用,并进一步提高堆肥腐熟程度,提高土霉素的降解率。WANG等^[42]在柑橘果皮堆肥中接种微生物,提高了细菌群落的丰度和多样性,延长堆肥高温期。嗜热脂肪芽孢杆菌的应用能改变堆肥升温 and 高温阶段的细菌群落结构,增加乳酸菌和硝化菌的相对丰度,提高温度,缩短升温期,显著降低pH值,促进硝化作用,减少氨的排放^[43]。

2.3.2 膨胀剂(疏松剂)

畜禽粪便等堆肥原料有机质含量和营养成分较高,质地较为细腻,透水透气性较差,会影响堆体中的氧气浓度,降低好氧微生物的活性和能量的利用,因此通常采用添加膨胀剂的方法,提高堆体的孔隙度,有助于堆体内部空气流通。目前大多数关于肥料堆肥过程中抗生素和ARGs的研究都是在添加膨胀剂的情况下进行的^[44-45]。它可以调节堆肥的湿度和C/N,使其达到最佳的堆肥条件,从而使堆肥温度迅速升高。膨胀剂的原料

种类较多,常用的膨胀剂有锯末、作物秸秆等。ZHOU等^[46]则将两种不同粒径(35、50 mm)的可回收塑料填充剂作为堆肥膨胀剂,研究该膨胀剂的品质和粒径对污泥堆肥温度、氧气浓度、含水率和植物毒性的影响。QIU等^[47]向肥料中添加稻草和锯末,增加了磺胺类抗生素的去除率。木屑、稻壳和蘑菇渣也是堆肥过程中常用的膨胀剂,它们的加入使猪粪中抗生素去除率提高14.9%~33.4%。

2.3.3 特定功能调理剂

LASHARI等^[48]发现农作物秸秆生物炭与畜禽粪便复合堆肥的产物能加固土壤抵御盐度胁迫的能力。AGEGNEHU等^[49]在大麦农田施用了包括生物炭在内的4种有机堆肥和无机肥,其中,所有施用了包含生物炭肥料的试验田组别均表现出更好的农学指标。ZHANG等^[50]通过在污泥堆肥中施用生物炭的研究指出,生物炭所表现出的积极影响,是因为它的高孔隙度和比表面积。ZHANG等^[51]发现天然沸石有效抑制了污泥堆肥过程中残留抗生素的抗性基因的表达。CHAN等^[52]利用沸石控制食品废物堆肥形成鸟粪石时带来的高电导率,并最终发现10%沸石的调节提高了堆肥腐熟度,也提高了对铵离子的吸附,减少了18%的氨气排放,控制了氮损失,从而使最终堆肥成品有更高的氮含量。

以上各条件因子不仅直接影响着堆肥的效果,且各影响因子之间的互相作用也影响堆肥效果。例如作为微生物生存的基本条件,堆肥过程中水分含量至关重要。水分过多压缩堆体孔隙,减少氧气供应,进而形成不利于好氧发酵的厌氧环境。调控各堆肥参数的平衡,选择最优化的堆肥条件是堆肥产生最佳肥效、对环境负面影响最小的关键所在。

3 抗生素在环境中的降解过程

非生物降解和生物降解是抗生素在环境中降解的主要过程^[53]。

3.1 抗生素的非生物降解

3.1.1 光降解

光降解是抗生素在环境中降解的一条重要途径。土霉素和金霉素都能够发生光解反应^[54]。光降解过程发生时,分子吸收光能转化为激发状态,从而引发各种反应^[55]。ZHANG等^[56]制备 $\text{Ag}_2\text{PO}_4/\text{NG/g-C}_3\text{N}_4$ 复合催化剂在100 W 氙灯光照下,四环素降解率最高为93.6%。HUANG等^[57]制备 $\text{AgBr}/\text{Bi}_2\text{WO}_6$ 光催化剂,在可见光照射下对四环

素具有良好的光催化降解性能。

3.1.2 水解

水和土壤中抗生素的水解也是一种关键的降解方式^[55]。HUANG等^[57]研究发现四环素的水解过程是四环素转化的重要途径。金霉素具备较强的水解倾向。pH值与温度是调控其水解速率的关键因素,碱性或中性条件会加速水解,而高温环境下的水解速率也远超室温。另外,金霉素一旦进入自然水体环境,其降解速度会更快,这归因于多种降解过程的共同作用^[58]。

3.1.3 氧化降解

抗生素也可以在强氧化剂的作用下降解。通过电话化处理过硫酸盐氧化可以有效去除废水中的盐酸四环素(Tetracycline Hydrochloride, TCH)^[59]。除了强氧化剂之外,电化学降解也是四环素降解的一种途径。智丹等^[60]研究证实了电化学降解过程对水中四环素的去除效果。

3.2 抗生素的生物降解

3.2.1 微生物降解

在微生物的作用下,抗生素残留物最终转变为水和二氧化碳,从而发生结构和理化性质的变化。能够降解四环素类抗生素的微生物主要是担子菌门和子囊菌门的真菌物种,少数为窄食单胞菌属和鞘氨醇菌属的细菌菌株。

3.2.2 体外酶降解

体外酶对抗生素降解的研究近年来也备受关注。李胜等^[61]研究表明黄素酶能够降解四环素类抗生素,同样能使四环素类抗生素失活。WEN等^[62-63]从病原菌和土壤中发现了一种黄素酶,对四环素具有破坏性。木素降解酶也被证实可用于四环素和土霉素的去除。

3.2.3 植物降解

植物降解在去除环境中有毒污染物方面也具有较好的应用前景,是一种绿色环保的降解技术,但在去除四环素类抗生素方面应用研究较少^[64]。

4 结论

中国是农业和畜牧业大国,抗生素使用量大、利用率低,大部分抗生素作为活性化合物排入环境中,是环境中抗生素的重要来源之一,抗生素污染问题有待进一步解决。堆肥是降解抗生素及抗性基因的环境友好手段,多种条件会影响堆肥对抗生素的降解,微生物活动对堆肥过程至关重要,随着生物技术的进步,关于堆肥微生物及其酶活性的研究也会持续深化。可以综合利用多种

途径对抗生素进行降解,生物降解是一种降解抗生素的重要途径,在堆肥中添加外源微生物可以提高堆肥效率和质量,促进抗生素的降解。

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