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芘、镉单一及复合污染胁迫下土壤生态功能稳定性的 影响机制

陈欣瑶1,杨惠子1,李敏2,牛晓丛1,苏雨轩1,张园1*

(1. 苏州科技大学环境科学与工程学院,苏州 215009; 2. 中国科学院南京地理与湖泊研究所,南京 210008)

摘要:当前我国土壤污染的形势已十分严峻,各种污染物进入土壤导致农产品生态安全问题已不容忽视,土壤污染问题亟待解决.本研究将土壤的抵抗力、恢复力及稳定性作为土壤质量的评价指标,对土壤施加不同含量(含量比)梯度的花(PYR)、镉(Cd)单一及花/镉(PYR/Cd)复合污染,分别从微生物活性、多样性和丰富度方面对土壤生态系统进行系统的描述,并构建模型来描述 Sb 与 PYR、Rt 与 Cd 的剂量-效应关系.结果表明,不同类型污染物在土壤的扩散过程中均会对土壤 DOC 含量造成一定程度的削减,且在 PYR/Cd 复合污染中污染物的含量比值与 DOC 含量的降低速度呈反比例关系; PYR、Cd 单一污染处理下,土壤微生物量和微生物菌落数均随着污染物含量的增加而降低; 在 PYR/Cd 复合污染中,PYR/Cd 比值大小与 Rt 呈负相关且其中 Cd 对微生物的抑制作用占有主导地位;此外本研究对显著相关的两组数据:PYR 含量与稳定性、Cd 含量与抵抗力构建了回归预测模型,其中二项式模型均可较好描述 PYR、Cd 胁迫下与上述稳定性参数的剂量-效应关系.

关键词:土壤稳定性;土壤微生物;剂量-效应;芘;镉

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Mechanism of Soil Eco-Functional Stability Under Pyrene/Cadmium Simplex and Combined Pollution Stress

CHEN Xin-yao¹, YANG Hui-zi¹, LI Min², NIU Xiao-cong¹, SU Yu-xuan¹, ZHANG Yuan^{1*}

(1. School of Environmental Science and Engineering, Suzhou University of Science and Technology, Suzhou 215009, China; 2. Nanjing Institute of Geography & Limnology, Chinese Acadamy of Sciences, Nanjing 210008, China)

Abstract: In current scenario, the soil pollution has become very severe and its effects on agricultural and ecological security issues cannot be ignored as various contaminants are discharged into soil. Thus, the soil pollution is exigent and has to be solved. This research took soil resistance(Rt), resilience (Rl) and stability (Sb) as evaluation indexes for judging soil quality by exerting different concentration (concentration ratio) gradient of pyrene (PYR), cadmium (Cd) and pyrene/cadmium (PYR/Cd) combined pollutants. A sympathetic description was showed from the aspects of microbial activity, diversity and abundance of soil ecosystem, and the models were constructed to describe the dose-response relationship between PYR-Sb and Cd-Rt. The research showed that different types of pollutants had certain inhibition on soil DOC content. In Cd and PYR simplex pollution, soil microbial mean biomass and colony number decreased with increasing concentration of pollutants. In PYR/Cd combined pollution, the ratio of PYR and Cd had a negative correlation with the decreasing rate of DOC and resistance, meanwhile Cd had a prominent influence on the above-mentioned correlations, in other words, the soil with higher concentration of Cd had lower DOC decrease rate and resistance, and Cd would have dominant inhibition effect on microorganisms under PYR/Cd combined pollution. In addition, this study found the significant correlation of $c_{\rm PYR}$ -Sb and $c_{\rm Cd}$ -Rt, and built the binomial forecasting model to describe the dose-response relationship of $c_{\rm PYR}$ -Sb and $c_{\rm Cd}$ -Rt.

Key words: soil stability; soil microbe; dose-response; pyrene; cadmium

土壤是人类社会生产活动的重要物质基础,是不可缺少、难以再生的自然资源. 当前,我国土壤污染的形势十分严峻,各种污染物进入土壤导致农产品质量安全问题已不容忽视^[1,2]. 由于土壤生态系统具有一定的稳定性,在经受污染或者剧烈环境变化后,系统的一些组成和功能随着时间的推移可能会部分或者完全恢复. McCann^[3]从生态学角度说明可用土壤的抵抗力和恢复力来评价土壤生态系统的稳定性^[4],其大小能够反映土壤质量及土壤健康的高低^[5,6].

环境中不同类型的污染物可分为有机和无机污染物,其中无机污染物主要以重金属为主,环境中重金属无法自然降解且不易被微生物利用,进入土壤很难被消除,而多环芳烃类是有机污染物中污染较

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作者简介: 陈欣瑶(1995~), 女, 硕士研究生, 主要研究方向为土壤 污染修复, E-mail:18896584296@163.com

* 通信作者, E-mail: yuanzhang_1001@ mail. usts. edu. cn

严重且研究较为广泛的一类污染物,其具较高的辛醇-水分配系数(K_{ow}),易在土壤中累积^[7].故各种污染物对土壤生态系统造成深刻影响可归纳为以下两点:一方面污染物通常具有致毒作用,会直接影响土壤生物活性,改变土壤生物系统结构,进而影响土壤生态功能;另一方面,土壤生物通过一系列代谢途径消纳、转化或富集污染物,影响污染物的环境行为和归趋^[8].大量研究表明土壤微生物对各种污染物的胁迫响应较植物(作物)更灵敏,土壤微生物承受暂时性扰动或长期胁迫的能力,可在一定程度上反映生态系统各个过程的稳定性^[9],所以研究微生物对外界环境胁迫的响应是十分必要的.

目前,已有研究表明环境胁迫和扰动作用对微生物的基础呼吸量有一定的影响. Chander 等^[10]通过对土壤重金属污染进行研究发现,土壤中的重金属会影响土壤微生物的代谢过程,而这一过程往往通过呼吸强度来表现. 而对于多环芳烃,其本身不易被微生物利用,微生物需从其他地方获得碳源和氮源进行呼吸作用从而改变多环芳烃的结构和性质. 因此土壤微生物活性可以作为评价土壤生态系统的稳定性的指标,以科学评价污染物胁迫下土壤修复能力的高低^[4].

本文以苏州地区农田土为研究对象,对其施加不同的含量(含量比)梯度的芘(PYR)、镉(Cd)单一及芘/镉(PYR/Cd)复合污染. 本研究充分考虑土壤稳定性,分析不同种类、不同含量的污染胁迫对微生物活性、丰富度和多样性的影响程度,从微生物角度清晰地描述重金属和有机污染物胁迫下土壤生态功能稳定性的变化情况,以期为土壤系统的污染治理提供理论支持.

1 材料与方法

1.1 土壤样品采集与制备

本次实验所采集的土壤样品取自地处亚热带季风气候区的苏州农田土(N31°18′, E120°6′),其理化性质、污染水平均具有典型代表性[11,12]. 取样时,随机选择周边彼此相距 10 m 的 3 个地块(范围0.5 m×0.5 m)采集土壤,除去表层 1 cm 左右的浮土,采集表层 1~20 cm 深度的洁净土壤样品,收集混合后进行风干并研磨过 2 mm 筛网备用[13].

1.2 理化性质指标测定方法

本实验采用称重法(NY/T 52-1987)测定含水率; 电位法(NY/T 1121.2-2006)测定 pH 值; 吸管法(LY/T 1225-1999)测定粒径; 乙酸铵交换法和氯

化铵-乙酸铵交换法(LY/T 1243-1999)测定阳离子交换量(CEC);碳氮元素分析仪(Vario MAX CN)测定碳氮比(C/N);水提法预处理后,总有机碳分析仪(TOC-V_{CPH},SHIMADZU,Japan)测定溶解性有机碳(DOC).本研究所测定土壤的基本理化性质见表 1.

表 1 土壤基本理化性质指标

Table 1 Soil physico-chemical properties

	1 1
理化性质	测定结果
рН	6. 37 ± 0. 26
粒径	黏粒: 2.97%,粉砂: 11.21%,砂粒: 85.82%
C/N	11.70
CEC	26. 32 cmol·kg ⁻¹
DOC	350.00 mg·kg ⁻¹

1.3 土壤中镉和芘含量的测定

1.3.1 土壤中镉含量的测定

土壤样品采用 $\mathrm{HF\text{-}HNO_3\text{-}HClO_4}(1:1:4)$ 进行消解,消解完全后的酸液用超纯水定容到 50 mL,经过 0.45 $\mu \mathrm{m}$ 水相滤头过滤后用电感耦合等离子体质谱仪(ICP-MS)进行 Cd 含量的测定 $^{[14]}$.

1.3.2 土壤中有效芘的测定

本研究选用土壤中总有效态 PYR(可解吸态和非解吸态之和)作为检测对象,采用化学分级提取的方法进行分析[即分别用 HPCD、二氯甲烷-丙酮混合溶剂(1:1)以及强碱(NaOH 溶液)进行分级提取],采用气相色谱-质谱法(GC-MS)进行测定^[14], 芘的检出限为 0.1 mg·kg⁻¹.

1.4 底物诱导呼吸速率实验

1.4.1 预实验

通过相关研究确定预实验对 Cd 及 PYR 所设置 5 个 污染含量梯度: 50、100、200、350、500 mg·kg^{-1[15,16]},复合污染 PYR/Cd 的污染物含量比例为50: 50、50: 200、50: 500、200: 50、500: 50,其中土壤被充分混合后随机分配进行两种处理:①对照组,未施加污染胁迫;②污染胁迫组,对其进行底物诱导呼吸速率实验,培养3d.采用 OriginPro 8.0软件分析计算重金属胁迫下土壤微生物的半数效应(EC50),其中 PYR、Cd 系列浓度梯度下的 EC50 分别为 192. 09 mg·kg⁻¹、98. 35 mg·kg⁻¹.

1.4.2 培养实验

土壤充分混合后被随机分配进行 4 种处理:①对照组,未施加污染胁迫;②Cd 胁迫组;③PYR 胁迫组;④PYR/Cd 复合胁迫组,对其进行底物诱导呼吸速率(SIR)实验^[16],此外每组处理设置 3 个

平行. 基于上述预实验所获得的 EC50,培养实验中将 PYR、Cd 的添加含量梯度均设置为 $10 \times 20 \times 50 \times 100 \times 200 \times 350 \text{ mg} \cdot \text{kg}^{-1}$, PYR/Cd 复合污染的含量

比例为 50: 50、50: 100、50: 200、100: 50、200: 50. 在 25℃恒温下保持 18% 的含水率培养 30 d,将以上处理分类编号,见表 2.

表 2 培养实验不同处理中污染物含量列表

Table 2	Concentration	list of	pollutants	using	different	treatments	in th	e incubation	experiment
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AL THE AM DIT	污染物含量		污染类型		污染物含量
处理类别	/mg·kg ⁻¹	芘(PYR)	镉(Cd)	复合(PYR/Cd)	添加比例
对照组	0		NPS (nor	n-polluted soil)	
	10	PYR-1	Cd-1	PYR/Cd-1	50: 50
	20	PYR-2	Cd-2	PYR/Cd-2	50: 100
污染胁迫组	50	PYR-3	Cd-3	PYR/Cd-3	50: 200
(polluted soil, PS)	100	PYR-4	Cd-4	PYR/Cd-4	100:50
	200	PYR-5	Cd-5	PYR/Cd-5	200:50
	350	PYR-6	Cd-6	_	_

本研究定义加入胁迫的当天为第 0 d,分别在第 1、3、7、15、30 d 测定土壤底物诱导呼吸速率,通过公式(1) [13,17] 计算污染胁迫组和对照组的呼吸强度. 为量 化土壤 微生物 对抗胁迫的抵抗力 (resistance,Rt)即呼吸量计算中对应的数值 f(1)、恢复力(resilience,Rl)即 f(30),从而确定添加胁迫后所测得第 1~30 d 的 Rt 和 Rl. 研究还定义反应第 1~30 d 的弹性曲线面积为稳定性(stability,Sb),计算见公式(2) [13,17].

$$f(t) = \frac{\text{CO}_{2\text{-stressed}}(t)}{\text{CO}_{2\text{-control}}(t)}$$
 (1)

$$Sb = \int_0^{60} f(t) dt \tag{2}$$

式中,f(t) 为第 t d 污染胁迫组与对照组 CO_2 的浓度比值; Sb 为土壤稳定性.

1.5 微生物量测定

准备 2 个干净平板,从处理组土壤样品中分别称取 5 g 土样放至平板①,不作任何处理,记为对照组;分别称取 5 g 土样放至平板②,然后高压灭菌锅中灭菌 10 min,温度设置为 105℃,记为灭菌组.将2 个平板中土样分别加至三角瓶内,再加入 50 mL浓度为 0.5 mol·L⁻¹的 K_2SO_4 溶液,摇床上振荡 20 min 后用滤纸过滤,滤液立即用碳自动分析仪测定有机质含量,通过公式(3) 计算微生物量.

微生物量
$$(mg \cdot kg^{-1}) = \frac{\overline{\chi}$$
 菌组 – 对照组
土壤干重 (3)

1.6 微生物菌落数测定

样品稀释液的制备: 称取 10 g 待测样品于 250 mL 三角瓶中, 再加入 90 mL 无菌水, 密封完好后置于摇床上振荡 20 min 制成悬菌液; 取 6 只试管分别加入 9 mL 无菌水, 依次编号为-1、-2、-3、4、-5、-6; 然后 10 倍稀释法将悬菌液稀释至 10⁻¹、10⁻²、

10⁻³、10⁻⁴、10⁻⁵、10⁻⁶等一系列稀释度于对应试管中. 每土样做 3 个平行样, 所有操作步骤必须保持无菌操作.

稀释涂布平板法:准备 6 只无菌平板,依次编号-1、-2、-3、4、-5、-6. 无菌条件下将 LB 培养基倒人 无菌平板中,待培养基凝固后,从试管中吸取 100 μL 菌液对号接种在平板上,再用涂布棒(已灭菌) 将菌液涂抹均匀,培养箱中 30℃条件下培养 3 d,利用 CFU 计数法进行计数.

1.7 数据分析处理

使用 SPSS 22.0 统计软件对污染物含量与底物诱导呼吸速率实验所得抵抗力、恢复力及稳定性数据进行相关性分析(F-检验),从而探讨不同含量、不同类型污染对土壤抵抗力、恢复力和稳定性的影响程度.

2 结果与讨论

2.1 不同污染胁迫对土壤可溶性有机碳的影响

可溶性有机碳(DOC)是土壤有机碳形成和矿化的重要中间形态,土壤中 DOC 的含量越高,决定着土壤肥力高低的有机质含量就越高. Griffiths 等^[18]研究有机质含量较低的土壤与有机质含量较高的土壤在受到铜胁迫和热胁迫时,土壤中的有机质有利于维持土壤中的微生物活性和稳定性.

从整体上分析,土壤中的 DOC 含量随时间推移基本呈逐渐变小的趋势,这可能是由于外源有机质(葡萄糖)的施入刺激土壤中微生物的繁殖,促进了对葡萄糖的分解,使 DOC 含量逐渐升高.由表 3 具体分析可知不同含量、不同类型污染物胁迫下土壤DOC 含量基本呈先下降后上升再下降的变化趋势,分别呈"W"型与"倒置 N"型,即前期均为"V"型,而

对应的呼吸曲线(见图1)也为"V"型,即有机碳含量曲线与呼吸曲线变化趋势在前期保持一致,这是由于培养前期污染物会对土壤呼吸有一定的促进作用,当土壤中微生物呼吸作用增强时,微生物所需的碳源增多,消耗量变大,从而使得土壤中剩余的可溶性有机碳含量下降. 此外所有污染胁迫下,第30d(D30)的DOC含量均小于第1d(D1),这说明不同类型污染物在土壤的扩散过程中均会对土壤DOC含量造成一定程度的削减.

由表 3 可知,当土壤受到 PYR 污染时,其 DOC 含量在培养期内要高于 Cd 和复合污染处理,且在第 1 d 接近原始土样的 DOC 含量 0.35 mg·g⁻¹,说明 PYR 污染对土壤 DOC 的危害相对较小,且 PYR-3 时土壤中 DOC 含量在培养周期内基本稳定在较高的水平,这符合彭静静等[19]研究中指出的 PAHs

污染的中值效应. 而在 Cd 污染胁迫下, DOC 的增加会导致重金属形成的络合物含量增多进而降低Cd 的有效性,这一点王艮梅等^[20]于土壤中水溶性有机物对重金属活性的影响实验为其提供了理论依据,此外研究发现土壤在添加 Cd 污染物后其DOC 含量急剧降低并在培养期内始终保持降低水平,说明 Cd 污染对土壤 DOC 的影响更剧烈,危害更大. 对于 PYR/Cd 复合污染,从 PYR/Cd-1、PYR/Cd-2、PYR/Cd-3 中发现,随着 Cd 含量的增加, DOC 含量的下降幅度随时间逐渐变大;从 PYR/Cd-1、PYR/Cd-4、PYR/Cd-5 中发现,随着PYR 含量的增加, DOC 含量的下降幅度则逐渐减小,说明 PYR/Cd 复合污染中, Cd 含量升高会加速DOC 含量的降低,而 PYR 含量的升高会减缓 DOC 含量的降低.

表 3 不同含量污染胁迫下 DOC 含量随时间变化趋势/mg·kg-1

污染物类型	处理方式	$D1^{1}$	D3	D7	D15	D30
	PYR-1	458. 8	350. 8	153. 6	292. 1	35. 6
	PYR-2	326. 7	189. 4	298. 3	157. 8	105. 2
芘污染	PYR-3	321.7	104. 9	287. 6	84. 8	41.7
(PYR)	PYR-4	331. 2	243. 2	278. 6	227. 6	170. 7
	PYR-5	207. 4	341. 9	169. 6	175. 3	121. 2
	PYR-6	195. 8	182. 8	124. 4	126. 7	108. 3
	Cd-1	181. 5	202. 6	201. 5	131.7	30. 9
	Cd-2	98. 7	86. 8	170. 9	123.4	32. 5
镉污染	Cd-3	113. 0	98. 5	206. 8	52. 5	26. 8
(Cd)	Cd-4	36. 6	24. 9	75. 1	68. 3	17. 4
	Cd-5	20. 0	18. 5	28. 3	23.4	12. 5
	Cd-6	50. 2	13. 9	15. 1	14. 0	12. 1
•	PYR/Cd-1	458. 8	350. 8	153. 6	292. 1	35. 6

189.4

104.9

243. 2

341.9

298.3

287.6

278.6

169.6

Table 3 Trends of DOC over time under stress with different concentrations of pollutants/mg·kg⁻¹

复合污染

(PYR/Cd)

2.2 不同污染胁迫下土壤的微生物呼吸曲线

326.7

321.7

331.2

207.4

PYR/Cd-2

PYR/Cd-3

PYR/Cd-4

PYR/Cd-5

土壤的基础呼吸被用来衡量污染物对于土壤生物活性的影响,本研究通过培养实验结果作相应呼吸曲线,即f(t)曲线,取f(1)为抵抗力、f(30)为恢复力.有研究表明,芘、镉及复合污染(PYR/Cd)的呼吸曲线均呈先下降再上升趋势,符合土壤受到污染胁迫后的污染扩散模式^[21].研究同时发现,在同种土壤应对不同类型污染物的胁迫过程中,f(t)曲线的转折点全部分布在第7或15d,说明土壤在7~15d内会基本完成污染物的扩散过程,并会在扩散完全后实施土壤的自我修复.

比较土壤中不同含量 PYR 胁迫的呼吸曲线(图 1), f(1)在不同含量下差异性较小,这可能是在添加 PYR 污染胁迫后由于可挥发性丙酮在培养前期未完全挥发从而导致土壤微生物呼吸偏差较小的现象. 此外呼吸曲线中f(15)在 PYR-6 时最小,即该含量下 PYR 对土壤微生物的抑制作用最大,说明在污染扩散完全且污染物含量较高时,其呼吸强度会大幅度降低,这和姜睿玲等^[22]研究得出 PYR 污染浓度越大对土壤微生物的抑制作用越大的结果一致.

157.8

84.8

227.6

175.3

105.2

41.7

170.7

121.2

对于 Cd 单一胁迫,低含量时其呼吸曲线差异性不明显,且 f(t) 值随时间变化也相对稳定,对此可

¹⁾ D_i 指土壤培养实验的第 i d

以解释为土壤微生物在低含量 Cd 环境下形成一种耐性机制,能抵抗外界干扰并很好地维持这种机制,这主要是因为低含量 Cd 对土壤中的微生物有一定的刺激作用,提高了其活性和生物量,改善其组成结构^[23].

对于 PYR/Cd 复合污染,其呼吸曲线变化规律较为复杂,但均呈先下降后上升的趋势,这可能是由于不同比例的两种污染物相互之间会产生抑制或促进的作用. 从 PYR/Cd-1、PYR/Cd-2、PYR/Cd-3 可

发现,Cd含量与抵抗力[f(1)值]呈负相关关系,这是由于 Cd 能够被吸附到土壤微生物周围,阻碍其生长代谢过程;另一方面可能是 Cd 进入土壤后生成具有更大毒性的物质,长时间内对微生物的活性产生损害.从 PYR/Cd-1、PYR/Cd-4、PYR/Cd-5 发现, f(7)与 PYR 含量呈正相关,即在污染物扩散基本均匀后污染物质 PYR 可刺激土壤微生物活性增强;同时部分微生物可将 PYR 作为碳源,从而促进二氧化碳产生量的增加.

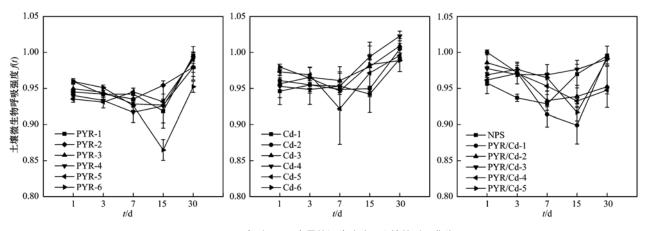


图 1 不同类型、不同含量的污染胁迫下土壤的呼吸曲线

Fig. 1 Respiration curves of soil under the stress with different concentrations of pollutants

2.3 污染物含量对土壤稳定性参数的相关性分析 及预测模型

2.3.1 相关性分析

为了解土壤中污染物含量对微生物抵抗力、恢复力和稳定性的影响程度,本研究对添加污染后的土壤进行了污染物实际含量的测定(见表4),并将不同污染物的测定含量与其抵抗力、恢复力及稳定性采用 SPSS Statistics 22.0 软件进行相关性分析.

由相关性分析结果(见表 5)可知, PYR 的含量与稳定性呈显著性负相关(R=0.825, P<0.05, n=6),即随着 PYR 含量增加土壤稳定性逐渐减小,在

多环芳烃(PAHs)污染的环境中,微生物降解 PAHs 的能力远远高于未受污染区域,但高浓度 PYR 具有很强的毒性,会破坏微生物的内部平衡,影响其生存环境的稳定,故导致土壤稳定性降低^[23]. 相关性分析还发现 Cd 的浓度与抵抗力呈显著性正相关(*R* = 0.887,*P* < 0.05, *n* = 6),即随着添加 Cd 浓度增大土壤抵抗力快速升高,这可能是因为低浓度 Cd 对土壤中的微生物有一定的刺激作用,提高了其活性和生物量,改善其组成结构^[22],而在高浓度 Cd 环境下,微生物由于长期的变异和适应会产生 Cd 的耐性菌种,该耐性菌种对土壤微生物呼吸强度的促进作用要大于高浓度Cd对其它微生物(除该耐性菌

表 4 土壤中不同污染物的测定含量(含量比)

Table 4 Measured concentration (or concentration ratio) of pollutants in soil

芘(PYR)	镉	(Cd)	复合(PYR/Cd)
处理方式	含量/mg·kg ⁻¹	处理方式	含量/mg·kg ⁻¹	处理方式	测定含量比例
NPS	ND^{1}	NPS	0. 017	_	_
PYR-1	7. 961	Cd-1	9. 772	PYR/Cd-1	40. 448: 48. 801
PYR-2	15. 422	Cd-2	19. 653	PYR/Cd-2	39. 977: 97. 513
PYR-3	40. 885	Cd-3	48. 907	PYR/Cd-3	39. 251: 195. 442
PYR-4	80. 197	Cd-4	98. 544	PYR/Cd-4	97. 513: 40. 126
PYR-5	158. 472	Cd-5	195. 381	PYR/Cd-5	194. 341: 38. 179
PYR-6	263. 359	Cd-6	342. 778	_	_

¹⁾ ND 表明未检出,检出限为 0.01 mg·kg-1

表 5 污染物含量(含量比)与抵抗力、恢复力及稳定性的显著性分析1)

Table 5 Significance analysis between the concentration (or concentration ratio) of pollutants and Rt/Rl/Sb

项目	相关性参数		芘(PYR)			镉(Cd)		复	合(PYR/C	Ed)
供目	相大任多奴	Rt	Rl	Sb	Rt	Rl	Sb	Rt	Rl	Sb
公共400	Pearson 相美性	0.082	-0.692	-0.812*	0.887*	-0.414	0.222	-0.588	0.124	-0.139
污染物含量 (含量比)	显著性(双侧)	0.877	0.128	0.050	0.019	0.415	0.673	0.297	0.843	0.824
(百里比)	N	6	6	6	6	6	6	5	5	5

1)* 为 P < 0.05

种外)呼吸的抑制作用,从而导致土壤微生物的整体呼吸强度升高,因此可以从 Cd 污染土壤中富集培养并筛选分离出抵抗能力较好的微生物种类,驯化培养后用于污染土壤的生物修复.

2.3.2 构建回归模型

对相关性显著的两组数据结果 PYR 浓度 (c_{PYR}) 与稳定性(Sb)、Cd 浓度 (c_{Cd}) 与抵抗力 (Rt)进行回归拟合. 结果发现: PYR 污染胁迫下

土壤稳定性的剂量-效应关系采用二项式回归模型(见表 6),其模拟精度较高,预测值与实测值的相关系数可达0.9025;二项式模型同样可以很好描述 Cd 胁迫下土壤抵抗力的剂量-效应关系(R=0.8774).基于模型构建结果,数据拟合图(图 2)中实线代表拟合曲线,虚线代表 95% 置信区间,全量预测值和实测值在回归图中均处于 95% 的置信区间内.

表 6 土壤稳定性参数与污染物浓度的剂量-效应关系

Table 6 Dose-response relationship between pollutant concentration and soil stability parameter

	1	1 1	7 I	
污染物	稳定性参数	回归曲线	R^2	F
芘(PYR)	稳定性(Sb)	Sb = $-2.532.8 \times 10^{-5} c_{PYR}^2 + 0.003.2 c_{PYR} + 27.443.8$	0. 814 6	6. 590 4
镉(Cd)	抵抗力(Rt)	Rt = 1.317 6 \times 10 ⁻⁷ c_{Cd}^2 + 3.304 7 \times 10 ⁻⁵ c_{Cd} + 0.954 0	0.7698	5. 015 5

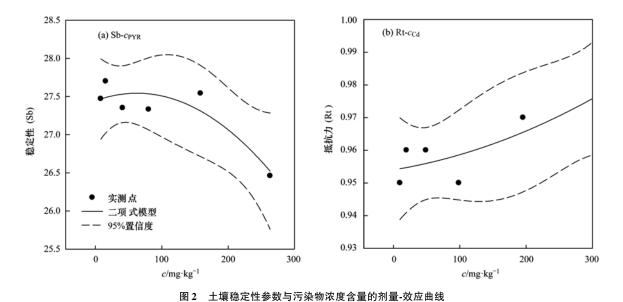


Fig. 2 Dose-response curves between pollutant concentration and soil stability parameter

2.4 不同污染物胁迫下土壤稳定性与土壤微生物的影响关系

本实验通过添加不同含量(含量比)的 PYR/Cd单一及复合污染,从微生物量及微生物菌落数两个方面对不同含量、种类的胁迫前后土壤微生物的变化情况进行了描述,从而对土壤稳定性与微生物的影响关系进行分析.本研究综合考虑 PYR、Cd系列

污染梯度下的 EC50 值及各污染物添加一定含量时 所对应呼吸曲线,选择 200 mg·kg⁻¹和 50 mg·kg⁻¹ 作为基准含量进行设置,对 3 种污染物共设置 7 种 处理(各处理方式见表 7).

添加污染物后,土壤的微生物量、菌落数均呈下降趋势(见图 3~4),这可能是因为微生物受到污染胁迫后其细胞功能遭到破坏,为了维持正常的细

胞功能需要一定的能量,从而导致生长受阻.但对于不同种类污染物及不同浓度含量的同种污染物胁迫,微生物量及菌落数的下降程度不同.

表 7 不同实验处理下的污染物含量/mg·kg-1

Table 7	Pollutant	concentrations	in	different	treatments/	mg•	kg -	I
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处理方式	Т0	T1	T2	Т3	T4	T5	Т6
芘(PYR)	0	200	50	0	0	200	50
镉(Cd)	0	50	200	200	50	0	0

2.4.1 微生物量

由图 3 可得:与 TO(对照组)相比,T1、T2、T3、 T4、T5、T6(胁迫组)土壤的微生物量分别下降 33.0%, 60.9%, 47.7%, 54.0%, 58.4%, 46.9%. 结果表明土壤微生物量随着重金属 Cd 污 染水平的提高而逐渐下降(比较 T3 和 T4 处理),程 金金等[24]研究也发现 Cd 会对土壤微生物有一定的 危害作用,这种危害作用的直接表现就是使土壤微 生物量和种群有所减少. 比较不同 PYR 含量的污 染胁迫(T5 和 T6)下对土壤微生物量的影响,发现 微生物量会随着 PYR 含量的增加而降低,这与 PAHs 浓度增加会导致土壤微生物量碳含量呈指数 下降趋势的研究结论相符^[25]. 而对于 PYR/Cd 复 合污染,T1 处理下的微生物量要高于T2,这可以解 释为在有机和无机复合污染中重金属 Cd 表现出更 明显的毒作用^[26]. 此外比较 T5 与 T3、T6 与 T4 发 现 PYR 对土壤微生物量的影响较小,这可能是因为 多环芳烃可作为某些微生物的碳源,从而刺激土壤 微生物的数量增加. 从 T1 与 T4、T2 与 T3 也可证 明上述结果.

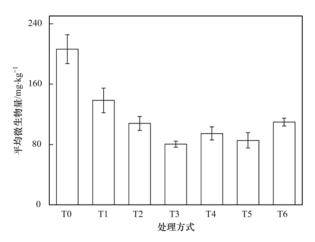


图 3 不同处理方式下土壤的微生物量变化情况

Fig. 3 Changes of soil microbial biomass in different treatments

2.4.2 微生物菌落数

本研究测定 TO 处理下土壤微生物的平均菌落

数为 $1.77 \times 10^7 \text{CFU} \cdot \text{g}^{-1}$,以此为基准从图 4 可发现 胁迫污染处理(T1~T6)下土壤微生物菌落数的下 降幅度均超过一个量级,说明污染物对土壤菌落数 的抑制作用较为明显. 从 T3、T4 可发现土壤微生 物菌落数随着 Cd 含量的升高而逐渐下降,韩桂琪 等[27] 用菌落计数法也发现土壤细菌、真菌等菌落 数会随着有效 Cd 含量的增加而降低. 比较不同浓 度的有机污染物 PYR(T5、T6)在单一污染情况下 对土壤微生物菌落数的影响,发现土壤微生物菌落 数与PYR浓度呈现负相关关系,这说明高浓度的 PYR 会对微生物表现更明显的抑制作用. 对于 PYR/Cd 复合污染胁迫来说,T1 比 T2 的微生物菌 落数更大,这可能是因为重金属对微生物存在着毒 害作用,导致微生物活性下降,故像海旋菌属等此类 可降解多环芳烃的微生物也受到影响,即对多环芳 烃的生物降解遭到破坏,从而影响了微生物菌落数, 此外研究发现 Cd 单一污染相对于其他类型污染对 土壤微生物菌落数的抑制作用更加明显,这与沈国 清等[26]得出的在有机和无机复合污染中 Cd 占有主 导地位的结论基本一致.

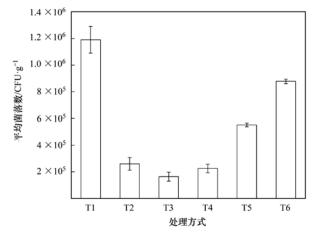


图 4 不同处理方式下土壤微生物的平均菌落数变化情况

Fig. 4 Changes of soil microbial average colonies number in different treatments

综上分析发现, PYR、Cd单一污染会导致微生物量及菌落数呈下降趋势, 而高浓度污染胁迫下其微生物量及菌落数的下降幅度较大, 而土壤稳定性较小(见图1), 这说明单一污染对土壤微生物的生物量、菌落数及活性产生的影响基本一致, 故本文定量描述的稳定性(Sb)可作为判断单一污染物胁迫下土壤微生物变化趋势的宏观指标. 而 PYR/Cd复合污染对微生物量、菌落数及活性的作用效果并不一致, 这可能是由于 Cd 含量较高时, 土壤微生物量及菌落数虽明显较低, 但抗 Cd 胁迫微生物相对

于抗 PYR 胁迫微生物的呼吸更加旺盛,导致土壤生态系统的整体微生物活性表现得更加明显.

3 结论

- (1)不同类型污染物在土壤的扩散过程中均会对土壤 DOC 含量造成一定程度的削减. 且在 PYR/Cd 复合污染中,Cd 含量升高会加速 DOC 含量的降低,而 PYR 含量对 DOC 含量则起到相反的作用.
- (2)PYR、Cd 单一污染处理下,土壤微生物量和微生物菌落数均随着污染物含量的增加而降低.
- (3)在 PYR/Cd 复合污染中, PYR/Cd 比值大小与f(1)呈负相关,与f(7)呈正相关,且 Cd 对微生物的影响作用占有主导地位.
- (4)本研究对显著相关的两组数据: PYR 浓度与稳定性、Cd 浓度与抵抗力构建了回归预测模型,其中二项式模型可较好描述 PYR、Cd 胁迫下与不同稳定性参数的剂量-效应关系.
- (5)本研究充分考虑土壤生态功能的自我恢复能力,从微生物活性、丰富度、多样性多角度地描述了重金属和有机污染物胁迫下土壤生态系统的变化情况,并通过建立土壤稳定性的剂量-效应预测方程,以达到 PYR、Cd 污染胁迫对土壤生态功能稳定性的危害及影响做出准确的预判的目的,为土壤系统的污染治理提供理论支持.

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