固定化微生物对水胺硫磷降解机理的研究*

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摘要 采用气相色谱-质谱联机和有关分析方法,给出了精品水腔硫磷的质谱图;证实固定化微生物可使有机磷 农药水胺硫磷的环链断裂,其最终产物为 CO₂、H₂O、NH₃、H₂S、H₃PO,等简单无机化合物;初步推断水杨酸异 丙酯等有机化合物是降解过程的中间产物。

关键词 固定化微生化,水胺硫磷,降解机理。

以往的研究结果表明,用固定化微生物降 解高毒有机磷农药水胺硫磷,其 COD 去除率可 保持较高的水平^[1],且反应速度符合 Michaelis-Menten 公式^[2]。本文拟采用气相色谱-质谱 (GC-MS)联机和有关分析方法,以识别水胺硫 磷降解过程的中间产物和最终产物。

1 材料和方法

1.1 材料

(1)水胺硫磷精品 由湖北沙市市农药厂 提供。

(2) 萃取剂 市售二氯甲烷(分析纯)和石 油醚(分析纯,沸程 30--60°C),均重蒸处理。

1.2 仪器及测定条件

(1) 气相色谱仪 GC-9A, 日本岛津。

(2) 气相色谱-质谱联用仪 GC/MS-QP-1000A,日本岛津。

(3) 测定条件 电子能量: 70 eV; 质量范 围: 40-350; 色谱柱: OV-17; 进口样和传输线 温度: 220℃; 升温程序: 初始温度 150℃, 保持 5 min, 以 5℃/min 的速度升温,终止温度 210℃, 保持 20 min; 流动相: He; 离子源温度: 250℃; 分离器温度: 250℃; 门槛值 5000。

1.3 试验方法

1.3.1 固定化微生物小球的制备和水胺硫磷降 解试验

按文献[1]的配方和工艺制备固定化微生物 小球,并置于反应器中,加入体积为小球松装 容积3倍的精品水胺硫磷水样,在室温(20°C) 下用压缩空气曝气,进行好氧降解试验。

1.3.2 GC-MS 分析样品的预处理

(1)萃取 取水样 20 ml 于 250 ml 分液漏
斗中,加 NaCl 5g,溶解后加入 100 ml 1:1(V/
V)的二氯甲烷和石油醚混合萃取剂,充分振摇,
待静置分层后收集有机相。再用 50 ml 混合萃
取剂重复萃取 2次;

(2) 脱水 将合并的萃取液通过盛有无水 Na₂SO₄的砂芯漏斗,然后用二氯甲烷洗涤 Na₂SO₄,收集洗涤液并与萃取液合并;

(3)浓缩 在收集的有机溶液中加入 2 ml 乙酸乙脂,并用 KD 浓缩器在 55-60℃下浓缩 至 2 ml 待测。

1.3.3 无机磷测定

采用分光光度法测磷酸盐标准液的吸光度, 并制得标准曲线,然后分别测降解前后水胺硫 磷水样的吸光度,从标准曲线上查得无机磷含 量。测量时采用1:1的10%钼酸铵和3%偏钒 酸铵作显色剂^[3],波长420 nm。

1.3.4 溶解性无机硫化物测定

分别取降解前后的水胺硫磷水样,用标准 碘量法进行测定^[4]。

1.3.5 氨的定性分析

将固定化微生物小球与水胺硫磷水样同时 放入试管内,借助棉塞将石蕊试纸和奈氏试纸 固定悬挂于试管上部,观察试纸颜色的变化。

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2 结果与讨论

2.1 生物降解前水胺硫磷色谱分析和质谱图

将含量 99.99%精品固体水胺硫磷用乙醇 溶解后加入蒸馏水,测得该水样的 COD 为 4812 mg/L。将该水样(样品 1)进行预处理,分别用气 相色谱仪和 GC-MS 联用仪测得如图 1 所示的 总离子流图,得出水胺硫磷的出峰时间为 22.08





图 1 生物降解前水胺硫磷水样的总离子流图

min, 峰面积为 83.5493%。

水胺硫磷的质谱图如图 2 所示。由于水胺 硫磷的分子量和结构式已知,故对其图谱可作 如下解析: *m/z* 289 为水胺硫磷分子离子峰, M +1和 M+2 为同位素峰。基峰 *m/z* 136 推断为 (C₆H₄OCOO)²⁻离子。*m/z* 43、64、78、94、110、 121、203、230、247 分别推断为 C₃H₇、PSH、 CH₅PNO、CH₃PSO、CH₅PSNO、C₇H₅O₂、C₇H₁₀ PSNO₂、C₈H₉PSNO₃、C₈H₁₀PSNO₄等碎片。

2.2 降解后水胺硫磷总离子流图及质谱解析

将初始 COD 为 4812 mg/L 的精品水胺硫 磷水样进行降解试验, 24h 后取第1次水样(样 品 2), 测得 COD 去除率为 61.5%; 48 h 后第2 次取样(样品 3),其 COD 去除率为 89.5%。将 样品 2 与 3 进行预处理,测总离子流图(图 3)。

由图 3(a)可见,出峰时间为 22.028 min 的 水胺硫磷峰,其峰面积为 6.5248%,但在图 3 (b)中,该分子离子峰已不存在,说明驯化好的 混合菌最终可将水胺硫磷完全分解。



图 3 总离子流图

为了鉴别水胺硫磷降解过程的中间产物, GC-MS 联机给出了图 3 所示主要分子离子峰的 质谱图。根据计算机检索和图谱解析,可初步 推断其主要中间产物为水杨酸异丙酯($C_{10}H_{12}$ O_{-})、水杨酸铵($C_7H_9NO_3$)、水杨酰胺 ($C_7H_7NO_2$)、水杨酸($C_7H_6O_3$)、甲氧基羟基硫代 磷酰胺酯(CH_6PSNO_2)及烷基(C_nH_{2n+1})、醛、酮 类($C_nH_{2n+1}CO$)化合物。

2.3 水样中无机磷含量

取水胺硫磷浓度为 200 mg/L 水样 30 ml, 生物降解前测得其中的无机磷为 0.8 mg/L,说 明在配制的水样中有少量水胺硫磷已被水解。 将同样浓度和体积的水样用固定化微生物在恒 温摇床内(30℃,200 r/min)降解 24 h,测得其中 的无机磷含量为 7.6 mg/L,证实无机磷是水胺 硫磷生物降解的最终产物之一。

2.4 水样中溶解性无机硫化物含量

取水胺硫磷浓度为 600 mg/L 的水样 80 ml,生物降解前未检出无机硫化物,但用固定 化微生物使其在恒温摇床内(30 C,200 r/min) 降解 24 h,测得溶解性无机硫化物为 2.0 mg/ L,证实无机硫化物是水胺硫磷生物降解的最终 产物之一。

2.5 氨的定性分析结果

试验经过 24 h 即可见石蕊试纸变蓝, 奈氏 试纸变黄, 说明有氨气逸出, 即氨亦是水胺硫 磷生物降解的最终产物。

2.6 水胺硫磷的生物降解机理

根据本文的试验结果,并参阅有关资料^[5], 对水胺硫磷生物降解的主要历程可作如下推论:



3 结语

(1)试验证实,高毒有机磷农药水胺硫磷的 环链结构可在微生物作用下断裂,并最终形成 CO₂、H₂O、H₂S、H₃PO₄、NH₃等简单无机化合物;

(2)根据质谱解析,初步推断水杨酸异丙 酯、水杨酸等有机化合物是水胺硫磷生物降解 过程的中间产物。

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alleviation of aluminum toxicity to wheat . Silicon was added as solutions of sodium slilicate with a pH value of 4.8 and the unamended soil was used as control. Plant biomass and contents of some elements in wheat seedlings were measured. The concentrations of some species of active aluminum in rhizosphere soils were also determined. It was found that the biomass of wheat seedlings increased and the uptake of Ca, Mg etc. by wheat seedlings was improved with 0.3 mmol/kg of Si added. When the amount of Si added reached 0.9 mmol/kg, more significant effects can be observed for the growth and uptake of nutrients by wheat seedlings. Moreover, silicon tretments slowed down the pH decline and the increment of soluble and exchangeable aluminum content in rhizosphere soils. Considering the resources and the effects for mitigating aluminum toxicity, application prospect of silicon materials such as some industrial waste products and crop residues in acid soils were discussed.

Key words: silicon. acid soil, aluminum toxicity, alleviating effect, wheat, rhizosphere.

Study on Degradation Mechanism of Organophosphorus Pesticide Isocarbophos by Immobilized Microorganism. Luo Qifang et al. (Institute of Environmental Medicine, Tongji Medical University, Wuhan 430030): Chin. J. Environ. Sci. 16(6), 1995, pp. 14-16

Gas chromatography/mass spectrometry and other analytical methods were applied in this study. The test has given a mass-spectrum of refined isocarbophos. Analytical results demonstrated that the cyclic structure of isocarbophos might be broken into simple inorganic compounds such as CO_2 , H_2O , NH_3 , H_2S and H_3PO_4 by immobilized microorganism. Organic compounds such as isopropyl salicylate were products of degradation process by inference. The primary research on the degradation mechanism of isocarbophos has also been done.

Key words: immobilized microorganism, isocarbophos, degradation mechanism.

Comparison of the Mutagenicity of Water Samples Disinfected with Several Disinfectants. Tang Fei et al. (Inst. of Environ. Med., Tongji Medical University, Wuhan 430030), Cheng Maoyi et al. (Shashi Station of Sanitation and Antiepidemics. Shashi 434000); Chin. J. Environ. Sci., 16(6), 1995, pp. 17-20

In this study, the mutagenicity was detected and compared after treatment of the Wuhan East Lake water with chlorine, chlorine dioxide or ozone. The concentrates of untreated and ozonetreated water at all sample collection dates were nonmutagenic in the Ames test. Both chlorine and chlorine dioxide treatment of the water resulted in increases of TA98 mutagenicity. Comparative analysis of these treated waters for mutagenicity showed a consistent pattern of mutagenic potency, with decreasing activity in the order: chlorine > chlorine dioxide > ozone. The study also found that pretreatment of water with potassium permanganate prior to chlorination was effective in reducing the level of mutagenicity which formed during chlorination.

Key words: chlorine, chlorine dioxide, ozone, mutagenicity of drinking water.

Biological Control of Nitrite in Fish Farming Ponds by Photosynthetic Nonsulfur Bacteria. Liu Shuangjiang et al. (Institute of Microbiology, Chinese Academy of Sciences, Beijing 100080), Sun Yan et al. (Institute of Water Research, Chinese Academy of Environmental Science, Beijing 100012); Chin. J. Environ. Sci., 16(6), 1995, pp. 21-23

Nitrite, a toxic factor for most aquatic animals and causing a great loss for fishery if it was kept at a higher level in water, was found to be utilized and removed screened and isolated strains of photosynthetic nonsulfur bacteria. Experiments indicated that the isolated strains removed nitrite by more than 80% when the nitrite in the bulk ranged from 0. 01 to 5. 0 mmol/L, regardless they were cultured under illumination or in dark. Results also show that acetate and lactate were the optimal C sources for nitrite removal. Field surveys on the nitrite concentration in fish farming ponds in Beijing area found that it was about 0.006 - 0.008 mmol/L. By using the isolated strains in the ponds, their nitrite concentrations decreased by 50% - 80%.

Key words: photosynthetic bacteria, nitrite, fishery.

Study on the Motor Soot Combustion Perfermance over Different Catalysts. Chen Min et al. (Catalytic Institute, Hangzhou University, Hangzhou 310028): Chin. J. Environ. Sci. 16 (6), 1995, pp. 24-25

In this paper, the soot collected from automobile