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Environmental Biological Effects of Chlorpyrifos based on Big Data Technology

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Abstract: Chlorpyrifos is one of the major pesticides used in rice fields in China at present. It has great toxicity to benthos, and its half-life of hydrolysis and photolysis is nearly one month. Affected by the expansion of rice field area, the concentration of chlorpyrifos poisoning in rice field drainage is increasing, so it is urgent to effectively degrade chlorpyrifos poisoning in salinized rice field. Therefore, this paper analyzes the environmental biological effects(EBE) of chlorpyrifos based on big data technology(BDT); The degradation ability of chlorpyrifos degrading bacteria in soil was measured, and the influence of chlorpyrifos degrading bacteria on soil enzyme activity and the number of soil microorganisms was discussed. The quantitative analysis of chlorpyrifos and TCP was carried out through BDT, which is of great significance for the improvement of pesticide contaminated soil in the future.

1. Introduction

Pesticide formulation is a pesticide dispersion form with various specific physical and chemical properties, which is researched and developed to make pesticides safe, convenient and reasonable for practical production. Chlorpyrifos is a typical organophosphorous pesticide, which is widely used. Its sales volume ranks first in the world of organophosphorous pesticides. It is one of the main pesticide components in the retreating water of rice fields. It can directly affect the existing form and performance of pesticides in the environment. Pesticides of different dosage forms not only have different control effects, but also often have different toxicological behaviors. It is of great significance to explore the environmental toxicological behavior of pesticides in different dosage forms for a comprehensive, scientific and reasonable evaluation of their biological effects and environmental safety. This paper makes an experimental analysis of the EBE of chlorpyrifos based on BDT.

Many scholars at home and abroad have studied and analyzed the EBE of chlorpyrifos based on

BDT. Papagiannis I et al. Found that chemical bonds such as C-C, C-O and C-N in chlorpyrifos chemical molecules are very easy to break and decompose into small molecular substances after absorbing photons of a certain wavelength and becoming excited molecules, which degrade slowly under natural light, but photosensitizers such as humus can significantly accelerate their photolysis speed [1]. Natascha et al. Found that the half-life of chlorpyrifos was shortened after repeated treatment of soil with the same concentration of chlorpyrifos for 3 times, and then a bacterium that can reduce and detoxify chlorpyrifos was isolated from the soil. After continuous use for 3 years, the control effect of chlorpyrifos granules on sugarcane underground pests decreased significantly. At the same time, the isolated microorganisms were also used to degrade chlorpyrifos residues in vegetables and soil, and achieved good results [2].

Chlorpyrifos is a moderately toxic organophosphorus pesticide, which can be effectively applied to the prevention and control of agricultural and urban pests, but it will cause a series of problems such as soil pollution if it remains in the soil for a long time. Based on the relevant research experience of scholars at home and abroad, this paper screened and obtained soil bacteria that can effectively degrade organophosphorus pesticides. Based on the BDT, the EBE of chlorpyrifos were experimentally analyzed, and the effects of contaminated soil improvement and plant growth promotion were studied in combination with PGPR bacteria. These studies are of great significance for the improvement of pesticide contaminated soil in the future [3-4].

2. Analysis of EBE of Chlorpyrifos

2.1. Introduction to Chlorpyrifos

Toxicity of chlorpyrifos: the toxicity of chlorpyrifos to humans depends on the exposure dose and time. People will have symptoms such as nausea, dizziness and unconsciousness after high-dose exposure. High concentration intake can cause respiratory paralysis and death; Long term low-dose exposure will lead to brain, eye, ear, genital and other organ defects in infants, and brain development or function abnormalities; Long term or repeated sub-toxic dose effects will also make cells and nerve axons pathological changes, and even cause behavior changes. Chlorpyrifos may also have endocrine disrupting effects. During the national health and nutrition survey, it was found that the levels of serum T4 and TSH were correlated with the metabolites of chlorpyrifos poisoning in urine. In addition, chlorpyrifos also has high toxicity to aquatic organisms and other non target organisms, which can significantly reduce the population of earthworms and termites in the soil, and inhibit the number and function of bacteria, fungi, actinomycetes and other microorganisms in the soil [5-6].

Current situation of chlorpyrifos pollution: continuous and excessive use of chlorpyrifos has caused serious environmental pollution. Chlorpyrifos not only pollutes the direct application areas, but also has the highest detection frequency of pesticide residues in the environment in some countries. Serious chlorpyrifos pollution has posed a threat to human health [7].

Degradation of Chlorpyrifos in the environment: chlorpyrifos is easy to hydrolyze, photolysis and be decomposed by microorganisms in the environment. Under certain conditions, the phosphate bond in chlorpyrifos molecule is easy to be hydrolyzed and broken, resulting in the decomposition of the compound. In water, temperature, pH value, initial concentration and ionic strength of water affect the hydrolysis rate of chlorpyrifos. The hydrolysis of Chlorpyrifos in weak acidic water is not obvious, but when the pH value is 9.0, the 7-day degradation rate of chlorpyrifos is 40.8%.

2.2. Analysis of EBE of Chlorpyrifos

2.2.1. Determination of Degradation Ability of Chlorpyrifos Degrading Bacteria in Soil

Determination of strain degradation ability: 50g soil sample was added to a 200ml beaker, 25ml bacterial solution (109cfu/ml) (25ml sterile water was added to the control group) and 2ml chlorpyrifos solution (500mg/l) were added, stirred and mixed well, and then stood still. Soil samples were taken on the day of the test and the 21st day after that to determine the residue of Chlorpyrifos in the soil. In order to ensure that the soil moisture content is basically unchanged during the test, weigh the soil samples every three days and supplement the moisture [7-8].

Identification of chlorpyrifos degrading bacteria: 11 strains of chlorpyrifos degrading bacteria were classified and identified. Through the extraction of genomic DNA, PCR amplification reaction and agarose gel electrophoresis detection, the fragment length of about 1500bp sequence was obtained [9]. The isolation, identification and degradation characteristics of chlorpyrifos degrading bacteria were studied. The strains with the highest homology with the screened strains were obtained, and the types of screened strains were determined. The 11 strains of chlorpyrifos degrading bacteria screened mainly belong to Burkholderia and Klebsiella, among which the CD5 and CD7 strains selected in the subsequent test belong to Burkholderia [10-11].

2.2.2. Effect of Chlorpyrifos Degrading Bacteria on the Growth of Brassica Campestris Seedlings

The root length, seedling height, fresh weight and dry weight of young cabbage were measured respectively. Wpsexcel was used for data processing, and SPSS18.0 software was used for one-way ANOVA ($p<0.05$). The results are shown in Table 1.

Table 1. Effects of chlorpyrifos degrading bacteria on the growth of Brassica campestris seedlings

Processing group	Average root length (CM)	Average seedling height (CM)	Average fresh weight (g)	Average dry weight (mg)
CK	1.642±0.107a	3.778±0.127a	0.048±0.003a	0.0173±0.002a
CD5	1.854±0.120a	4.012±0.234a	0.053±0.006a	0.0257±0.004a
CD7	1.832±0.086a	4.233±0.275a	0.059±0.004a	0.1123±0.003a

The results of pot experiment showed that irrigation with CD5 or CD7 bacterial solution had no effect on the growth of Brassica chinensis seedlings. Compared with the control group, the root length and seedling height of Brassica chinensis increased by 12.91%, 11.57% and 6.19%, 12.04% respectively [12].

2.2.3. Effect of Chlorpyrifos Degrading Bacteria on Soil Enzyme Activity

Using one-way ANOVA and least significant difference analysis, it can be seen from the measured soil enzyme activity data (Table 2) that watering small green vegetables with CD5 or CD7 bacterial suspension will have different effects on soil enzyme activity. CD5 bacterial

suspension will inhibit soil urease and sucrase activities, while CD7 bacterial suspension can improve soil urease, cellulase and catalase activities, and there is a significant difference with the control group, It shows that strain CD7 has no significant effect on soil enzyme activity, which lays a foundation for the selection of strains in subsequent experiments [13-14].

Table 2. Data sheet of soil enzyme activities

Processing group	Urease	Sucrase	Cellulase	Catalase
CK	0.849±0.004b	745.835±0.721a	4.473±0.950a	0.253±0.025b
CD5	0.549±0.002c	715.326±0.472b	5.333±0.943a	0.473±0.007a
CD7	1.073±0.002a	518.381±0.001c	2.164±0.946a	0.522±0.009a

2.2.4. Effect of Chlorpyrifos Degrading Bacteria on the Number of Soil Microorganisms

Soil microorganism is an important part of soil ecosystem, and its composition and structural changes will affect the physical and chemical properties and fertility of soil [12]. Generally speaking, soil containing more microorganisms has high fertilizer efficiency and is more conducive to the growth of plants [15]. It can be seen from Figure 1 that watering CD5 bacterial suspension will reduce the number of soil microorganisms, indicating that there may be mutual inhibition between this strain and soil microorganisms; The addition of CD7 suspension can better maintain the number of fungi and bacteria in the soil, while the number of actinomycetes decreased slightly compared with the control group. To study the effect of CD5 and CD7 strains with high chlorpyrifos degradation capacity on the number of soil microorganisms is to provide a basis for further strain screening [16].

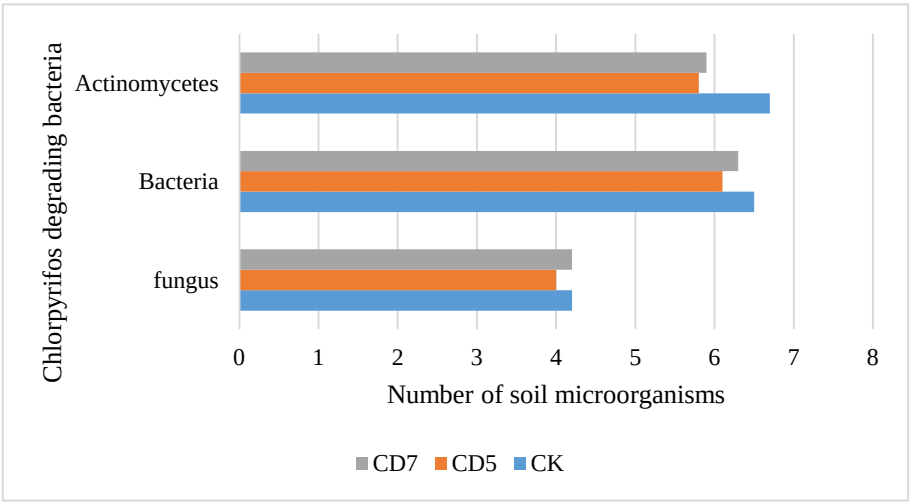

Figure 1. Effect of chlorpyrifos degrading bacteria on the number of soil microorganisms

After 16SrDNA sequencing and blast comparison, these strains were preliminarily identified as Burkholderia, Klebsiella, raoulia and Enterobacter. The degradation ability of 11 strains of chlorpyrifos degrading bacteria screened was determined. The results showed that the degradation ability of Burkholderia CD5 and CD7 strains to chlorpyrifos in culture medium and soil was higher

than that of other strains within 72 hours. It is further confirmed that the two strains have high degradation capacity in the culture medium and soil, so they have the potential and practical application value for the remediation of pesticide contaminated soil [17-18].

3. Application of BDT in Environmental Biological Effect Analysis

BDT substitutes the adsorption / desorption data of most substances into the bet equation to obtain the specific surface area of each matrix. Larger specific surface area means that there are more sites that may have adsorption characteristics. Only gravel, expanded vermiculite and Fe-C have pore sizes <100nm. From the data of specific surface area, vermiculite and Fe-C are ideal matrix materials.

$$\frac{f}{s(f_0-)}=\frac{1}{S_n\cdot H}+\frac{H-1}{S_n\cdot H}\cdot(f/f_0) \tag{1}$$

Where, f represents the partial pressure of adsorbate; F0 represents the saturated vapor pressure of adsorbent; S refers to the actual adsorption amount of the sample; Sn refers to the saturated adsorption capacity of single layer; H is a constant.

The adsorption kinetic characteristics of matrix can reflect the boundary adsorption behavior of matrix and chlorpyrifos. Based on BDT, this paper analyzes the pseudo first-order dynamics model and pseudo second-order dynamics model, and their expressions are as follows:

$$IN(G_{e1}-G_t)=InG_{e1}-K_1t \tag{2}$$

$$\frac{t}{G_t}=\frac{1}{k_2G_{e2}^2}+\frac{t}{G_{e2}} \tag{3}$$

Where, t is the adsorption time (min); Adsorption GT and Ge are the adsorption capacity of chlorpyrifos at adsorption time t and the adsorption capacity at adsorption equilibrium, respectively(μ gg-1); K1 represents the reaction constant of the pseudo first-order model (lmin-1); K2 represents the reaction constant of the pseudo second-order model (G(μ gmin)-1)

4. Analysis of EBE of Chlorpyrifos based on BDT

4.1. Experimental Scheme

4.1.1. Experimental Materials and Devices

The experiment was carried out in a greenhouse with room temperature of 24 ± 3 °C and humidity of 50 ± 20%. The main wetland plants tested in the experiment include Canna (plant height is about 50cm, wet weight is about 75g), reed (plant height is about 60cm, wet weight is about 25g) and cattail (plant height is about 50cm, wet weight is about 30g). The slag (irregular block, particle size 50 ± 20mm) is selected as the substrate of the constructed wetland. The experimental water is tap water.

The experimental device is made of plexiglass with an overall dimension of 150 (side length) × 150 (side length) × 600 (height) cm. The bottom of the device is equipped with water intake, and the front of the device is equipped with two matrix sampling ports with a diameter of 8cm, 15 and 30cm away from the bottom respectively. The filling height of the substrate of the constructed

wetland is set at 40cm, which is covered with 10cm soil. Three groups were set up in the experiment, with three in each group in parallel. Group 1 transplanted 1 Canna; Group 2 transplanted 5 reeds; Group 3 transplanted 3 cattails. The outer wall of the device is covered with tin foil to avoid light. The specific structure of the experimental device is shown in the figure below.

4.1.2. Experimental Design

Before the experiment, the nutrient solution (glucose 50mgl-1, urea 10mgl-1, potassium dihydrogen phosphate 10mgl-1) was injected into the constructed wetland, and the nutrient solution was changed every 7 days for 28 days. The purpose is to make the newly transplanted plants adapt to the new growth environment and allow microorganisms to multiply and attach to the substrate surface. At the beginning of the experiment, chlorpyrifos simulated wastewater was injected into the artificial wetland simulation device, and no water was replenished during the experiment. The HRT was set as 8D in the experiment. At 0h, 2h, 5h, 8h, 18h, 1D, 2D, 3D, 4D, 6D and 8D after the beginning of the experiment, 600ml water samples were taken from each row of water intakes on the side of the constructed wetland device, and stored in 1L Brown sample bottles after 0.45 μ After filtration with M filter membrane, the pH, conductivity, ammonia nitrogen, nitrate nitrogen, inorganic phosphorus, na+, chlorpyrifos and TCP concentrations of water samples were measured. The influent water quality parameter is: chlorpyrifos concentration is 300 μ GL-1, the concentration of nitrogen and phosphorus is total nitrogen 4mgl-1 (ammonia nitrogen 3.0mgl-1; nitrate nitrogen 1.0mgl-1), inorganic phosphorus 0.4mgl-1, and the saline alkali level is PH9, hco3-800mgl-1.

4.2. Quantitative Analysis Method of Chlorpyrifos and TCP

Solid phase extraction was used to extract chlorpyrifos and TCP from water samples. Water oasis series (6cc/500mg) solid phase extraction column is selected as the extraction column. When in use, place the extraction column on the solid-phase extractor, activate and balance the extraction column with 5ml methanol (chromatographically pure) and 5ml ultra pure water (flow rate 1mlmin-1). Measure 500ml water sample, adjust pH3.5 with hydrochloric acid, and then load the sample. The flow rate of solid phase extraction should be controlled at 8 – 10mlmin-1. Until the volume of eluent is less than 1ml. Pass the concentrated solution through 0.22 μ M organic filter membrane, transfer it to the chromatographic injection bottle, and finally fix the volume to 1ml with methanol (chromatographic purity). The recovery rate of Chlorpyrifos in this method is 83.61 – 101.75% (n=6), and the recovery rate of TCP is 81.13 – 98.63% (n=6). The limit of quantitation was 2.0 – 4000ng and 0.02 – 2000ng. Mass spectrum parameters are shown in Table 3.

Table 3. Optimization parameters of mass spectrometry conditions for chlorpyrifos and TCP

Test object	Detection mode	Parent ion(da/z)	Daughter ion(da/z)	DP(V)	CE(V)	CXP(V)
chlorpyrifos	ESI(+)	349.9	197.9	50	35	3
		351.9	199.9	50	35	3
TCP	ESI(-)	195.7	36.1	-35	-40	-5
		197.7	36.1	-36	-36	-5

4.3. Substrate and Plant Screening for Reducing and Detoxifying Chlorpyrifos in Constructed Wetlands

The saturated adsorption capacity of vermiculite and Fe-C was significantly higher than that of the other four matrices. Gravel has the worst adsorption performance. The findings of the two groups are consistent with the comparison results of specific surface area and scanning electron microscope. The fitting of the first-order model and the second-order model of the adsorption kinetics of chlorpyrifos on the six substrates is shown in Figure 2.

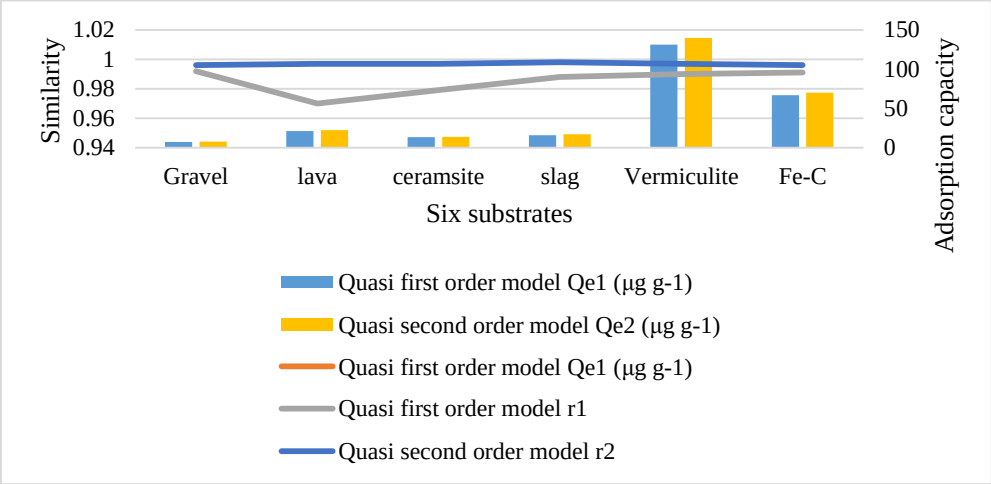

Figure 2. Saturated adsorption capacity and similarity of six substrates by kinetic fitting

The results showed that the six substrates fitted the two kinetic models well, and the fitting degree of the pseudo second-order model was higher ($r>0.99$). Degrading bacteria exist in contaminated soil, plants and microbial reactors for treating pollutants. The degrading bacteria can use chlorpyrifos or TCP as carbon source, phosphorus source and energy, and completely degrade the two pollutants through separate metabolism or co metabolism. Some degrading bacteria cannot use chlorpyrifos or TCP as the only carbon source, phosphorus source or energy source. The degradation of chlorpyrifos by microorganisms involves a variety of complex biochemical reactions, such as hydrolysis, ring opening, chain breaking, monolipid production, dephosphorization and dechlorination, which eventually convert it into inorganic phosphate. Specifically, the hydrolysis of chlorpyrifos to TCP and diethyl thiophosphate is mainly realized by aerobic bacteria. The hydrolysate TCP needs alkylation to produce 3, 5, 6-trichloro-2-methylpyridine (TMP) or gradual dechlorination to dihydro-2-pyridone and tetrahydro-2-pyridone. Tetrahydro-2-pyridinone forms maleimide semialdehyde and maleic acid through ring opening reaction, and is finally degraded into water, carbon dioxide and ammonium ions. Dihydro-2-pyridone has another degradation pathway, that is, it is oxidized to hydroxypyridines with different hydroxyl positions, which will eventually be oxidized to maleic acid. Degrading bacteria with different functions constantly use a variety of complex intermediates produced in the degradation process as carbon and phosphorus sources for their own metabolism, promoting the degradation downward, and finally realizing the complete degradation of chlorpyrifos.

5. Conclusion

With the help of BDT, this paper analyzes the EBE of chlorpyrifos, explores and reveals for the

first time the degradation law, influencing factors and degradation mechanism of chlorpyrifos and its toxic hydrolysate TCP in the treatment of paddy field retrogradation in the saline alkali environment in the constructed wetland, analyzes and discusses the change law of microbial community structure in the constructed wetland under the saline alkali condition, and greatly improves the operation efficiency of the constructed wetland, And provide important technical support for the treatment of chlorpyrifos pollution in constructed wetlands. However, there are also shortcomings. This study obtains the best application time of soil conditioner through pot experiment. The best time and method of field application and the real role of each component in soil conditioner need to be further studied.

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Data Availability

Data sharing is not applicable to this article as no new data were created or analysed in this study.

Conflict of Interest

The author states that this article has no conflict of interest.

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