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Research on World Agricultural Economy
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Study on the Degradation Effect of Plant-Derived Active Ingredients on Organophosphorus Pesticides

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ARTICLE INFO

Article history
Received: 19 March 2020
Revised: 26 March 2020
Accepted: 24 April 2020
Published Online: 30 April 2020

Keywords:
Organophosphorus pesticides
Plant-Derived active ingredients
Pesticide degradation
Chinese medicine agriculture
Soil remediation

ABSTRACT

In order to explore new ways and methods for the degradation of organophosphorus pesticides, the degradation effects of plant-derived active ingredients on three organophosphorus pesticides were studied. Rhubarb, Pittosporum bark, Hibiscus bark, and Chinese gall in 9:4:3:2 parts were mixed by mass, ground and soaked in water, and the concentration changes of organic phosphorus pesticide before and after the test were compared through GC-MS quantitative detection method and rapid pesticide residue measurement method to clarify its degradation effect on organophosphorus pesticides. The results showed that the degradation rates of chlorpyrifos and parathion were 93.2% and 92.9% respectively in the extract within 2 minutes; the degradation rate of dichlorvos was 66.67% in the extract within 17 hours, and the degradation rate of chlorpyrifos was 48.69% within 11 hours. This study shows that the extracts of rhubarb, sea tongs bark, hibiscus bark, and gallnut have significant degradation effects on chlorpyrifos, parathion, dichlorvos and other organophosphorus pesticides.

1. Introduction

As an important means of agricultural production, chemical pesticides play an important role in curbing plant diseases, insect pests and weeds and reducing agricultural production losses. However, long-term excessive and unreasonable application of chemical pesticides has also caused serious impacts on human production, life and the ecological environment. The resulting quality and safety of agricultural products, decline in the quality of cultivated land, agricultural non-point source pollution, sudden increase in malignant diseases and environmental hormone problems have caused pay close attention to [1-5].

Organophosphorus pesticides are widely used, especially in the field of pesticides, accounting for more than 70% of the total pesticides [6]. The utilization rate of pesticides applied in farmland is 38.8% [7]. Most of the pesticides diffuse into the environment, enter the human body through the water vapor cycle and the food chain, endangering health, and because a large amount of pesticides remain in the soil, they become agricultural non-point source pollution. One of the important factors [8-9].

Pesticide degradation can be divided into physical methods, chemical methods and biological methods. However, due to factors such as application conditions, use costs, and technological maturity, large-scale promotion and application have not been carried out [8,10],

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DOI: http://dx.doi.org/10.36956/rwae.v1i1.162

especially in agricultural production. There is no precedent. At present, biological methods are considered to be an ideal method to reduce pesticide residues due to their significant effect of degrading pesticide residues and low environmental impact. In particular, the screening and application technology research and development of pesticide degrading bacteria have achieved significant research results [8,10- 12], but these technologies are still in the laboratory development stage.

Based on the principle of enzymatic degradation of pesticides, this research has developed a unique approach, based on the theory of traditional Chinese medicine, prescriptions and years of application practice, to screen and develop plant-derived active ingredients that have rapid and efficient degradation effects on organophosphorus pesticides. After indoor testing and field verification, The effect is remarkable, realizing the cross-border integration of modern agriculture and traditional Chinese medicine, and providing a good technical path and basic data support for exploring and applying Chinese medicine concepts to promote the harmonious development of "production, life, and ecology". It has important scientific research and application value.

2. Materials and Methods

2.1 Preparation of Plant-Derived Active Ingredients

(1) Mix and crush the raw materials of traditional Chinese medicine such as 180 g of rhubarb, 80 g of sea tongs bark, 60 g of hibiscus bark, 40 g of gallnut, and sieving through 200 mesh.

(2) Put the powder in a stainless steel container, add 2000 g of water, and heat to 100°C for 30 min.

(3) Under the condition of full contact with air, cool down naturally (not lower than 5℃ , and let stand for 48 h.

(4) Filter out the drug solution and dilute it with 5,000 g of clean water (the water temperature is room temperature).

2.2 Detection of Degradation Effect

2.2.1 Gas Chromatography-Mass Spectrometry (GC-MS Method)

March 10, 2018, in the laboratory of the Institute of Agricultural Quality Standards and Testing Technology, Shandong Academy of Agricultural Sciences, using Agilent 7890A gas chromatography-mass spectrometry, the analytical method quantitatively detects the rapid degradation effect of plant-derived components on organophosphorus pesticides (chlorpyrifos, parathion) [13-15].

(1) Prepare a 1 μg/mL mixed standard sample of chlorpyrifos and parathion.

(2) Take 1 mL of the standard sample and place it in a 20 mL pointed glass test tube, then add 1 mL of plant-derived active ingredient solution, and vortex for 1 min.

(3) Add 5 mL acetonitrile and 1 g NaCl, and vortex for 1 min.

(4) Centrifuge at 3,000 r/min for 2 min, take it out and place it in a test tube rack.

(5) Take 1 mL of the supernatant solution after centrifugation in a glass test tube, blow with nitrogen in a 40°C water bath (until it blows dry), and add 2 mL of acetone to make the volume constant.

(6) On-machine (Agilent 7890A) detection, the injection volume is 2 μL. Set 3 repeats.

2.2.2 Pesticide Residue Rapid Detection Method

With leek and chrysanthemum chrysanthemum as the test objects, the NC-800 6-channel intelligent pesticide residue rapid detector is used to detect the degradation effect of plant-derived components on organophosphorus pesticides (dichlorvos, chlorpyrifos).

(1) Leek-Dichlorvos

Test conditions: On May 6, 2018, the experiment was carried out at the leek base in Caiyuan Village, Zhanglu Town, Xin County, Liaocheng City, Shandong Province. The base was planted with leek for 2 years, covering an area of 0.33 hm^2, with a variety of Hanzhong winter leek and a plant height of 15-20 cm., The border is east-west, 25 m long, 2 m wide, and leeks are planted horizontally from north to south. The experiment was carried out on May 6, with real-time temperature 22-25°C, wind force 2-3; 16-17 hours later, sampling and testing.

Experimental procedure:

① From 15:00 to 16:00 on May 6th, select a leek plot of 20 m^2, and spray the leek plants and the ground between rows with 775 mg/L dichlorvos 2,000 mL evenly;

② Randomly select 10 m^2 of them for test treatment, immediately dilute with plant-derived active ingredients and room temperature water by 4 times, take 400 mL and spray evenly on the leek plants and the ground between rows; the other 10 m^2 is used as control I;

③ The same planting conditions of adjacent plots, without spraying dichlorvos and plant-derived active ingredients of leek in the leek border as control II;

④ From 9 am to 10 am on May 7th, use the 5-point mixed sampling method to collect 1 kg of leek samples in the experimental treatment area, control I area, and control II area, and put them into plastic sealed bags; at 11 o'clock, use the rapid test method Detection of dichlorvos residues

 DOI: http://dx.doi.org/10.36956/rwae.v1i1.162

in leek samples. Let 2 repeats.

(2) Garland chrysanthemum-Chlorpyrifos

Test conditions: On May 9, 2018, the experiment was carried out at the Chrysanthemum chrysanthemum export base in Shiziyuan Town, Xin County, Liaocheng City, Shandong Province. The base planted Chrysanthemum chrysanthemum 2.67 hm^2, the growth period was 45 days, the plant height was 30-50 cm, and the border was east-west., 80 m long and 5 m wide, Chrysanthemum vulgare was planted vertically from east to west. It was harvested on May 10th as planned. Due to accidental spraying during flight, chlorpyrifos exceeded the standard by 16 times. Emergency treatment should be taken upon request. The test was carried out at night on May 9. The real-time temperature is 16-18°C, without wind; it will be detected after 10-11 hours.

(3) Experimental Procedure

① Set up 3 communities in Qitou and Qizhong, each of which is about 20 m^2;

② At 22:00 on the evening of May 9th, dilute 4 times with plant-derived active ingredients and room temperature water, and spray 1,000 mL evenly on Chrysanthemum chrysanthemum plants in the test plot;

③ Take the adjacent section on the west side of each plot as a control;

④ At 8:00 on May 10, use the 5-point mixed sampling method to collect 500 g of Chrysanthemum chrysanthemum (middle and upper part) samples in the experimental treatment area and the control area, and put them into plastic bags; at 9:00, use the rapid measurement method Detection of chlorpyrifos residues in Chrysanthemum chrysanthemum samples.

3. Results and Analysis

3.1 The Degradation Effect of Plant-Derived Active Ingredients on Chlorpyrifos and Parathion

T-test analysis was performed by SPSS, and the results are shown in Table 1. Taking the test concentration of the standard sample as the initial concentration (ck), after mixing the chlorpyrifos and parathion standard sample solution with the plant-derived active ingredients for 2 minutes, the degradation rate of chlorpyrifos is 93.6%, the degradation rate of parathion is 92.9%, and the degradation rate of chlorpyrifos and parathion is 92.9%. Phosphorus concentration decreased significantly ($P<0.01$), and the degradation effect was significant; the degradation effects of chlorpyrifos and parathion were equivalent, and the difference did not reach a significant level ($t=2.347$, $P=0.079$)

Table 1. Degradation effects of plant-derived active ingredients on chlorpyrifos and parathion (indoor)

species	Test repeat	Concentration/(μg/mL)	Mean/(μg/mL)±standard deviation	Degradation rate/%
samples	1	0.9635	0.9653±0.0047A	
	2	0.9706		
	3	0.9618		
Chlorpyrifos	1	0.0983	0.0962±0.0032B	93.6
	2	0.0978		
	3	0.0925		
Parathion	1	0.1039	0.1031±0.0039B	92.9
	2	0.1065		
	3	0.0988		

3.2 The Field Degradation Effect of Plant-Derived Active Ingredients on Dichlorvos and Chlorpyrifos

It can be seen from Table 2 that the control II leek sample is 1.01%, the control leek sample is 19.00%, and the experimental treatment leek sample is 6.11%. The results show that the plant-derived active ingredients have a significant effect on dichlorvos under natural conditions in the field, within 17 hours the degradation rate reached 66.67%.

Table 2. Degradation effect of plant-derived active ingredients on dichlorvos (field)

species	Test repeat	Inhibition rate/%	Mean/%	Degradation rate/%
CKI	1	0.48	1.01	
	2	1.54		
CKII	1	20.41	19.00	
	2	17.59		
Test treatment	1	11.89	6.11	66.67
	2	0.32		

It can be seen from Table 3 that after emergency treatment, the concentration of chlorpyrifos on Chrysanthemum chrysanthemum was significantly reduced. After 10 to 11 hours, the degradation rate exceeded 48.7%.

Because 100% of the pesticide residue concentration has exceeded the instrument range, it is impossible to obtain a relatively accurate value. Although it is impossible to perform an accurate significance test and analysis, it is inferred from the data distribution that there should be a significant difference between the two.

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Table 3. Degradation effect of plant-derived active ingredients on chlorpyrifos (field)

species	Test repeat	Inhibition rate/%	Mean/%	Degradation rate/%
Test treatment	1	52.16	46.83	48.69
	2	46.41		
	3	41.92		
CK	1	73.77	91.26	
	2	100		
	3	100		

4. Conclusion

This study shows that the extract obtained from rhubarb, sea tongs bark, hibiscus bark, and Chinese gall according to the fixed mass ratio has a significant degradation effect on chlorpyrifos, parathion, dichlorvos and other organophosphorus pesticides, and the degradation rate is extremely fast. Especially when organophosphorus pesticides are fully mixed, the degradation efficiency exceeds 90% in 2 minutes.

In view of the rapid and significant degradation effect of this plant-derived component on organophosphorus pesticides, it has broad application prospects in farmland soil remediation, elimination of pesticide residues in fresh fruits and vegetables, production of organophosphorus toxic compounds and emergency treatment of leakage, clinical poisoning rescue, etc.

5. Discussion

Organophosphorus pesticides are widely used. With the increase of pest resistance, the amount and frequency of application continue to increase, which has a greater impact on the environment and poses an acute risk of poisoning to the human body [16]. Pesticide biodegradation technology is a hotspot of global technology research in the field in recent years [17-18]. Obligate and broad-spectrum microbial species with obvious enzymatic degradation of organophosphorus have also been screened from soil and water, including bacteria, Fungi, actinomycetes and algae. Among them, the research on bacteria is the most in-depth [17], such as the more representative Pseudomonas in bacteria, which has good degradation of malathion, dichlorvos, phorate, and methyl parathion effect. Liu Yuhuan et al. [19] conducted a detailed study on methamidophos-degrading fungi, Wang Baihui et al. reviewed the mechanism of soil enzyme degradation of pesticides by microorganisms [20], Zhao Renbang et al. [21] degraded methylamine by Penicillium oxalicum ZHJ6 The application method of phosphorus was studied. Biazyme is a fermentation product produced by microorganisms selected from the soil. As a result of the "863 Program" project, it has been exclusively produced by Beijing Sengenbia Biotechnology Co., Ltd. since 2012. It is used for civilian purposes. It is against dichlorvos, chlorpyrifos, and Mala. The average degradation effect of thion, fenthion, and methyl parathion reaches 70%, and a number of patented technologies such as "methyl parathion hydrolase mutant with improved enzymatic performance and its application" have been developed [22].

Up to now, there are few research literatures on the application of pure plant-derived ingredients to degrade organophosphorus pesticides and other types of pesticides. As a brand-new concept in the development of modern agriculture, TCM agriculture is of great significance to promoting the sustainable development of agriculture [23]. In traditional Chinese medicine, there are many formulas and cases of applying Chinese medicine to detoxify. Edible proteases are extracted from pineapple, papaya and other fruits to develop detergents that can degrade vegetable pesticide residues, such as methamidophos, omethoate, dichlorvos, and chlorpyrifos. The degradation rate of other pesticides reaches more than 90% [24]; different concentrations of tea tree oil and water-soluble tea tree oil are used to soak and clean the cowpea sprayed with pesticides. The results show that 0.8% of water-soluble tea oil has the best effect in removing pesticide residues. Excellent, the pesticide removal rates for triazophos, malathion, and chlorpyrifos are 82.79%, 94.54%, and 84.58%, respectively [25].

Although many kinds of microorganisms and plant extracts that degrade pesticides have been screened out, it can be seen from the existing literature that these research results have not yet been applied on a large scale. As the most stable and effective organophosphorus pesticide degradation enzyme at present, Biazyme is affected by factors such as raw materials, production methods, yield and cost, and its application range is limited. The plant-derived ingredients in the traditional Chinese medicine prescriptions used in this study have a wide range of sources and have equivalent functional alternatives, which can be mass-produced at low cost. After 11 years of field application, the effect is significant and stable. For the first time in this study, the principles and techniques of detoxification of traditional Chinese medicine were applied to the degradation of chemical pesticides. The prescriptions of traditional Chinese medicines that can efficiently degrade organophosphorus pesticides were screened out, and more systematic production processes, products and supporting application technologies were developed, which can be applied to farmland. Degradation of pesticide residues in contaminated soil, degradation of pesticide residues on agricultural products, water environment disinfection, and emergency treatment of organophosphorus pesticides (factories) leakage. The field application initially shows that this plant-derived ingredient has a certain degradation effect on carbamates, sulfonylurea pesticides and herbicides, and scientific experiments are needed to prove it; at the same time, the mechanism of action of this ingredient needs further research.

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  DOI: http://dx.doi.org/10.36956/rwae.v1i1.162