

RESEARCH ARTICLE

Analysis of the residual digestion rules of arylpyridine carboxylate herbicides based on Q-TOF technology

Xian Wu¹, Yanwei Liu¹, Hao Zhang², Hongxin Li¹, Zongzhi Lu^{1,*}

¹Institute of Plant Protection, Jilin Academy of Agricultural Sciences (Northeast Agricultural Research Center of China), Gongzhuling, Jilin, China. ²College of Plant Protection, Jilin Agricultural University, Changchun, Jilin, China.

Received: August 21, 2024; accepted: December 10, 2024.

With the modernization of agriculture, herbicides are widely used in rice fields to increase crop yields and reduce competition from weeds for nutrients. However, using herbicides also brings hidden dangers of environmental pollution, especially the problem of residues in soil and water bodies. As a new type of herbicide, the degradation behavior of arylpyridine carboxylic acid ester herbicides in the environment is still unclear. Therefore, to study the degradation process of these herbicides in paddy field environments, an analysis was conducted on the degradation law of residues of arylpyridine carboxylic acid ester herbicides in paddy field environments. This study used high-resolution mass spectrometry Quadropole-Time-of-Flight (Q-TOF) technology to qualitatively and quantitatively analyze the arylpyridine carboxylic acid ester herbicides and their residues in the samples. In addition, the study utilized Q-TOF technology combined with gas chromatography-mass spectrometry and liquid chromatography-mass spectrometry techniques to track metabolic changes and infer herbicide degradation pathways. The results showed that two new differential compounds with mass-to-charge ratios of 427.023 and 405.082 were generated in soil degradation experiments, which were speculated to be degradation products of fluoropyridine esters. Two degradation products, chlorofluoropyridine ester and chlorofluoropyridine acid, were identified in the hydrolysis metabolite experiment. In the roots of rice, the highest concentration of chloropyridine ester reached 399.37 $\mu\text{g}/\text{kg}$ with a degradation rate of up to 99.98%. In the soil, the highest concentration of chlorofluoropyridine acid reached 282.56 $\mu\text{g}/\text{kg}$ with a degradation rate of up to 99.99%. The analysis of the degradation law of arylpyridine carboxylic acid ester herbicide residues based on the Q-TOF technology revealed the main degradation pathways and degradation products of this type of herbicide. This research provided important support and reference for the pollution monitoring and risk assessment of arylpyridine carboxylic acid ester herbicides in paddy field environments.

Keywords: herbicide; arylpyridine formate ester; Q-TOF technology; rice; residue digestion.

*Corresponding author: Zongzhi Lu, Institute of Plant Protection, Jilin Academy of Agricultural Sciences (Northeast Agricultural Research Center of China), Gongzhuling 136100, Jilin, China. Email: wuxianjass@163.com.

Introduction

With the rapid development of industrialization, the application of chemical technology in rice cultivation has become increasingly common [1].

Due to the favorable environment for weed growth in paddy fields, arylpyridine carboxylic acid ester herbicides (ACAEH) have been widely used for weed control, leading to serious weed damage problems. The degradation dynamics of

herbicides in soil are not only related to their physical and chemical properties but also related to soil type, pH value, humidity, organic matter content, and microbial community composition in the soil [2]. However, improper use of chemical herbicides can easily lead to phytotoxicity, resulting in problems such as rice plant dwarfism, abnormal root growth, and reduced tillering [3]. With the growth of population and rapid social development, the sharp increase in demand for agricultural products has led to increasingly serious problems of herbicide residues in the environment. However, the use of herbicides has brought challenges such as environmental pollution and health risks while increasing crop yields.

ACAHEH mainly includes fluoropyridine ester (FE) and chlorofluoropyridine ester (CFE) [4]. Among them, FE is a new hormone type ACAHEH developed, which can simulate the action of plant growth hormone on plant somatic cells, accelerating cell division and over-consumption of nutrients until death [5]. CFE is the second ACAHEH developed after FE with the same mechanism of action as FE but different target sites [6]. Although the application of chemical herbicides effectively reduces agricultural labor intensity and improves agricultural production level and efficiency, the selective effect of chemical herbicides is influenced by multiple factors and has high technical requirements [7]. Studies have shown that residues of ACAHEH may enter ecosystems through pathways such as soil and water, causing adverse effects on the environment and biological health. The accumulation and diffusion of these residues in the soil may affect crop growth, reduce soil fertility, and cause a series of problems such as water pollution. Therefore, analyzing the degradation patterns of these herbicides in the environment is of great significance for assessing their ecological risks.

In recent years, Quadropole-Time-of-Flight (Q-TOF) technology has been widely applied in pesticide residue detection and has achieved significant results. This technology has the

advantages of high resolution, high sensitivity, high precision, and fast analysis. On the other hand, the disadvantages include high environmental requirements and relatively complex operation and maintenance. Singh *et al.* found that the dispersion of herbicides in the environment could lead to a decrease in crop yields, a decrease in agricultural product quality, a decline in soil fertility, and water pollution, thereby threatening the lives of humans and animals [8]. Escolano *et al.* believed that the process of bridging aromatic feedstocks with aliphatic ring structures when applied to pyridine, quinoline, and isoquinoline would form partially or completely saturated nitrogen heterocycles, and these transformations would make the de-aromatization process thermodynamically unfavorable [9]. In addition, these chemicals might enter ecosystems through pathways such as soil and water bodies, which could have adverse effects on the environment and human health. Therefore, analyzing the degradation patterns of herbicides in the environment is of great significance for assessing their ecological risks. Traditional herbicide residue analysis methods often rely on gas chromatography-mass spectrometry (GC-MS) or liquid chromatography-MS (LC-MS) techniques [10]. Although these technologies have significant advantages in detection sensitivity and specificity, they also have disadvantages such as complex operation, high cost, and long analysis time when facing multiple metabolites in complex environmental samples [11, 12]. Q-TOF technology has provided an effective means for the detection of trace compounds in complex matrix samples due to its high-resolution and high-precision qualitative and quantitative capabilities. Chang *et al.* used Q-TOF technology to quickly determine 482 pesticide residues in the traditional Chinese medicine *Codonopsis pilosula*, demonstrating the effectiveness of Q-TOF technology in chemical residue detection [13]. Liu *et al.* also used Q-TOF technology for high-throughput screening and quantitative analysis of pesticides in chili peppers, effectively detecting pesticide residues in chili peppers and once again confirmed the role of this technology

in chemical residue detection [14].

This study innovatively utilized Q-TOF technology to analyze and investigate the digestion law of ACAEH residues to explore the main factors that affected their digestion rate.

Materials and methods

Sample resources

The soil samples were collected from the Fuyang Base of the China Rice Research Institute (30.0490 N, 119.9606 E) (Hangzhou, Zhejiang, China) and included paddy soil, brown soil, red soil, and black soil. After removing large impurities, the soil samples were stored in labeled sample containers at room temperature.

Soil physical and chemical properties

The study used various methods to determine the physical and chemical properties of different tested soils. A HI2211 pH meter (Hanna Instruments, Woonsocket, Rhode Island, USA) was applied to measure soil pH using a standard glass electrode. The Walkley Black method was used to determine soil organic matter contents using potassium dichromate (Hach, Loveland, CO, USA). The soil cation exchange capacity (CEC) was measured using the ammonium acetate method. A Malvern Panalytical Mastersizer 3000 laser diffraction particle size analyzer (Malvern Panalytical Ltd., Malvern, Worcestershire, UK) was used to analyze the particle size distribution.

Soil degradation

The impurities of the collected soil samples were removed followed by air drying the samples and exposing it to sunlight. The soil was then screened through a 16-mesh sieve and stored in bottles with 200 g soil in each bottle. For each experimental group, 6 soil sample bottles were used to test the soil degradation of CFE (Dr. Ehrenstorfer GmbH, Augsburg, Bavaria, Germany). Briefly, 40% ultrapure water was added to each sample soil bottle and then incubated at room temperature for 14 days before adding the CFE at 1.0 mg/L concentration

to 3 soil culture bottles, stirring and mixing thoroughly. The remaining 3 soil bottles were served as blank control group. All 6 experimental soil sample bottles were placed at 25°C for 14 days before sampling analysis were conducted to evaluate the degradation status in the soil using Agilent 6460 liquid chromatography tandem mass spectrometer (LCTMS) with EC-C18 chromatographic column (Agilent Technologies, Santa Clara, California, USA) following manufacturer's instructions.

Soil hydrolysis

The buffer solutions were prepared according to industry standards for pesticide physicochemical property determination [15, 16]. Luria Bertani (LB) media (Sigma Aldrich, St. Louis, MO, USA) were prepared at pH 4.0 by mixing with 0.1 mol/L potassium citrate and 0.1 mol/L sodium hydroxide, pH 7.0 by mixing 0.1 mol/L potassium dihydrogen phosphate and 0.1 mol/L sodium hydroxide, and pH 9.0 by mixing 0.1 mol/L boric acid and 0.1 mol/L sodium hydroxide. The medium solutions with different pH values were sterilized, mixed with CFE, diluted to 400 mL at a concentration of 1.0 mg/L, and then placed in a 50°C incubator for dark incubation. Three additional solutions of the same volume without herbicide were taken as blank control groups and incubated under the same conditions.

Experiment on adding recycling rate

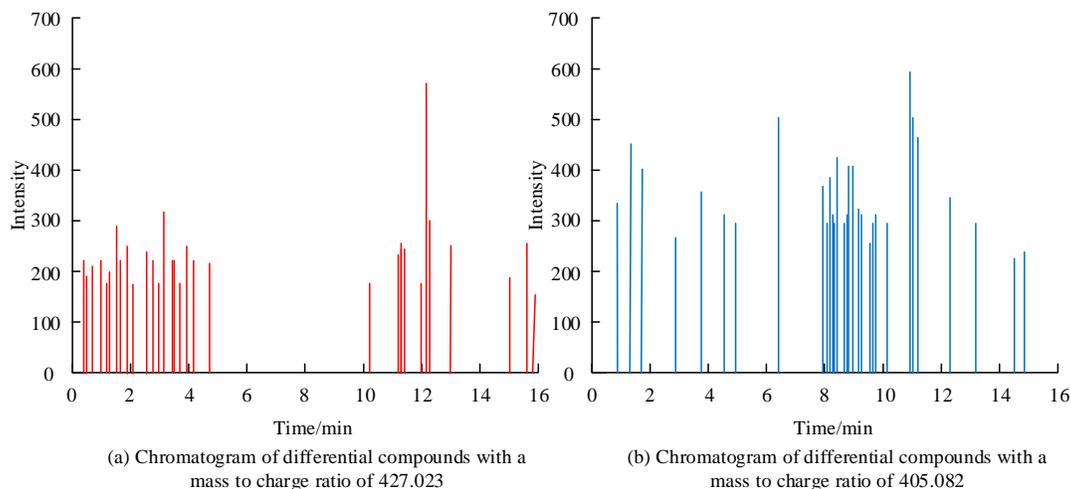
To analyze the recovery rate of herbicides added to soil and water, 3 different levels of herbicides including 0.05 mg/kg, 0.20 mg/kg, and 10 mg/kg were added to the soil culture bottle. The same amount of herbicide was also added to the aqueous solution. The experiments were repeated 5 times for each level of addition. The relative standard deviation was calculated.

Data analysis

The data were recorded and processed using Microsoft Excel 2003 (Microsoft, Redmond, WA, USA). SPSS V26.0 (IBM, Armonk, NY, USA) was employed for statistical analysis of independent samples and variance with a significance level at $P < 0.05$.

Table 1. The physicochemical properties of different tested soils.

Physicochemical properties	Paddy soil	Black soil	Red soil	Brown soil
PH	6.2	7.4	4.6	6.5
Total nitrogen (mg/kg)	1.3	1.5	0.8	1.1
Organic carbon (g/kg)	37.1	33.8	8.3	13.7
Organic matter (mg/kg)	298.5	290.4	71.4	117.8
Effective phosphorus (mg/kg)	12.4	15.0	8.1	10.5
Quick acting potassium (mg/kg)	68.3	72.1	50.0	59.4

**Figure 1.** Chromatograms of soil degradation metabolites of ACAEH.

Results

The physical and chemical properties of different tested soils

The physical and chemical properties of different tested soils were determined and shown in Table 1.

Analysis of ACAEH soil degradation metabolites

The ACAEH soil degradation metabolites were analyzed. Two new differential compounds were generated in the soil degradation experiment including one with a mass-to-charge ratio (MCR) of 427.023 and the peak time of 12.40 minutes, which was completely consistent with the peak time of sodium ions with an MCR of 405.81 (Figure 1a), and another one with an MCR of 405.082 and the peak time of 11.00 minutes, which perfectly matched the peak time of chloride ions with an MCR of 405.81 (Figure 1b).

The results suggested that the compound with an MCR of 427.023 was a metabolite of ACAEH degraded in soil, while the differential compound with a new MCR of 405.082 was also a metabolite of ACAEH after degradation in soil.

Analysis of ACAEH hydrolysis metabolites

The ACAEH hydrolysis metabolites demonstrated a new differential compound with an MCR of 439.042 and a peak time of 11.86 minutes (Figure 2a). According to the comparison to the mass spectrometry library, the compound was identified as CFE with a chemical formula of $C_{20}H_{14}Cl_2F_2N_2O_3$, a theoretical mass number of 438.035, and a mass deviation of 1.46 ppm. In addition, there was another new differential compound with an MCR of 348.995 and a peak time of 9.54 minutes, which was identified as chlorofluoropyridine acid (CFA) with a chemical formula of $C_{13}H_8Cl_2F_2N_2O_3$, a theoretical mass

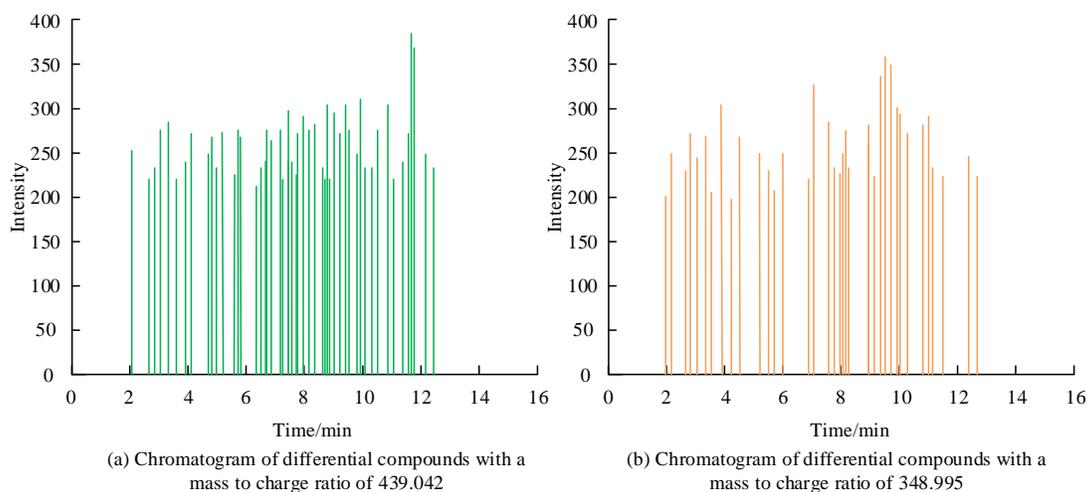


Figure 2. Chromatograms of hydrolysis metabolites of ACAEH.

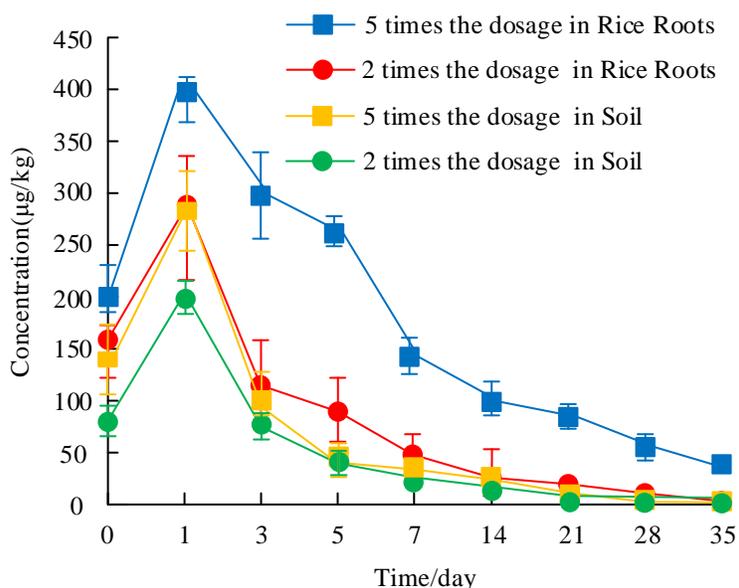


Figure 3. Degradation dynamics of CFE.

number of 347.988, and a mass deviation of 0.80 ppm (Figure 2b). The peak times of CFA and CFE were completely consistent with CFE standard. Therefore, they both were hydrolysis metabolites of ACAEH.

Degradation dynamics of CFE

The results of degradation dynamics of CFE showed that, under different doses of ACAEH, the concentration curves of CFE in rice roots and soil were consistency, reaching their maximum

values after one day. After doubling the dose of ACAEH to the roots of rice, the concentration of CFE reached its maximum value of 284.46 µg/kg one day later, while, under the application of five times the dose, the maximum concentration was 399.37 µg/kg. Afterwards, the CFE concentration in the roots of rice gradually decreased and reached its minimum value after 35 days. At the double dose, the minimum concentration of CFE was 0.04 µg/kg, and the degradation rate reached 99.98%. At 5 times the dose, the

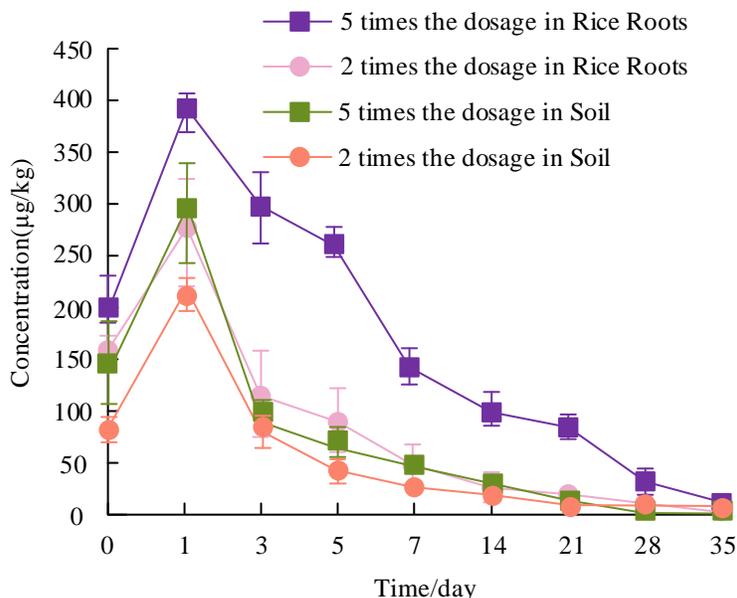


Figure 4. Degradation dynamics of CFA.

minimum concentration was 42.17 $\mu\text{g}/\text{kg}$, and the degradation rate reached 89.44%. After applying double dose of ACAEH in soil, the concentration of CFE reached its maximum value of 214.83 $\mu\text{g}/\text{kg}$ one day later, while, under the application of five times the dose, the maximum concentration was 282.56 $\mu\text{g}/\text{kg}$. After 35 days, the CFE concentration in the soil approached zero at the double dose treatment, approximately 0.01 $\mu\text{g}/\text{kg}$, and the degradation rate reached 99.99%. The minimum concentration at 5 times the dose was also close to 0, and the degradation rate reached 99.99% (Figure 3). Overall, CFE demonstrated a better degradation effect in soil.

Degradation dynamics of CFA

The degradation dynamics of CFA showed that the concentration variation curves of CFA in rice roots and soil under different doses of herbicides were consistent, reaching their maximum values within one day after application. After applying 2- and 5-times doses of ACAEH to the roots of rice, the concentration of CFA reached its maximum values of 278.67 $\mu\text{g}/\text{kg}$ and 483.25 $\mu\text{g}/\text{kg}$, respectively, after one day. Afterwards, the CFA concentration in the roots of rice gradually

decreased and reached its minimum value after 35 days. At doses of 2 and 5 times, the minimum concentrations of CFA were 0.04 $\mu\text{g}/\text{kg}$ and 11.23 $\mu\text{g}/\text{kg}$ with the degradation rates of 99.98% and 97.67%, respectively. After applying 2- and 5-times doses of ACAEH in soil, the concentration of CFA reached its maximum values of 209.65 $\mu\text{g}/\text{kg}$ and 287.93 $\mu\text{g}/\text{kg}$, respectively after one day. After 35 days, the CFA concentration in the soil approached zero at the double dose, approximately 0.01 $\mu\text{g}/\text{kg}$, and the degradation rate reached 99.99%, while the minimum concentration at the 5 times of the dose was also close to 0 with the degradation rate reaching 99.99% (Figure 4). CFA showed a better degradation effect in soil.

Residual ACAEH in soil

The residual levels of CFE and CFA in soil after 35 days of treatment were shown in Figure 5. The degradation products of CFE had a molecular unsaturation characteristic of 1,552. The compounds of DBE still existed in the soil. Therefore, CFE and its metabolite CFA had a relatively fast degradation rate in soil, but their degradation products were 1,552. DBE might have a longer environmental residual time.

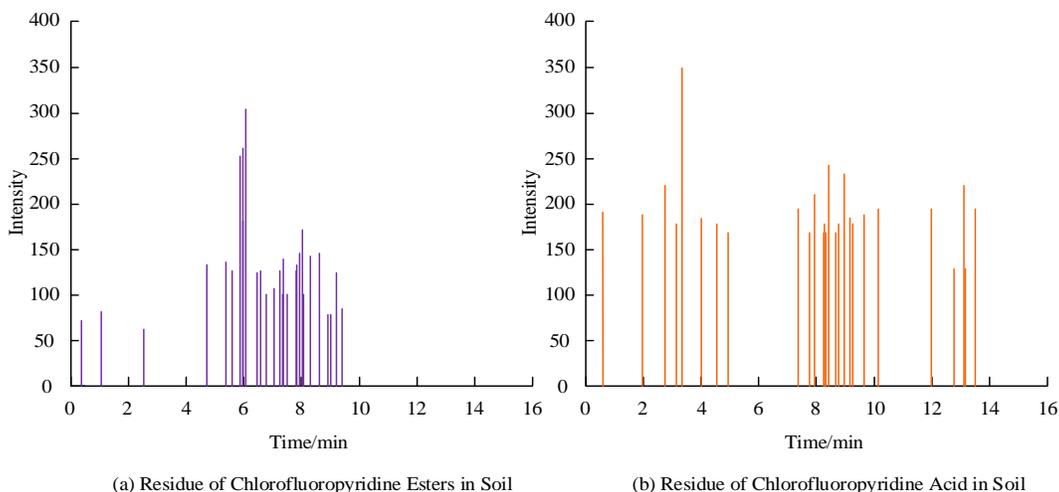


Figure 5. Residues of ACAEH in soil.

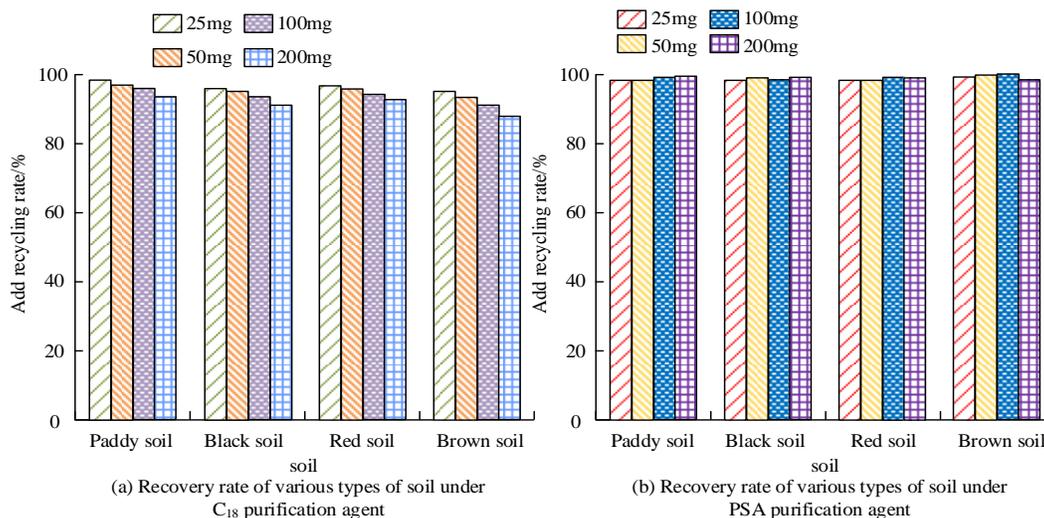


Figure 6. The effect of different purifying agents on the residues of ACAEH.

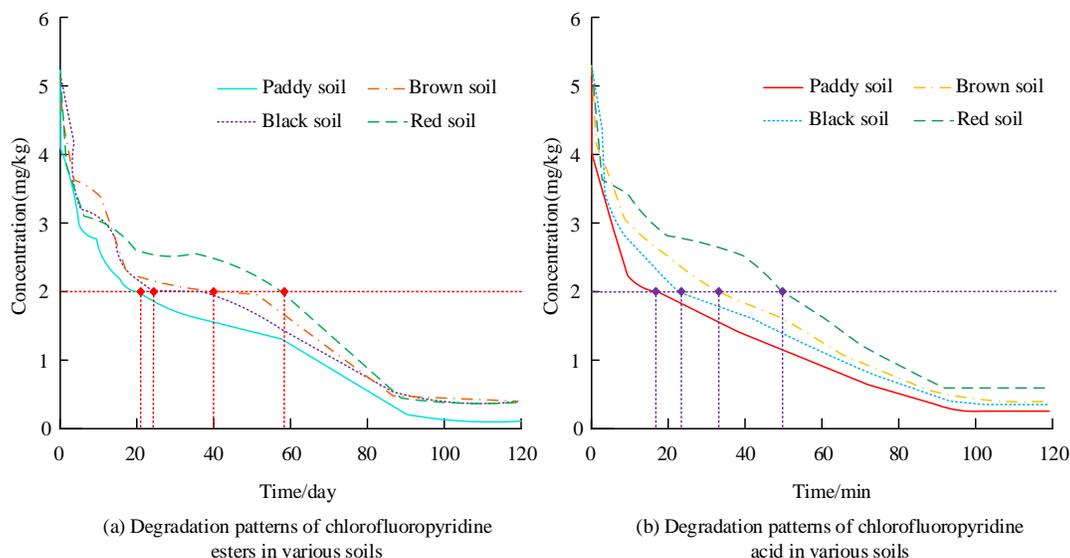
The effect of different purifying agents on ACAEH residues

The effects of different purifying agents on ACAEH residues demonstrated that, under different doses of C₁₈ purification agent, the recovery rates of various soil samples were similar and slightly decreased with the increase of C₁₈ purification agent dosage. Among them, the recovery rates of rice soil with 25, 50, 100, and 200 mg C₁₈ purification agents were 98.09, 96.54, 96.00, and 93.98%, respectively (Figure 6a). Under different doses of PSA purification

agent, the recovery rate of soil samples showed high stability. The recovery rates of rice soil with 25, 50, 100, and 200 mg PSA purifiers were 98.10, 98.10, 99.24, and 99.25%, respectively (Figure 6b). The recovery rate of PSA purification agent being added at different doses was higher than 98% and exhibited high stability. The recovery rate of C₁₈ purification agent decreased with the increase of dosage. Therefore, considering all factors, using PSA purification agent was more optimal.

Table 2. The effect of different extractants on the residues of ACAEH.

Extraction agent	Recovery rate of adding different soils (%)			
	Paddy soil	Black soil	Red soil	Brown soil
Pure acetonitrile	76.00	74.02	80.05	78.28
0.1% FAA	88.03	83.65	86.60	85.65
0.5% FAA	91.11	90.97	90.15	91.76
1% FAA	92.04	91.28	91.97	92.53
2% FAA	93.16	92.36	92.34	92.62

**Figure 7.** Degradation patterns of ACAEH residues in various soils.

The effect of different extractants on ACAEH residues

The effects of different extractants on the residues of ACAEH were shown in Table 2. When 0.5% formic acid acetonitrile (FAA) was used as an extraction agent, it significantly improved the recovery rate of herbicide residues in soil. When the extraction agent was 0.5% FAA, the recovery rate of various soils exceeded 90% including 91.11% for paddy soil, 90.97% for black soil, 90.15% for red soil, and 91.76% for brown soil. In addition, concentrations of FAA exceeding 0.5% had little effect on the recovery rate. Therefore, considering all factors, 0.5% FAA was determined as the optimal extraction agent.

Degradation patterns of ACAEH residues in various soils

The degradation curve of CFE gradually decreased over time. Among them, paddy soil had the fastest degradation rate with herbicide concentration dropping to 2.0 mg/kg after 21 days followed by black soil with a concentration dropping to 2.0 mg/kg at 24 days. At 40 and 58 days, the concentrations of herbicides in brown soil and red soil decreased to 2.0 mg/kg (Figure 7a). The degradation curve of CFA was consistent with the trend of CFE, and the degradation rates were in the order of paddy soil > black soil > brown soil > red soil. The times required for each soil type to degrade to 2.0 mg/kg were 18, 22, 32, and 49 for paddy, black, brown, and red soils (Figure 7b). Overall, the degradation efficiencies of paddy soil and black soil were relatively high, while the degradation rates of brown soil and red soil were relatively slow.

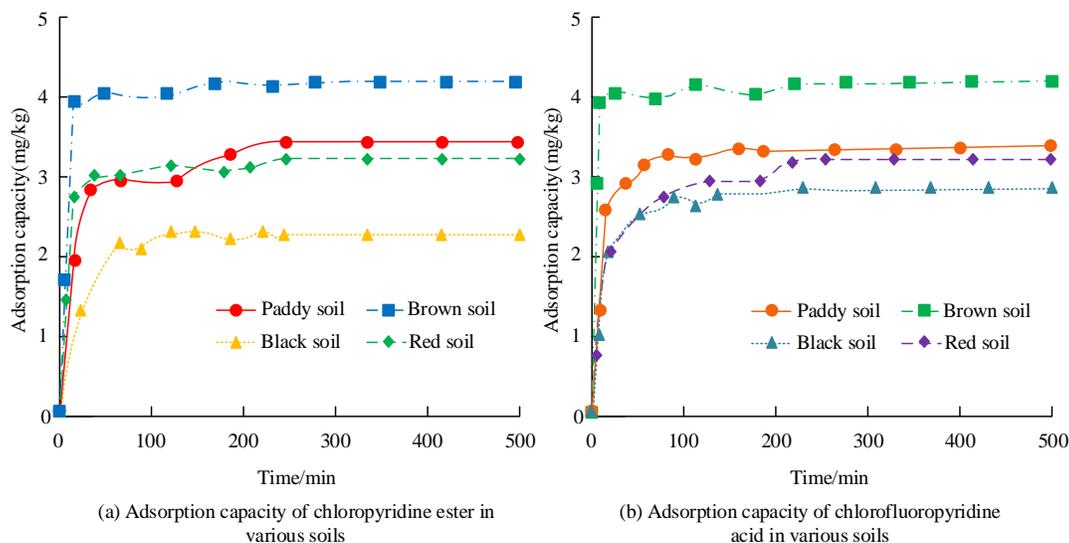


Figure 8. Adsorption capacity of ACAEH residues in various soils.

Table 3. Hydrolysis kinetic parameters of ACAEH at different pH values.

pH	Kinetic equation	Rate constant (k/d)	Half-life period ($T_{1/2}$, day)	Correlation coefficient (r)
4	$C_t = 0.8762e^{-0.02t}$	0.020	35	0.967
7	$C_t = 1.0594e^{-0.04t}$	0.041	17	0.960
9	$C_t = 1.6585e^{-0.373t}$	0.374	2	0.964

Adsorption capacity of ACAEH residues in various soils

The adsorption capacity of CFE rapidly increased to its highest point within 30 minutes followed by a decrease in adsorption rate and finally stabilized at about 250 minutes. The adsorption capacity of paddy, black, brown, and red soils were 3.45, 2.23, 4.19, and 2.28 mg/kg, respectively (Figure 8a). The adsorption trend of CFA was like that of CFE with the CFA adsorption capacity of each soil as 3.32, 2.85, 4.17, and 3.26 mg/kg for paddy, black, brown, and red soils, respectively (Figure 8b). Overall, ACAEH residues had the highest adsorption capacity in brown soil among all soils.

Hydrolysis kinetic parameters of ACAEH at different pH values

The hydrolysis kinetics parameters of ACAEH at different pH values showed that, when the pHs were 4, 7, and 9, the half-life times of this type of

herbicide in water were 35, 17, and 2 days, respectively (Table 3). A half-life of ≤ 30 days indicated that the herbicide was easily degradable, while a half-life greater than 30 days indicated that it had moderate degradation characteristics. Therefore, when $\text{pH} > 4$, the hydrolysis effect was significantly improved. From the perspective of hydrolysis rate, the hydrolysis rates at pH values of 4, 7, and 9 were 0.020, 0.041, and 0.374, respectively, which indicated that the hydrolysis rate of ACAEH in water significantly increased with the increase of pH.

Discussion

Researchers believe that the adoption and use of synthetic herbicides in food production have led to the evolution of resistance in various weeds to many herbicide chemical classes, making weed

management more difficult [17]. Gaines *et al.* suggested that the use of chemical synthetic herbicides had led to strong and widespread selection pressure on weeds, resulting in hundreds of weed species evolving herbicide resistance [10]. In addition, the long-term use of chemical herbicides has resulted in a series of environmental and resistance issues. In recent years, the concept of ecological civilization has been deeply rooted in people's hearts, and people are now not only pursuing food and clothing, but also paying more attention to health and environmental protection. Therefore, developing green technology to cultivate pollution-free agricultural products is very popular [18]. It is particularly important to study the degradation law of ACAEH in soil. This study used Q-TOF technology to analyze the residual degradation patterns of ACAEH in various soil types including paddy, black, brown, and red soils and water. Chromatographic analysis revealed the production of two new differential compounds in soil degradation with MCRs of 427.023 and 405.082, respectively. The peak times of these two compounds perfectly matched that of sodium and chloride ions, indicating that they were metabolites of ACAEH after degradation in soil. The results of hydrolysis metabolite found two new differential compounds with MCRs of 439.042 and 348.995, and were identified as CFE and CFA, respectively, by comparing them to the mass spectrometry library. CFA was a metabolite of CFE hydrolysis, and its peak time was consistent with that of CFE standard. Therefore, CFE and CFA were hydrolysis metabolites of ACAEH.

In the degradation dynamic study of CFE, the concentration change curves of CFE in rice roots and soil were consistent under the different doses of ACAEH treatment, both reaching their maximum values after one day, which might be due to the adsorption of CFE in soil and its deposition in water. The results were consistent with the research results of Ghosh *et al.* [19]. Under the application of the double doses of herbicide in rice roots and soil, the degradation rates of CFE in soil reached 99.98% and 89.44%,

respectively, while the degradation rates at five times of the dose were both 99.99%. The degradation dynamics of CFA showed that it was consistent with the degradation dynamics of CFE in soil and rice roots with a degradation rate of 99.99%, which indicated that CFE and CFA had better degradation effects in soil and were consistent with the results of Gautam *et al.* [20]. The residual situation of ACAEH in soil showed that CFE and CFA were not detected over time in both the 2-fold and 5-fold dose treatment groups. The results showed that the metabolites in the soil after application had been degraded, but the molecular unsaturation characteristics of the degradation products still existed in the soil, which was the same as the results of Biale *et al.* [21]. The degradation rates of different soil types showed the order as paddy soil > black soil > brown soil > red soil. Bai *et al.* reported that the organic carbon content in soil was high, and there was more microbial activity, which could accelerate the soil degradation of pesticides, which also confirmed the results of this study [22]. The results of the adsorption capacity of ACAEH in various soils showed that CFE rapidly reached its maximum adsorption capacity within 30 minutes, then the adsorption rate slowed down, and finally stabilized around 250 minutes. Among different soils, brown soil had the highest adsorption capacity, which was because, after the adsorption sites on the soil surface were occupied, drugs entered the soil and were adsorbed inside. In addition, the pore size and internal micropores of soil also affected the adsorption rate, which was consistent with the research of Yang *et al.* and Gaines *et al.* [23, 24]. The hydrolysis kinetics at different pH values showed that the hydrolysis effect was better when the pH was greater than 4, and the hydrolysis rate increased with the increase of pH. Different soil types and environmental conditions had a significant impact on the degradation rate and residue of herbicides, providing support and reference for optimizing herbicide use and reducing environmental pollution.

With the advancement of agricultural modernization, the application of chemical

herbicides in rice fields has become increasingly widespread. ACAEH is widely used due to its high efficiency, but its environmental residue issue has also attracted people's attention. The main purpose of this study was to explore the degradation patterns and residue dynamics of ACAEH in different soil types to better understand the impact of herbicides on the environment and provide scientific basis for the rational use of herbicides and the reduction of environmental pollution. To analyze the degradation patterns of residues of ACAEH in paddy field environments, this study used Q-TOF technology to analyze FE herbicides and their metabolites in paddy field environments, thereby exploring the degradation patterns of ACAEH in different soil types. The degradation rate data of different soil types showed that soil organic carbon content and microbial activity had a significant impact on degradation. The adsorption capacity of brown soil was the highest one, and the adsorption rate tended to stabilize after 250 minutes. The analysis of the degradation law of ACAEH residues based on Q-TOF technology revealed the decomposition changes of ACAEH residues in soil, providing support for the detection of such herbicides in soil environmental residues. However, this study was limited to the analysis of ACAEH in soil degradation, adsorption, and hydrolysis, and did not cover all possible soil types and environmental conditions. Future research should be further expanded to more soil types and climatic conditions to more comprehensively evaluate the impact of herbicide residues on the environment.

Acknowledgements

The research was supported by Jilin Academy of Agricultural Sciences Innovation Project "Study on the effects of 3% chlorofluoropyridine ester residues on the physical and chemical properties and microbial community structure of black soil" (Grant No. KYJF2021ZR111).

References

1. Dimaano NG, Iwakami S. 2021. Cytochrome P450-mediated herbicide metabolism in plants: Current understanding and prospects. *Pest Manag Sci.* 77(1):22-32.
2. Vázquez DE, Balbuena MS, Chaves F, Gora J, Menzel R, Farina WM. 2020. Sleep in honeybees is affected by the herbicide glyphosate. *Sci Rep.* 10(1):10516-10527.
3. Liao H, Li X, Yang Q, Bai Y, Cui P, Wen C, *et al.* 2021. Herbicide selection promotes antibiotic resistance in soil microbiomes. *Mol Biol Evol.* 38(6):2337-2350.
4. Wellendorf A, Tichelmann P, Uhl J. 2023. Performance analysis of a dynamic test bench based on a linear direct drive. *Archives of Advanced Engineering Science.* 1(1):55-62.
5. Hepburn HB, Donohoe TJ. 2020. Reductive hydroxymethylation of 4-heteroarylpyridines. *Chem Eur J.* 26(9):1963-1967.
6. Pang Y, Moser D, Cornella J. 2020. Pyrlium salts: Selective reagents for the activation of primary amino groups in organic synthesis. *Synthesis.* 52(04):489-503.
7. Marinic B, Hepburn HB, Grozavu A, Dow M, Donohoe TJ. 2021. Single point activation of pyridines enables reductive hydroxymethylation. *Chem Sci.* 12(2):742-746.
8. Singh S, Kumar V, Gill JPK, Datta S, Singh S, Dhaka V, *et al.* 2020. Herbicide glyphosate: toxicity and microbial degradation. *Int J Environ Res Public Health.* 17(20):7519-7523.
9. Escolano M, Gaviña D, Alzuet-Piña G, Díaz-Oltra S, Sánchez-Roselló M, Pozo CD. 2024. Recent strategies in the nucleophilic dearomatization of pyridines, quinolines, and isoquinolines. *Chem Rev.* 124(3):1122-1246.
10. Gaines TA, Duke SO, Morran S, Rigon CA, Tranel PJ, Küpper A, *et al.* 2020. Mechanisms of evolved herbicide resistance. *J Biol Chem.* 295(30):10307-10330.
11. Duke SO, Dayan FE. 2020. The search for new herbicide mechanisms of action: Is there a 'holy grail'? *Pest Manag Sci.* 78(4):1303-1313.
12. Ito K, Doi T, Tsukamoto H. 2023. *De novo* synthesis of polysubstituted 3-hydroxypyridines via "Anti-Wacker"-type cyclization. *Catalysts.* 13(2):319-322.
13. Chang Q, Ge L, Li J, Qiu G, Wu F, Zhang H, *et al.* 2021. Automated QuEChERS for the determination of 482 pesticide residues in *Radix codonopsis* by GC-Q-TOF/MS and LC-Q-TOF/MS. *Anal Methods.* 13(46):5660-5669.
14. Liu X, Fan Y, Chang CP, Lo CK, Wang X. 2021. High-throughput screening and quantification of pesticides in paprika by UHPLC-Q-TOF/MS. *Food Anal Methods.* 14(10):2186-2198.
15. Qu RY, He B, Yang JF, Lin HY, Yang WC, Wu QY, *et al.* 2021. Where are the new herbicides? *Pest Manag Sci.* 77(6):2620-2625.
16. Jia L, Zhao LX, Fu Y, Ye F. 2022. Research progress in the design and synthesis of herbicide safeners: A review. *J Agric Food Chem.* 70(18):5499-5515.
17. Ofosu R, Agyemang ED, Márton A, Pásztor G, Taller J, Kazinczi G. 2023. Herbicide resistance: Managing weeds in a changing world. *Agronomy.* 13(6):1595-1598.
18. Suzuki H, Liao Y, Kawai Y, Matsuda T. 2021. Rhodium-catalyzed additive-free C–H ethoxycarbonylation of (Hetero) arenes with

- diethyl dicarbonate as a CO surrogate. *Eur J Org Chem.* 2021(35):4938-4942.
19. Ghosh AK, Shahabi D, Yadav M, Kovala S, Anson BJ, Lendy EK, *et al.* 2021. Chloropyridinyl esters of nonsteroidal anti-inflammatory agents and related derivatives as potent SARS-CoV-2 3CL protease inhibitors. *Molecules.* 26(19):5782-5786.
 20. Gautam R, Geniza I, Iacono ST, Friesen CM, Jennings AR. 2022. Perfluoropyridine: Discovery, chemistry, and applications in polymers and material science. *Molecules.* 27(5):1616-1623.
 21. Biale G, La Nasa J, Mattonai M, Corti A, Vinciguerra V, Castelvetro V, *et al.* 2021. A systematic study on the degradation products generated from artificially aged microplastics. *Polymers.* 13(12):1997-2003.
 22. Bai Y, Ma L, Degen AA, Rafiq MK, Kuzyakov Y, Zhao J, *et al.* 2020. Long-term active restoration of extremely degraded alpine grassland accelerated turnover and increased stability of soil carbon. *Glob Chang Biol.* 26(12):7217-7228.
 23. Yang H, Huang K, Zhang K, Weng Q, Zhang H, Wang F. 2021. Predicting heavy metal adsorption on soil with machine learning and map** global distribution of soil adsorption capacities. *Environ Sci Technol.* 55(20):14316-14328.
 24. Gaines TA, Busi R, Küpper A. 2021. Can new herbicide discovery allow weed management to outpace resistance evolution? *Pest Manag Sci.* 77(7):3036-3041.