A PRELIMINARY STUDY OF ISOPROTHIOLANE DEGRADATION IN WATER COLUMN OF PADDY FIELD FROM PENAGA VILLAGE, SEBERANG PERAI UTARA, PULAU PINANG, MALAYSIA

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ABSTRACT

Rice field is one of the agriculture areas in Malaysia that is exposed to the usage of pesticide before the paddy planting phase, during vegetative phase and the post- harvesting phase. Isoprothiolane is one of the fungicides that is used during the vegetative phase to kill the fungi on the paddies. This study of isoprothiolane degradation in water samples was conducted in four different conditions, namely autoclaved water sample exposed to sunlight, autoclaved water sample kept in dark, non-autoclave water sample exposed to sunlight and non-autoclaved water sample kept in dark via laboratory batch experiment in a course of 30 days. The extraction and determination of the isoprothiolane in the water samples were done by solid phase extraction method (SPE) and analysed by Gas Chromatography–Mass Spectrometry (GC-MS). The study revealed that isoprothiolane degraded very slowly in all four conditions within 30 days of observation, and the degradation rates ranged from 0.0003 to 0.0037 day$^{-1.}$ The study also proved that the residue of isoprothiolane remained at least a month after its application.

Keywords: Degradation, GC-MS, isoprothiolane, paddy field and surface water.

INTRODUCTION

Pesticides are heterogeneous chemical compound. They are toxic and non-biodegradable substances that can harm our environment. Pesticides are designed as substances that can control, prevent, destroy, repel or mitigate any pests, including insects, animals, fungi and plants [1,2]. Moreover, the application of pesticides on the crops is the easiest way to protect the crops and the crop yields from air-borne diseases and pests, increase the economic potential of the agricultural sector, and also increase the quality of yields [3,4]. Apart from that, the reaction pathways and the degradation of pesticides play important roles in determining the process involved, the nature of the reactive intermediates, and their final products to control the dispersion of pesticides in the environment [5].

Isoprothiolane (Diisopropyl 1,3-dithiolan-2-ylidenemalonate) is one of the active ingredients in pesticides. It has been categorized as a fungicide, and used in Malaysian paddy fields [3]. Besides, it is also known as choline biosynthesis inhibitor (CBI) and is used to control the rice blast fungus such as leafhoppers and planthoppers on paddies. Some of the synthetic pesticides may have been banned in some countries because of their toxicity persistence in the environment and ecosystem such as food grain and non-target animals, reduction in the insect population, and contamination in soil and water [6].

The movement and degradation of pesticides are influenced by many factors, such as a country's climate and the condition of the soil, including pH, moisture content, type of soil, organic matter and clay contents. Moreover, the environmental condition where pesticides were applied on the crops, and the physical-chemical properties of the pesticides themselves also play important factors that determine how pesticides move and degrade in the environment [1]. A pesticide that enters the soil can degrade through various degradation processes or dissipation mechanisms. It can degrade via hydrolytic degradation, photolysis degradation, surface run-off, leaching, volatilization, oxidation or reduction (usually occurs in aerobic or anaerobic condition), and microbial degradation (the pesticide compound is degraded by the available microorganisms in the soil) [7].

A study by Farahani et al. (2012) has shown that photochemical degradation and hydrolysis are the main pathways for carbofuran degradation in water. Biological degradation and the pH of the water are some of the main factors affecting the degradation of carbofuran in water [8]. Lartiges and Garrigues, (1995) study of degradation kinetics of organophosphorus (OP) and organonitrogen (ON) pesticides in water under different environmental circumstances found that the half-lives of OP pesticides could be more than several months which could lead to environmental pollution [9].

The degradation rate of pesticides is different between countries with a temperate climate and countries with a tropical climate. The important elements of climate that can affect the degradation rate of pesticides are the average amount of rainfall, the average temperature in the region, the average emission of sunlight, and the presence of microorganisms [10]. In general, tropical countries have heavier rainfall compared to countries with a temperate climate. Based on previous studies, rainfalls can influence the contamination of pesticides in soil and water by diluting the concentrations of pesticides in the environment or producing pesticide runoffs that could spread to the watersheds and irrigation channels. Besides, paddy fields in tropical countries have been identified to have adopted the practice of pesticides applications intensively, which has contributed to high pesticide contaminations in the agriculture sectors and the environment [11,12].

Isoprothiolane is used in paddy fields to kill fungi that harm the crops and affect the production of rice during the planting season. This fungicide has been used in Malaysian paddy fields [3]. However, there have been limited studies on the degradation rate of isoprothiolane in soil and water in a tropical paddy field ecosystem, especially in Malaysia where the intensity of sunlight (UV) and the presence of microorganisms could lead to more rapid degradation. The purpose of this study is to determine the isoprothiolane degradation rate within 30 days in paddy field water which is subjected to four different conditions, namely autoclaved water sample exposed to sunlight, autoclaved water sample kept in dark, non-autoclaved water sample exposed to sunlight, and non-autoclaved water sample kept in dark.

MATERIALS AND METHODS

Sampling Site

The location of water sampling for this study was in a paddy field at Padang Tembusu Village. The village is located at Penaga Mukim 5, Seberang Perai Utara, Pulau Pinang, which is situated in the northern part of Peninsular Malaysia at the latitude of 5^{0}34’0.01’’ and longitude of 100^{0}22’0.01’’. It has an annual average range of about 27^{0}C to 32^{0}C for temperature, 100mm to 400mm for rainfall and 60% to 75% for relative humidity [13]. Figure 3.1 shows the location map of Padang Tembusu Village.

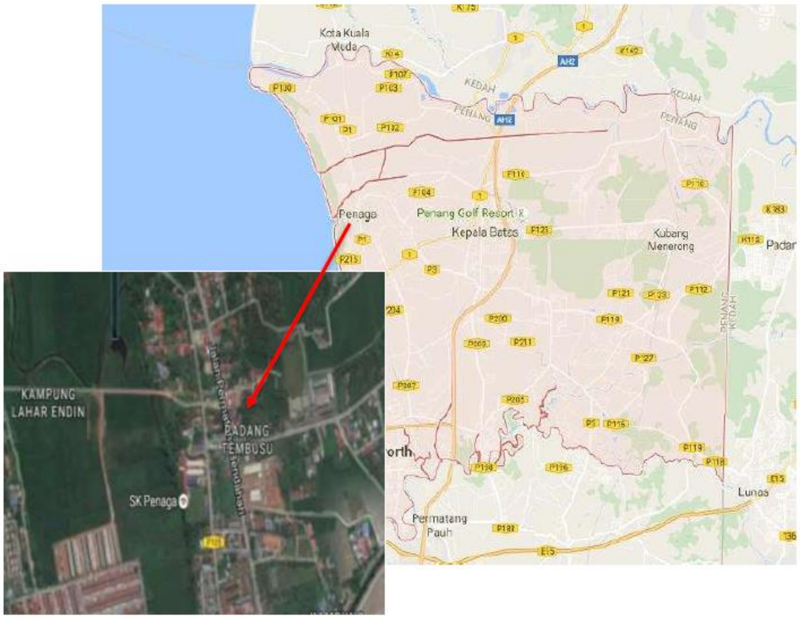

Figure 1. Map of Padang Tembusu Village, Penaga Mukim 5, Seberang Perai Utara, Pulau Pinang, Malaysia.

Degradation Study

In this study, the degradation course of isoprothiolane in the paddy field water under four conditions, namely autoclave, non-autoclave, with sunlight and without sunlight was followed for 30 days. The surface water from the river water near to the paddy field was taken and put into a 1L glass bottle for each condition. 40 sample bottles and 10 blank bottles were used in the experiment. All the bottles were wrapped in aluminium foil and stored in the refrigerator prior to the degradation study. The 1L sample was dosed with 4μL pesticide according to the recommendation provided by the manufacturer to the farmers. Half of the sample bottles were autoclaved, and while the other half were not autoclaved before the pesticide dosing. Similarly, half of the sample bottles were exposed to sunlight, and while the other half were kept in the dark in a drying cabinet with an average temperature of 28°C. The sample bottles that were kept in the dark were covered with an aluminium foil to ensure that no light could penetrate through to the samples. The batch of study was followed for 30 days. The data on the degradation rate was taken, starting from two weeks after the pesticide dosing until the 30th day. A blank sample was also prepared for each condition. The samples of water were extracted by using solid-phase extraction (SPE) and analysed with Gas Chromatographic Mass Spectrometric (GC-MS). The experiment was conducted in triplicate.

Solid-Phase Extraction of water samples

The samples were extracted by solid-phase extraction (SPE) before being sent for GC-MS analysis. The solid-phase extraction (SPE) cartridges, DSC-C18 (500 mg/6 mL) were obtained from Supelco (Milan, Italy) supplier, and the solvents used were pesticide GC-grade. SPE and GC-MS were used to quantify the pesticide content in the water samples. Before undergoing the SPE process, the samples were filtered by 0.45-mm filter to remove any particulate matter. Throughout the SPE process, 10 mL of acetone was used to wash the C-18 cartridge under vacuum, followed by 3 mL of acetonitrile and 3 mL of distilled water. All the samples were mixed well and passed over the discs with a 1.5 mL/min flow rate under vacuum. After all the SPE tubes had been filled with water samples, the vacuum pump was turned on. When the suction occurred, the water samples were drawn into the C18 cartridges. The pressure knob was used to control the water flow through the SPE cartridge to fall in droplets. When the extraction was finished, the pesticide compound that was trapped in the cartridge was collected by using 6 mL of acetone as the eluting solvent. The pesticide compound that was collected by acetone was evaporated to 1 mL by rotavapor and stored in GC vial before being analysed by GC-MS [14].

Gas Chromatographic Mass Spectrometric (GC-MS) Analysis

The identification of Isoprothiolane pesticide was done by a gas chromatographic mass spectrometer. It was performed on a gas chromatograph mass spectrometer-2010 plus with a Shimadzu model, and a capillary column BPX-5 (30 m x 0.25 mm, 0.25 μm film thickness). The carrier gas that was used was nitrogen at 1 mL min-1. Data acquisition and control of the GCMS-2010 Plus was carried out by GC-MS solution software and data system software, and conducted with an injector temperature of 250ºC, a temperature detector (280ºC) and a splitless time of 0.75 min. The initial temperature of 70°C, was increased at a rate of 10 ºC min^{-1} up to 160, followed by a rate of 3ºC min^{-1} up to 240ºC. The 1µ volume of the sample solution was injected automatically in split mode [15].

RESULTS AND DISCUSSION

Degradation of Isoprothiolane in water samples

In this study, the degradation of isoprothiolane was followed in four conditions to mimic the field conditions, when water samples were exposed to sunlight, during night time, with and without the enhancement from existing microorganisms (non-autoclaved and autoclaved samples). Thus, the different study conditions might illustrate the rate of degradation of the isoprothiolane by hydrolysis, photodegradation, and biodegradation.

Non-autoclaved water samples with exposure to sunlight

The degradation of Isoprothiolane after the water samples was dosed with pesticide according to the recommended usage in field condition, in non-autoclaved samples with exposure to sunlight is shown in Figure 2. The rate of degradation calculated from the data was 0.0003 day^{-1}, which was the slowest amongst the four conditions. It was noticed that the least degradation occurred only after 20 days of dosing.

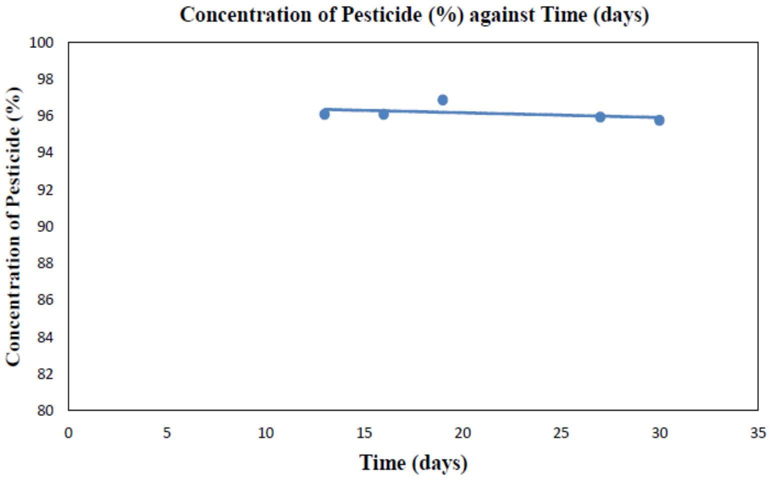

Figure 2. The concentration of Pesticide (%) against Time (days) in Non-autoclaved water samples with exposure to sunlight.

The concentrations of pesticide in water samples on the 19th, 27th and 30th day were 96.87%, 95.95% and 95.77% respectively. During the experiment, algae growths were observed after 10 days of incubation, as shown in Figure 3.

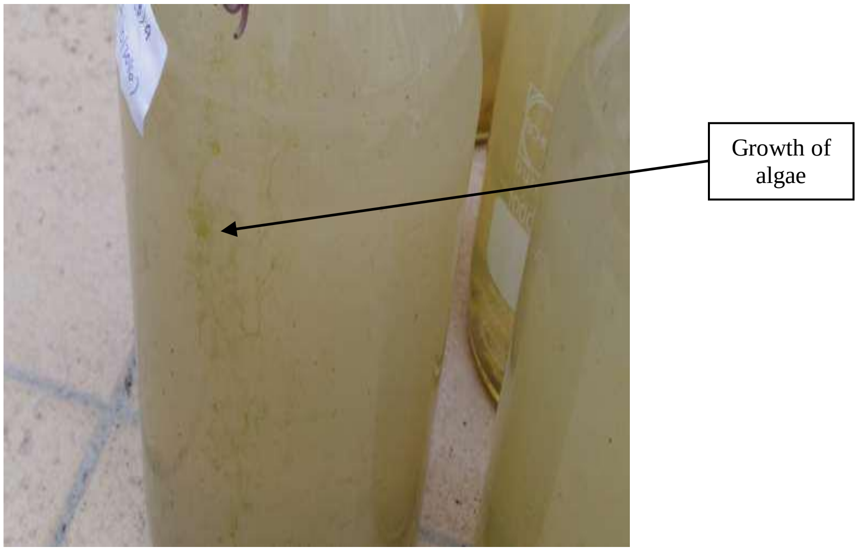

Figure 3. The growth of algae in the non-autoclaved water samples exposed to sunlight.

Degradation of Isoprothiolane in non-autoclaved water samples kept in dark.

Based on Figure 4, the degradation of Isoprothiolane in non-autoclaved water samples which were kept in the dark started to degrade on the 19th day. The degradation rate was also very slow at 0.001 day^{-1}. The concentrations of pesticide on the19th, 24th, 27th and 30th day were 96.06%, 95.29%, 94.43% and 94.9% respectively.

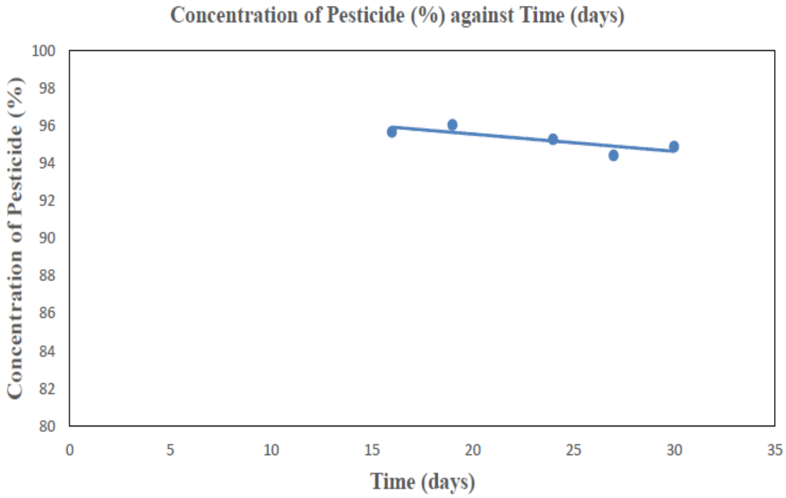

Figure 4. The concentration of Pesticide (%) against Time (days) in non-autoclaved water samples kept in dark.

Degradation of isoprothiolane in autoclaved water samples exposed to sunlight.

The degradation plot of isoprothiolane in autoclaved water samples with exposure to sunlight is shown in Figure 5. It was noticed that the rate of degradation in this condition was faster compared to the former condition, at a rate of 0.0037 day^{-1}. It began to degrade on the 15th, 24th, 27th and 30th day after the application of the pesticide. The concentrations of isoprothiolane on the 24th, 27th and 30th day were 95.77%, 94.54% and 90.59% respectively. The presence of algae growth in this condition was noticed after the 23rd day of pesticide dosing, as shown in Figure 6.

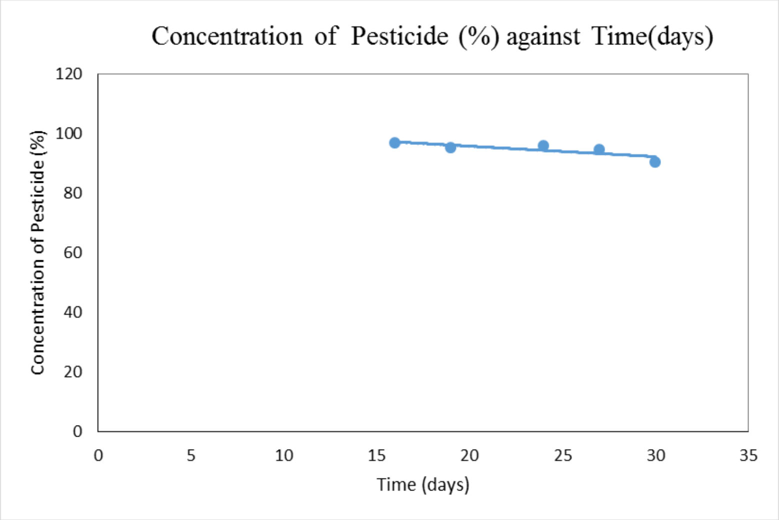

Figure 5. The concentration of Pesticide (%) against Time (days) in autoclaved water samples exposed to sunlight.

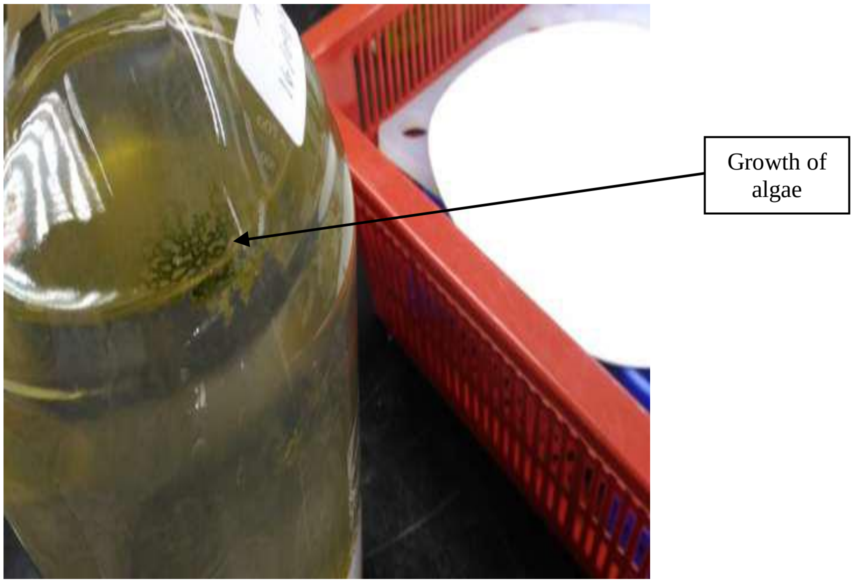

Figure 6. The growth of algae in the autoclaved water samples exposed to sunlight.

Degradation of Isoprothiolane in autoclaved water samples kept under dark condition.

The degradation rate of isoprothiolane in the autoclaved water samples under the dark condition was 0.002 day^{-1}. Figure 7 shows that the trend of the isoprothiolane degradation within the experimental period is clear.

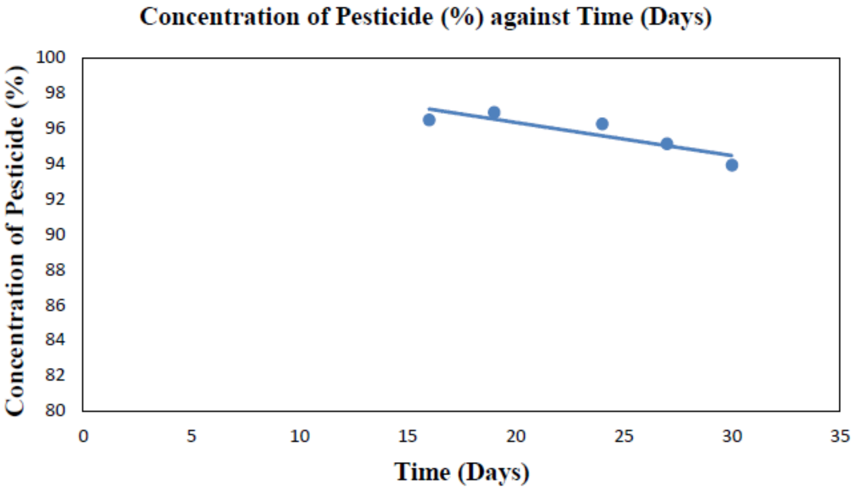

Figure 7. The concentration of Pesticide (%) against Time (Days) in autoclaved water samples kept in dark condition.

Based on the results shown in Figures 2, 4, 5 and 7, it can be concluded that the rate of isoprothiolane degradation in the water samples in all four conditions was very slow within the 30 days after the pesticide application. The presence of pesticide residual that was trapped in the higher concentration of suspended matter, such as algae and other particles with a mixture of the low organic content of sediment, might have led to the smaller ‘disappearance’ in the batch of study [15].

Throughout the experiment, the growth of algae was noticed in the water samples that were exposed to sunlight. Additionally, the growth of algae in non-autoclaved water samples was faster compared to autoclaved water samples without the presence of the microorganisms. In the non-autoclaved water samples, the algae began to grow after 10 days of isoprothiolane pesticide dosing. Meanwhile, in the autoclaved water samples without the presence of microorganisms, the algae started to appear after 23 days of the experiment. The factors that caused the growth of algae in the water samples were the presence of enough sunlight and nutrients that promoted its growth [16].

CONCLUSIONS

In this degradation study, based on 30 days of observation, the range of isoprothiolane degradation rate can be considered to be very slow. It was observed that the rate of degradation of isoprothiolane was much higher, at 0.0037 day^{-1} when the water sample went through the autoclave process and was exposed to the sunlight, compared to water samples in the dark which had a rate of degradation at 0.002 day^{-1}. The residual of isoprothiolane was the least after 30 days of application and was not susceptible to hydrolysis, photolysis and biodegradation. As a recommendation, the duration of the degradation study needs to be extended to more than 30 days to get a more significant trend of the degradation rate.

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