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Application of ECO₂FUME[®] Phosphine Fumigant for the Complete Control of Major Stored Product Insect Pests in Milled Rice in Thailand

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Abstract

ECO₂FUME[®] phosphine fumigant was used to fumigate milled rice in a commercial plastic bag (5 kg) and milled rice in a jumbo bag (1,000 kg) under gas-proof sheets to assess its performance against a mixed-age culture of *Sitophilus zeamais*, *Tribolium castaneum* and *Oryzaephilus surinamensis*. The trials were divided into 2 groups: 1) milled rice in a commercial plastic bag (packed rice) treated with a 50 g/m³ ECO₂FUME[®] application rate (700 ppm phosphine) for 2 days with two bag stacks of 46 m³ and 55 m³ and for 3 days with two bag stacks of 50 m³ each; and 2) milled rice in a jumbo bag (raw material rice) with stack size of 314 m³ treated with a 35 g/m³ ECO₂FUME[®] application rate (500 ppm phosphine) for 3 days and a stack size of 435 m³ treated with 50 g/m³ ECO₂FUME[®] application rate for 2 days. Gas sampling lines were installed in the stack to measure the phosphine concentrations during the fumigation period. The results of the fumigation trials showed that mixed-age cultures of the three insect species in packed rice stacks were completely controlled at 2 and 3 days when applied with an ECO₂FUME[®] application rate of 50 g/m³, whereas most insects in untreated control cages remained alive. ECO₂FUME[®] was also 100% effective in raw material rice stacks with complete control of mixed-age cultures of the three insect species using 35 g/m³ of ECO₂FUME[®] for 3 days and 50 g/m³ of ECO₂FUME[®] for 2 days. Commercial tarp fumigation of milled rice with ECO₂FUME[®] can be fumigated successfully without “top up” with good sealing procedures. Gas monitoring at regular intervals throughout the whole fumigation period is part of best fumigation practice to ensure that the minimum recommended phosphine concentration is maintained for complete control of all stages of target insect pests.

Keywords: ECO₂FUME[®] phosphine fumigant, fumigation, stored-product insect pests, milled rice, commercial plastic bag (5 kg), jumbo bag (1,000 kg).

1. Introduction

One of the largest rice-producing countries in the world, Thailand has an estimated 1.06 million hectares of cultivated land. In 2016, Thailand exported 9.88 million tons of rice with a total value of 154,434 million Baht (Office of Agricultural Economics, 2016).

Milled rice is prone to damage due to insect infestation. When the grain has no protection, the insect population will build up rapidly. Therefore, the losses and damage to stored milled rice due to insect

pests are related to the storage duration. The percentage of loss is very difficult to determine as the figure varies from 1% to as high as 25%. In 1977, FAO reported that the loss of rice ranged from 8 to 14% (Champ and Dyte, 1967) within the post-harvest system in Thailand. Suprakarn (1985) reported that 70 species of beetles and moths had been recorded in association with grains and other agricultural products in Thailand. Fortunately, only a handful of these insects have caused major economic damages. The major insect pests adversely affecting stored rice in Thailand are *Sitophilus zeamais*, *Tribolium castaneum*, and *Oryzaephilus surinamensis*, among others. Considered as an individual factor, the number of insect pests is not significant to the health of the harvested rice; however, preventing contamination is imperative as these pests can impact the quality and weight losses of stored rice. If contamination via insects occurs in the exported product, the rice may be rejected and returned to the country of origin, negatively affecting the reputation and revenue of the grain producer and exporter. Future exporting issues can occur as a result.

ECO₂FUME[®] fumigant gas is a cylinderized formulation of 2% PH₃ and 98% CO₂ by weight. It is packaged in high pressure aluminum or steel cylinders with net content of 31 kg of PH₃/CO₂ mixture and an equivalent phosphine amount of 620 g (Tumaming et al., 2012). ECO₂FUME[®] was first registered and commercialized in Australia during the 1990s. It was then registered in the USA in 2000, and approved for a shorter fumigation time of 24 hours for 1,000 ppm phosphine concentration at 27 °C or higher temperature for susceptible insect species (Cavasin et al., 2001).

There are several advantages when applying ECO₂FUME[®]. The dose of ECO₂FUME[®] phosphine fumigant applied to the commodity is rapid delivery, easy maintenance of the required dose during the fumigation period, shorter exposures and ease of application. ECO₂FUME[®] does not require the applicator to enter the fumigation space. The ready-to-use cylinders can be dispensed from outside of silos or structures being fumigated. This eliminates the need for entry into confined spaces to apply fumigants and solid waste disposal (Bonjour, 1998; Phillips, 1998).

The controlled application of fumigant gas resulting in less fumigant was introduced in stored product instead of the traditional solid formulation. It relies on the generation of a high initial phosphine concentration followed by a slow deterioration to ensure that the phosphine concentration - time product (CT) - will result in an effective fumigation. With ECO₂FUME[®] fumigant gas, the concentration can be easily controlled by the applicator to maintain an efficacious concentration and can be precisely measured by adding the required amount of gas when needed. Disposal of solid waste products from tablets is becoming more difficult every day. ECO₂FUME[®] fumigant gas eliminates the concern associated with deactivating unspent metal phosphide residue and disposal of the waste product (Cavasin et al., 2001).

The objective of this research is to study the application of ECO₂FUME[®] phosphine fumigant on milled rice in a packaged plastic bag and as a raw material in a jumbo bag for the control of major stored-product insect pests under commercial fumigation conditions of selected Thai rice companies.

2. Materials and Methods

Test insects and preparation of mixed-age cultures

Strains of *Sitophilus zeamais*, *Tribolium castaneum* and *Oryzaephilus surinamensis* were investigated. All insects used in this study were from the stored-product insect colonies maintained at the Post-harvest and Processing Research and Development Division Lab of the Thai Department of Agriculture (DOA). The mixed-age population was prepared by adding 50 young adults (2-3 week olds) of each strain of each of *S. zeamais*, *T. castaneum* and *O. surinamensis* into the cage of a glass bottle containing 200 g of culture medium and kept for 3 weeks. The culture medium was different for each species: brown rice for *S. zeamais*, rice bran for *T. castaneum* and grinded rice for *O. surinamensis*. Afterwards, all adults were removed and kept in the laboratory for 4 weeks at a temperature of 30±2°C and relative humidity of 65±5% before fumigation. All of the life stages were examined for their presence in the mixed-age culture glass bottle prior to fumigation. Before the

experiment was carried out, the test insects were transferred from glass bottles to the spun-bonded bags.

Fumigant

ECO₂FUME[®] phosphine fumigant is manufactured at Solvay's Niagara Falls, Canada, facility (known also by its associated legal entity name, Cytec Canada).

Fumigation of mixed-age culture

The experiment was conducted in a concrete warehouse of Patum Rice Mill and Granary Public Company Limited Group of Companies, located at Sikhiu district, Nakhon Ratchasima province, Thailand, in 2015. The trials were divided into 2 groups, as outlined below.

1 Milled rice in a commercial plastic bag of 5 kg capacity (packed rice) was treated with a 50 g/m³ ECO₂FUME[®] application rate (700 ppm phosphine) for 2 days with 2 bag stacks of 46 m³ and 55 m³, and for 3 days with 2 bag stacks of 50 m³ each (Figure 1).

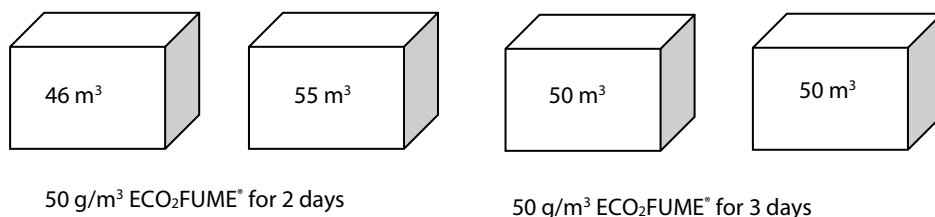


Fig. 1 Bag stack arrangement of milled rice in commercial 5-kg plastic bags

2) Milled rice in a jumbo bag of 1,000 kg capacity (raw material rice) and a stack size of 314 m³ was treated with a 35 g/m³ ECO₂FUME[®] application rate (500 ppm phosphine) for 3 days, and a stack size of 435 m³ was treated with 50 g/m³ ECO₂FUME[®] application rate (700 ppm phosphine) for 2 days (Figure 2).



Fig. 2 Bag stack arrangement of milled rice in 1,000 kg - jumbo bags

The raw material rice stacks and packed rice stacks were piled on the floor. Afterwards, 15 spun-bonded bags of test insects in a culture medium were placed in the stacks at 5 different locations: top (left and right), middle and bottom (left and right). Each spun-bonded bag of test insects contained a mixed-age culture of each insect species; *S. zeamais*, *T. castaneum* and *O. surinamensis*. For the fumigation of both packed rice and raw material rice, the spun-bonded bags of mixed-age test insects in the culture medium were placed inside the bag (Figures 3 and 4).

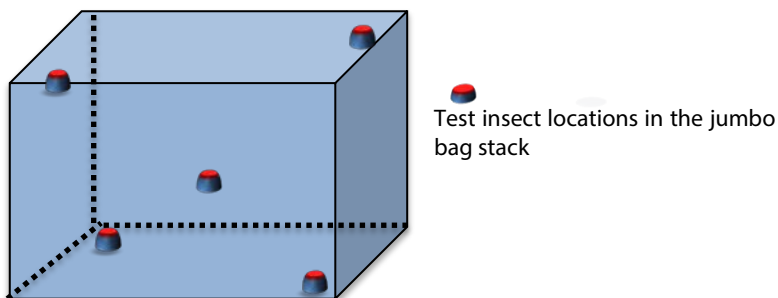


Fig. 3 Placement of test insects inside the jumbo bag stack



Fig. 4. Test insect placement inside the commercial 5-kg plastic bag.

The commercial 5-kg plastic bag of milled rice in the market of Thailand was punched 4 holes with diameter 1.0 mm. On the front and back of the bag, 2 holes were punched for each side. These holes allowed the air transfer which the bag could be stacked on the shelf during the distribution. Additionally, phosphine gas would penetrate into the plastic bag through these holes.



Two holes on the front of the plastic bag

Two holes on the back of the plastic bag

Fig. 5 Position of holes on the plastic bag

Gas sampling lines were installed at 3 locations in the rice stack (top, middle and bottom). Gas sampling lines were also installed inside the packed rice bag and the raw material bag to measure the phosphine concentration during the fumigation period.

Each milled rice stack with test insects was then covered with a clear polyvinyl chloride (PVC) sheet (0.2 mm thickness) as a fumigation enclosure to keep the phosphine gas inside the stack. The sheets were sealed to the concrete floor with sand snakes and masking tape.

The equivalent amount of phosphine from ECO₂FUME[®] was injected inside the tarp using a stainless steel quick-dispensing hose and gas injector with a gas flow rate of 6.8 kg/min. The exact amount of dispensed ECO₂FUME[®] was determined by the weight change of the cylinder on the top of a digital weighing scale accurate to 0.01 kg or 10 g.

Monitoring of gas concentration

Phosphine concentration was monitored at each of the following time intervals: 1) 1, 18, 24 and 42 hours for 2-day exposure time; 2) same as item 1 plus 48 and 66 hours for 3-day exposure time. The two phosphine target concentrations of 500 ppm and 700 ppm were maintained during the whole fumigation period. When the phosphine concentration fell below the target concentration, ECO₂FUME[®] dosing was topped up to bring the concentration back to or above the target level. Phosphine concentration was monitored with calibrated SILOCHEK phosphine monitor (0 - 2000 ppm).

Fumigation was terminated at 2 and 3 day exposure times followed by aeration of the slightly opened enclosure until the phosphine concentration reached the threshold limit value (TLV) of 0.3 ppm or lower. The plastic cover sheets were completely removed afterwards.

Assessment of insect mortality

Effectiveness of ECO₂FUME[®] against the test insect was determined by mortality of the mixed-age culture. After fumigation, the spun-bonded bags that contained the test insects were retrieved from each stack. The mortality of adults from each experiment was recorded after the removal of the fumigation sheet cover. Dead and alive insects were separated and culture media were returned to the bottles and then kept in the laboratory at 30±2°C and 65±5% for 6 weeks and were observed weekly for any newly emerged adults. This period was sufficient for emergence of all insects in the treatment as well as the control. The occasional death from the control was corrected by Abbott's formula (Abbott, 1925).

Monitoring of temperature, relative humidity and moisture content

The temperature and relative humidity in the warehouse were monitored by a thermo-recorder every day during the fumigation period. The moisture contents of maize were measured before and after treatment by applying the samples to an SB 900 Steinlite moisture meter.

3. Results

The effectiveness of ECO₂FUME[®]

The results of the fumigation on packed rice stacks shown in Table 1 demonstrated that mixed-age *Sitophilus zeamais*, *Tribolium castaneum* and *Oryzaephilus surinamensis* were completely controlled at 2 and 3 days when applied with an ECO₂FUME[®] application rate of 50 g/m³ (700 ppm phosphine). No live insects were observed immediately after fumigation or 6 weeks later. Most insects in non-fumigated control cages remained alive. ECO₂FUME[®] was also 100% effective in raw material rice stacks, completely controlling the mixed-age cultures of the three insect species with concentrations of 35 g/m³ (500 ppm phosphine) for 3 days and 50 g/m³ (700 ppm phosphine) for 2 days (Table 2).

The concentration of phosphine

Fumigation on packed rice in stacks at 50 g/m³ ECO₂FUME[®] (700 ppm) phosphine concentration was achieved at 2 and 3 days of exposure period times, as shown in Table 3. One hour after gas injection, the phosphine concentrations in the rice stacks were higher than 2,000 ppm, yet phosphine gas did not penetrate the plastic bag of packed rice. After fumigation for 18 hours, the phosphine concentration decreased but was still higher than the target concentration of 700 ppm in all rice stack fumigations. Phosphine concentration was monitored throughout the fumigation period. There were small reductions in phosphine concentrations, but all were still over the target concentration. The phosphine concentrations inside the plastic bag were nearly equal to the phosphine concentrations inside the fumigated stacks. This indicated that phosphine penetrated well into the plastic bag.

Phosphine concentrations on raw material rice fumigated with 35 g/m³ ECO₂FUME[®] for 3 days and 50 g/m³ ECO₂FUME[®] for 2 days from the rice stacks were shown in Table 3. The phosphine concentration of the fumigated raw material rice was quite similar to the fumigated packed rice stacks. After fumigation for an hour, the phosphine concentrations in raw material rice stacks were higher than 2,000 ppm. However, phosphine concentration could not be found inside the jumbo bag, indicating that there was no gas penetration yet.

After fumigation for 18 hours, the concentrations of phosphine decreased but were still well above the target concentration in the rice stacks. All phosphine concentrations were above the target concentrations of 500 and 700 ppm throughout the fumigation period. These results show that phosphine concentrations inside the jumbo bags were close to the concentrations of the fumigated stacks, indicating that phosphine could also penetrate well into the jumbo bag.

Monitoring of temperature, relative humidity and moisture content

The moisture content of maize was 11.8-13.5%. Temperature and relative humidity in the warehouse were 29.0-30.3°C and 66.9-68.2%, respectively.

4. Discussion

The commercial fumigation trials of milled rice in 5 kg plastic bags and 1,000 kg jumbo bags using ECO₂FUME[®] showed complete control of all stages of the target major grain insects at selected rice mills in Thailand. Results were achieved using phosphine fumigation protocols of 35 g/m³ ECO₂FUME[®] (500 ppm phosphine) for 3 days, and 50 g/m³ ECO₂FUME[®] (700 ppm) for 2 days at a daily temperature fluctuation of 26 - 33°C.

The results showed that the amount of phosphine gas absorbed by milled rice is minimal. The good sealing of the stack also minimized the gas loss due to gas leaks, and the higher phosphine concentration achieved at the start of fumigation remained higher than the target phosphine concentration throughout the entire fumigation period. In this study, top up of ECO₂FUME[®] was unnecessary to maintain the target concentration. Hence, the commercial fumigation of milled rice during storage of both raw material rice and packed rice with ECO₂FUME[®] before distribution could be completed successfully without top up.

Despite the fact that the target phosphine concentration was maintained throughout the duration of the fumigation period, it is still necessary to monitor phosphine concentrations at regular intervals. If the phosphine concentration is not regularly monitored during the entire exposure period, there could be a risk of greater gas loss due to gas leak and gas sorption, which can render the fumigation ineffective.

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Tab. 1 Mortality of 3 insect species after fumigation on packed rice stacks with ECO₂FUME[®] fumigate at different exposure times.

Dosages (g/m ³)	Time (Days)	Stack size (m ³)	Location	%Mortality of insect species					
				Sitophilus zeamais		Tribolium castaneum		Oryzaephilus surinamensis	
				Immediately after fumigation	6 weeks after fumigation	Immediately after fumigation	6 weeks after fumigation	Immediately after fumigation	6 weeks after fumigation
50 g/m ³ (700 ppm)	2	55	Top-left	100	100	100	100	100	100
			Top-right	100	100	100	100	100	100
			Middle	100	100	100	100	100	100
			Bottom-left	100	100	100	100	100	100
			Bottom-right	100	100	100	100	100	100
			Control (Unfumigated)	0 ^{1/3}	0	0	0	0	0
	46	Top-left	100	100	100	100	100	100	
		Top-right	100	100	100	100	100	100	
		Middle	100	100	100	100	100	100	
		Bottom-left	100	100	100	100	100	100	
		Bottom-right	100	100	100	100	100	100	
		Control (Unfumigated)	0 ^{1/3}	0	0	0	0	0	
	50	3	Top-left	100	100	100	100	100	100
			Top-right	100	100	100	100	100	100
			Middle	100	100	100	100	100	100
			Bottom-left	100	100	100	100	100	100
			Bottom-right	100	100	100	100	100	100
			Control (Unfumigated)	0 ^{1/3}	0	0	0	0	0
50	3	Top-left	100	100	100	100	100	100	
		Top-right	100	100	100	100	100	100	
		Middle	100	100	100	100	100	100	
		Bottom-left	100	100	100	100	100	100	
		Bottom-right	100	100	100	100	100	100	
		Control (Unfumigated)	0 ^{1/3}	0	0	0	0	0	

^{1/3} Mean of 3 replications

Tab. 2 Mortality of 3 insect species after fumigation on raw material rice stacks with ECO₂FUME[®] fumigate at different dosages and exposure times.

Dosages (g/m ³)	Time (Days)	Stack size (m ³)	Location	%Mortality of insect species					
				Sitophilus zeamais		Tribolium castaneum		Oryzaephilus surinamensis	
				Immediately after fumigation	6 weeks after fumigation	Immediately after fumigation	6 weeks after fumigation	Immediately after fumigation	6 weeks after fumigation
35 g/m ³ (500 ppm)	3	314	Top-left	100	100	100	100	100	100
			Top-right	100	100	100	100	100	100
			Middle	100	100	100	100	100	100
			Bottom-left	100	100	100	100	100	100
			Bottom-right	100	100	100	100	100	100
			Control (Unfumigated)	0 ^{1/3}	0	0	0	0	0

50 g/m ³ (700 ppm)	2	435	Top-left	100	100	100	100	100	100
			Top-right	100	100	100	100	100	100
			Middle	100	100	100	100	100	100
			Bottom-left	100	100	100	100	100	100
			Bottom-right	100	100	100	100	100	100
Control (Unfumigated)	3			0 ^{1/2}	0	0	0	0	0

^{1/2} Mean of 3 replications**Tab. 3** Phosphine concentrations inside the stacks during milled rice fumigations with ECO₂FUME[®] fumigation at different dosages and exposure times.

Applicat	Dos	Time	Stack	Volume of	Location	Phosphine concentration (ppm)						
						Hours						
						1	18	24	42	48	66	
5-kg in plastic bag	50 g/m ³ (700 ppm)	2	55	2.73	Top	>2000	1,325	1,289	1,213	1,195		
					Middle	>2000	1,293	1,295	1,225	1,197		
					Bottom	>2000	1,284	1,297	1,255	1,199		
		46	2.32	Top	>2000	853	860	829	819			
				Middle	>2000	864	849	837	826			
				Bottom	>2000	867	855	845	830			
					Inside plastic bag	-	805	875	812	811		
	50	3	50	2.48	Top	>2000	1,365	1,369	1,283	1,144	1,060	
					Middle	>2000	1,360	1,361	1,281	1,147	1,065	
					Bottom	>2000	1,359	1,380	1,279	1,148	1,069	
		50	2.50	Top	>2000	1,145	1,125	1,070	1,069	1,075		
				Middle	>2000	1,163	1,112	1,085	1,065	1,078		
Bottom				>2000	1,173	1,120	1,095	1,075	1,085			
				Inside plastic bag	-	999	1,001	1,104	1,101	1,127		
1,000-kg jumbo bag	35	3	314	11.22	Top	>2000	1,239	1,225	1,230	1,171	910	
					Middle	>2000	1,227	1,231	1,242	1,189	926	
					Bottom	>2000	1,244	1,237	1,259	1,236	940	
		50	2	435	21.77	Top	>2000	1,492	1,494	1,150	986	
						Middle	>2000	1,505	1,510	1,146	1,087	
						Bottom	>2000	1,512	1,514	1,192	1,098	
					Inside jumbo bag	-	1,490	1,534	1,483	1,493		

Residual behaviour of phosphine in different commodities

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Abstract

Phosphine is one of the most common active substances used in storage protection worldwide. As it is very efficient amongst a broad range of living organisms, it has become the favoured product after phasing out methyl bromide in 2010, as it can be used in many commodities.

In 2005, the regulation 396/2005 was enacted and came into force in 2008. With this, the European commission started to evaluate residues arising from the use of a pesticide and to set maximum residue levels (MRLs) for safe and regulated food trade.

To proof residue levels are below MRL and therefore far below concerning concentrations of phosphine in food or feed, residue studies are permanently conducted. In addition to support MRL settings, the intention of these trials is to determine withholding periods needed in storage protection, corresponding to PHI (pre harvest interval) for field and glasshouse treatments.

Results of those studies show different levels and differences in decrease of residues after defined time periods. Thus, withholding periods for various commodities can differ. Residue trials with repeated exposure were conducted as well to determine possible additive effects.

Keywords: Phosphine, maximum residue level (MRL), withholding period