

# Comparison of Methyl Bromide and Phosphine for Fumigation of *Necrobia rufipes* (Coleoptera: Cleridae) and *Tyrophagus putrescentiae* (Sarcoptiformes: Acaridae), Pests of High-Value Stored Products

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## Abstract

Fumigation with methyl bromide has been a long established and effective method for controlling many pests of stored products, including the key major pests that infest dry-cured hams, aged cheese, and other value-added durable stored products. Methyl bromide had been widely used for the disinfestation of dry-cured ham facilities in the United States, but is now phased out of use since it is an ozone-depleting substance. This paper reports laboratory studies to evaluate the efficacies of methyl bromide and phosphine for controlling two of the key arthropod pests of dry-cured hams and aged cheeses. Larvae of the red-legged ham beetle, *Necrobia rufipes* (Fabricius), were the most tolerant life stages when treated with either phosphine or methyl bromide for 48 h exposure at 23°C, whereas eggs of the mold mite, *Tyrophagus putrescentiae* (Schränk), were slightly more tolerant than mobile stages for both compounds. Under laboratory conditions, complete control was achieved for the both species with concentrations of 0.85 and 4.0 g/m<sup>3</sup> of phosphine and methyl bromide, respectively, at 48 h exposure. The results give new information for judicious use of the existing stocks of methyl bromide, whether for pest mitigation or to help in developing a quarantine treatment schedule with that gas. Phosphine shows good potential as an effective alternative to methyl bromide, but if it was to be adopted as a fumigant in the dry-cured ham industry, methods to prevent metal corrosion would need to be designed and effectively implemented.

**Key words:** lethal concentration, hydrogen phosphide, ham beetle, ham mite, country ham

Fumigation is a critical tool for the mitigation of arthropod pest infestations of durable postharvest stored products such as grains and many other bulk raw or processed food products (Hagstrum et al. 2012). Methyl bromide (MB) is the most effective fumigant insecticide available for use, and it has been used for both fresh and durable commodities because of its high level of toxicity against all life stages of key target pests (Thoms and Phillips 2004). The short application times and its toxic activity at relatively low temperatures has made MB the required fumigant for many quarantine treatments. MB was the fumigant of choice for structural treatments of mills and other food processing facilities for its efficacy, the minimal impacts on equipment and electronics, and its fast treatment times that allowed processors to resume manufacturing operations. Despite its value to agriculture and food safety, MB is among a group of halogenated

compounds implicated in the destruction of the earth's protective atmospheric ozone layer and it is now banned under the international agreement known as the Montreal Protocol on Ozone Depleting Substances (United States Department of State 2019).

Signatories to the Montreal Protocol agreed to decrease and stop the use of all MB, with the exception of critical use exemptions and cases in the context of a use that has a "quarantine and pre-shipment" need. Once the ban is complete in a country the Montreal Protocol allows for fumigations to continue only until the existing stocks of the properly labeled fumigant are depleted. At this writing, MB is banned in the United States, the EU and many other countries as a fumigant for any use except for required quarantine treatments. The phase-out and ban of methyl bromide has stimulated substantial research activity into developing other fumigants or pest control

methods as alternatives and replacements to methyl bromide (e.g., Fields and White 2002, Phillips et al. 2012).

A variety of durable stored products from animal origins such as dried fish, cheese, and dried cured meats are subject to insect pest infestation during processing and storage, and often require pest management that includes fumigation (Arbogast 1991, Rajendran and Parveen 2005). The dry-cured ham industry in the United States and elsewhere has relied on methyl bromide for many years to mitigate infestations of debilitating arthropod pests (EPA 2015). Dry-cured ham products and their production and aging facilities are infested by arthropods such as the cheese skipper *Piophilidae casei* L. (Diptera: Piophilidae), the red legged ham beetle *Necrobium rufipes*, other beetles in the genus *Dermestes* (Coleoptera: Dermestidae), and the mold mite *Tyrophagus putrescentiae* (Hasan et al. 2016). Methyl bromide was the only known fumigant that was effective at controlling these arthropod infestations of dried cured ham in the United States for many years (e.g., Marriott and Schilling 2004). Until recently there has been very little research reported on the use of methyl bromide alternatives in the fumigation of dry-cured ham. The potential for using controlled atmosphere treatments of low oxygen or high carbon dioxide to control aged ham pests was studied by Hasan et al. (2016). Nonfumigant and food-safe compounds such as propylene glycol and other food preservatives have the potential to protect aged pork from *T. putrescentiae* infestations (Abbar et al. 2016). Sulfuryl fluoride is one commercially available fumigant that is proposed as an alternative or replacement for methyl bromide (Bell et al. 2004, Mueller et al. 2006), but experiments with arthropod pests of dried ham showed it to be ineffective against eggs of *T. putrescentiae* within its allowable application levels (Phillips et al. 2008). Hydrogen phosphide, commonly known as phosphine, is the most common fumigant used for bulk-stored cereal grains (Thoms and Phillips 2004), but its use may be limited for some food processing facilities due to its corrosive nature causing serious damage to valuable machines and electrical systems (e.g., Zhao et al. 2015) if electrical protections are not made.

Methyl bromide is not currently allowed for fumigation of dry-cured hams in the United States as requests to the international Methyl Bromide Technical Options Committee for yearly continuing use exemptions have stopped (e.g., US EPA 2019). Nevertheless, dried cured hams and similar products can continue to be fumigated with methyl bromide in the United States into the foreseeable future while existing stocks of the gas as labeled for use on hams still exist. Similarly, it is likely that dried animal products could continue to be fumigated with methyl bromide in other countries where the use of the gas has not been fully discontinued. Dry-cured hams are special meat products of which there are over 3 million sold yearly in the United States at a value over US\$400 million (Hanson et al. 2014). Although methyl bromide has a long history of use as a postharvest fumigant for pests of stored products and fresh commodities (reviewed in Bond 1984), we are not aware of scientific accounts of its efficacy for pests of dry-cured hams. The lack of information on toxic effects of phosphine and methyl bromide on arthropod pests of dry-cured hams, as well as the economic impacts on high-value food products, justifies the study of these common fumigants for control of these pests. The objective of the work described below was to investigate the mortality of all life stages of *T. putrescentiae* and *N. rufipes*, two of the most serious pests of dry cured ham, exposed to concentrations of methyl bromide and phosphine in controlled laboratory experiments.

## Materials and Methods

### Mite and Beetle Cultures

The *T. putrescentiae* and *N. rufipes* that were used in these experiments were from cultures that are maintained at the Department

of Entomology, Kansas State University. The laboratory diet for *T. putrescentiae* was the same as that used in our recent work (Abbar et al. 2016) and was composed mostly of water (475 ml), ground pet food (160 g), wheat germ (5 g), brewer's yeast (5 g), a vitamin mix (5 g), agar (5 g), glycerol (25 ml), and methyl-p-benzoate (25 ml of 15.8 g ml<sup>-1</sup>) as an antifungal agent. Mite colonies were maintained in 500-ml glass canning jars with filter paper ventilated lids and mites were transferred using metal probes and small brushes. *Necrobium rufipes* was reared in small plastic boxes on mixed foods of equal amounts of dried fish, pet food, and ham pieces as described in detail by Hasan et al. (2016). Both cultures were maintained in an incubator at 27°C and 70% RH with a photoperiod of 16:8 (L:D) h.

### Fumigant Gases

All fumigant gases were applied to test samples in 3.8-liter gas-tight glass jars (25 cm tall by 15 cm diameter, with a neck diameter of 11.4 cm) equipped with a port on the screw-on lid to accommodate a septum for introduction and sampling of gas using gas-tight syringes (Hamilton; Fisher Scientific, Pittsburg, PA). Phosphine, PH<sub>3</sub>, used in these fumigation experiments was in a pressurized cylinder from Matheson Tri-Gas at a concentration of 1% PH<sub>3</sub> (10,000 ppm) in nitrogen (Linweld Inc., Waverly, NE). A second pressurized cylinder contained 200 ppm PH<sub>3</sub> in nitrogen and was prepared by Matheson Tri-Gas and purchased from the same supplier for use as an analytical standard to quantify gas concentrations in the laboratory fumigation chambers. Phosphine gas was drawn from a cylinder and held briefly in a 3-liter Tedlar gas bag (CEL Scientific Corp, Santa Fe Springs, CA) that had been fully evacuated prior to use. For the methyl bromide (MB) experiments, a small pressurized cylinder of Meth-O-Gas 100 (100% purity; Great Lakes Chemical Corporation, West Lafayette, IN) was used. MB gas was also drawn from the cylinder and held in a previously evacuated Tedlar bag for further use in experiments that same day. A quantitative standard of MB was prepared at 5000 ppm in air in a Tedlar bag to quantify MB concentrations in fumigation chambers.

The experimental concentrations of PH<sub>3</sub> were determined for a given fumigation chamber by injection of three samples from each chamber into a gas chromatograph, GC, equipped with a flame photometric detector (Shimadzu GC 17A, Shimadzu Scientific Instruments, Columbia, MO) set in the phosphorus mode at the beginning and end of each exposure period. A GS-Q column (30 m long × 0.53 mm i.d., 0.25-μm film thickness; J and W Scientific, Folsom, CA) with a helium carrier gas at 4.0 ml/min run isothermally at 150°C was used to quantify PH<sub>3</sub> concentrations based on the peak area outputs from a HP-3390A integrator-recorder and a retention time of approximately 50 s. MB concentrations were determined using a coupled GC-mass spectrometer (Shimadzu GC 17A with model QP-5050A MS, Shimadzu Scientific Instruments, Columbia, MO) from three samples of each chamber taken at the beginning and end of gas exposure. The GC-MS had a DB-1 column (30 m long × 0.250 mm ID, w/0.25 μm film thickness; Agilent Technologies, Santa Clara, CA) held isothermally at 150°C, and MB was quantified from the peak area generated from the data system at a retention time of about 22 s. The average starting and ending phosphine concentrations were targeted within 0 to 0.468 g/m<sup>3</sup> for *N. rufipes*, and 0 to 0.850 g/m<sup>3</sup> for *T. putrescentiae*, whereas the range of MB concentration was 0 to 4.0 g/m<sup>3</sup> and 0 to 6.8 g/m<sup>3</sup> for *N. rufipes* and *T. putrescentiae*, respectively. The concentrations targeted for each experiment were selected to encompass the dose-response range ensuring, where possible, that a 100% kill was achieved at the maximum concentration tested. Fumigants were initially measured as ppm relative to air in a chamber, but were then reported as g/m<sup>3</sup> after

conversion. Phosphine at  $1 \text{ g/m}^3$  is equivalent to 714.8 ppm while methyl bromide at  $1 \text{ g/m}^3$  is equivalent to 257.7 ppm.

### Prefumigation Bioassay Preparation

#### Ham Beetle

Bioassay vials of *N. rufipes* containing a given life stage were placed separately within the fumigation chambers. The following age *N. rufipes* life stages were prepared for the bioassay: adults at 7–14 d posteclosion; pupae at 2–3 d old; mature larvae at 35–40 d old; and eggs at 2–3 d old. All stages of *N. rufipes* were placed in glass shell vials (18 ml, Fisherbrand, USA) with 0.5 g of standard diet, and the vials were closed with a fine netted cloth to allow gas entry while retaining the insects. Based on available beetles from the colony, each vial contained 10 randomly selected individuals of one of the four *N. rufipes* life stages and three vials for each life stage were fumigated in each chamber.

#### Mold Mites

Mite eggs less than 2 d old and large mobile mite stages (a mixture of nymphs and adults) of *T. putrescentiae* were selected for fumigation. Colony handling allowed for 10 eggs and 20 mobile stage mites to be placed in separate ventilated shell glass vials with approximately 0.2 g of standard diet. The fumigation chambers containing the experimental ham beetle and mite vials were held in an incubator (Thermo Scientific, USA) at  $23 \pm 2^\circ\text{C}$  and  $70 \pm 5\%$  R.H. for 48 h under fumigation.

### Postfumigation Bioassay Data Analysis

After the 48 h fumigation, each chamber was opened in a fume hood and ventilated in clean air for at least 1 h prior to evaluating the bioassays for mortality. The larval mortality in *N. rufipes* was determined based on the pupal formation after 7 d while the adult mortality was assessed within 10 d postfumigation. Fumigated pupae were stored in Petri dishes and observed for more than 14 d or until no additional air-control pupae eclosed to adults. Adequate standard diet was added into the vials that contained eggs to minimize cannibalism. Egg-hatch was visually checked through the vial for 7 d until no further hatch was observed. The fumigated eggs and mobile stages of *T. putrescentiae* were transferred to a 10-liter glass desiccator that was maintained at 70% RH with a saturated NaCl

solution at  $23 \pm 2^\circ\text{C}$  for mortality assessment (failure of egg hatch to larvae and immobile death or desiccation of mobile stages) following a minimum of 3 d postfumigation.

### Statistical Analyses

Mortality within shell vials for a given species and life stage group were pooled for a given fumigation chamber. The means procedure (SAS institution 2002) was used to summarize average mortalities of *N. rufipes* (eggs, larvae, pupae, and adults) and *T. putrescentiae* by their life stages (egg and nymphs/adults) and by the various target concentration of each gas. Analyses of variance were conducted to evaluate the role of fumigant concentration for the mortality of each life stage (PROC GLM; SAS 2002). Mortality data for a given jar at its gas calculated concentration determined via quantitative GC were subjected to probit analyses (PROC PROBIT; SAS 2002) to estimate doses needed for 50% ( $\text{LC}_{50}$ ) and 99% ( $\text{LC}_{99}$ ) levels of mortality.

## Results

### Necrobia rufipes

Both phosphine and methyl bromide were effective at killing all life stages of the red-legged ham beetle within the conditions of our experiments. ANOVA results for mortality as affected by concentration of phosphine as the main effect were highly significant for each life stage as follows: eggs with  $F_{1,31}=14.30$  and  $P = 0.0007$ ; larvae with  $F_{1,31}=30.67$  and  $P = 0.0001$ ; pupae with  $F_{1,31}=24.94$  and  $P = 0.0001$ ; and adults with  $F_{1,31}=10.52$  and  $P = 0.0028$ . Figure 1 shows the average mortality for groups of each life stage exposed to 10 different phosphine concentrations. All larvae, pupae, and adults of *N. rufipes* were killed when exposed to target phosphine concentrations of  $0.159 \text{ g/m}^3$  and higher, whereas eggs required the highest tested concentration of  $0.468 \text{ g/m}^3$  for 100% mortality. Probit analyses of *N. rufipes* mortality as a function of phosphine concentration (Table 1) estimated that the concentration needed to kill 99 % of a test population of eggs ( $\text{LC}_{99}$ ) was  $0.534 \text{ g/m}^3$ , which was over 5-fold more than the  $\text{LC}_{99}$  for adults estimated at  $0.095 \text{ g/m}^3$ . The  $\text{LC}_{99}$  for larvae and pupae were intermediate at  $0.212$  and  $0.165 \text{ g/m}^3$ , respectively. The  $\text{LC}_{50}$  values for mortality of the *N. rufipes* life stages to phosphine showed less variation among life stages than

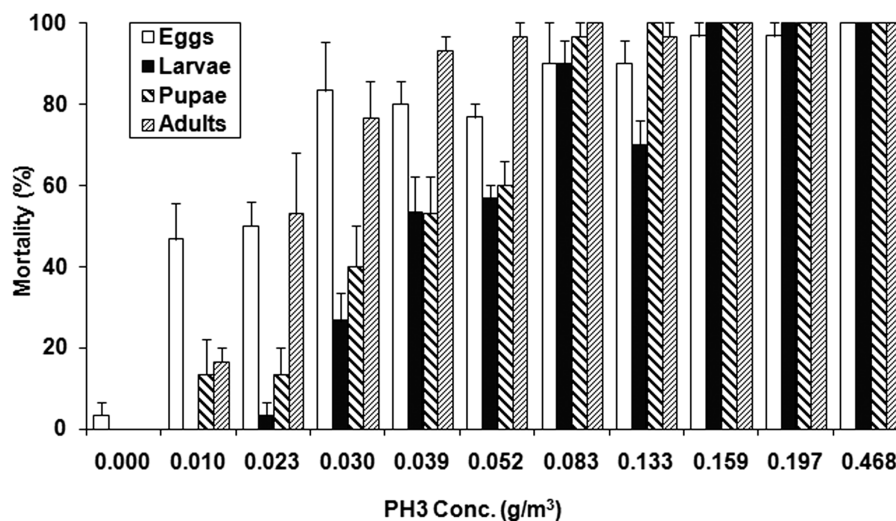


Fig. 1. Average mortality ( $\pm$  SE of mean) of *Necrobia rufipes* life stages exposed to increasing concentrations of phosphine gas,  $\text{PH}_3$ , for 48-h exposure at  $23^\circ\text{C}$ .

the  $LC_{99}$  values, with the largest difference at about a 2.6-fold when comparing larvae ( $LC_{50} = 0.049$  g/m<sup>3</sup>) as the most tolerant life stage, to adults ( $LC_{50} = 0.019$  g/m<sup>3</sup>) as the most susceptible stage, with eggs and pupae being more intermediate in tolerance. The mortality data we collected for phosphine fit the probit regression equation generated for each life stage as indicated by the  $\chi^2$  analyses finding no significant differences for fit of the data (Table 1).

Mortality of *N. rufipes* life stages exposed to doses of methyl bromide was directly related to gas concentration: eggs with  $F_{1,49} = 14.92$  and  $P = 0.0003$ ; larvae with  $F_{1,34} = 108.57$  and  $P = 0.0001$ ; pupae with  $F_{1,31} = 127.74$  and  $P = 0.0001$ ; and adults with  $F_{1,34} = 70.87$  and  $P = 0.0001$ . Figure 2 shows the mean levels of mortality for groups of each life stage exposed to 15 different target concentrations of methyl bromide. *Necrobia rufipes* eggs were the most susceptible life stage as all eggs were killed at 0.93 g/m<sup>3</sup>, a dose at which other life stages had low levels of mortality, whereas the maximum concentration tested of 3.98 g/m<sup>3</sup> was required to kill all life stages in these experiments (Fig. 2). The probit analyses (Table 2) for mortality of *N. rufipes* to methyl bromide also show that eggs were very susceptible with a  $LC_{50} = 0.316$  g/m<sup>3</sup>, whereas pupae and adults were intermediate in methyl bromide tolerance at  $LC_{50}$  of 1.659 and 1.745 g/m<sup>3</sup>, respectively, and larvae were the most tolerant at  $LC_{50} = 2.687$  g/m<sup>3</sup>. Probit regression models generated for mortality data of each life stage were found to fit the data very well as indicated by the  $\chi^2$  analyses (Table 2).

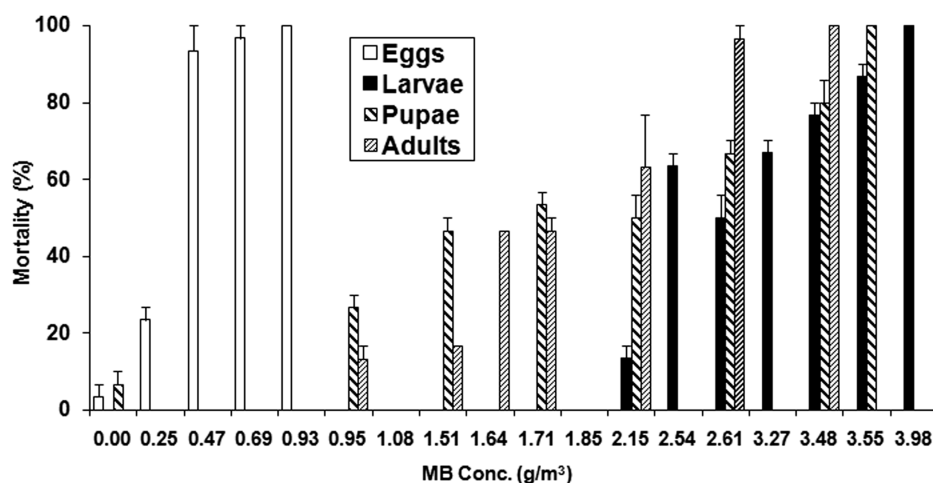
### Tyrophagus putrescentiae

Phosphine was very effective at killing eggs and the mobile stages of *T. putrescentiae*, with no survivors at 0.85 g/m<sup>3</sup> (Fig. 3). Mortality of *T. putrescentiae* varied significantly by concentration of phosphine as revealed by ANOVA for eggs at  $F_{1,18} = 199.10$  and  $P = 0.0001$  and the mobile stages at  $F_{1,19} = 104.06$  and  $P = 0.0001$ . Tolerance of the mites to phosphine based on probit analyses (Table 3) was similar between the life stages as the  $LC_{50}$  for eggs was 0.193 and the  $LC_{50}$  for mobile stages was 0.122 with broad overlap for their fiducial limits. Chi-square analyses showed that the data for mite mortality from phosphine had a weak fit with the calculated probit regression equation for eggs and a very poor fit for the mobile stages and mobile stage for phosphine, and this may be due to large increases in mortality associated with small changes in gas concentration.

Methyl bromide elicited 100% mortality of eggs and mobile stages of *T. putrescentiae* at 2.15 g/m<sup>3</sup> (Fig. 4). Mortality of *T. putrescentiae* varied significantly by concentration of methyl bromide as shown in ANOVA for eggs at  $F_{1,43} = 61.75$  and  $P = 0.0001$  and for the mobile stages at  $F_{1,43} = 53.62$  and  $P = 0.0001$ . Probit analyses for mortality from methyl bromide showed that the life stages of mites had similar levels of tolerance, with the  $LC_{50}$  for eggs at 0.961 g/m<sup>3</sup> and  $LC_{50}$  for mobile stages at 0.845 g/m<sup>3</sup>. As with phosphine for mites, the mortality data with methyl bromide did not fit the calculated probit regression equations as indicated by significant  $\chi^2$  values, and estimates of LC values are therefore of low quality. As with the probit analyses for phosphine, this may be due to large changes in mite mortality between closely spaced concentrations of methyl bromide (Fig. 4).

**Table 1.** Probit analyses of mortality for *Necrobia rufipes* life stages fumigated with Phosphine for 48-h exposure periods at 23°C

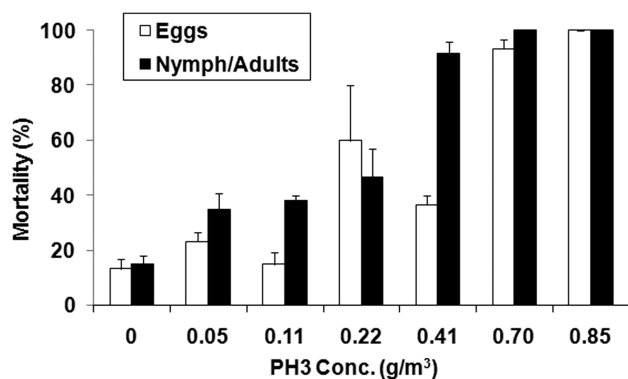
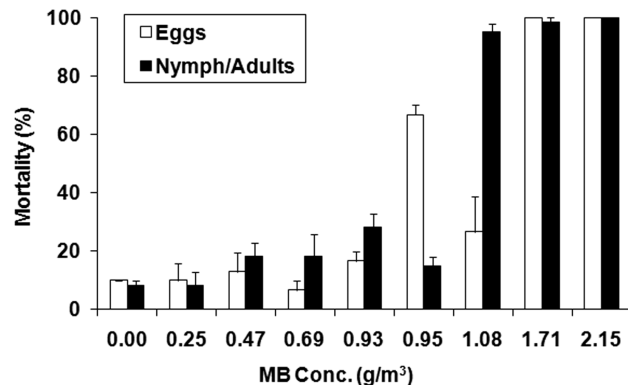
Insect stages	<i>n</i>	$LC_{50}$ g/m <sup>3</sup> (95% fiducial limits)	$LC_{99}$ g/m <sup>3</sup> (95% fiducial limits)	Intercept	Slope $\pm$ SE	$\chi^2$ values
Eggs	300	0.029 (0.010–0.047)	0.534 (0.215–11.822)	–0.059	0.032 $\pm$ 0.004	1.44 ( $P = 0.837$ )
Larvae	270	0.049 (0.040–0.062)	0.212 (0.136–0.553)	–0.001	0.013 $\pm$ 0.001	1.42 ( $P = 0.964$ )
Pupae	240	0.033 (0.025–0.042)	0.165 (0.099–0.529)	0.001	0.008 $\pm$ 0.001	4.61 ( $P = 0.465$ )
Adults	270	0.019 (0.015–0.026)	0.095 (0.058–0.291)	–0.003	0.006 $\pm$ 0.001	1.30 ( $P = 0.861$ )



**Fig. 2.** Average mortality ( $\pm$  SE of mean) of *Necrobia rufipes* life stages exposed to increasing concentrations of methyl bromide gas (MB) for 48-h exposure at 23°C.

**Table 2.** Probit analyses of mortality for *Necrobia rufipes* life stages fumigated with methyl bromide for 48 h exposure periods at 23°C

Insect stages	<i>n</i>	LC <sub>50</sub> g/m <sup>3</sup> (95% fiducial limits)	LC <sub>99</sub> g/m <sup>3</sup> (95% fiducial limits)	Intercept	Slope ± SE	χ <sup>2</sup> values
Eggs	210	0.316 (0.273–0.356)	0.692 (0.568–0.990)	0.010	0.072 ± 0.009	11.53 ( <i>P</i> = 0.318)
Larvae	240	2.687 (2.546–2.817)	4.777 (4.306–5.596)	1.731	0.207 ± 0.016	18.15 ( <i>P</i> = 0.697)
Pupae	300	1.659 (1.389–1.901)	9.177 (6.207–19.372)	-0.105	0.390 ± 0.025	12.89 ( <i>P</i> = 0.844)
Adults	210	1.745 (1.603–1.892)	3.976 (3.337–5.282)	-0.105	0.390 ± 0.025	12.88 ( <i>P</i> = 0.844)

**Fig. 3.** Average mortality (+/- SE of mean) of *Tyrophagus putrescentiae* life stages exposed to increasing concentrations of phosphine gas, PH<sub>3</sub>, for 48-h exposure at 23°C.**Fig. 4.** Average mortality (+/- SE of mean) of *Tyrophagus putrescentiae* life stages exposed to increasing concentrations of methyl bromide gas, MB, for 48-h exposure at 23°C.

## Discussion

Results indicated that fumigation of *N. rufipes* and *T. putrescentiae* with either phosphine or methyl bromide can result in 100% mortality when applied for 48 h at 23°C. Examination of the gas concentrations needed for mortality reveals that phosphine is more effective than methyl bromide based on a g/m<sup>3</sup> concentration basis. For example, all life stages of *N. rufipes* were killed at 0.468 g/m<sup>3</sup> of phosphine, but about 7 times as much methyl bromide, 3.98 g/m<sup>3</sup> was required for 100% kill, and for *T. putrescentiae* all mites were killed with phosphine at 0.85 g/m<sup>3</sup>, but over 2 times as much methyl bromide, 2.15 g/m<sup>3</sup>, was required for the same level of kill. This same trend of phosphine being more toxic than methyl bromide is

in agreement with the results of [Hole \(1981\)](#) who studied seven different insect species of stored product beetles that were subjected to phosphine and methyl bromide treatments. The tolerance pattern of *T. putrescentiae* eggs to both fumigants was similar to the tolerance of the mobile stages; this observation is in agreement with the results of [Barker \(1967\)](#) who reported eggs to be only slightly more tolerant to MB compared with mobile stages. MB required nearly a two-fold higher concentration to kill 99% of mites when compared with estimates for mortality from phosphine ([Table 3](#)). Several factors influence fumigant activity in controlling mites ([Boczek 1975](#), [Bowley and Bell 1981](#), [Jian et al. 2000](#)). [Amaro \(1963\)](#) found that the tolerance of the eggs of *Acarus siro* L. to MB greatly varied with age. He also reported that the tolerance of eggs increased by a factor of five during the first 2 d and then returned to initial levels. [Boczek \(1975\)](#) reported that the integument of the MB treated eggs of *A. siro* became nonelastic, sticky, and soft. [Lewis \(1948\)](#) reported that MB clearly inhibits the activity of papain, urease, and the respiratory enzymes which profoundly affect embryonic development. [Jian et al. \(2000\)](#) investigated the toxic action of phosphine on the adults of *T. putrescentiae* and concluded that the uptake of phosphine by the adult mite increased as concentration increased.

Fumigation of food products accompanies the concerns over food safety and also any changes in food quality with regard to consumer acceptance. Earlier work indicated that phosphine did not affect the flavor or smell of the stored animal origin products at the dosage of 1.5 g/m<sup>3</sup> for 5 d ([Friendship 1990](#)). It has been also found that the fumigation of dried fish with phosphine did not pose any residue problem with proper aeration. In a laboratory study, [Rajendran and Muthu \(1978\)](#) determined an effective methyl bromide dosage of 24 g/m<sup>3</sup> for dried salted fish with a 24-h exposure period. The earlier work by [Harris and Halliday \(1988\)](#) using a method described by [Scudamore and Goodship \(1986\)](#) has shown that residues of phosphine in dried 'bombay duck' (*Herpadon nehereus* Hamilton-Buchanan), fumigated in accordance with normal fumigation practice at the rate of 2.8 g/m<sup>3</sup>, were 0.11 mg/kg immediately after completion of the 5-d fumigation period. These residues of phosphine subsequently reduced during exposure to air to 0.04 mg/kg after 24 h, 0.02 mg/kg after 48 h, and 0.01 mg/kg after 14 d. These residue data confirmed the absence of any hazard to consumers arising from fumigation of animal origin products with phosphine and indicated a possible maximum residue limit of 0.1 mg/kg. Fumigation with MB results in the formation of residues of inorganic bromide due to reaction of the fumigant with constituents of the produce following physical sorption ([Friendship 1990](#)). Research from our group indicated that residue levels from either methyl bromide were well below maximum allowable regulatory level of 10 ppm in dry cured hams, and fumigation of these hams had no significant impact on their market quality when evaluated

**Table 3.** Probit analyses of mortality for *Tyrophagus putrescentiae* life stages fumigated with phosphine and methyl bromide for 48-h exposure periods at 23°C

Fumigants	<i>n</i>	Insect stages	LC <sub>50</sub> g/m <sup>3</sup> (95% fiducial limits)	LC <sub>99</sub> g/m <sup>3</sup> (95% fiducial limits)	Intercept	Slope ± SE	χ <sup>2</sup> values
Phosphine	140	Eggs	0.193 (0.127–0.288)	2.126 (1.035–9.605)	–0.097	0.089 ± 0.005	20.107 ( <i>P</i> = 0.065)
	360	Nymphs/adults	0.122 (0.083–0.163)	1.405 (0.784–4.215)	–0.183	0.042 ± 0.004	41.61 ( <i>P</i> = 0.001)
Methyl Bromide	120	Eggs	0.961 (0.615–2.111)	4.776 (2.151–205.73)	0.225	0.149 ± 0.022	36.74 ( <i>P</i> < 0.001)
	300	Nymphs/adults	0.845 (0.649–1.096)	3.555 (2.293–8.668)	0.099	0.095 ± 0.006	48.515 ( <i>P</i> < 0.001)

through consumer tasting panels (Sekhon et al. 2010, Zhao et al. 2015). Therefore, both fumigants can be effective for pest control with little to no residues or negative impact on the acceptability of dry-cured ham.

Methyl bromide is now banned in many countries for use in fumigating food products and related storage or processing structures, but our results with application of methyl bromide to these two pests of dry cured hams can be of value for those who can continue its use. Methyl bromide is approved and required for quarantine treatments, and existing stocks can be used for nonquarantine treatments until depleted in the United States (US EPA 2019). Therefore, a schedule of treatments for either *N. rufipes* or *T. putrescentiae* on any commodities could be written based on our results. Bond (1984) prescribes a treatment of 32 g/m<sup>3</sup> of methyl bromide for any stored products. Our results suggest that a treatment of either 20 g/m<sup>3</sup> for *N. rufipes* or about 10 g/m<sup>3</sup> for *T. putrescentiae* could control infestations of these pests, and perhaps save fumigation costs and lower methyl bromide emissions.

A key finding reported here is that phosphine could serve as an effective alternative fumigant to MB for *N. rufipes* and *T. putrescentiae* when they are pests of dry cured hams and numerous other postharvest commodities. This is very important because the fumigant sulfuryl fluoride is being promoted as a methyl bromide replacement, but it is ineffective at killing eggs of *T. putrescentiae* (Phillips et al. 2008). Although we recently reported on efficacy of various controlled atmospheres for controlling *N. rufipes* and *T. putrescentiae* (Hasan et al. 2016), such methods are not practical for current commercial applications to buildings, but would work best in chambers that can achieve low oxygen or high CO<sub>2</sub> atmospheres. Phosphine fumigation products are already registered for use in facilities like ham-production and aging plants and there are several methods to apply phosphine to achieve the concentrations needed for good kill (Thoms and Phillips 2004). One challenge for fumigating buildings like ham facilities with phosphine is to avoid or protect against the damaging corrosion that can occur from phosphine to electrical and other metallic fixtures (Zhao et al. 2015). A combination treatment of low phosphine applied with carbon dioxide under high temperature has been tested and proposed for preventing corrosion during phosphine treatments of food processing facilities (Mueller 1998). Further work is needed to develop corrosion-safe fumigation practices for phosphine to verify that phosphine can be utilized in dry-cured ham facilities.

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## References Cited

- Abbar, S., B. Amoah, M. W. Schilling, and T. W. Phillips. 2016. Efficacy of selected food-safe compounds to prevent infestation of the ham mite, *Tyrophagus putrescentiae* (Schränk) (Acarina: Acaridae), on southern dry-cured hams. *Pest Manag. Sci.* 72: 1604–1612.
- Amaro, J. P. P. 1963. Mortality and delayed development caused by methyl bromide applied to eggs of the flour mite, *Acarus siro*. Doctoral thesis, Univ. of Reading, England. pp. 258.
- Arbogast, R. T. 1991. Beetles: coleoptera, pp. 122–131. In J. R. Gorham (ed.), *Ecology and management of food-industry pests*. FDA Technical Bull. 4. Virginia.
- Barker, P. S. 1967. Susceptibility of eggs of *Tyrophagus putrescentiae* (Schränk) (Acarina: Acaridae) to methyl bromide. *J. stored Prod. Res.* 2: 247–249.
- Bell, C. H., N. Savvidou, T. J. Wontner-Smith, S. K. Cardwell, and C. Bodle. 2004. Development of sulphuryl fluoride as a fumigant for the milling industry. HGCA Project Report No. 333. Home-Grown Cereals Authority, London, UK.
- Boczek, J. 1975. Effect of methyl bromide on the embryonic development of *Acarus siro* L. (Acarina: Acaridae). *J. stored Prod. Res.* 11: 41–46.
- Bond, E. J. 1984. Manual of fumigation for insect control. FAO Plant Production and Protection Paper 54, Food and Agriculture Organization of the United Nations, Rome, Italy.
- Bowley, C. R., and C. H. Bell. 1981. The toxicity of twelve fumigants to three species of mites infesting grain. *J. Stored Prod. Res.* 17: 83–87.
- Environmental Protection Agency. 2015. Protection of Stratospheric Ozone: The 2016 Critical Use Exemption From the Phaseout of Methyl Bromide. Code of Fed. Reg. 40 CFR Part 82. <https://www.federalregister.gov/documents/2015/10/15/2015-26301/protection-of-stratospheric-ozone-the-2016-critical-use-exemption-from-the-phaseout-of-methyl>
- Fields, P. G., and N. D. White. 2002. Alternatives to methyl bromide treatments for stored-product and quarantine insects. *Annu. Rev. Entomol.* 47: 331–359.
- Friendship, R. 1990. The fumigation of dried fish. *Trop. Sci.* 30: 185–195.
- Hagstrum, D. W., T. W. Phillips, and G. W. Cuperus. 2012. Stored Product Protection. Kansas State University, Manhattan, KS. KSRE Publ; S–156.
- Hanson, D. J., G. K. Rentfrow, M. W. Schilling, W. B. Mikel, K. Stalder, and N. L. Berry. 2014. Ripened meat products: U. S. products. Ch. 40, pp. 347–354. In F. Toldra (ed.), *Handbook of Fermented Meat and Poultry*. 2nd ed. Wiley Press, West Sussex, UK.

- Harris, A. H., and D. Halliday. 1988. Residue of Fumigants in Dried Fish. Unpublished Report, ODNRI, London.
- Hasan, M. M., M. J. Aikins, W. Schilling, and T. W. Phillips. 2016. Efficacy of controlled atmosphere treatments to manage arthropod pests of dry-cured hams. *Insects*. 7: 44.
- Hole, B. D. 1981. Variation in tolerance of seven species of stored product coleoptera to methyl bromide and phosphine in strains from twenty-nine countries. *Bull. Ent. Res.* 71: 299–306.
- Jian, F., D. S. Jayas, and N. D. G. White. 2000. Toxic action of phosphine on the adults of the copra mite *Tyrophagus putrescentiae* (Astigmata: Acaridae). *Phytoprotection*. 81(1): 23–28.
- Lewis, S. E. 1948. Inhibition of SH enzymes by methyl bromide. *Nature*. 161: 692.
- Marriott, N. G., and M. W. Schilling. 2004. Dry Cured Pork Research Review. White Paper. National Country Ham Association, Inc. National Country Ham Association Annual Meeting. April 2–4, 2004. Morehead City, NC. pp. 1–62.
- Mueller, D. K. 1998. Stored Product Protection. A Period of Transition. *Insect Limited Inc.*, Westfield, IN. p. 352.
- Mueller, D. K., P. J. Kelley, and A. R. VanRyckeghem. 2006. Factors affecting the efficacy of sulphuryl fluoride as a fumigant. In I. Lorini, B. Bacaltchuk, H. Beckel, D. Deckers, E. Sundfeld, J. P. dos Santos, J. D. Biagi, J. C. Celaro, L. R. D'A. Faroni, L. de O.F. Bortolini, M. R. Sartori, M. C. Elias, R. N. C. Guedes, R. G. da Fonseca, V. M. Scussel (eds.), *Proceedings of the Ninth International Working Conference on Stored-product Protection*, 15–18 October 2006, Campinas, São Paulo, Brazil, Brazilian Post-harvest Association, Passo Fundo, RS, Brazil. pp. 1117–1122.
- Phillips, T. W., M. M. Hasan, M. J. Aikens, and M. W. Schilling. 2008. Efficacy of sulfurlyl fluoride to control ham mites and red-legged ham beetles. In: *Annu. Int. Res. Conf. Methyl Bromide Altern. Emiss. Reduction*, Orlando, FL.
- Phillips, T. W., E. M. Thoms, J. DeMark, and S. Walse. 2012. Chapter 14. Fumigation, pp. 157–177. In Hagstrum, D. W., T. W. Phillips, and G. Cuperus (eds.), *Stored Product Protection*. Kansas State University, Manhattan, KS.
- Rajendran, S., and M. Muthu. 1978. A laboratory study on the minimum dosage requirements for effective methyl bromide fumigation of food commodities. *International Pest Control*. 20: 5–9.
- Rajendran, S., and K. M. H. Parveen. 2005. Insect infestation in stored animal products. *J. Stored Prod. Res.* 41: 1–30.
- SAS. 2002. User's Guide, v. 8. SAS Institute, Cary, NC.
- Scudamore, K. A., and G. Goodship. 1986. Determination of phosphine residues in fumigated cereals and other foodstuffs. *Pesticide Sci.* 17: 385–395.
- Sekhon, R. K., M. W. Schilling, T. W. Phillips, M. J. Aikens, M. M. Hasan, A. Corzo, and W. B. Mikel. 2010. Effects of phosphine and methyl bromide fumigation on the volatile flavor profile and sensory quality of dry cured ham. *Meat Sci.* 86: 411–417.
- Thoms, E., and T. W. Phillips. 2004. Chapter 20. Fumigation, pp. 1165–1261. In S. Hedges, ed. *Mallis Handbook of Pest Control*, 9th ed. G.I.E. Media Inc., Valley View, OH.
- United States Department of State. 2019. The Montreal Protocol on Substances That Deplete the Ozone Layer. <https://www.state.gov/key-topics-office-of-environmental-quality-and-transboundary-issues/the-montreal-protocol-on-substances-that-deplete-the-ozone-layer/>. Accessed 30 August 2019.
- United States Environmental Protection Agency. 2019. Methyl Bromide <https://www.epa.gov/ods-phaseout/methyl-bromide>
- Zhao, Y., S. Abbar, T. W. Phillips, and M. W. Schilling. 2015. Phosphine fumigation and residues in dry-cured ham in commercial applications. *Meat Sci.* 107: 57–63.