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POTENTIAL FOR OZONE FUMIGATION AGAINST ANOBIID BEETLES INFESTING STORED PRODUCTS AS AN ALTERNATIVE TO METHYL BROMIDE

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ABSTRACT

Most anobiids are wood boring beetles, but two species, the cigarette beetle, *Lasioderma serricornis* (F.) and the drugstore beetle, *Stegobium paniceum* (L.), are serious pests of stored products. They are the most common storage insect pests found in botanicals. Both species infest dry plant or animal materials, they have been recorded from a wide range of foods, but their distributions are more temperate than tropical. We investigated the efficacy of ozone as a methyl bromide alternative for controlling two anobiids viz. *L. serricornis* and *S. paniceum*. The ozone fumigation showed promise for controlling both the species. Ozone treatment at concentrations ranging from 150 to 195 ppm was highly effective against adults of both species after 36 h of exposure at room temperature (24°C). Generally *S. paniceum* was more tolerant to ozone treatment than *L. serricornis*. Results showed ozone concentration requires 144.73 ppm to kill 99% of adult *S. paniceum* while, 107.42 ppm was needed for *L. serricornis*. Ozone treatment at the low concentration of 24 ppm showed no effective control against both species since the mortality rate was less than 50%, even at 48 h exposure. The work reported suggests that the ozone treatment could be a fumigant alternative to environmentally hazardous chemicals for controlling two key botanical insect pest species.

Key words: Botanicals, anobiids, non-chemical alternatives, ozone, fumigation, cigarette beetle, drugstore beetle, stored products.

INTRODUCTION

Stored-product pests are responsible for tremendous damage and economic losses to post-harvest products, stored grains and seeds, packaged food products, and animal and plant derived items and other durable commodities. There are over 1000 described species of Anobiidae (Coleoptera). Most anobiids have wood boring larvae, but two, the cigarette beetle, *Lasioderma serricornis* (F.) and drugstore beetle, *Stegobium paniceum* (L.) are important pests of stored products. The most common insects found in botanicals are *L. serricornis* and *S. paniceum* (Abdelghany et al. 2010). *L. serricornis* is also the most serious pest of stored tobacco, tobacco products, cereal grains and processed foods (Maroof and Phillips 2008) while *S. paniceum* frequently infests dry plant or animal materials, it has been recorded from wide range of food, but its distribution is more temperate than tropical (Abdelghany et al. 2010). Control of both species around the world is primarily dependent upon continued

applications of fumigation including phosphine and methyl bromide (MB) (White and Leesch, 1995; Abdelghany et al. 2010). Although effective, repeated use of fumigants has disrupted biological control by natural enemies and led to outbreaks of insect species, development of resistance to the chemical, undesirable effects on non-target organisms, and environmental and human health concerns (Champ and Dyte, 1976; Phillips and Throne, 2010). Therefore, it is essential to develop alternative pest control techniques for protecting stored commodities.

Ozone (O₃) has received recent attention in disinfecting stored foodstuffs, particularly durable products as an alternative to MB (Sousa et al. 2008; Işikber and Öztekin, 2009; Bonjour et al. 2011). There are only a few published data on the effectiveness of ozone as an insecticide against stored products insect pest (Erdman, 1980; Mason et al. 1999; Kells et al. 2001; Mendez et al. 2003). Therefore, this study was undertaken to investigate the effect of ozone toxicity on the adult annobids *L. serricornis* and *S. paniceum*.

MATERIALS AND METHODS

Insects

The adult beetles *L. serricornis* and *S. paniceum* used in the ozone experiment were obtained from the Center for Grain and Animal Health Research, USDA-ARS, Manhattan, USA. Subsequent cultures were routinely maintained on white flour (95%) and brewers yeast (5% by weight) at 27°C and 60–70% relative humidity (r.h.), in a constant 16 h light: 8 h dark lighting regime in the Department of Entomology, Kansas State University, Manhattan, USA.

Experimental procedures

To determine the ozone efficacy against *S. paniceum* and *L. serricornis*, mixed sex adults of both species were collected from the stock culture. For each species of annobids, ten insects were placed in cylindrical glass vials (4.5 cm in height by 1.2 cm in diameter), and then five glass vials for each species were placed in an Erlenmeyer glass flask (1L) for ozone treatment. The vials had lids with the inside top edge coated with liquid Fluon (polytetrafluoroethylene; Northern Products, Woonsocket, RI) to inhibit insect climbing and with 0.42-mm openings in the caps to permit air and ozone entry yet prevent beetles from escaping. A small quantity of diet was added to each vial. Lids were screwed on tightly after insects and the diet had been placed in the vials. The ozone concentrations evaluated were 24, 59, 96, 128, 146, 153 and 195 ppm and exposures were 3, 6, 12, 18, 24, 30, 36 and 48 h. The adult mortality was assessed 24 h later. Ozone concentration–time exposure response data were subjected to probit analysis (SAS, 2004), for generating mortality curves. Ozone application was carried out in an ambient temperature of approximately 25°C and 55–60% r.h., while the control chambers were placed in atmospheric air under the same conditions.

Ozone generation and system functioning

An ozone generator of laboratory scale was provided by the Adaptive Ozone Solutions Inc., Kansas, USA (<http://adaptiveozone.com/>). Ozone gas was generated from ambient room air using an electric discharge system. The air flow rate was adjusted to 2L min⁻¹. The generated ozone was initially injected into and stored in a large plastic chamber (approx. 48 L) to facilitate uniform distribution of the air-ozone mixture into the insect treatment chambers (Fig 1). The amount of generated ozone was regulated by adjusting the electric tension through a voltage regulator (dosing button). The exhaust of the ozone-treated chamber was connected to a Tygon tube and then passed through an Erlenmeyer glass flask (1L) containing water, before releasing into the insect chamber, to humidify the gas mixture and minimize the desiccation of test insects. The ozone carrier flow rate was adjusted to 2 L min⁻¹ while entering into the Erlenmeyer glass flasks (1L) containing insect samples. The output of the insect treatment flasks were directly connected into the ozone monitoring instrument through

a filter chamber for cleaning the generated ozone. The ozone concentration indicated by the generator was checked with a continuous UV ozone monitor (model IN-2000 LoCon Ozone Analyzer, USA) and the data were directly recorded onto a laptop computer equipped with appropriate software (Taltalk ver. M5000).

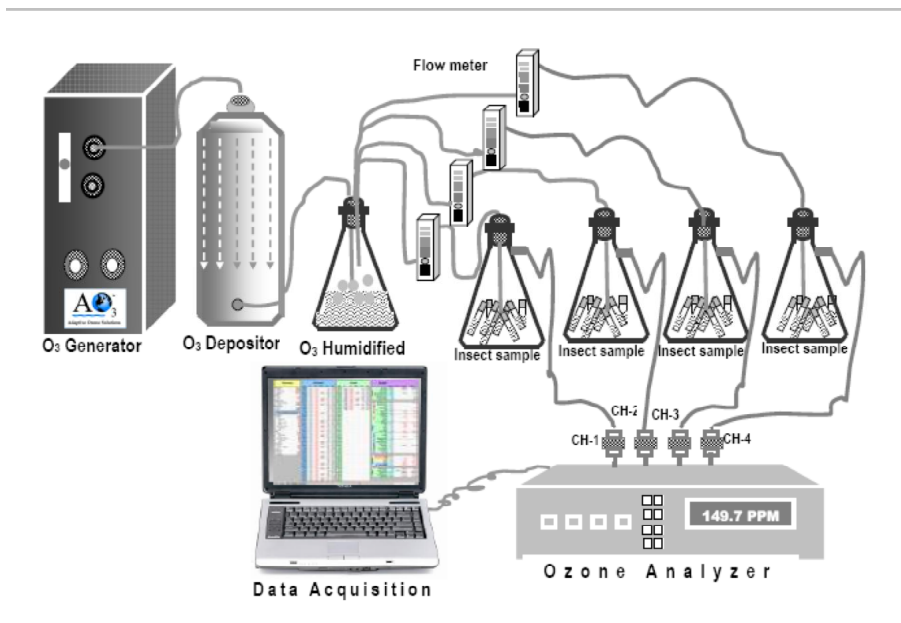


Fig. 1- Schematic of ozone generation and monitoring system.

RESULTS AND DISCUSSION

The probit estimates derived from mortality responses of adult *S. paniceum* and *L. serricornis* exposed to ozone at different exposure periods and gas concentrations are summarized in Tables 1 and 2. In general, adult *S. paniceum* were found to be more tolerant than *L. serricornis*. An ozone concentration of 195 ppm caused 100% mortality in *L. serricornis* at 12 h exposure (Fig. 2), while in *S. paniceum* the same concentration required 24 h exposure (Fig. 3). At longer exposures, i.e. 48 h, 100% kill was achieved with 24 ppm and 59 ppm ozone, for *L. serricornis* (Fig. 2) and *S. paniceum* (Fig. 3) respectively.

Table 1. Probit analyses of mortality for *S. paniceum* fumigated with ozone at different exposure periods.

Exposure h	N	LC ₅₀ ppmv (95% fiducial limits)	LC ₉₉ ppmv (95% fiducial limits)	Slope ± SE	Intercept±SE	χ ² values (df)
12	55	87.86 (73.98 – 100.85)	344.12 (258.61 – 561.62)	13.26 ± 0.91	33.89 ± 6.14	177.53 (53) P=0.0001
18	30	72.94 (65.59 -80.82)	276.75 (220.65 – 382.54)	16.13 ± 0.83	8.65 ± 4.97	35.46 (28) P=0.15
24	50	65.76 (57.13 – 73.64)	215.69 (179.65 – 281.13)	14.09 ± 0.75	11.15±5.75	85.14 (48) P=0.008
30	25	50.64 (35.99 – 64.55)	296.15 (176.21 – 1093.00)	11.34 ± 1.25	8.75 ± 8.15	66.98 (23) P=0.001
36	40	37.14 (31.77 - 42.10)	144.73 (119.75 - 188.76)	14.13± 1.05	-13.19 ± 8.67	31.81 (38) P=0.75

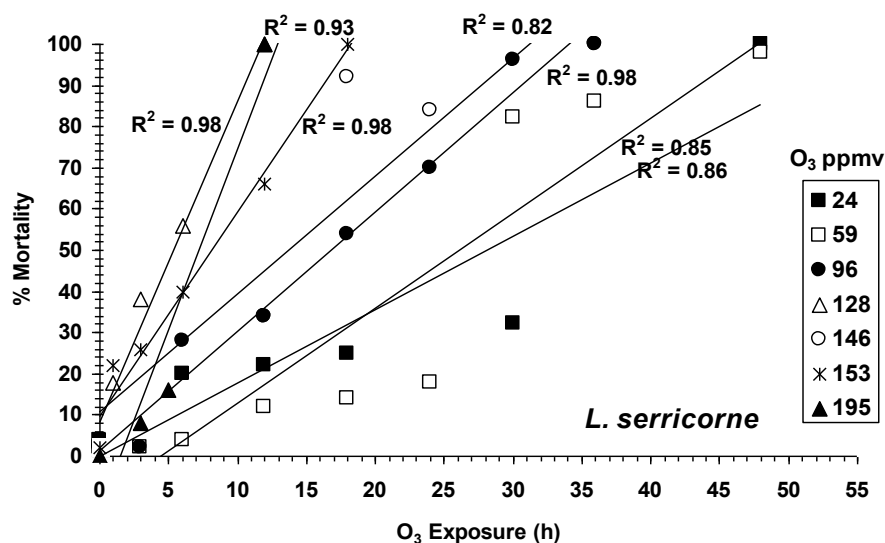


Fig. 2- Mortality of adult *L. serricorne* fumigated with ozone at different concentrations and exposures.

Table 2. Probit analyses of mortality for *L. serricorne* fumigated with ozone at different exposure periods

Exposure h	N	LC ₅₀ ppmv (95% fiducial limits)	LC ₉₉ ppmv (95% fiducial limits)	Slope ± SE	Intercept±SE	χ ² values (df)
12	55	102.79 (81.29 – 127.46)	631.73 (373.05 – 2033.00)	11.62 ± 1.21	52.37 ± 7.38	264.05 (53) P=0.001
18	30	78.36 (60.43 – 102.25)	351.16 (217.18 – 1017.00)	14.85 ± 1.04	19.62 ± 6.09	120.89 (28) P=0.0001
24	50	70.09 (54.65 – 84.26)	285.98 (207.81 – 513.17)	14.09 ± 0.75	11.15±5.75	215.50 (48) P=0.0001
30	25	22.14 (9.01 – 31.88)	195.85 (118.51 – 766.05)	10.69 ± 1.65	-7.48 ± 13.15	43.36 (25) P=0.006
36	40	31.17 (26.36 – 35.56)	107.42 (88.70 – 141.77)	14.13 ± 1.05	-13.19 ± 8.67	24.57 (38) P=0.95

Mortality of adults of both the species was significantly ($P > 0.001$) higher at the medium (>128 ppm) and high (>153 ppm) ozone levels than at the low (> 24 ppm) levels after 24 h exposure. However, 100% mortality was never reached, even at the high ozone level at lower (< 24 h) exposure. The lethal time estimated for 50% and 99% mortality trended longer in *S. paniceum* than in *L. serricorne*, corresponding to the respective ozone concentrations (Figs. 2 and 3). These trends clearly indicate that adults *S. paniceum* were more tolerant to ozone fumigant than *L. serricorne*. The mortality data of adults of both species tested with ozone fumigation agreed with those of several authors who observed toxicity of ozone against different stored products insect pests (Erdman 1980; Mason et al. 1999; Kells et al., 2001; Sousa et al. 2008; Bonjour et al. 2011).

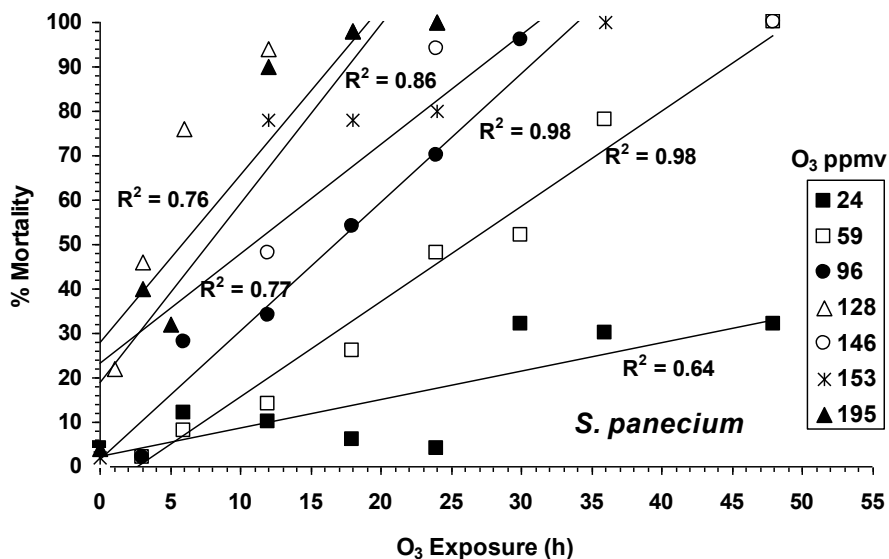


Fig. 3- Mortality of adult *S. paniceum* fumigated with ozone at different concentrations and exposures.

Erdman (1980) investigated the efficacy of ozone against *T. castaneum* and *T. confusum* by mortality of larvae. Maize treated with 50 ppm ozone for 3d resulted in 92–100% mortality of adult *T. castaneum*, *Sitophilus zeamais* and *Plodia interpunctella* larvae and 63% reduction in the fungus *Aspergillus parasiticus* Speare on the kernel surface (Kells et al., 2001). In a follow-up study, Mendez et al. (2003) investigated the effects of ozone exposure on the food end-use properties of maize, popcorn (*Z. mays* L. var. *everta*), soybean (*Glycine max* (L.) Merr.), wheat (*Triticum aestivum* L.), and rice (*Oryza sativa* L.). Ozone has been demonstrated as an effective fumigant insecticide and fungicide to extend the storage life of dried maize; however, ozone has not been examined as a method to increase the allowable storage time of high-moisture maize. Sousa et al. (2008) assessed ozone toxicity to phosphine-resistant pests of stored products: no tested populations showed resistance to ozone. All these results reveal that ozone is a potential fumigant for stored products.

Therefore, the use of ozone against *S. paniceum* and *L. serricornis* seems a promising management alternative to MB. The species *L. serricornis* is generally much more susceptible to gaseous ozone than *S. paniceum*. Furthermore, detailed studies are required to determine mortality responses of immature life stages of anobiids and to show the products' quality that are frequently infested by the anobiids after exposing ozone fumigation.

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