Controlled Release Fumigation of the Greater Wax Moth[1,2]

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ABSTRACT

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Polyethylene film controlled release packets utilizing ethylene dibromide (EDB) and p-dichlorobenzene (PDB) were designed and tested against late instar larvae of *Galleria mellonella* (L.). EDB controlled release packets gave 100% control of larvae in 48 h under experimental conditions at 26.6° and 32.3°C. PDB controlled release packets gave 13.3 and 17.2% control of larvae in 96 h under similar temperature regimes.

Permeation constants for the EDB and PDB controlled release systems were derived from experimental rates observed in 4 and 6 mil polyethylene dispenser packets at temperatures of 10.0°–35.0°C.

Damage to honey combs caused by the greater wax moth, *Galleria mellonella* (L.), resulted in an estimated $4 million loss to U.S. beekeepers in 1976 (Williams 1976). The nature of the damage and cost of replacement has been reported by Oertel (1969) and Williams (1976). The effectiveness of 1,2 dibromoethane (EDB) and p-dichlorobenzene (PDB) fumigation against greater wax moths has been reported by Vorwohl (1965) and Lehnert and Shimanuki (1967). Posttreatment reinfestation is a problem, especially in the southern U.S. where the greater wax moth is commonly multivoltine.

Controlled release delivery systems exhibit characteristics for insect control and are well suited for control where multiple generations and reinfestations are common.

The rationale behind controlled release wax moth fumigation includes long term protection against infestation and reinfestation, reduced operator exposure to noxious and potentially toxic insecticides, and relative ease of application. Several other features of the controlled release system make it appealing for wax moth control. For example, permeation and duration of the toxicant can be modified to match environmental requirements through manipulation of surface area, film type, thickness, and dosage. Also, permeation and larval activity are both influenced by ambient temperature, which means toxicant emission will parallel larval development. In addition, it may be possible to incorporate fumigant mixtures in controlled release delivery systems which would act in 2 phases, i.e., a primary rapid release of toxicant such as EBD followed by a secondary long term release of PDB. An overview of controlled release pesticide systems was presented by Knapp (1977). A review of polymer permeability and the factors controlling permeation was presented by Lebovits (1966). The efficacy of a controlled release dispenser modeled after Toba et al. (1969) and St. Clair et al. (1972) utilizing EDB and PDB for greater wax moth fumigation is reported here.

Materials and Methods

Heat sealed dispenser packets were constructed with inner dimensions of 8.9×10.2 cm and a surface area of 181.6 cm^2. Test compounds were sealed into dispenser packets either as free crystals or as liquid sorbed onto pieces of pressed fiberboard. Permeation rates for test compounds were determined over a range of temperatures, 10.0°–35.0°C, and in 4 and 6 mil polyethylene tubing (actual thickness, 3.5 and 5.5 mil). Samples were hung in Percival® controlled environment chambers, and weight losses were determined gravimetrically with a Mettler H64 analytical balance. Three test packets were monitored simultaneously at each temperature regime. A minimum of 5 weighings was recorded for each packet. Permeation rates were computed by dividing fumigant weight loss by elapsed time and then averaged.

Mixtures of EDB and PDB in the ratio of 1:2 were prepared with the addition of dichloromethane as an organic solvent. These mixtures were similarly monitored for permeation rates in controlled release packets. In a separate test, vapors emanating from a controlled release packet containing the EDB-PDB mix were captured in ice-packed bubble traps containing redistilled hexane. Aliquots taken periodically from the traps were examined with a Varian Aerography (Series 2700) gas chromatograph for traces of the mixture components.

Larvae were reared on a nutrient medium adapted from the original formula of Marston and Campbell (1973). The proportions of glycerine and water were modified to maintain ca. 80% RH in the test chambers.

Fumigation tests were conducted in standard 5 frame nucleus hive boxes (nucs) with an internal volume of 0.023 m^3 placed into controlled environment chambers. Four test frames, each holding 45 larvae, were placed in each nuc with a controlled release delivery packet. Each frame held 9 circular cages in rows of 3. Each cage contained 5 larvae and ca. 20 g of nutrient medium. Cages were fabricated from 2 standard Mason® jar screw caps with plastic screen in place of the metal sealing lid. Two of these caps when taped together, open side inward, created a cage with 2-way circulation and a volume of 34.9 cm^3. At specified times after the start of each test, i.e., 6, 12, 24, and 48 h for EDB and 24, 72, and 96 h for PDB, a frame was randomly selected from each nuc and the larvae were observed for mortality. Any remaining live larvae were placed on fresh medium, held for 72 h and again observed for postexposure mortality.

Results and Discussion

Table 1 presents permeation data for EDB and PDB in 4- and 6-mil dispenser packets held at constant tem-

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Table 1.—Permeation data for EDB and PDB in 4- and 6-mil polyethylene test packets.

°C	EDB[a] g/h	p[b] constant	PDB[a] g/h	p[b] constant
		6 mil		
10.0	0.061	57.51	0.007	6.60
12.8	0.057	53.74	0.008	7.54
15.5	0.093	87.69	0.012	11.31
18.3	0.099	93.34	0.018	16.97
21.1	0.130	122.57	0.059	55.63
23.9	0.201	189.51	0.072	67.89
26.6	0.242	228.17	0.076	71.66
29.4	0.327	308.31	0.095	89.57
32.2	0.490	462.00	0.142	133.89
35.0	0.583	549.69	0.172	162.17
		4 mil		
10.0	0.168	100.80	0.014	8.40
15.5	0.403	241.80	0.038	22.80
21.1	0.574	344.40	0.060	36.00
26.2	0.834	500.40	0.141	84.60
32.2	1.556	933.60	0.253	151.80

[a] Permeation rates for EDB and PDB from test packets with a surface area of 181.6 cm^2. Each value represents the grand mean of 15 observations on 3 test packets.

[b] The permeation constant (p) is defined as the rate of permeation multiplied by the thickness of the membrane and divided by the product of its area and the difference in concentration in the adjacent compartments separated by the plastic membrane, i.e., g/24 h/645.2 cm^2/mil film thickness.

Table 2.—Greater wax moth larval mortality following exposure to controlled release packets containing EDB and PDB.

°C	% larval mortality 72 h following treatment	
	Test	Control
	EDB (6 mil) - 48-h exposure	
10.0	18.3	7.2
15.5	57.8	5.6
21.1	77.8	2.8
26.6	100.0	1.1
32.2	100.0	2.2
	PDB (4 mil) - 96-h exposure	
10.0	1.7	1.7
15.5	3.9	3.3
21.1	2.8	1.1
26.6	13.3	1.7
32.2	17.2	2.2

perature. Rate of permeation increased exponentially with increasing temperature. Dispenser packets were also tested under changing temperature regimes, and it was found that a mathematical weighted average (estimating permeation from the proportional amount of time spent at different temperatures) approximated (±5%) the actual observed weight loss of toxicant. Mixtures of EDB, PDB, and the dichloromethane solvent were tested, and although, permeation data were inconclusive, GC separation of transmitted vapors revealed that all mixture components were present in the emission.

Over 3600 greater wax moth larvae were tested, 1800 each for EDB and PDB dispenser packets. Table 2 summarizes the mortality data. Analysis of variance indicated a significant difference in mortality ($P<0.05$) between fumigated and nonfumigated larvae. Test temperatures and exposure time also had significant effects on larval mortality. In regression analyses for both EDB and PDB, only the variable chemical and the interactions of chemical × temperature × time, chemical × temperature, and chemical × time, significantly affected mortality in the test larvae. All other interactions and independent effects were dismissed. In PDB analyses, the interaction of chemical × temperature narrowly missed being significant at the 5% level but is included in the discussion because of the overall similarity between EDB and PDB regressions models and the lower volatility of PDB compared to EDB. For EDB, these 4 variables accounted for 81% ($R^2 = 0.81$) of the variation in larval mortality; for PDB, 21% ($R^2 = 0.21$). In both test situations, there appeared to be a critical temperature and exposure time above which mortality rose rapidly. For EDB, these were 21.1°C and 12-h exposure; for PDB, 21.1°C and 72-h exposure. The difference in time may be a reflection of the differences in volatility and subsequent activity of PDB vs. EDB packets. The similar temperature suggests that at 21.1°C both fumigants begin to permeate rapidly enough to effect the larvae and that at 21.1°C larval metabolism increases to a point where the fumigant is assimilated rapidly enough to become toxic. With both treatments, cumulative mortality increased in proportion to toxicant dosage, which reflected the exposure time and the permeability of the delivery system at different temperatures.

We believe that the incorporation of EDB in a controlled release delivery system offers potential advantages in the commercial application of fumigants for the control of honeycomb depredators. The advantages of a PDB controlled release system do not appear to warrant the additional cost of materials and preparation.

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