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Dermal and Respiratory Exposure of Mixers/Sprayers to Acephate, Methamidophos, and Endosulfan During Tobacco Production

J. A. Lonsway, M. E. Byers, Hl. A. Dowla, M. Panemangalore. G. F. Antonious

Community Research Service, Atwood Research Facility, Kentucky State University, Frankfort, Kentucky 40601, USA

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Exposure of tobacco farm workers to pesticides, plant growth regulators and nicotine can occur during the various stages of tobacco production. Exposure occurs during mixing-loading, setting, spraying, "topping", cutting, hanging and stripping. Often, the same person performs all of these procedures. Kentucky's farmers grow more than 175,000 acres of tobacco annually (Anonymous 1994). Acute illness in tobacco workers in Kentucky has become a concern. This has been termed green tobacco sickness (GTS) and probably results from exposure to nicotine from wet tobacco leaves (Gelbach 1979; Ghosh 1991).

Formulations of acephate, an organophosphorus systemic insecticide and endosulfan, an organochlorine insecticide and acaricide are commonly used to spray reproductive stage tobacco in Kentucky. Each is mixed with the plant growth regulator, maleic hydrazide.

Dermal and respiratory routes are the major avenues of exposure to insecticides and natural products for agricultural workers (Durham and Wolfe 1962). Exposure has been quantified for various mixer/sprayer and field worker situations (Byers et al. 1992; Ghosh et al. 1991; Lavvy and Mattice 1985). However, such exposure to tobacco farm workers is undefined and health risks associated with tobacco production remain in question.

The objectives of this research were to quantify the dermal and respiratory exposure of the mixer/sprayer to commonly used insecticides during the mixing and spraying processes and to assess the potential risk to the mixer/sprayer.

This report is part of a larger study measuring exposure of tobacco farm workers to these insecticides and nicotine during field reentry, cutting and hanging. Another aspect of this research project involved biomarker determination through correlation of enzyme inhibition in farm worker blood with exposure to agricultural chemicals (Dowla et al. 1996).

Correspondence to: M. E. Byers

MATERIALS AND METHODS

In 1994 and 1995, a mixture of acephate (Orthene 75, O,S-dimethyl acetylphosphoramidothiate; Valent Chemical Inc., Walnut Creek, CA) and maleic hydrazide (Royal MH-30; 6-hydroxy-2H-pyridazin-3-one; Uniroyal Chemical Inc., Middlebury, CT) was prepared and applied to reproductive stage burley tobacco in Hart County, Kentucky at rates of 1 and 2 kg active ingredient (AI) per hectare, respectively. Application was done by local farmers using typical methods. Chemicals were poured by hand into a 568 L spray tank which was half full of water. The remainder of the tank was then filled with water using a garden hose. Applications were made to adjacent rectangular plots (2.025 ha) by means of a tractor mounted boom sprayer. The boom extended half the width of each plot. Tobacco plant density was approximately 3900 plants per ha and row spacing was 1.02 m. In 1995 a mixture of endosulfan (Golden Leaf Tobacco Spray; 1,4,5,6,7,7-hexachloro-8,9,lO-trinorborn-5-en-2,3-ylenebismethylene sulphite; FMC Corporation, Philadelphia, PA) and maleic hydrazide was prepared and sprayed in the same manner. In addition, a mixture of endosulfan and maleic hydrazide was prepared and applied to similar tobacco plots via an open air highboy in Franklin County, Kentucky. All plots were chosen randomly. The tasks of the mixer/sprayer in all cases were to pour, mix, and spray acephate and maleic hydrazide or endosulfan and maleic hydrazide.

Exposure times for each event were recorded. Weather data including temperature, humidity, wind speed, wind direction, and general conditions were collected. One mixer/sprayer was monitored for dermal and inhalation exposure in 1994 through three events each for mixing and spraying and four participants were monitored in 1995 through four mixing and seven spraying events.

The mixer/sprayer's dermal exposure to acephate, it's metabolite methamidophos, and endosulfan was monitored using a modified adsorbent gauze pad technique (Durham and Wolfe 1962). Gauze pads were constructed according to Byers et al. (1992). The exposed surface area of each pad was 6.4 by 6.4 cm. Eight pads were used to monitor dermal exposure. Five pads with outer cotton/polyester swatch were attached to front trunk (chest), back trunk (back), upper arm, thigh, lower leg. Penetration of chemicals through the swatch onto the gauze pad represented penetration of chemicals through a clothing layer to bare skin. Three exposure pads without outer swatch layer (gauze exposed) were attached to face/front neck, head/back neck, and forearm. These gauze pads represented exposure to unclothed skin. Cotton gloves were used to determine hand exposure. Respiratory

exposure was measured using personal type air samplers (Gilian Instrument Corp., W. Caldwell, New Jersey). The air sampler pumps were attached to the waist of the mixers/sprayers. Each pump was connected to a Tygon tube which extended to an air sampler intake tube containing carbon filled air sampler cartridges (Supelco ORBO-32, Bellefonte, PA) which was placed in the breathing zone of the mixers/sprayers. Each air sampler was field calibrated for each exposure event to operate at an intake flow rate of 2 L/min. The mixers/sprayers wore exposure pads, gloves, and air sampler units for the duration of each mixing and spraying event. Immediately following mixing and spraying, all pads, gloves, and charcoal adsorbent air sampler cartridges were removed, placed in individual ziploc bags, stored on ice, transported to the laboratory, and stored frozen until extraction.

Percent transfer from outer cotton layer to gauze was calculated by dividing the insecticide quantity penetrated to the gauze (through the swatch) by total quantity on swatch plus gauze and multiplying by 100. Total hand exposure was determined using cotton gloves.

To determine insecticide stability during handling and storage of samples, gauze pads (n=3) were fortified with 10 ug technical grade acephate, methamidophos, and endosulfan on each day of sample collection. Field fortified samples were extracted and analyzed in the same manner as the exposure pads.

For extraction, exposure pads were cut diagonally in half. One half was extracted for GC analysis of acephate, methamidophos, endosulfan, nicotine, and cotinine while the other half was stored. Gauze pads and cotton swatch portions were extracted separately in 25 and 50 mL HPLC grade methanol. Gloves were extracted in a similar manner using 75 mL methanol. Fleakers were closed with duraseal-lined polypropylene caps and mechanically agitated for 60 minutes on a wrist-action shaker. Extracts were poured through filter paper (Whatman no. 40) and 3 g anhydrous sodium sulfate. All extracts were concentrated by rotary evaporation and a nitrogen stream, reconstituted with ethyl acetate to 1mL and stored at -20° C until chemical analysis was performed. Cleanup of glove extracts was accomplished by pouring them through a Cl8 Solid Phase Extraction column (Varian, Harbor City, CA) and eluting with methanol. Residues were extracted from the air sampler charcoal adsorbents by sonicating the contents of the air sampler cartridge two times each for 15 minutes in 3 mL methanol, acetone, and ethyl acetate, respectively for those events where acephate was the sprayed product. Air sampler cartridges containing endosulfan residues were extracted

by sonicating the contents for 15 minutes each in 3 mL methanol, ethyl acetate, methylene chloride, and hexane respectively.

Acephate and methamidophos were analyzed using gas liquid chromatography (GC, Hewlett Packard Company, Model 5890 Series II, Palo Alto, California) equipped with a nitrogen phosphorus detector (NPD). Endosulfan was analyzed using GC mass spectrometry (MS). GC/MS operated in selective ion monitoring mode was used to confirm acephate and methamidophos (HP Model 5971A mass selective detector). The GC column used was a capillary RTX-5 (5% diphenyl-95% dimethyl polysiloxane), 30 m x .53 mm-id. GC run parameters included: 250° C injection temperature, 260° C detector temperature; oven program was 130° C for 7 minutes, 5.0° C/min increase to 160° C, this was held for 1 minute, 25 deg/min until a final temperature of 225° C was reached. Equilibrium time was set at 3 minutes. Gas flows were set at 15, 20, 120, and 5 mL/min for carrier (He), auxiliary (He), air and hydrogen respectively. Retention times for acephate, methamidophos, and endosulfan were 15.4, 9.6, and 21.6 minutes, respectively. Minimal detectable levels for acephate, methamidophos, and endosulfan were .05, .0l, and .25 ppm, respectively. Quantitation was based on average peak areas from two consecutive injections obtained from pure external pesticide standards and regression analysis.

Insecticide quantities were converted from ug/mL of analyzed sample to ng/cm^2 based on pad area (20.5 cm^2 gauze pad) and then to mg/body region/hr. The body surface area (cm^2) for the mixers/sprayers was calculated from the mixer/sprayer's height and mass (Dubois and Dubois 1916). Using the procedure of Berkow (1931), the total body surface area was divided into regions. Total dermal exposure was determined by combining the exposure of clothed and unclothed body regions with the hand exposure. Exposure to unclothed body regions (face plus "v"-front neck, head plus back of neck and forearms) was estimated by multiplying the area of each region with appropriate exposure rates for exterior pads. Hand exposure was determined by adding the insecticide amounts detected on the right and left gloves. Respiratory exposure was calculated from the insecticide extracted from the charcoal contents in the air sampler (ug/mL) and multiplying this amount by the average respiration rate of a man engaged in light work 1740 l/hr (Durham and Wolfe 1962; U.S. EPA 1989). The acute percutaneous LD_{50} for acephate, methamidophos, and endosulfan are 2000, 130, 359 mg/kg, respectively (Tomlin 1994). No observed adverse effect level (NOAEL) values for acephate, methamidophos, and endosulfan are 30, 2, and 3 mg/kg/day, respectively (Tomlin 1994). The percentage of acute

toxic dose per hour (PATDH) and margin of exposure (MOE) were calculated for each event, mixing and spraying according to Byers et al. (1992).

RESULTS AND DISCUSSION

Recovery rates of fortified gauze pads for acephate, methamidophos, and endosulfan were 97%, 99%, and 102%, respectively. Recoveries of fortified air sampler cartridges were 99%, 98%, and 95%, respectively. Mean total exposure times for mixing and spraying were 5.64 and 29.56 min, respectively. Weather data collected in 1994 and 1995 during mixing and spraying included: mean temperature 30° C; mean humidity 69%; and mean wind speed 2.10 m/set.

The EPA recommended human exposure value of 70 kg was used to calculate mg/kg/day values (US EPA 1989). The average body weight of the mixers/sprayers 102.3 kg. In general, during mixing events, the hands were the primary source of dermal contamination for all three insecticides (Tables 1 and 2). For acephate, methamidophos, and endosulfan the hands received 72%, 51%, and 82% of overall dermal exposure. The thighs and lower legs were among the most exposed body regions during spraying. This was probably due to the orientation of the boom in relation to the legs. For

Table 1. Estimated mean dermal exposure to workers involved in mixing insecticides in Hart and Franklin Counties, Kentucky in 1994 and 1995.

		Insecticide mean (±SD)		
Body Region	Area* (cm^2)	Acephate	Methamidophos	Endosulfan
		mg/hr**		
Hands	1144.2	38.9 ±27.2	1.1 ±1.2	133.5 ±113.4
Front Trunk	4123.5	.5 ± 1.0	.2 ± .4	.3 ± .1
Back Trunk	4576.4	.1 ± .1	.3 ± .8	.1 ± .2
Upper Arms	1620.8	.2 ± .4	.1 ± .1	.1 ± 0.0
Back neck	667.4	.1 ± .1	0.0 ±0.0	0.0 ± 0.0
Face+V-neck	1096.4	.1 ± .1	0.0 ±0.0	.2 ± .1
Forearms	1668.5	.7 ± .5	.1 ± .1	.1 ± .1
Thighs	4409.6	1.1 ± 2.6	.3 ± .5	.9 ± .8
Lower legs	4528.7	.7 ± 1.7	.1 ± .3	.1 ± .1
Total	23,835.5*	42.4 ±55.6	2.2 ±1.6	135.3 ±181.8

* Average body region surface area for the mixers.
** Mean quantity of residue (mg/hr) for acephate, methamidophos, endosulfan n=5, n=5, n=2, respectively

Table 2. Estimated mean dermal exposure to workers involved in spraying insecticides in Hart and Franklin Counties, Kentucky via a tractor mounted boom sprayer and highboy in 1994 and 1995.

Body Region	Area* (cm^2)	Insecticide mean (±SD) Acephate	Methamidophos	Endosulfan
		mg/hr**		
Hands	1144.1	.4 ± .3	0.0 ±0.0	40.1 ±39.1
Front trunk	4576.4	.3 ± .7	.9 ±1.4	18.6 ±40.7
Back trunk	4123.5	.0 ± .0	.1 ± .2	.5 ± .7
Upper arms	1620.8	.1 ± .3	.4 ± .5	.4 ± .7
Back neck	667.4	.0 ± .1	.0 ± .0	12.9 ±28.5
Face+v-neck	1096.4	.3 ± .6	.7 ±1.6	25.4 ±55.8
Forearms	1668.5	.1 ± .2	.4 ± .5	1.6 ± 2.4
Thighs	4409.6	.7 ±1.5	1.0 ±1.4	2.4 ± 4.7
Lower legs	4528.7	.1 ± .3	1.0 ±1.5	.8 ± 1.6
Totals	23,835.5*	2.0 ±3.2	4.5 ±5.3	102.7±130.6

* Average body region surface area for the sprayers
** Mean quantity of residue (mg/hr) for acephate, methamidophos, endosulfan n=7,n=7,n=5, respectively

acephate, methamidophos, and endosulfan, lower legs and thighs combined received 38%, 45%, and 12%, respectively. For spraying, hands received 34%, 26%, and 56%, for acephate, methamidophos, and endosulfan, respectively. The hands, front trunk, and face + v-of chest (front neck), and back of neck (head) also received some exposure during spraying.

Exposure to acephate was greater for mixing than for spraying as indicated by the high concentrations found on the gloves. Exposure to methamidophos was greater for spraying likely due to the transformation to methamidophos during mixing and spraying (Nigg et al. 1981).

The mean insecticide penetration through the 50:50 cotton/polyester blend swatches was 32%, 56%, and 42% for acephate, methamidophos, and endosulfan, respectively. This transfer was random and ranged from 0% to 100% for all three insecticides.

Where acephate was the applied product, respiratory exposure only occurred to methamidophos. Respiratory exposure to endosulfan occurred only during mixing.

The mean estimated percentage of toxic dose per hour

(PTDPH) ranged from 0.002 to .541. The maximum PTDPH values of .541 and 0.413 were observed for endosulfan during mixing and spraying, respectively (Table 3).

Table 3. Exposure of mixers/sprayers to insecticides applied to tobacco via a tractor mounted boom sprayer and an open air highboy in Hart and Franklin counties, Kentucky in 1994 and 1995.

Insecticide EVENT	Dermal (w/W/d)***	Respiratory (mg/kg/d)	PTDPH* %	MOE**
ACEPHATE				
Mixing	5.13	0	.030	59
Spraying	.24	0	.002	1250
METHAMIDOPHOS				
Mixing	.26	.01	.085	56
Spraying	.54	.04	.302	21
ENDOSULFAN				
Mixing	16.18	.01	.541	1.843
Spraying	8.06	0	.413	3.722

* PTDPH=Percentage of toxic dose per hr (Durham and Wolfe 1962).
** MOE=Margin of exposure (insecticide NOAEL value divided by 10% of dermal plus 100% respiratory exposure amounts).
*** Based on average body weight 70 kg (U.S. EPA 1989).
**** Dermal LD_{50} for acephate, methamidophos, endosulfan= 2000, 130, and 359, respectively. NOAEL for acephate, methamidophos, and endosulfan=30, 2, and 3, respectively.

Due to the hazards inherent in this practice of mixing and spraying chemicals as well as differing tolerance levels of individuals, caution is advised. Limited resource farmers are generally not well equipped, particularly regarding respiratory exposure. Endosulfan exposure was notably high. According to Auslander (personal communication to MEB in 1997), a 1993 death was attributed in part to endosulfan exposure while spraying under conditions similar to this scenario. Although considered adequate by law, following the pesticide label directions is the minimal precaution which should be taken.

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