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## Effects of Four Lights on Malathion Residues on Glass Beads, Sorghum Grain, and Wheat Grain<sup>1</sup>

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### ABSTRACT

Different regions of light have different photodegradative effects on malathion deposits on glass beads, sorghum grain, and wheat grain. Temperature effects of IR light produced the most rapid disappearance of malathion, whereas far UV light, plant GRO, and near UV light exhibited a decreasing order of photodegradative activity.

Identified degradation products of malathion were: mala-oxon, malathion monoacid, malathion diacid, *O*-demethyl malathion, *O,O*-dimethyl phosphorodithioate, *O,O*-dimethyl phosphorothioate, dimethyl phosphate, and phosphoric acid. Several unidentified compounds, both thiophosphates and phosphates also were detected.

Several workers reported that malathion undergoes chemical changes after exposure to light. Cook and Pugh (1967) reported for UV light initially degraded malathion to less-polar compounds, but that prolonged exposure caused formation of more-polar compounds than the parent compound. Although not highly susceptible to photodegradation induced by far UV light, malathion breaks into several different compounds (Mitchell 1961). El-Refai (1960)<sup>4</sup> identified these degradation products in residues of malathion exposed to far UV light: mala-oxon, malathion diacid, *O,O*-dimethyl phosphorodithioic acid, *O,O*-dimethyl phosphorothioic acid, and phosphoric acid.

Most studies of malathion photodegradation have used UV light. We used IR, solar visible region, near UV, and far UV lights on deposits of malathion applied to glass beads, sorghum grain, and wheat grain. We analyzed irradiated malathion residues at

various time intervals of exposure to determine how each light affected malathion's rate of disappearance and degradation products formed.

**MATERIALS AND METHODS.—Apparatus and Reagents.**—Light Sources—Westinghouse F15T8/GRO plant GRO lamp (6550 A), General Electric F15T8/BL blacklight lamp (3660 A), General Electric G15T8 germicidal lamp (2537 A), and General Electric R-40 reflector IR bowl lamp (11000 A) were used. Each was mounted in a box 16 in. above the samples.

**Thin-layer Chromatographic Plates.**—Plastic thin-layer chromatographic plates coated with silica gel (MN silica gel N-HR) (Brickman Instruments Inc., Westbury, N. Y.) and Eastman silica gel plates (Type K301R2 without fluorescent indicator) (Distillation Products Industries, Rochester, N. Y.) were used for TLC analysis.

**Chromatographic Chambers.**—Half-gallon jars were used as chromatographic chambers. Chromatographic plates (20×10 cm) were supported in a cylindrical fashion in the jar by 2 circular wires and were developed by the ascending method.

**Chromogenic Reagents.**—DCQ (2,6-dibromo-*N*-chloro-*p*-quinoneimine) (Fisher Scientific Co., St. Louis, Mo.) in acetone (Kadoum 1970), Silver nitrate-bromophenol blue (Getz 1962), and the Hanes-Isherwood reagent (Hanes and Isherwood

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<sup>4</sup> A. R. El-Refai, 1960. Nature of chemical changes occurring in residues of malathion. Ph.D. thesis, Entomology Department, Kansas State University.

1949) were used to detect malathion and its degradation products. Detection of malathion and its thiophosphate degradation products with DCQ at sub-microgram levels was accomplished by spraying developed chromatograms with 0.5% wt/vol DCQ, heating for 5 min at 100°C, then exposing them to bromine vapor until reddish or yellow spots formed on a white background. Table 1 indicates different colors noted for the various malathion degradation products. Other chromogenic reagents were used without modification.

**Standard Solutions.**—Analytical grade standards of malathion, malathion monoacid, malathion diacid, potassium salt of *O*-demethyl malathion, potassium *O,O*-dimethyl phosphorodithioate, potassium *O,O*-dimethyl phosphorothioate, sodium dimethyl phosphate, and phosphoric acid (American Cyanamid Corp., Princeton, N. J.) were used as received. Standard solutions of 1 µg/µliter were prepared in redistilled acetone.

**Procedure.**—Thin-Layer Chromatographic Analysis.—Thin-layer chromatographic separation of malathion and its degradation products was accomplished with 4 solvent systems. Varying quantities of cleaned samples were spotted 3 cm from the bottom of the TLC plate and developed to a premarked line 15 cm from the origin. Quantities of compounds detected in light-exposed and dark (control) samples were estimated by comparing spot size and color intensity with a series of standards on the same chromatogram. Solvent systems were: (1) hexane:benzene:acetic acid (40:40:20) used with Brinkman silica gel plates; (2) petroleum ether:ethyl ether:acetic acid (60:40:1) used with Brinkman silica gel plates; (3) chloroform:methanol:30% ammonium hydroxide (75:25:4) used with Eastman silica gel plates; and (4) isopropanol:30% ammonium hydroxide (85:15) used with Brinkman silica gel plates. Reagent grade solvents for solvent systems were redistilled before use. Systems 1 and 2 most effectively separated less-polar malathion

degradation products, whereas better solution of polar compounds was obtained with systems 3 and 4. Table 1 shows  $R_f$  values.

Washing and reactivating silica gel TLC plates were not necessary for plates developed with systems 1, 2, or 3. However, plates developed with system 4 were washed with acetone and water (1:1) according to El-Refai and Hopkins (1965) and dried 30 min at 100°C. Plates deactivated by atmospheric moisture were reactivated by heating 30 min at 100°C.

**Malathion Treatment and Light Exposure.**—A standard solution of 10.0 mg/ml was prepared by dissolving analytical grade malathion (99.3%) in redistilled acetone. Twenty grams of sorghum or wheat grain, 12.5±0.05% moisture content, were treated in a 50-ml beaker by slowly pipetting 0.2 ml of 10.0 mg/ml malathion solution over the surface. The grain then was mixed thoroughly with a glass stirring rod to distribute the malathion. An initial deposit of 100 ppm was thus attained. Glass beads (3-mm diam) were treated the same way. After treatment, each sample was transferred to a petri dish (9-cm diam) and immediately placed under the appropriate light. Samples of sorghum and wheat grains and glass beads were exposed to one of the 4 lights and analyzed by TLC for malathion and malathion degradation products after exposures of 0, 1, 5, 10, 15, 25, and 30 days. Control samples kept in darkness were analyzed for the same periods.

**Removal of Malathion Residues from Glass Beads.**—Light-exposed and control glass-bead samples were transferred to 125-ml Erlenmeyer flasks and shaken with 3 successive 50-ml aliquots of redistilled acetone. After the solvent evaporated to near dryness under vacuum, malathion and its degradation products were transferred quantitatively to a 15-ml graduated centrifuge tube with the aid of 15 ml of redistilled acetone. The sample solution was then concentrated to 1–2 ml under a stream of nitrogen for TLC spotting.

Table 1.— $R_f$  values for malathion and its degradation products, and colors produced with DCQ detection reagent.

Compound	System* 1	System 2	System 3	System 4
1. Unknown (pink-red)	0.98±0.1 <sup>b</sup>	0.92±.02	0.98±.01	0.95±.02
2. Unknown (pink-red)	.90±.02	.80±.02	.98±.01	.89±.02
3. Unknown (bright yellow)	.87±.02	.77±.02	.98±.01	.89±.02
4. Unknown (pink-red)	.84±.02	.74±.02	.98±.01	.89±.02
5. Unknown (bright yellow)	.80±.02	.72±.02	.98±.01	.89±.02
6. Malathion (bright red)	.77±.02	.69±.02	.98±.01	.89±.02
7. Malaoxon (bright yellow)	.72±.02	.62±.02	.96±.01	.88±.02
8. Malathion monoacid (bright red)	.59±.02	.33±.02	.73±.02	.68±.02
9. Unknown (bright yellow)	.43±.02	.27±.02	.96±.01	.82±.02
10. Malathion diacid (bright red)	.34±.02	.14±.01	.07±.01	.11±.01
11. Unknown (bright yellow)	.28±.02	.14±.01	.96±.01	.88±.01
12. <i>O,O</i> -dimethyl phosphorodithioate (pink-red)	.23±.02	.08±.01	.57±.02	.61±.02
13. Unknown (bright yellow)	.12±.01	.02±.01	.51±.02	.57±.02
14. <i>O,O</i> -dimethyl phosphorodithioate (pink-red)	.18±.01	.06±.01	.45±.02	.51±.02
15. <i>O</i> -demethyl malathion (brown-red)	.08±.01	.02±.01	.82±.02	.76±.02
16. Unknown (bright yellow)	.02±.01	.02±.01	.39±.02	.45±.02
17. Unknown (bright yellow)	.00	.00	.77±.02	.72±.02
18. Dimethyl phosphate <sup>c</sup>	.00	.00	.24±.02	.25±.02
19. Unknown <sup>c</sup>	.00	.00	.02±.01	.03±.01
20. Unknown (brown-yellow)	.00	.00	.00	.00
21. Phosphoric acid <sup>c</sup>	.00	.00	.00	.00

\* Systems: (1) hexane:benzene:acetic acid (40:40:20) used with Brinkman silica gel plates; (2) petroleum ether:ethyl ether:acetic acid (60:40:1) used with Brinkman silica gel plates; (3) chloroform:methanol:30% ammonium hydroxide (75:25:4) used with Eastman silica gel plates; (4) isopropanol:30% ammonium hydroxide (85:15) used with Brinkman silica gel plates.

<sup>b</sup> Mean deviation.

<sup>c</sup> Reacts to produce color only with Hanes-Isherwood reagent; DCQ and silver nitrate-bromophenol blue reagents are insensitive.

Extraction and Cleanup of Malathion and its Degradation Products in Stored Grains.—Malathion and its degradation products in wheat and sorghum grains were extracted and cleaned by the method of Kadoum (1969). After cleanup, residues were dissolved in 1–2 ml of redistilled acetone for TLC spotting.

RESULTS AND DISCUSSION.—Fig. 1, 2, 3 shows the effects of 6 different light or dark conditions on the rate of disappearance of malathion on glass beads, sorghum grain, and wheat grain. Malathion disappeared from glass beads, sorghum, and wheat grains most rapidly after exposure to heat-producing IR light. Control samples kept in darkness at 119°C indicated that although IR light may accelerate disappearance of malathion, the high temperature produced by the IR light lamp ( $119 \pm 2^\circ\text{C}$ ) accounted for more acceleration. Others showed that losses of malathion by evaporation (Awad et al. 1967), isomerization, and decomposition (O'Brien 1956, McPherson and Johnson 1956) were related directly to temperature.

Unlike IR light-exposed samples, temperature effects were less significant in samples exposed to the far UV, near UV, and plant GRO. All samples contained intact malathion residues after all exposure periods.

Differences in rates at which malathion disappeared because of exposure to varying wavelengths of light were noted more readily. Far UV light caused malathion to disappear fastest, whereas plant GRO

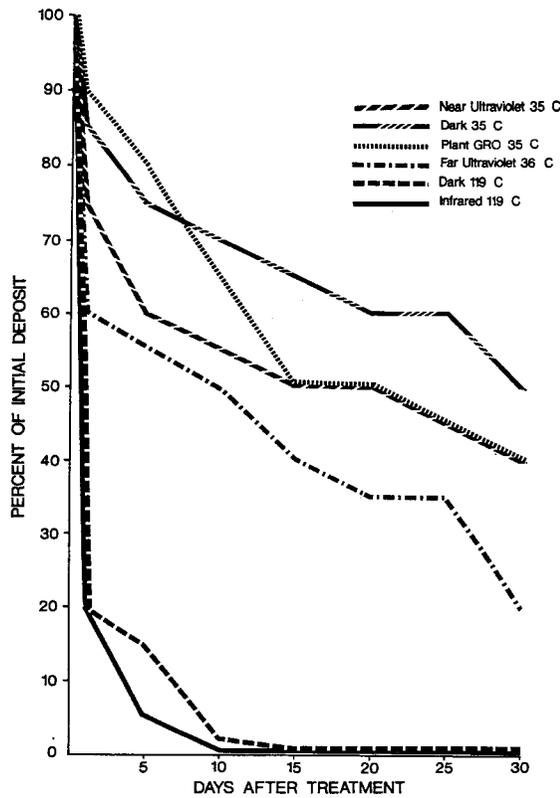


FIG. 2.—Effects of 6 light or dark conditions on the rate malathion disappeared from sorghum grain, estimated by thin-layer chromatographic analysis.

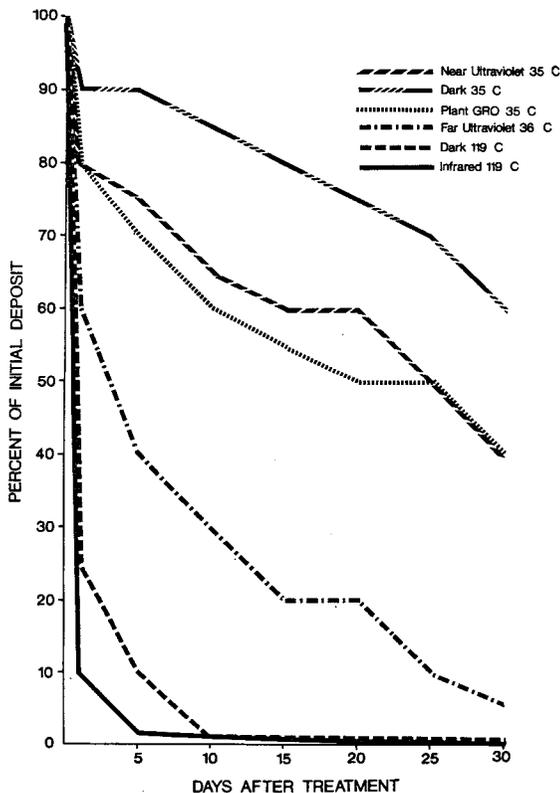


FIG. 1.—Effects of 6 light or dark conditions on the rate malathion disappeared from glass beads, estimated by thin-layer chromatographic analysis.

and near UV light caused nearly equal, but less rapid, disappearance.

Generally, malathion disappeared more rapidly from glass beads, less from sorghum grain, and least from wheat grain. Its penetration into sorghum and wheat grains may have influenced its evaporation and degradation; however, our data provide no evidence of penetration effects. Evaporation is a possible explanation for the rapid loss of malathion during the 1st day of exposure. Under all exposures, light and dark, malathion residues declined sharply at first, then leveled off for the remainder of the exposure periods. Evaporation also might account for more rapid loss of malathion from glass beads than from grains. Malathion evaporates from smooth glass surfaces more rapidly than from porous plant surfaces (Matsumura 1960, Awad et al. 1967).

Thin-layer chromatographic identification and estimation of malathion degradation products showed a relationship between malathion's disappearance and an increase in degradation products. The solvent systems enabled us to separate 20 degradation products ( $R_f$  values in Table 1), 12 of which are not identified. The greatest quantities of malathion-degradation products were detected in samples exposed to high temperatures ( $119^\circ\text{C}$ ). Light-exposed and control samples subjected to lower temperatures ( $35^\circ$  or  $36^\circ\text{C}$ ) contained smaller quantities of degradation products.

Temperature effects of IR light were exhibited further in that control samples kept in darkness at

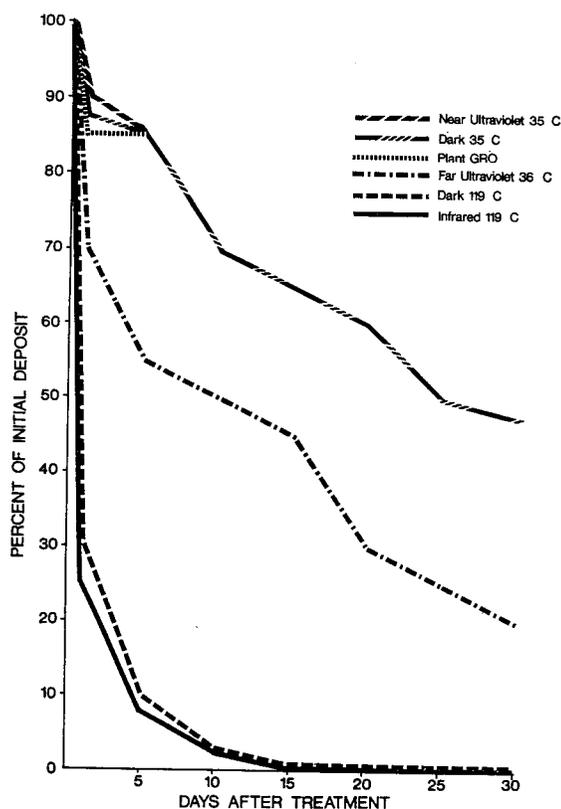


FIG. 3.—Effects of 6 light or dark conditions on the rate malathion disappeared from wheat grain, estimated by thin-layer chromatographic analysis.

the same experimental temperature contained the same degradation products at slightly lower concentrations. The principal degradation products detected after IR light exposure were malaoxon and an unknown (no. 9 in Table 1) which is less polar than malaoxon ( $R_f$  0.43 in system 1), both of which produced yellow spots with DCQ (Table 1). Smaller quantities of *O,O*-dimethyl phosphorothioate, malathion monoacid, malathion diacid, *O*-demethyl malathion, and phosphoric acid also were identified during the 30-day, IR-light exposure. Four unknowns (no. 13, 17, 19, 20 in Table 1), all less polar than the unknown no. 9 previously mentioned, all persisted after malathion and malaoxon had disappeared completely.

Far UV light initially degraded malathion into malaoxon and 3 "less polar than malaoxon" unknown compounds (no. 1, 2, 4 in Table 1). The same unknowns were detected in samples exposed to the other 3 lights, but greatest quantities of them followed far UV light exposure.

Awad et al. (1967) noted that malathion exposed to a combination of far UV and near UV light disappeared more rapidly than when exposed to visible (fluorescent) light and darkness. Because the quantities of degradation products which they detected did

not differ significantly, they considered the differences to be caused by temperature and evaporation rather than by UV light.

Greater quantities of degradation products were detected in the far-UV-light-exposed samples than samples held in the visible-light (plant-GRO), near-UV light, or in the dark. In addition to the malathion degradation products identified by El-Refai (1960),<sup>4</sup> we identified malathion monoacid, *O*-demethyl malathion, and dimethyl phosphate in far-UV-light-exposed residues.

Plant GRO and near UV lights produced smaller quantities of degradation products such as malaoxon, malathion monoacid, malathion diacid, *O,O*-dimethyl phosphorodithioate, and *O,O*-dimethyl phosphorothioate.

In general, all lights induced formation of more-polar malathion degradation products. The initial breakdown products of malathion after far UV light exposure were less polar than malathion; however, longer exposure resulted in the formation of compounds more polar than malathion. Far UV light produced greater quantities of polar compounds such as *O,O*-dimethyl phosphorodithioate, *O,O*-dimethyl phosphorothioate, dimethyl phosphate, and phosphoric acid than did other lights.

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# Degradation of Malathion on Viable and Sterilized Sorghum Grain<sup>1</sup>

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### ABSTRACT

The effect of the biological activity of the kernel on the rate of malathion degradation was determined during 9 months of storage. The migration of malathion residue to the endosperm and embryo of sterilized sorghum grains was greatest during the 1st month of storage; however, the amounts recovered during that time accounted for only 41.4% of the remaining total malathion residue. After 3 months' storage, the total residue gradually decreased, until 34.3% of the initial total deposit remained at the end of 9 months. At all time

intervals during the 9 months' storage, more malathion residue was detected on, and in, the sterilized sorghum kernels than on the live, viable kernels, but the patterns of residue decline were similar. Only 14% of the initial malathion deposit remained on the viable sorghum after 9 months' storage. These findings indicated that during a normal storage period enzymes contribute to the breakdown of malathion applied to sorghum grain as an insect protectant.

Sorghum grain production, which has increased considerably during the past few years, has become a major crop for animal food. About 80% of the yearly production is sold through normal grain-marketing channels and is stored for many months in commercial warehouses. During storage, sorghum grain is readily attacked by a large number of stored-product insect pests. Malathion, the principal insecticide used to treat stored grains, does not protect sorghum grain (LaHue 1967) to the extent that it does corn (LaHue 1966) and wheat (LaHue 1965).

The influence of hybrid varieties, foreign material, moisture content, and storage temperature on the rate of dissipation of malathion residue on sorghum grain has been determined (Kadoum and LaHue 1969). Effects of grain-moisture contents and storage temperatures on the insecticidal effectiveness of malathion on wheat have been demonstrated (Lindgren et al. 1955, Strong and Sbur 1960, Minett et al. 1968,

Watters 1959). Papers on effectiveness of malathion as an insect protectant on sorghum have been valuable in elucidating optimum conditions for application (King et al. 1962; LaHue 1967, 1969). But, except for the report by Kadoum and Sae (1970), studies on the fate of malathion residue on sorghum grain are nonexistent. Therefore, we attempted to determine the amount of malathion that penetrates the kernel at different times during storage and the effect of the kernel's biological activity on the rate of malathion degradation.

**MATERIALS AND METHODS.—Adjusting Moisture Content.**—Distilled water was added gradually to increase the moisture content of the 'Taylor-Evans 66' hybrid sorghum grain from 11.86% to 14.0%. After each addition, the sorghum was turned on a wheel mixer for 20 min, then left to equilibrate for 1 week at 26.7°C. When the desired 14.0% moisture level was obtained (within limits of ±0.1%), the sorghum was sealed in 1-gal glass jars until the malathion was applied. The moisture content was determined (at 26.7°C) with a Steinlite Model 512 RC moisture tester.

**Effect of Biological Activity of Sorghum.**—Part of the sorghum grain was autoclaved at 10 psi steam

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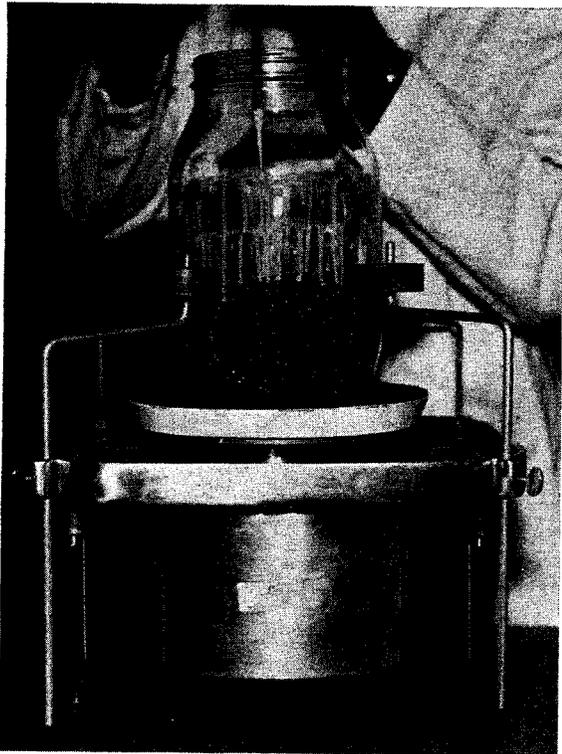


FIG. 1.—Applying 1 ml of malathion emulsion to 1000 g sorghum on a turntable.

pressure for 20 min. The sterilized grain was allowed to equate before the moisture content was adjusted to the 14.0% level. Tests showed that  $95 \pm 2\%$  of the source grain was viable; none of the sterilized kernels germinated. Malathion degradation caused by physical and chemical changes in the nonviable sterilized sorghum was compared with that caused by enzymic action in the living, viable grain.

**Malathion Treatment.**—Malathion was applied at the rate of 1.0 pint of 57% EC/1000 bu. The concentration was diluted with distilled water to make an emulsion for 1-ml applications of 10 ppm technical malathion. One ml of the emulsion—applied with a 1-ml volumetric pipette to the inside wall of a 1-gal, large-mouthed glass jar containing 1000 g of sorghum—was discharged above the grain while the jar turned at 33 rpm on a turntable (Fig. 1). The jars were sealed immediately, shaken by hand for 30 sec, then mixed for 15 min at 33 rpm on a mechanical

tumbler. The emulsion, prepared immediately before use, was agitated continuously (at  $26.7 \pm 0.6^\circ\text{C}$ ) for application to  $26.7^\circ\text{C}$  grain. Immediately after treatment the 1000-g samples were maintained at  $26.7^\circ\text{C}$  in a storage room.

**Extraction and Cleanup Procedures.**—Malathion residues were extracted and cleaned, using a modification of Kadoum's methods (1968, 1969), for gas-chromatographic analysis. Four replicates of a 20-g sample were shaken vigorously with 100 ml of distilled acetone for 50 sec to remove the malathion from the surface of the grain. The extract was filtered off and transferred, with the aid of 50 ml of acetone wash, to a 250-ml, round-bottom flask. (During the experimental testing of the extraction procedure, no additional malathion residue was recovered when the treated sorghum was shaken longer than 50 sec.) The 20-g sample of sorghum was then blended with 100 ml of distilled acetone in a covered Sorvall Omnimixer at top speed for 5 min. The extract was filtered and transferred (with an additional 50 ml of acetone wash) to another 250-ml, round-bottom flask. The extracts were then concentrated to 2–3 ml under vacuum, and the residues were transferred, using 10 ml hexane, to 250-ml separatory funnels containing 100 ml of water. Each funnel was vigorously shaken for 30 sec. After the 2 layers separated, the lower (aqueous) layer was drawn off and discarded. The upper (hexane) layer was partitioned with 80% acetonitrile in water for analysis. Total recovery of malathion was obtained with the aforementioned extraction procedure.

**Gas-Chromatographic Analysis.**—An electron capture detector was used to analyze malathion under these operating conditions: column—6-ft glass column of 3% DC-11 on 60 mesh silinized Gas Chrom. P; carrier gas—nitrogen, 36 ml/min; temperature—column  $200^\circ\text{C}$ , detector cell  $220^\circ\text{C}$ ; injector  $240^\circ\text{C}$ ; volume injected—4  $\mu$ liter of the extract in hexane.

**RESULTS AND DISCUSSION.**—Table 1 shows the malathion residues found in the sorghum grain during a 9-month storage period. It is evident that considerably more residue was retained by the sterilized than by the live sorghum grain. Thus, it appears that the kernel's viability greatly affected the persistence of malathion and that the biological activity enhanced the breakdown rate. It is reasonable to assume that enzymic hydrolysis contributed to the breakdown of malathion as well as chemical and physical factors which we studied previously (Kadoum and LaHue 1969). We found that increased moisture content of the grain enhanced breakdown of malathion. (The increased enzymic activity in grains with moisture above the critical level (Kadoum and LaHue 1969)

Table 1.—Malathion residue on live and sterilized sorghum grain following 10 ppm applications.

Residue age	Live sorghum (ppm)			Sterilized sorghum (ppm)		
	External	Internal	Total	External	Internal	Total
24 hr	7.13 <sup>a</sup>	1.14	8.27	7.80	2.12	9.92
1 month	4.25	1.71	5.96	4.79	3.38	8.17
1½ months	3.40	1.60	5.00	4.36	3.18	7.54
3 months	1.88	0.93	2.81	3.56	2.25	5.81
6 months	0.97	.62	1.59	3.08	1.23	4.31
9 months	.83	.33	1.16	2.72	0.68	3.40

<sup>a</sup> Avg 4 replicates.

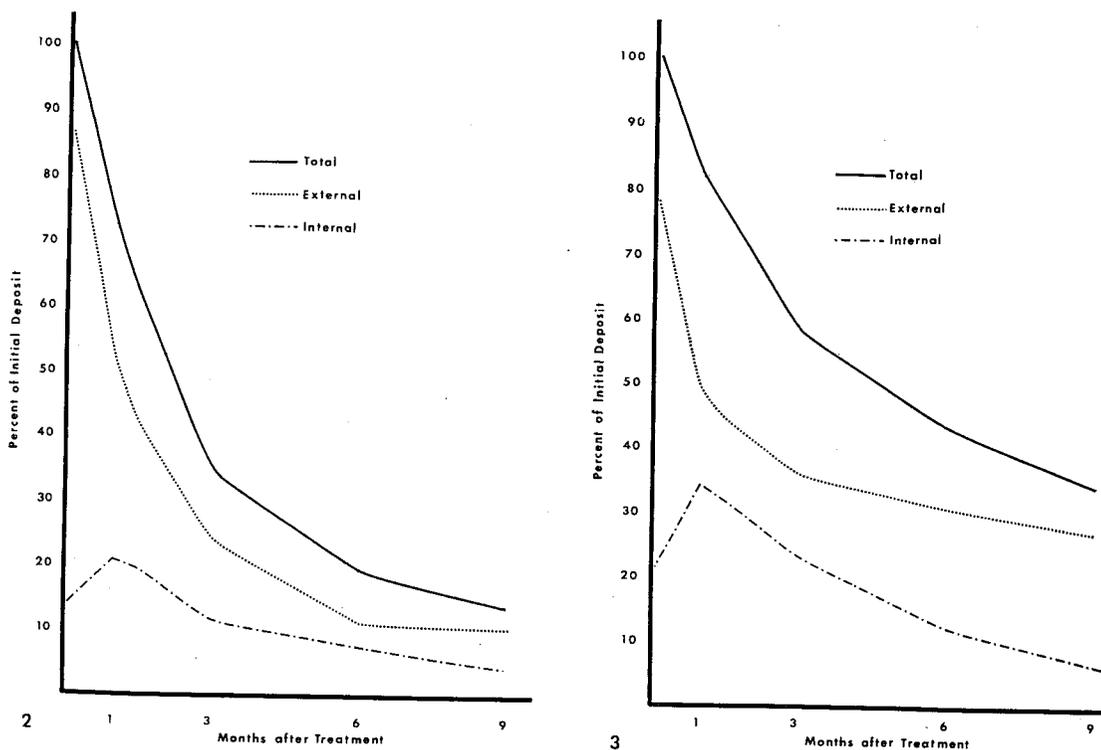


FIG. 2, 3.—Malathion recovery from live (Fig. 2) and sterilized (Fig. 3) sorghum grain during 9 months of storage. Percentage of recovery based on initial deposit determined 24 hr after application.

is due to the appearance of free water in the grain which is a necessary prerequisite for increased enzymic activity and biological change.)

Previous studies (Kadoum and Sae 1970) showed that malathion and its metabolites failed to inhibit sorghum-grain esterase, which fact indicates that malathion degradation could be affected by esterases as well. That conclusion agrees with the findings of Rowlands (1965); he concluded that wheat grain esterases oxidized malathion to malaosxon, which had no lasting inhibitory effect on those hydrolytic enzymes involved in malathion degradation and that oxidation contributed little malathion degradation. In a previous study, Rowlands (1964) concluded that malathion on wheat and maize broke down slowly by a chemical route and by enzymic hydrolysis by phosphatase to yield the same phosphoric acid derivatives. Enzymic oxidation did not appear to occur.

Malathion residues recovered from live and sterilized sorghum are shown graphically in Fig. 2 and 3, respectively. To determine the initial residue deposits, analyses were made 24 hr after treatment. Those initial analyses showed that 86.2% of the residue remained on the exterior of live samples and that 13.8% had penetrated the grain. After 1 month of storage, only 51.4% of the initial deposit remained on the exterior; internal residue had increased to 20.7%.

Analyses revealed that total residues recovered from sterilized sorghum were consistently greater than those from live grain. Initial analyses showed that 78.6% of the residue remained on the exterior and that 21.4% had penetrated the grain. After 1

month of storage, the sterilized grain had 48.3% and 34.1% of the initial malathion deposit remaining on the exterior and the interior of the grain, respectively.

Maximum penetration of malathion was observed after 1 month's storage in both live and sterilized sorghum with an increase of 13.4% in internal residue in sterilized compared with live sorghum.

Malathion penetrated the kernel fastest during the 1st few weeks of storage; thereafter rate of degradation masked rate of penetration which was always higher in the sterilized compared with that of live sorghum.

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## Dissemination of *Salmonella montevideo* Through Wheat by the Rice Weevil<sup>1</sup>

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### ABSTRACT

Adult *Sitophilus oryzae* (L.) in wheat contaminated with *Salmonella montevideo* Hormaeche & Peluffo (10<sup>7</sup> cells/g) were released at 1 end of a cylindrical metal chamber, 311.25×7.5 cm, filled with uncontaminated wheat. *S. montevideo*-positive wheat samples were recovered as far from the infestation point as 270 cm. *S. montevideo* numbers ranged from 0.92 to 1.60/g at 30 cm and from 0.0 to 0.004/g at 270 cm from the infestation point.

Flies and cockroaches have been investigated extensively as disseminators of *Salmonella*; stored-product insects have not. Lesser mealworm adults collected in poultry-brooder houses yielded 5 serotypes of *Salmonella* (Harein et al. 1970). Lesser mealworms, *Alphitobius diaperinus* (Panzer), fed

on dog food contaminated with *S. typhimurium* showed that whereas bacterial counts decreased 50% in dog food, they increased from 1 to 2.4×10<sup>6</sup>/insect (De las Casas et al. 1968). Large numbers and multiple doses of *Salmonella* were required for even a few cells to be transmitted through the alimentary canal of the hide beetle, *Dermestes maculatus* De Geer (Julseth et al. 1969).

Some rice weevils, *Sitophilus oryzae* (L.), exposed 7 days to wheat contaminated with ca. 10<sup>8</sup> *S. montevideo*/g, contaminated clean wheat up to 7 days later; others, exposed for 14 days, contaminated clean wheat up to 28 days later (Husted et al. 1969); weevils when put in contaminated wheat for 14 and 21 days and then transferred weekly to clean wheat, tested positive for *S. montevideo*, externally and internally, up to 5 transfers.

*S. montevideo* Hormaeche & Peluffo accounted

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for 2 of 3 *Salmonella* isolations from grain in 1963 (Anonymous 1964). Allred et al. (1967) reported that in several isolations in grain from basic feed mills, *S. montevideo* led the list, accounting for 63 of 544 isolations.

The objective of this study was to determine

whether rice weevils could transfer *S. montevideo* through wheat and, if they could, to what extent.

#### Materials and Methods

Weevil cultures were maintained in 1-qt fruit jars in a dark rearing room at ca. 27°C and 68%

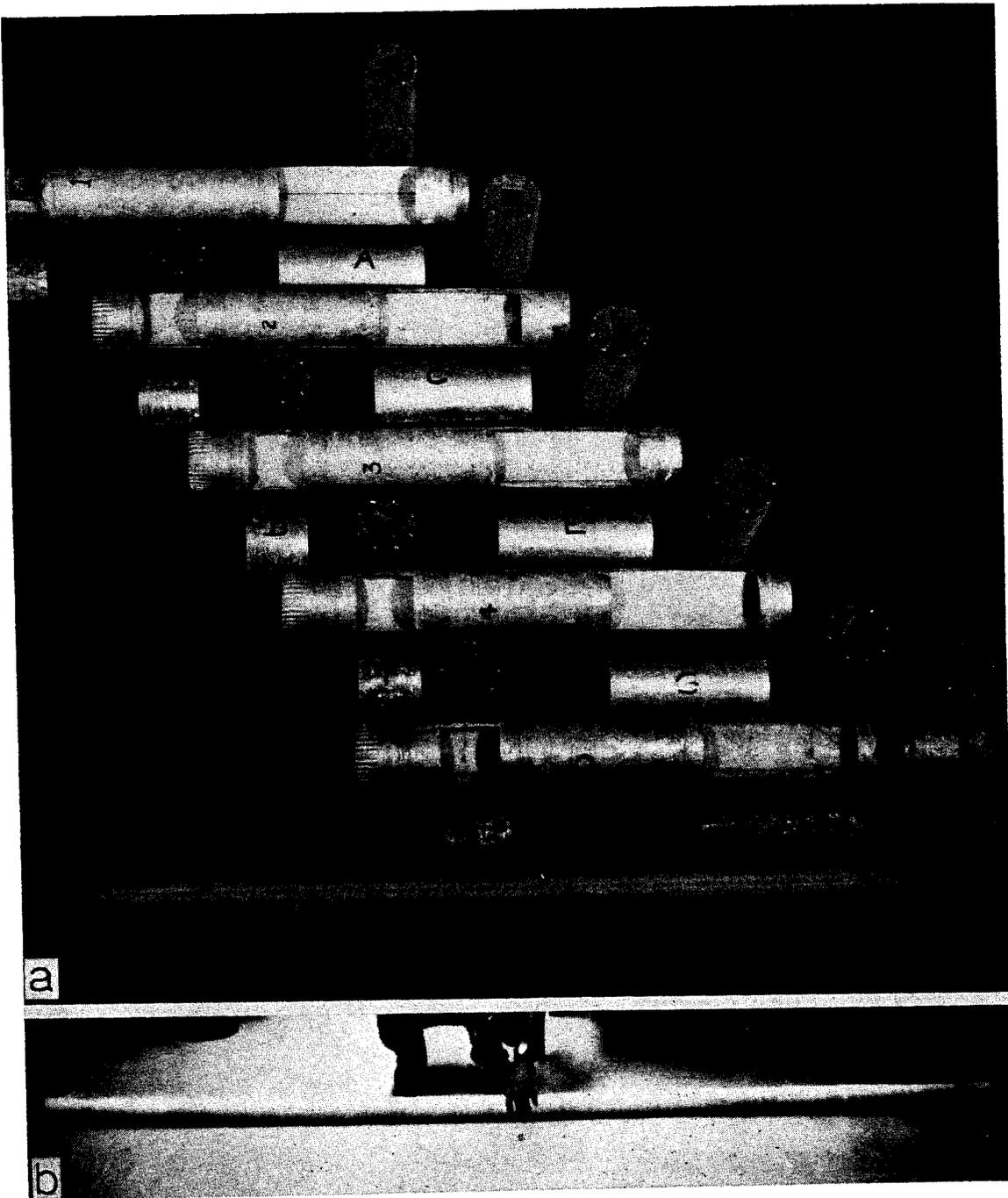


FIG. 1.—a, Disassembled elongated metal chamber. Numerals denote chamber sections; each letter denotes cover for the sample site above in the cylinder (E covered the contamination site). Sample cages A, C, E, G, and I were constructed larger than the others to permit enumeration of *S. montevideo* on the wheat. Note sample cages and bent metal covers for each sample location: b, Assembled, taped, and infested metal chamber.

Table 1.—Elongated metal-chamber test, to determine how far and in what numbers the rice weevil could transfer *S. monteideo* from contaminated wheat ( $10^7$  cells/g) to clean wheat (see Fig. 1).

		Samples and distance from contamination chamber in cm										
		X	A 30	B 60	C 90	D 120	E 150	F 180	G 210	H 240	I 270	J 300
Rep. no. 1	Wheat (+ or -)	+	100g 5+ <sup>a</sup>	+	100g 2+	+	100g 1+	-	100g 0+	-	100g 1+	-
			10g 5+		10g 1+		10g 1+		10g 0+		10g 1+	
			1g 3+		1g 0+		1g 0+		1g 0+		1g 0	
	No. <i>S. monteideo</i> /g		0.920		0.007		0.004		0.000		0.004	
	No. insects	105	156	18	20	9	9	1	7	0	0	0
	Insects (+ or -)	+	+	+	+	+	+	+	+			
Rep. no. 2	Wheat (+ or -)	+	100g 5+	+	100g 5+	+	100g 0+	-	100g 1+	-	100g 0+	-
			10g 5+		10g 4+		10g 0+		10g 1+		10g 0+	
			1g 4+		1g 2+		1g 0+		1g 0+		1g 0+	
	No. <i>S. monteideo</i> /g		1.600		0.220		0.000		0.004		0.000	
	No. insects	152	197	50	55	8	12	2	5	0	3	0
	Insects (+ or -)	+	+	+	+	+	+	+	+		+	
Rep. no. 3	Wheat (+ or -)	+	100g 5+	+	100g 5+	+	100g 3+	-	100g 0+	-	100g 0+	-
			10g 5+		10g 5+		10g 0+		10g 0+		10g 0+	
			1g 5+		1g 2+		1g 2+		1g 0+		1g 1+	
	No. <i>S. monteideo</i> /g		>1.600		0.540		0.013		0.000		0.002	
	No. insects	43	196	40	52	2	15	1	2	0	0	0
	Insects (+ or -)	-	-	-	+	-	-	+	+			

<sup>a</sup> No. positive subsamples (of 5) of each size.

RH. To assure *Salmonella*-free insects, culture-wheat and weevil samples were cultured in Brilliant Green Tetrathionate Broth (Difco) and then streaked on Brilliant Green Agar (Difco) plates. (No *Salmonella*-like organisms were found.) Culture and test wheat were withdrawn from the lot that was tested, adjusted to ca. 13.7% moisture content (determined by Steinlite Model G and Motomco instruments), and placed in the rearing room for 1 week to equilibrate to rearing-room conditions.

Wheat was contaminated for the test by these steps: (1) yeast-extract-enriched, proteose-peptone agar slants were inoculated from stock cultures of *S. monteideo* and incubated at 37°C for ca. 18 hr; (2) cells were then washed off the slants with sterile 0.1% tryptone water, and the turbidity of the suspension was standardized to ca. 200 times the Nephelometer no. 1 tube; (3) the suspension was added (1 cc for each 100 g) to the wheat, which was rolled for at least 10 min on a U.S. Stoneware Roller to uniformly distribute *S. monteideo*. This gave a contamination of ca.  $1 \times 10^7$  cells/g.

Previous tests using metal drums and wooden troughs indicated that the rice weevil was capable of transporting *S. monteideo* up to 145 cm through wheat. To determine the extent of *S. monteideo* dissemination, a test chamber (311.25 cm long) was constructed from 5 sections (60-cm long) of 7.5-cm galvanized-steel stovepipe (Fig. 1). The 3rd and 5th sections (+ 18.5 cm of a 6th section soldered to the 5th) were screened (3-mm hail) at 1 end. The opposite end of the 3rd remained open. That of the 5th was solder-sealed with a galvanized-steel disk, which had a 4.5-cm-diam hole (covered with 60-mesh/in. screen soldered to the disk, and filter paper taped over the hole). The 2nd and 4th sections had both ends screened. These steps allowed

the filled chamber to be disassembled without spillage for freezing and autoclaving. A 5-cm end portion of the 1st section was screened off to form an initial contamination and infestation cylinder (the end was solder-sealed with a galvanized-steel disk).

Beginning with the larger size, holes of alternate sizes (17.5 cm long  $\times$  10.0 cm around the circumference and 6.6 cm long  $\times$  10.0 cm around the circumference) were cut in the top of the chamber at 30-cm intervals (beginning at the contamination cylinder). Five cylindrical sample cages 6.9 cm diam  $\times$  17.8 cm long and 5 cages 6.9 cm diam  $\times$  6.25 cm were constructed of 3-mm hail screen. The larger sample sizes were for later enumeration of *S. monteideo* on the wheat. A flap (3.1 cm square) was cut in 1 end of each cage, to permit wheat to be funneled in the cages (560 g in the larger and 170 g in the smaller).

Each section of the chamber was held vertically and filled with wheat up to the 1st hole. A filled sample cage (of appropriate size) was inserted in each hole, which was then covered with a bent piece of galvanized steel lined with an adhesive-backed, rubberized cork tape and sealed with 5-cm masking tape. When all holes and all sections were filled, the chamber was assembled by slipping each section into the preceding one to a predetermined mark and sealing with masking tape.

One thousand weevils were put on 180 g of wheat contaminated with  $10^7$  *S. monteideo*/g and set in the rearing room for 1 week. Wheat and insects then were held at 4°C for 2 hr (to immobilize insects) before being funneled into the contamination cylinder and sealed. After the sealed chamber had been in the rearing room 1 week, it was separated in 2 places above a pan containing mercuric chlor-

ide to catch falling wheat and insects. The open ends were sealed with masking tape and the sections were placed in a freezer for 3 days (to kill insects), then the sampling holes were opened carefully. Sample cages were withdrawn with forceps and emptied into a white porcelain pan (sterilized between samples by rinsing with mercuric chloride). Insects were transferred with forceps from the samples to sterilized test tubes where they were counted and macerated. Wheat from the smaller sampling cages was put in sterile pint fruit jars; wheat from the larger cages was divided thus: five each of 100-g aliquots (qt fruit jars), 10-g aliquots (pint fruit jars), and 1-g aliquots (large test tubes). These steps were taken to determine the number of *S. montevideo* present per gram of sample by the Most Probable Numbers Method of Galton et al. (1968). The insect and wheat samples were inoculated individually with Brilliant Green Tetrathionate Broth and incubated at 37°C. After 48 hr, Brilliant Green Agar plates were streaked for each sample and incubated at 37°C for 24 hr. Two typical colonies, picked from each positive plate, were streaked and stabbed into Triple Sugar Iron Agar (Difco) slants, then incubated at 37°C for 24 hr. Cells from positive slants that agglutinated in both poly H and O Group C<sub>1</sub> antisera were considered *S. montevideo*. Three replicates were completed.

### Results

Wheat samples from the contamination cylinders of all 3 replicates were positive; insects were positive in only the 1st two (Table 1). All other insect samples from the 1st 2 replicates were positive at distances of from 30 to 270 cm. Only 3 of 8 insect samples in the last replicate were positive (C, E, and F), and only 1 colony was recovered from a plate streaked from sample C. No explanation is offered. Wheat samples in smaller cages (B, D, F, H, and J) were positive at least up to 120 cm from the contamination cylinders in all replicates and up to 180 cm in the last replicate. Larger samples (A, C, E, G, and I), in 2 replicates, had aliquots positive as far as 270 cm. Enumeration results showed that *S. montevideo* was present in small numbers, even at a distance of only 30 cm from the contamination cylinders (0.92–1.60 cells/g). Between 30 and 90 cm, numbers dropped rapidly; beyond that, recoveries leveled off and ranged from 0.013 to 0.0 cells/g at distances of 150 to 270 cm.

### Discussion

Thomas and Hobson (1955) showed that serotypes of *Escherichia coli* and *Aerobacter aerogenes* were present in 73% of the ears and panicles of growing cereals tested and that their numbers increased during the season. It was shown previously (Crumrine and Foltz 1969) that *S. montevideo* could survive at least 28 weeks on wheat held at moisture contents comparable to those in commercially binned wheat. Animals conceivably could contaminate

wheat in the field or in the bin or anywhere in between, with wheat remaining infective for a relatively long time.

The low numbers of *S. montevideo* transmitted in this study do not seem important from a human health standpoint, because larger dosages are normally required to produce disease symptoms in adults. Theoretically, though, 1 surviving organism is capable of causing disease. Also, rats were shown to excrete positive fecal pellets after ingesting 1–15 *Salmonella* and were able to contaminate other rats in their colony (Welch et al. 1941). They could ingest the low numbers of *Salmonella* organisms that insects might transfer, increase the numbers of bacteria in their digestive tracts, and recontaminate the wheat with their feces. Perhaps other animals, such as birds, have similar capabilities.

If rice weevils in farm or commercial bins could carry *Salmonella* 2.7 m in any direction from the source, they would be capable of contaminating nearly all parts of a cylindrical bin containing ca. 128 m<sup>3</sup> (3550 bu.) of wheat. When considering the possible recontamination by animals, the amount of wheat contaminated could be increased greatly. However, conditions of these tests and those in the field are quite different. In the tests, large numbers of weevils per gram of wheat were exposed to highly contaminated wheat for relatively long periods. In a natural situation, a weevil population of the size in these tests would quickly destroy the grain. The normally lower field populations likewise would decrease the dispersal pressure on the insects. Since weevils are not confined, their exposure to a concentration of contaminated wheat likely would be short. In some cases, it might be as little as the time necessary to move through the contaminated grain. Freedom to move in a much larger grain mass also would result in fewer insects being recovered at increased distances, thus lessening the possibility of *Salmonella* being deposited. Hence, the rice weevil probably plays a relatively small role in the cycle of salmonellosis, but its potential should not be overlooked.

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# Phoxim as an Insect Protectant for Stored Grains<sup>1</sup>

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## ABSTRACT

Phoxim was tested on hard winter wheat, shelled corn, and sorghum grain as a protectant against adult rice weevils, *Sitophilus oryzae* (L.); red flour beetles, *Tribolium castaneum* (Herbst); confused flour beetles, *T. confusum* Jacquelin duVal; and lesser grain borers, *Rhyzopertha dominica* (F.). Evaluations of results were made at intervals by determination of mortalities after aging of the deposits, by counting the number of progeny developing after toxicity test exposures, and by an assessment of progeny damage to the treated grain.

Phoxim applied at a rate of 5 ppm was superior to

the standard dosage of 10 ppm malathion in all tests. Protection against rice weevil damage was excellent throughout the 12-month storage. A lessening of toxicity effectiveness against the flour beetles and lesser grain borers was noted after 9 months' storage, and little progeny damage was inflicted.

Definite levels of phoxim residue were evident with the 3 dosages applied. Variations were found, but the recoveries seem representative for 3 scalar rates of application.

Routine, but detailed, laboratory evaluations of promising candidate compounds for the control of insects that attack stored grains are a continuing part of research leading to the development of acceptable grain-protectant materials. Such compounds should be low in mammalian toxicity, and should possess highly effective residual and vapor toxicant properties against a variety of insects attacking stored grain and grain products. An urgent need for new materials is indicated by development of resistance by some species and by the natural tolerance shown by others to materials now in use.

In preliminary laboratory studies, phoxim applied at 10 ppm to cleaned, uninfested sorghum grain containing 12.5% moisture killed all introduced adult rice weevils, *Sitophilus oryzae* (L.), and lesser grain borers, *Rhyzopertha dominica* (F.), and prevented establishment of an internal infestation in exposures made 1 and 3 months after treatment.

McDonald and Gillenwater (1967) found phoxim more effective than malathion as a direct-contact toxicant against adult confused flour beetles, *Tribolium confusum* Jacquelin duVal; cigarette beetles, *Lasioderma serricorne* (F.); larvae of Indian meal moths, *Plodia interpunctella* (Hübner); and black carpet beetles, *Attagenus megatoma* (F.). They concluded that phoxim appeared promising for further study in several areas of stored-product insect control work, and specifically as a protective treatment of bulk grain and other bulk-stored commodities.

Among a group of organophosphorus (OP) compounds about equal to or safer than malathion, Strong and Sbur (1968) found phoxim had potential for use as a commodity protectant, as a direct spray application, and as a residual surface treatment in tests with adults of the Angoumois grain moth, *Sitotroga cerealella* (Olivier); confused flour beetle; granary weevil, *Sitophilus granarius* (L.); sawtoothed grain beetle, *Oryzaephilus surinamensis* (L.); and larvae of *Trogoderma parabile* Beal (now *T. variabile* Ballion).

Strong (1969) found that phoxim along with dichlorvos ranked highest among 12 OP compounds selected for tests with applications of spray solutions made directly to the adult moths. He concurred with the McDonald and Gillenwater (1967) sug-

gestion that phoxim was sufficiently promising against stored-product insects for continued research on its use for protective treatments, space application, and residual surface treatments in storage facilities.

Speirs and Zettler (1969) found phoxim the most promising of 3 OP compounds tested against 2 strains of malathion-resistant red flour beetles, and concluded that it was a most promising candidate insecticide for further testing.

One of us (La Hue unpublished data) found that many Indian meal moth adults were killed when exposed to vapors above grain treated with phoxim, and that the F<sub>1</sub> generation was greatly reduced even after 12 months' aging of the treated grain.

The objectives of this study were (1) to determine the effectiveness of 3 dosage levels of phoxim against adult rice weevils, confused flour beetles, red flour beetles, and lesser grain borers when it is applied as an emulsion to wheat, shelled corn, and sorghum grain, and (2) to record the residues remaining on the grain at intervals during a 12-month period.

**MATERIALS AND METHODS.**—Cleaned, uninfested lots of hard winter wheat, shelled corn, and sorghum grain were tempered to 12.5±0.05% moisture. The source lots were held in covered fiberboard drums at 26.7±1.1°C and 60±5% RH for 21 days for the moisture to come to equilibrium. Test insects were reared in a room maintained at 26.7±1.1°C and 60±5% RH. The 14-day-old rice weevil and 7-day-old lesser grain borer adults used in the test were reared on hard winter wheat containing about 12.5% moisture. The 14-day-old red and confused flour beetles were reared on a standard laboratory culture medium containing 10 parts flour, 10 parts yellow cornmeal, and 1.5 parts brewer's yeast.

Lots of 1000 g of tempered grain were placed in 1-gal, widemouth glass jars. An emulsifiable concentrate containing 47.0% of phoxim (4.0 lb of technical material/gal) was diluted to make emulsions for 1-ml applications to the grain of 5, 10, and 20 ppm of actual phoxim. The emulsions, which were prepared immediately before use, were kept in constant agitation at 26.7±0.6°C.

Malathion was used as a standard for comparison. An emulsifiable concentrate containing 57.0% of premium-grade malathion was diluted to make an emulsion for 1-ml applications to the grain of 10 ppm of actual malathion.

Applications were made with a 1-ml volumetric pipette to the inside wall of the 1-gal glass jars above

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the grain level while the jars were turning on a 33-rpm turntable. Immediately after application, the jars were shaken by hand for 30 sec, and then rotated for 15 min on a mechanical tumbler to mix the phoxim with the grain. Each application was replicated 4 or 5 times. After the required aging period in covered jars, 250-g samples were removed and placed in 1-pint glass mason jars for the toxicity exposures. The test jars were fitted with rings, 40-mesh screens, and filter-paper lids.

The insects were exposed after the deposits had aged for periods of 1, 3, 6, 9, and 12 months. About 50 adult insects were placed in individual samples. After 21 days' exposure, the adult insects were removed by sifting, and counts were made of those alive and dead. The adults were discarded, but the grain and fine siftings were returned to the jars for a later record of progeny emergence. Counts of the  $F_1$  progeny were recorded following peak emergence as shown by counts made in untreated samples of the different grains. After the progeny readings were completed, the grain with the progeny was retained for at least 120 days for an assessment of progeny damage as described previously (La Hue 1969).

**RESULTS.**—Results were evaluated by determining mortality of the adult insects and counting the progeny, and by noting the amount of grain damage caused by progeny. All applications gave complete protection from damage in studies made 1 and 3 months after treatment, and only minimal damage was recorded at the 6- and 9-month periods on the

Table 2.—Mortality of adult red flour beetles and  $F_1$  progeny damage to 3 grains after 12 months' storage following treatment with phoxim.<sup>a</sup>

Insecticide applied	Adult % mortality	Progeny		
		Total no.	% dead	Visible damage <sup>b</sup> (rating)
<i>Wheat</i>				
Phoxim				
20 ppm	100.0	0		0
10 ppm	100.0	10.0	100.0	0
5 ppm	96.0	14.6	52.2	0.2
Malathion, 10 ppm	68.0	38.0	18.4	2.2
Untreated	0	79.0	0	3.2
<i>Corn</i>				
Phoxim				
20 ppm	100.0	0		0
10 ppm	100.0	0		0
5 ppm	100.0	0		0
Malathion, 10 ppm	57.3	11.0	0	1.2
Untreated	0	32.0	0	3.0
<i>Sorghum</i>				
Phoxim				
20 ppm	100.0	0		0
10 ppm	100.0	0		0
5 ppm	86.0	21.0	42.2	.2
Malathion, 10 ppm	22.0	56.0	25.0	2.0
Untreated	0	76.0	0	3.2

<sup>a</sup> All data are averages of 4 or 5 replications.

<sup>b</sup> Damage 150 days after infestation ranged from 0 (no visible infestation or damage) to 4 (heavy infestation and considerable damage).

Table 1.—Mortality of adult rice weevils and  $F_1$  progeny and damage to 3 grains after 12 months' storage following treatment with phoxim.<sup>a</sup>

Insecticide applied	Adult % mortality	Progeny		
		Total no.	% dead	Visible damage <sup>b</sup> (rating)
<i>Wheat</i>				
Phoxim				
20 ppm	100.0	0		0
10 ppm	100.0	0		0
5 ppm	100.0	72.0	25.0	0.6
Malathion, 10 ppm	81.0	202.2	70.4	1.4
Untreated	0	1792.0	3.2	5.0 <sup>c</sup>
<i>Corn</i>				
Phoxim				
20 ppm	100.0	0		0
10 ppm	100.0	0		0
5 ppm	96.0	60.0	66.7	.4
Malathion, 10 ppm	90.2	49.6	83.2	1.2
Untreated	0	389.2	1.6	5.0
<i>Sorghum</i>				
Phoxim				
20 ppm	100.0	0		0
10 ppm	100.0	0		0
5 ppm	100.0	98.0	42.9	1.0
Malathion, 10 ppm	39.2	831.0	31.4	5.0
Untreated	0	1414.2	0.4	5.0 <sup>c</sup>

<sup>a</sup> All data are averages of 4 or 5 replications.

<sup>b</sup> Damage 120 days after infestation ranged from 0 (no visible infestation or damage) to 5 (heavy infestation, damage, and grain spoilage).

<sup>c</sup> Damage at 90 days.

grain with the 5 ppm dosage. Phoxim was slightly more effective on corn than on wheat and sorghum.

Tables 1-4 compare the mortality of adult insects exposed for 21 days on wheat, corn, and sorghum after 12 months' storage following treatment with phoxim.

**Rice Weevils.**—The 20- and 10-ppm dosages gave complete protection for 12 months, but a few of the samples with the 5-ppm dosage showed some progeny damage (Table 1). Small infestations became established in all samples of sorghum treated with the 5-ppm dosage, and damage was greater than to comparable lots of corn and wheat. Damage to malathion-treated sorghum was heavy after 6 months, but fairly good protection to corn and wheat was obtained with malathion for 12 months.

**Red Flour Beetles.**—All dosages of phoxim protected the grains from damage for 12 months, even though a few live red flour beetles were found in samples of wheat and sorghum with the 5-ppm treatment (Table 2). Considerable red flour beetle damage occurred in most of the malathion-treated samples of wheat and sorghum in contrast to the small numbers of beetles and little damage in corn.

**Confused Flour Beetles.**—Complete kills of confused flour beetles were obtained in exposures to all 3 grains treated with the 20-ppm application throughout the 12-month storage period (Table 3). A small amount of damage was inflicted on wheat and sorghum treated with 10 ppm after 12 months, and many adults survived during exposures to the 5-ppm application. A few  $F_1$  progeny were found in

Table 3.—Mortality of adult confused flour beetles and  $F_1$  progeny damage to 3 grains after 12 months' storage following treatment with phoxim.<sup>a</sup>

Insecticide applied	Adult % mortality	Progeny		
		Total no.	% dead	Visible damage <sup>b</sup> (rating)
<i>Wheat</i>				
Phoxim				
20 ppm	100.0	0		0
10 ppm	80.4	6.0	50.0	0.2
5 ppm	33.2	12.0	25.0	.6
Malathion, 10 ppm	43.0	34.0	21.4	1.4
Untreated	0	49.2	0	3.0
<i>Corn</i>				
Phoxim				
20 ppm	100.0	0		0
10 ppm	98.0	2.0	100.0	0
5 ppm	71.4	6.0	50.0	.2
Malathion, 10 ppm	40.0	16.0	37.5	.6
Untreated	0	63.0	0	2.8
<i>Sorghum</i>				
Phoxim				
20 ppm	100.0	0		0
10 ppm	81.7	11.0	63.6	.2
5 ppm	20.2	23.0	26.1	1.0
Malathion, 10 ppm	18.4	60.0	0	1.8
Untreated	0	98.2	0	2.8

<sup>a</sup> All data are averages of 4 or 5 replications.

<sup>b</sup> Damage 150 days after infestation ranged from 0 (no visible infestation or damage) to 4 (heavy infestation and considerable damage).

Table 4.—Mortality of adult lesser grain borers and  $F_1$  progeny and damage to 3 grains after 12 months' storage following treatment with phoxim.<sup>a</sup>

Insecticide applied	Adult % mortality	Progeny		
		Total no.	% dead	Visible damage <sup>b</sup> (rating)
<i>Wheat</i>				
Phoxim				
20 ppm	100.0	0		0
10 ppm	100.0	11.0	100.0	0
5 ppm	91.2	43.0	58.1	0.4
Malathion, 10 ppm	58.0	289.6	16.7	2.6
Untreated	0	1114.2	2.9	5.0 <sup>c</sup>
<i>Corn</i>				
Phoxim				
20 ppm	100.0	0		0
10 ppm	100.0	0		0
5 ppm	100.0	43.0	33.3	.2
Malathion, 10 ppm	88.0	43.0	16.3	1.4
Untreated	0	116.0	3.4	4.0
<i>Sorghum</i>				
Phoxim				
20 ppm	100.0	0		0
10 ppm	100.0	0		0
5 ppm	100.0	42.0	50.0	.4
Malathion, 10 ppm	14.0	906.8	5.7	4.0
Untreated	0	1389.0	1.1	5.0 <sup>c</sup>

<sup>a</sup> All data are averages of 4 or 5 replications.

<sup>b</sup> Damage 120 days after infestation ranged from 0 (no visible infestation or damage) to 5 (heavy infestation, damage, and grain spoilage).

<sup>c</sup> Damage at 90 days.

1 replicate of corn with the 5-ppm dosage. In comparing results obtained from exposures of red flour beetles and confused flour beetles, it is quite evident that the phoxim residues were uniformly less toxic to the confused flour beetles. Malathion gave good protection to wheat and corn for 9 months and to sorghum for 6 months.

*Lesser Grain Borers.*—Phoxim residues were very toxic to lesser grain borers, and although some progeny developed in some of the samples of all 3 grains with the 5-ppm dosage, damage was minimal (Table 4). Malathion-treated sorghum was damaged after 6 months' aging, wheat after 9 months, and corn after 12 months.

*Residues.*—Definite levels of phoxim residue are evident at the 3 dosages (Table 5). Apparently little decomposition of the phoxim occurred during 12 months' aging of the treated grain.

A very significant loss of malathion residues occurred on all the grains during the 1st month of storage; thereafter, a more or less gradual decrease was evident. After 24 hr, malathion recovery from sorghum was slightly higher than from wheat or corn. Malathion residues on corn and sorghum declined more rapidly than residues on wheat during the storage period.

*Discussion.*—The so-called broad-spectrum insecticides, which are effective against a large number of important insects, should be used on stored grains, oil seeds, and their products as these are susceptible to attack by mixed populations of several species. Evaluation of an insecticide should be made on a

Table 5.—Mean residues of phoxim and malathion on 3 grains at given intervals during storage.<sup>a</sup>

Insecticide applied	24 hr	Interval after treatment				
		1 month	3 months	6 months	9 months	12 months
<i>Wheat</i>						
Phoxim						
20 ppm	16.6	14.1	12.7	13.5	4.2	8.4
10 ppm	7.5	6.0	7.0	5.1	2.4	3.0
5 ppm	4.6	2.8	4.1	2.8	1.6	2.0
Malathion, 10 ppm	8.4	5.4	4.2	3.5	2.5	2.0
<i>Corn</i>						
Phoxim						
20 ppm	15.3	13.6	11.8	10.9	9.8	7.3
10 ppm	5.1	6.0	6.4	5.5	6.2	4.7
5 ppm	2.8	2.7	2.6	1.4	2.1	1.9
Malathion, 10 ppm	7.9	5.2	3.2	3.0	1.2	0.6
<i>Sorghum</i>						
Phoxim						
20 ppm	13.2	16.6	16.8	14.7	6.7	16.1
10 ppm	8.1	8.0	7.5	6.1	9.5	9.1
5 ppm	2.8	4.3	4.4	4.6	4.0	5.7
Malathion, 10 ppm	9.6	4.3	2.4	1.7	0.6	.6

<sup>a</sup> Means based on 2 samples.

basis of performance against a large number of insect species as control measures are often necessary without specific identification of the insects comprising mixed populations infesting a commodity. From the results obtained in our studies and from those of other workers, phoxim appears to be a most promising insecticidal material. Its performance has been better than that of malathion, or comparable with it, in studies with several insect species. In all the tests reported here, phoxim applied at a rate of 5 ppm was superior to the standard dosage of 10 ppm malathion. Phoxim may be further tested in intermediate-type, small-bin studies.

Until complete assessment of the safety of phoxim and its degradation products is made, phoxim cannot be recommended for use as an insecticide in or around foods.

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