# Residual Nature of Certain Organophosphorus Insecticides in Grain Sorghum and Coastal Bermudagrass[1]

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Residue studies were conducted on several insecticides being evaluated for the control of the two-spotted mite, Tetranychus telarius (Linn.), on grain sorghum and the fall armyworm, Spodoptera frugiperda (Smith), on Coastal bermudagrass. On grain sorghum, the residual persistance of dimethoate, ethion, azinphos-methyl and methyl parathion was determined while similar studies were performed with Imidan, N-(mercaptomethyl) phthalimide-S-(O,C-dimethylphosphorodithioate, and trichlorfon on Coastal bermudagrass.

## Treatment

Grain Sorghum. The first applications were made when the sorghum grain was still in the dough stage. Emulsifiable formulations of the insecticides were applied at the rate of 0.5 lbs. of toxicant per four gallons of water per acre. A high clearance spray rig with No. 3 Teejet nozzles spaced at 20-inch intervals along a horizontal boom was used to apply the insecticides under a pressure of 60 psi. For this treatment, the nozzles were

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about six inches above the sorghum heads with one nozzle directly over the plants and another in the middle of each row.

A different nozzle arrangement was used for the second applications made on the same plots seven days after the first treatment. To obtain more thorough plant coverage, 0.5 lbs. of insecticide per nine gallons of water per acre were applied as described above except that the nozzle positioned between the rows was replaced by two nozzles at the end of a 20-inch drop extending between each row. One of the nozzles was directed to the side of the plants in one row and the other nozzle adjusted to spray the sides of plants in the adjoining row. Each plant, then, received spray from the top and directly from both sides.

Samples for residue analysis were collected at 0, 3, and 6 days after the first treatment and 0, 3, 6, and 14 days following the second application. Zero-day samples were taken within one hour after treatment. The heads were kept separate from the forage samples and the grain removed for analysis.

Coastal Bermudagrass. Trichlorfon and Imidan sprays were prepared from emulsifiable concentrate formulations and applied with a high clearance sprayer at a rate of 1.0 lb./a. and 0.75 lbs./a., respectively. Conditions under which they were applied were the same as described for the first treatment of the grain sorghum. Samples were collected at 0, 1, 3, 7 and 14 days following treatment by cutting the stems just above ground level.

## Analysis

Grain Sorghum. All of the residue analyses were based on described colorimetric methods with some modifications in the extraction and clean-up techniques. The detection of dimethoate depended on alkaline hydrolysis to yield thioglycolic acid which was determined colorimetrically using the original Folin's uric acid method as described by Giang and Schechter, (1). Ethion was determined spectrophotometrically as a copper salt complex of its hydrolysis product, diethyl phosphorodithioic acid (2). Techniques for clean-up and detection of azinphosmethyl were taken from Adams (3) and Anderson (4) which involved alkaline hydrolysis to anthranilic acid, diazotization and coupling with N-(1-naphthyl) ethylenediamine to produce color. Methyl parathion residues were detected using the colorimetric method of Averell and Norris (5).

Coastal Bermudagrass. Trichlorfon residues were determined with a Barber-Colman Series 5000 Gas Chromatograph equipped with a sodium thermionic detector (6). Chromatography conditions were as follows: A six foot, four mm i.d. glass column was packed with 30% XF-1150 on 60/80 mesh acid-washed Chromosorb W. A 10 mg. plug of glass wool was loosely packed into the injector port end of the column to achieve maximum thermal breakdown of trichlorfon to its phosphate moiety (7). Nitrogen was used as the carrier gas at a pressure of 20 psi. Air and hydrogen was supplied to the burner at a pressure of 44 and

20 psi, respectively. All supply cylinders were fitted with a length of capillary tubing and the expressed pressures represent the reading of a pressure regulator at the supply cylinder. Applied potential was obtained from a 300 volt battery and the baseline current was held constant at $5X10^{-9}$ with an electrometer setting of $10^{-8}$ ampere full scale. Temperatures were as follows: column 180°C, injector 280°C and detector oven 215°C. The recorder chart speed was 0.25 inches per minute. Under these conditions, trichlorfon had a retention time of two minutes.

Extraction of trichlorfon from the bermudagrass samples was accomplished using a procedure similar to the one described by Anderson (7). Frozen samples were run through a Hobart Food Chopper and 100 g. subsamples processed for analysis. Each subsample was ground in a Waring blender with 300 ml. of 0.1 N $H_2SO_4$ for five minutes and the homogenate filtered through cheesecloth. Following centrifugation of the filtrate for 10 minutes at 2000 rpm, the supernatant was decanted into a one liter separatory funnel. Sixty-eight g. of sodium chloride was added, dissolved by shaking and then the aqueous mixture extracted twice with 200 ml. portions of ether. The combined ether extracts were evaporated to about 10 ml., 50 ml. of benzene added and the water in the sample removed with anhydrous sodium sulfate. The dried benzene-ether mixture was reduced to a total volume of one ml. Two microliters of this solution was injected into the gas chromatograph.

Imidan residues were determined using colorimetric and gas chromatographic methods. One-hundred g. of chopped bermudagrass were homogenized in 400 ml. of benzene, filtered through glass wool into a one liter separatory funnel and the filtrate extracted with 100 ml. of water. The benzene phase was separated from the water layer, dried with anhydrous sodium sulfate, filtered and a volume equivalent to five g. of plant material removed for further clean-up. This involved passing the benzene extract through a column containing five g. of a charcoal adsorbent mixture (1:1 Darco G-60 and Hyflo Super-Cel) and eluting the Imidan with 70 ml. of a 1:1 benzene-chloroform mixture. The effluent was then evaporated to a volume of about 0.5 ml. and the residue transferred to a 60 ml. separatory funnel with two 10 ml. portions of hexane and five ml. of acetonitrile. After vigorous shaking, the acetonitrile layer was removed and the hexane re-extracted with 5 ml. of fresh acetonitrile. The acetonitrile extracts were combined.

Color was developed in the exact manner as described in the colorimetry section of the report by Batchelder and Patchett (8). Briefly, this method was based upon the conversion of Imidan to anthranilic acid which was then coupled with 3-methyl-2-benzothiazolone hydrazone to produce a magenta colored product.

For analysis using gas chromatography the 10 ml. of acetonitrile was reduced in volume to one ml. and two microliters injected into the instrument. Chromatographic conditions different than those described for trichlorfon analysis are as follows:

the column was packed with 1.5% SE 30 on 80/100 mesh acid-washed Chromosorb P and the nitrogen pressure was reduced to 16 psi. Air and hydrogen pressure were 44 and 25 psi, respectively. Operating temperatures were as follows: column 200°C, injector 250°C and detector oven 220°C. The retention time for Imidan was 2.5 minutes.

## Results and Discussion

Grain Sorghum. After slight modifications in some of the techniques, recovery of each insecticide from both forage and grain samples exceeded 80 percent. Practical sensitivity levels of the methods, based on the amount of interferring materials from untreated samples, were as follows: dimethoate, 0.20 ppm; ethion, 0.15 ppm; and methyl parathion, 0.20 ppm.

Although all four organophosphates were applied at the rate of 0.5 lbs./a., the initial residue deposits varied with each compound. This variation was more evident in the forage samples of the first treatment where the dimethoate and methyl parathion treated samples had residues of approximately 4.5 ppm immediately after treatment while the ethion and azinphosmethyl samples contained residue of 2 to 3 ppm (Table 1). Residues on the forage just after the second application varied from about 8 ppm for dimethoate to 14 ppm for azinphosmethyl. These variations in the actual amounts of active material deposited on the plant did not seriously affect the residue study since the rate of dissipation of the toxicant present was the major factor under consideration.

TABLE 1

P.P.M. Insecticide Residues on Sorghum Forage at Various Intervals After Treatment 1/

| Days After Treatment | P.P.M. Dimethoate | Ethion | Azinphos-methyl | Methyl Parathion |
|---|---|---|---|---|
| | First Treatment | | | |
| 0 | 4.65 | 2.13 | 2.90 | 4.80 |
| 3 | 2.21 | 0.79 | 0.76 | 0.36 |
| 6 | 0.56 | 0.55 | 0.64 | ND 2/ |
| | Second Treatment | | | |
| 0 | 8.13 | 11.45 | 13.60 | 12.56 |
| 3 | 2.96 | 4.60 | 4.41 | 1.61 |
| 6 | 1.07 | 1.32 | 1.71 | 0.49 |
| 14 | 0.64 | 0.36 | 0.99 | ND |

1/ All materials applied at a rate of 0.5 lbs./a. First application June 26, 1964; Second application July 3, 1964.
2/ None detected

Dissipation rates for dimethoate, ethion and azinphosmethyl from the forage were very similar. About 60 percent of the material deposited had disappeared by three days after either treatment. By six days, approximately 20 percent of the applied dose was still present and extending the persistance study to 14 days after the second treatment revealed that less than 10 percent of the insecticides were still on the plant portion of the treated

sorghum. The rate of dissipation did not appear to be maintained after the residues declined below the 1.0 ppm level. These small quantities of residues slowly disappeared and resulted in over 0.50 ppm in the plant six days after treatment.

Analysis of forage samples from methyl parathion-treated grain sorghum showed that the residual life of this insecticide was very short. Of those residues found immediately after the first or second treatment, 4.80 ppm and 12.56 ppm respectively, less than 10 percent was present after three days. No residues were detected six days after the first application or 14 days after the second.

More consistent residues were deposited on the grain (Table 2). Deposits were similar for each insecticide within a treatment and fairly consistent after both the first and second applications. In general the residues were on the order of 2 to 4 ppm. A major exception was grain from the second application of methyl parathion where only 0.80 ppm residues were detected on these 0-day samples and none at all at the later sampling dates. There was no apparent explanation for these low residues since the corresponding plant samples contained 12.56 ppm methyl parathion residues.

Dissipation of the four organophosphates was slightly slower from the sorghum grain than from the plant. Again, however, dimethoate, ethion and azinphosmethyl reacted in a similar manner. Residues of approximately 2 to 4 ppm of these insecticides deminished to almost half these levels in three days.

Unlike the forage samples, the residues on the grain did not appear to persist after levels of less than 1.0 ppm were present. This was shown by the fact that only ethion residues could be detected on the grain after 14 days.

TABLE 2

P.P.M. Insecticide Residues on Sorghum Grain at Various Intervals After Treatment. 1/

| Days After Treatment | P.P.M. Dimethoate | Ethion | Azinphos-methyl | Methyl Parathion |
|---|---|---|---|---|
| | First Treatment | | | |
| 0 | 2.50 | 3.93 | 3.13 | 3.14 |
| 3 | 1.60 | 2.02 | 1.45 | 0.36 |
| 6 | 0.68 | 0.99 | 0.95 | ND 2/ |
| | Second Treatment | | | |
| 0 | 1.78 | 2.96 | 2.44 | 0.80 |
| 3 | 0.95 | 1.54 | 0.96 | ND |
| 6 | 0.66 | 1.09 | 0.57 | ND |
| 14 | ND | 0.85 | ND | ND |

1/ All materials applied at a rate of 0.5 lbs./a. First application June 26, 1964; Second application July 3, 1964.
2/ None detected

Although the second application methyl parathion-treated grain samples contained unexplainably low residues, the results of the residual nature of this material on grain after the first

treatment clearly showed it to be of short residual life. Only the three-day samples had detectable amounts of residues present and these were less than 0.40 ppm.

Residues on sorghum forage after the first and second applications clearly demonstrated that the same amount of active material per acre resulted in different residue levels when applied in a different manner. Insecticide deposits were as much as four times greater in concentration after the second treatment.

The fact that the residues on the grain after both treatments were almost the same suggests that adjustment of nozzles could be important in preventing insecticide residues on certain portions of a crop. In a crop such as sorghum it is likely that insects attacking only the stalks and leaves could be controlled with minimum insecticide contamination of the sorghum grain.

Coastal Bermudagrass. Attempts were first made to determine trichlorfon using electron capture gas chromatography (7). However, the procedure could not be used in the present study because interfering materials with the same retention time as trichlorfon were present in untreated Coastal bermudagrass.

Since the electron capture method was dependent upon the quantitative thermal breakdown of trichlorfon to chloral, the compound actually detected when trichlorfon was injected, it appeared likely that the phosphate moiety could be determined using a sodium thermionic detector. This was actually the case as trichlorfon was easily detected upon injection of as little as

10 nanograms. At this concentration a peak height of six mm. was observed and the response was linear up to 300 nanograms. Injections of untreated bermudagrass extracts did not show any contamination peaks in the trichlorfon area. Unlike the electron capture detector, this method could not determine chloral or trichlorethanol residues.

Very good recoveries were obtained when trichlorfon was added to 100 g. of untreated bermudagrass at four different levels ranging from 0.1 to 3.0 ppm. The maximum recovery was 120 percent, a minimum of 88 percent and an average of all recoveries of 97 percent.

Table 3 shows the residues present on Coastal bermudagrass at intervals up to 14 days following trichlorfon treatment. Almost half of the material deposited at treatment, 59.12 ppm, had diminished after only one day. After this, the rate of dissipation declined and residues of slightly more than five ppm remained in the plants 14 days after treatment.

Imidan was determined using colorimetry and sodium thermionic methods of detection. By colorimetric analysis, recoveries were greater than 88 percent and exceeding 97 percent when the residues were detected by sodium thermionic gas chromatography. Several extraction steps necessary in the former method but eliminated in the latter probably accounted for the increased recovery. Certainly, the comparative recoveries indicated that the sodium thermionic method of detecting Imidan residues could be

used with confidence.

TABLE 3

Residues on Coastal Bermudagrass following Trichlorfon and Imidan Treatment 1/

| Days After Treatment | P.P.M. | |
|---|---|---|
| | Trichlorfon | Imidan |
| 0 | 59.12 | 37.93 (26.89) 2/ |
| 1 | 29.37 | 32.66 (24.91) |
| 3 | 16.02 | 23.89 (19.81) |
| 7 | 8.04 | 15.79 (14.15) |
| 14 | 5.74 | 8.31 (11.05) |

1/ Trichlorfon and Imidan applied at 1.0 and 0.75 lbs./a., respectively on September 20, 1964.
2/ Numbers in parentheses are ppm as determined colorimetrically.

Sodium thermionic and colorimetric determination of Imidan residues on Coastal bermudagrass reflected the same findings observed in the recovery experiments. In all but the 14 day samples, the gas chromatography method resulted in higher residues, indicating better recoveries (Table 3). Both procedures showed residues of a similar magnitude and served as a check for each other in demonstrating the residual persistence of Imidan in Coastal bermudagrass.

Imidan deposits on the plants at the time of treatment, approximately 38 ppm, dissipated slower than trichlorfon residues. With Imidan, however, the rate of dissipation continued fairly

constant throughout the 14 day test period, with a half-life of between five and six days.

## References


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