

PESTICIDE DRIFT AND ITS EFFECT ON  
CHOLINESTERASE LEVELS OF BIRDS IN NON-TARGET  
AREAS OF THE BUENA VISTA MARSH

by

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## ABSTRACT

Pesticide drift from the aerial spraying of agricultural chemicals and its effects on the cholinesterase levels of birds in non-target areas of the Buena Vista Marsh, Wisconsin, was investigated. Surface water samples were also surveyed for pesticide residues.

Brain cholinesterase activity was determined in birds from wildlife management lands adjacent to agricultural areas. Pesticide drift was sampled by the use of nylon collection screens and analyzed by gas chromatography for some of the commonly used cholinesterase inhibiting pesticides.

Residues of pesticides were found in 71% of the air samples collected on 2 study areas. Parathion and diazinon were found in the highest concentrations (up to 200 micrograms of pesticide/m<sup>2</sup> screen) with malathion, monocrotophos, dimethoate, disulfoton, fenthion, and phorate being present in lesser amounts.

A decline in the brain cholinesterase levels of red-winged blackbird (Agelaius phoeniceus) and savannah sparrow (Passerculus sandwichensis) was found throughout the spraying season, indicating possible exposure of the birds to cholinesterase inhibiting pesticides. Low cholinesterase levels were also found early in the season which may have resulted from the birds consuming treated seeds during the planting season.

Results indicate that pesticides are drifting over non-target areas of the Buena Vista Marsh and possibly in amounts

high enough to produce a measureable effect on native bird-life.

Preliminary study indicates that pesticides may also be moving through the ecosystem via surface water drainage.

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## INTRODUCTION

Organophosphate and carbamate compounds have replaced chlorinated hydrocarbons as the major pesticides used in the United States (Fig. 1). Chlorinated hydrocarbons are persistent and tend to accumulate in the ecosystem. They are long lived, build up in living organisms and are very mobile in the ecosystem. Chlorinated pesticides have therefore received considerable attention as environmental toxicants while little work has been done on organophosphates and carbamates (Stone 1979).

Organophosphates and carbamates are presently considered environmentally safer than chlorinated hydrocarbons due to their generally lower persistence and ability to be more readily metabolized. Long lasting organophosphates and carbamates can be manufactured but they are avoided by industry and discouraged by government (Stickel 1973). The degradability of the organophosphates and carbamates can be influenced by environmental factors. Sunlight, temperature, pH, and biological activity are known to affect the rate of breakdown, but this is highly variable. Malathion is hydrolyzed almost instantaneously at a pH of 12 but no detectable hydrolysis occurs for 12 days at a pH of between 5 and 7 (Spiller 1961). The chemical half-life of diazinon has been determined to be about 10 days at a pH of 10 but only 7 days at a pH of 5, the opposite relationship from malathion (Baughman 1973). The persistence of these compounds can vary from hours to months and usually ranges from days

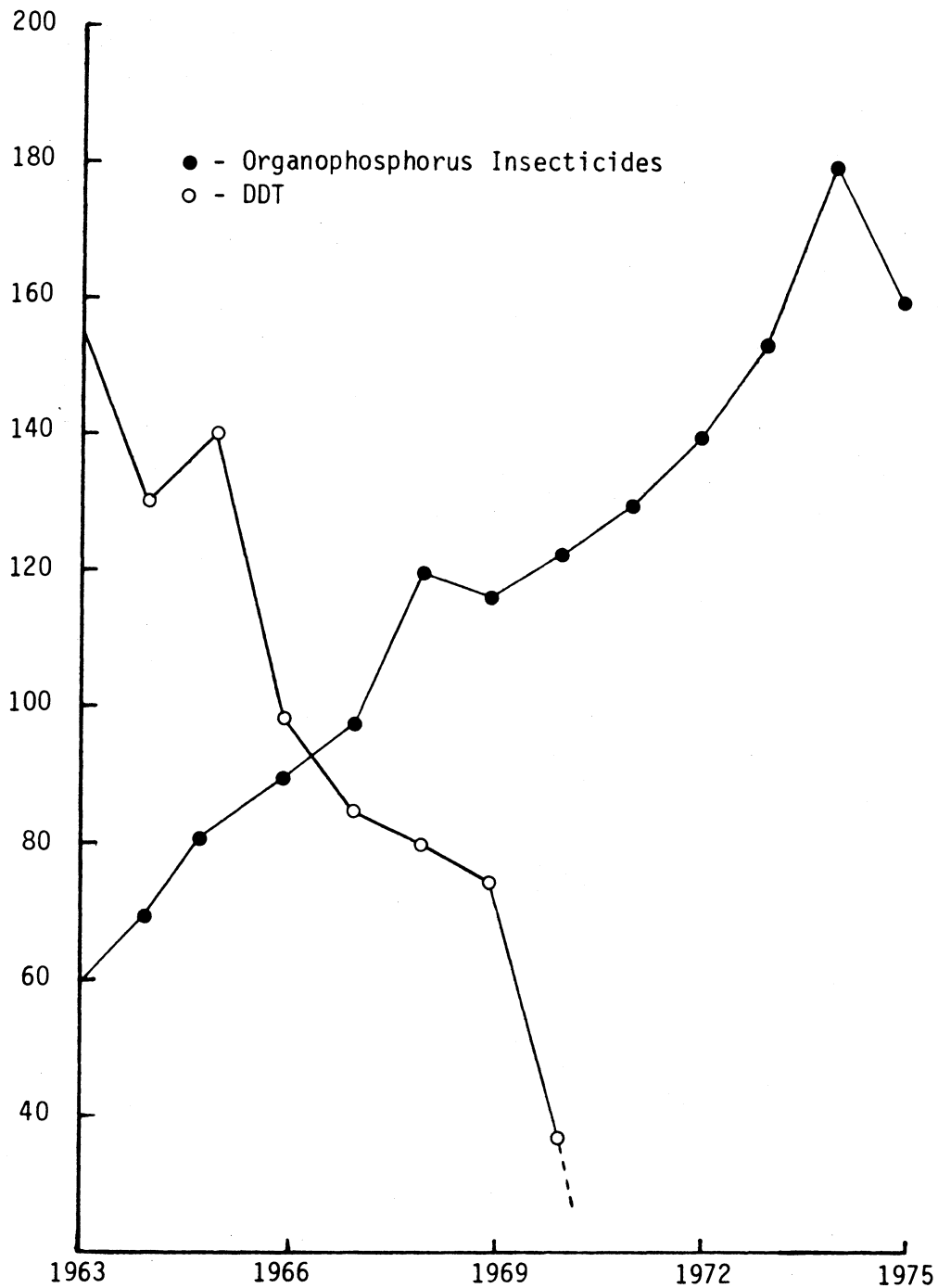


Figure 1. Use of organophosphate insecticides and DDT (a chlorinated hydrocarbon) in the United States, 1963-1975 (Brown 1978).

to a few weeks. Azodrin, an organophosphate, has been found to be undegraded in river water after 8 weeks while sevin, a carbamate, was 95% degraded after 1 week (Eichelberger and Lichtenberg 1971).

Organophosphates and carbamates are often applied more frequently than previously used pesticides because they are generally less persistent. They may be more prevalent in ecosystems than their persistence would indicate because of the short dissipation time between applications. Organophosphate insecticides have accumulated in marsh soils and were transferred to its drainage systems (Miles and Harris 1978). More frequent applications of organophosphorus and carbamate compounds probably also result in greater numbers of these compounds being applied, and little is known about the toxicity of pesticides which mix after application (Eaton 1973). Studies have shown some of them to be synergistic (Bender 1968).

While usually less persistent, organophosphate and carbamate pesticides are usually more acutely toxic than chlorinated hydrocarbons. Organophosphates and carbamates inhibit the neural enzyme cholinesterase. This allows acetylcholine to accumulate at the nerve junctions causing alteration of the transmission of nerve impulses across synapses. The parasympathetic nervous system and the neuro-myal junctions, as well as the central nervous system, are severely affected in acute poisoning (Stone 1979). Signs of such poisoning include pupil constriction muscle fascicu-

lation, decreased heart rate, respiratory difficulties leading to respiratory arrest, paralysis and convulsions (Davis et al. 1971). Acute toxicity expressed as the LD<sub>50</sub> based on oral feeding of male rats range from 1-5 mg/kg for phorate (thimet) to 560 mg/kg for sevin (carbaryl), 2 commonly used pesticides. LD<sub>50</sub> is the number of milligrams of the compound required per kilogram of animal weight to produce mortality in 50% of test animals (EPA 1978).

Pesticides enter and move through the ecosystem in a variety of ways. The most obvious one is through agricultural field applications but also through applications to forests, home lawns and gardens, water, urban trees and elsewhere. They are applied from the ground, aerially, as seed treatments or in irrigation water. Once in the ecosystem, pesticides have a varying mobility by aerial drift, runoff, groundwater or uptake by mobile organisms. Mobility is greatly influenced by the persistence, polarity, and physical state of the compound, and the climate, soil type and hydrology of its location. Drift from aerial spraying is affected by weather conditions, altitude of application, type of pesticide formulation, pilot experience, type of aircraft and spraying equipment.

That extensive drift can occur has been documented. Ware et al. (1970) reported that less than 50% of aerially sprayed insecticides were deposited on the intended agricultural target during summer months in Arizona. Under ideal conditions, and with correctly adjusted spray equipment,

total spray drift was 1.4% of applied spray, while under unfavorable conditions, with wrong adjustments of the sprayer, drift was 37% of the applied spray (Nordby and Skuterud 1974). Severe damage to susceptible crops from 2,4-D and related herbicides has been reported as far as 16 to 24 kilometers from the application site (Yates and Akesson 1963). Air over both urban and rural communities has been shown to contain pesticide residues that directly reflect pesticide applications in local agricultural and insect control operations (Tabor 1965). The amount of drift which occurs is not surprising when considering that during a 1.2 meter fall in a stable atmosphere, with only an 8 mph wind, a 50 micrometer diameter droplet of technical grade malathion will travel 46 meters, and a 5 micrometer diameter droplet will travel 4790 meters (Mount 1970). Droplet sizes of aerial sprays and aerosols range from <5 micrometer diameter to 700 micrometer diameter.

The direct and indirect toxicity to wildlife of many of the aerially applied pesticides has been documented. During a mosquito control operation in Grand Forks, North Dakota, more than 5,000 birds died from an aerial application of just 36.8 grams/acre of fenthion, an organophosphate (Seabloom et al. 1973). While the acute toxicity to some organisms of most organophosphates and carbamates has been studied, very little is known about sublethal poisoning and chronic effects of exposure to these pesticides. Effects could range from the undetectable to the serious impairment

of the ability to survive and reproduce. Indirect effects on survival could include such things as a decrease in available food supply. Pheasants (Phasianus colchicus) in nontreated areas were found to have consumed significantly more insects than those in pesticide treated areas; this paralleled the difference in the numbers of insects present in both areas (Messick et al. 1974).

There is a possibility of contamination of drinking water supplies and aquatic habitats because of the sandy soils and high water table in this region. Many organophosphorus pesticides were found to persist for over 6 months in a fine sand by Mulla et al. (1961). Shallow wells supplying water to a Florida town were contaminated for several months in 1962-1963 because of parathion seepage from a canal draining sprayed citrus groves (Brown 1978).

There were reportedly 410 million kilograms (active ingredient) of all types of pesticides used in the United States in 1974, with 94% of this being used in agriculture, 34% (141 million kilograms) were organophosphates and carbamates while 24% (98 million kilograms) were chlorinated compounds (Keil et al. 1977). In Wisconsin, 5 million kilograms of all types of pesticides were used in 1974 (Keil et al. 1977). The use of insecticides in Wisconsin has about doubled on every crop surveyed between 1970 and 1978. Of the total planted, 765,000 acres (28%) was treated in 1970 while 2,059,000 (55%) was treated in 1978 (Wisconsin Department of Agriculture 1979).

Irrigated acreage increased in Portage County, Wisconsin, from 17,000 acres in 1967 to 54,000 acres in 1977 with a parallel increase in aerially applied pesticides (Butler 1978). Much of this acreage is adjacent to wildlife management lands. The impact of this increased pesticide spraying has never been fully investigated.

This project was designed to determine if the present pesticide practices are a potential threat to wildlife on the Buena Vista Marsh, a wildlife management area surrounded by intensive agricultural lands.

The objectives of this study were:

1. To determine if drift of aerially applied cholinesterase inhibiting pesticides occurs over non-target wildlife management lands.
2. To determine if the incidence of drifting pesticides (should they occur) are correlated with a decline in cholinesterase levels of birdlife in the study area.
3. To determine if pesticides are present in drainage ditches.

## STUDY AREA

The Buena Vista Marsh, southwestern Portage County, Wisconsin, is typical of approximately 1,075,093 acres of other lands in 5 adjacent counties of Central Wisconsin undergoing a conversion from wild lands to irrigated agricultural lands (Fig. 2). These lands are of glacial outwash origin and consist of flat, sandy or shallow organic soils with a large, readily available groundwater supply close to the surface (United States Department of Agriculture 1974). These characteristics, which are appealing to the large scale crop producers, also provide a unique habitat for numerous forms of wildlife.

The Buena Vista Marsh Wildlife Area, approximately 12,000 acres of public land which is managed by the Wisconsin Department of Natural Resources is a haven for the threatened prairie chicken (Tympanuchus cupido) and other wildlife species (Fig. 3).

Two study areas were selected for the project. One study area (SA-S) is located in the town of Plover, Portage County, Wisconsin, (T22N R8E section 7) and is immediately adjacent to agricultural lands to the west and north. This section consists of permanent grasslands which are managed for grassland wildlife (Fig. 4). The other study area (SA-C) is located in the town of Buena Vista, Portage County, Wisconsin, (T22N R8E sections 28 and 29) approximately 3.2 kilometers to the southeast of SA-S. This area is identical to the study area but has no agricultural lands closer than

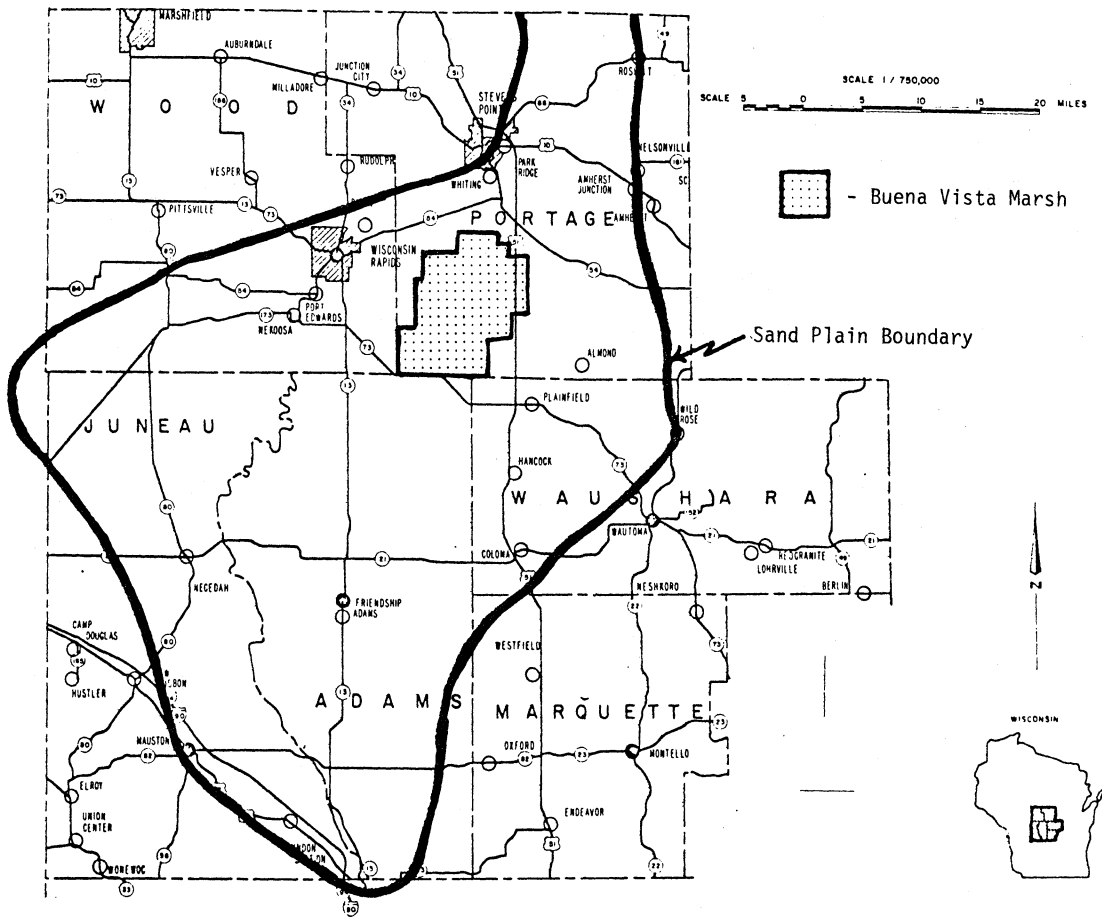


Figure 2. Sand plain region of Central Wisconsin (University of Wisconsin 1964).

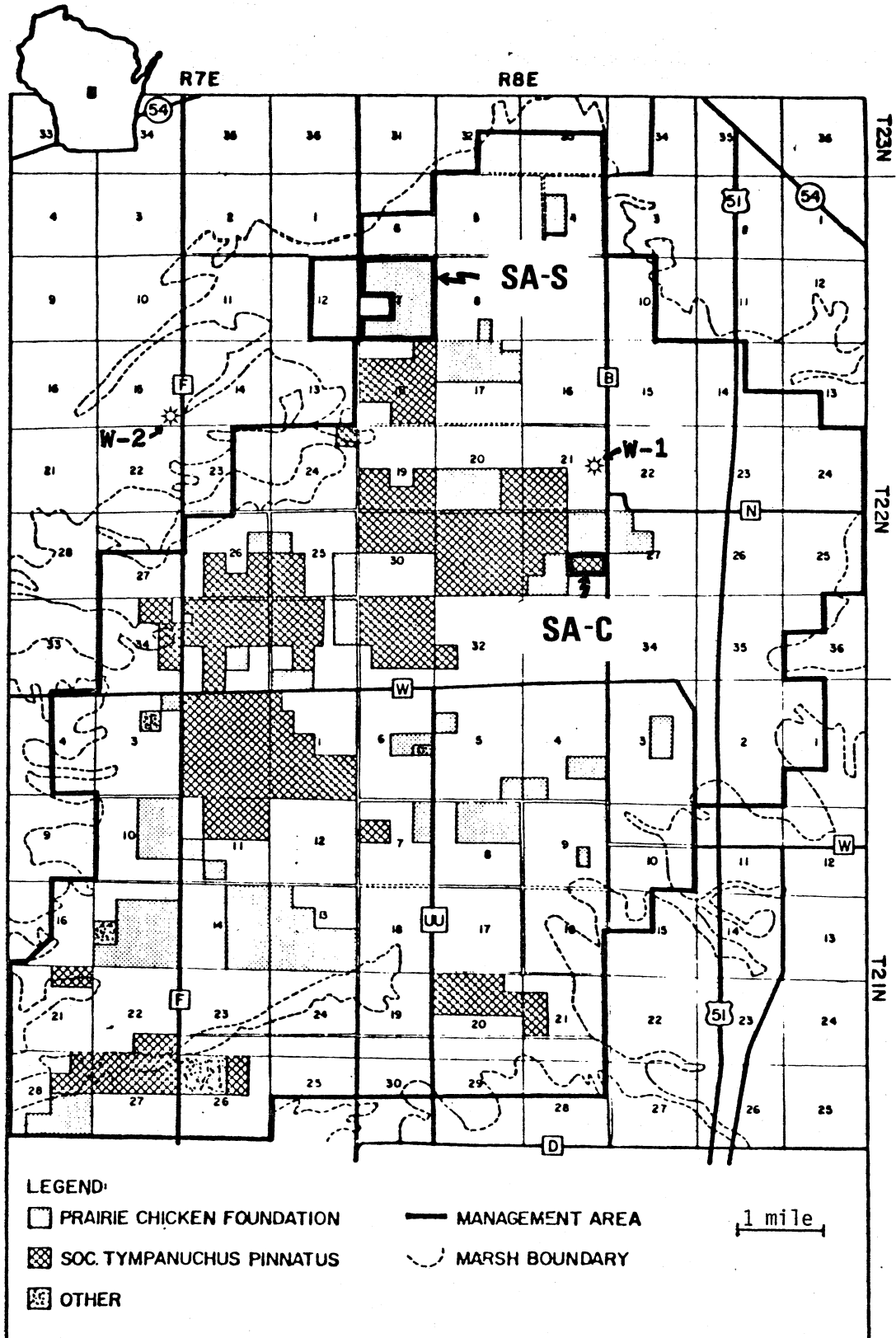
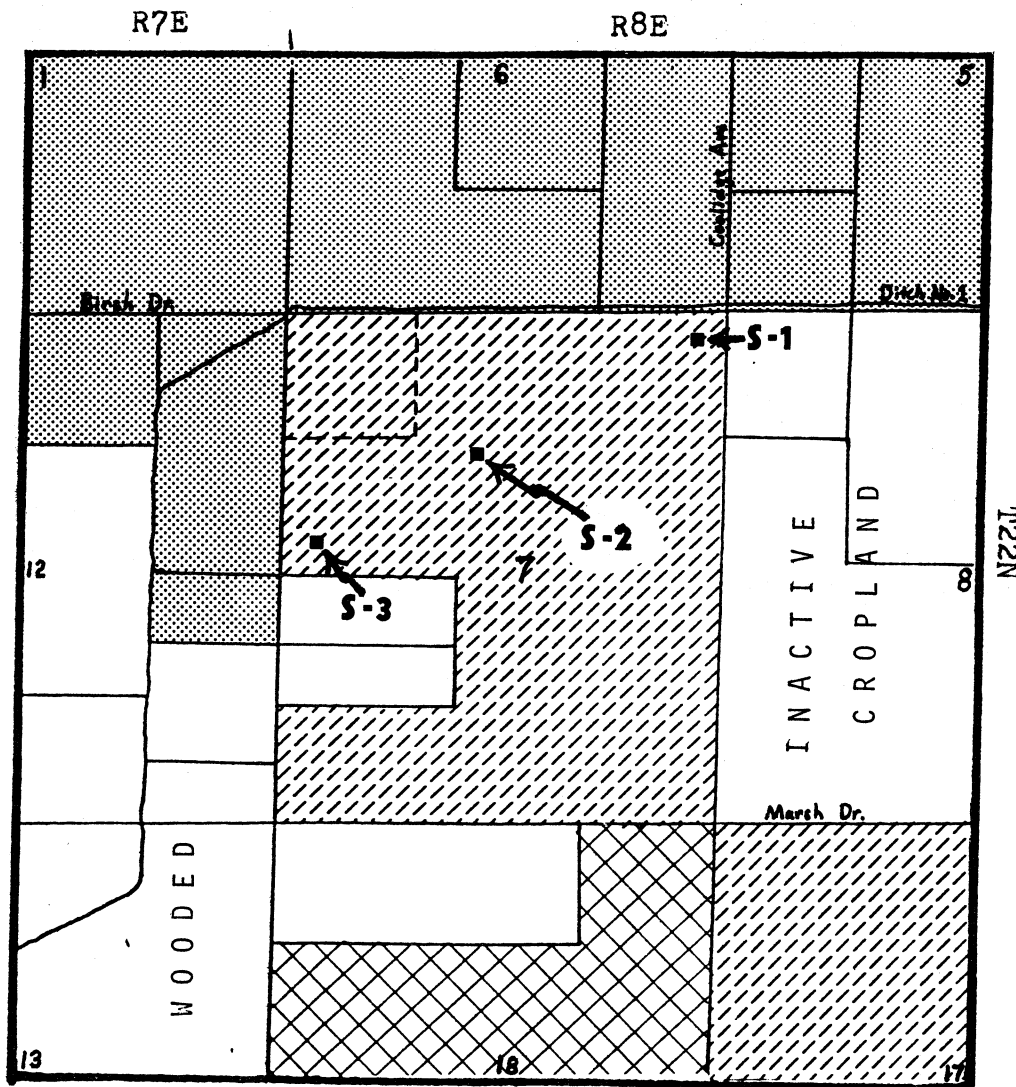


Figure 3. Buena Vista Marsh and location of study areas.





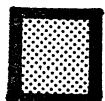
-  - SOCIETY TYMPANUCHUS CUPIDO PINNATUS
-  - PRAIRIE CHICKEN FOUNDATION
-  - ACTIVE CROPLAND

Figure 4. SA-S and locations of sample sites; S-1, S-2, and S-3.

2.4 kilometers to the west. One agricultural field is present to the east (Fig.5).

SA-S is potentially subject to continuous exposure from repeated applications of various chemicals.

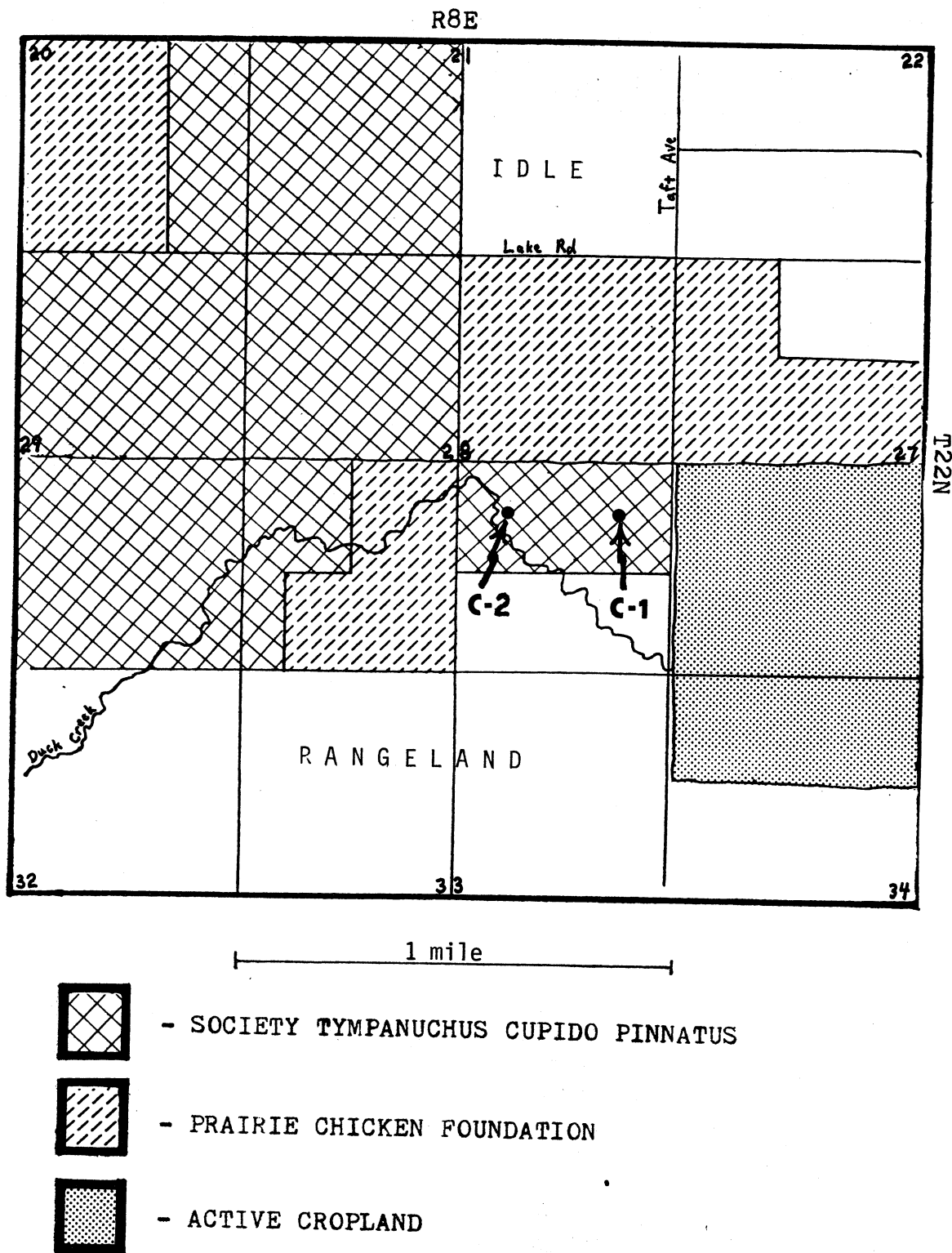


Figure 5. SA-C and locations of sample sites, C-1 and C-2.

## METHODS

### Air Sample Collection

Five air sampling sites were established, 3 on SA-S (S-1, S-2, S-3) and 2 on SA-C (C-1, C-2). These were placed downwind of the prevailing southwesterly winds (Fig. 4 and 5).

Pesticide fallout was collected on an ethylene glycol coated nylon cloth (43 cm x 43 cm) suspended horizontally in a wooden frame 60 cm above the ground at each site (Tessari and Spencer 1971).

The nylon cloth was first cleaned by Soxhlet extracting with acetone-hexane (1+1) for 4 hours, then saturated by storing in a solution of 10% ethylene glycol in acetone. Screens were ready for sampling upon removal from the solution. Prepared screens were suspended in the frame with alligator clips and exposed to air for the desired length of time. After the sampling period the screens were removed from the frames, wrapped in aluminum foil, placed on ice and taken to the College of Natural Resources, University of Wisconsin-Stevens Point. Samples were frozen until analyzed.

Samplers were left in the field for 3 to 7 day intervals from 4 May 1979 until 2 August 1979 after which it was discovered that a majority of the pesticides were degrading on the samplers over these sampling periods. Samplers were therefore collected at shorter intervals in early August.

### Air Sample Preparation

Samples were analyzed according to Tessari and Spencer

(1971). Reagents used in extraction and gas chromatographic analysis were pretested for purity. Anhydrous sodium sulfate was pretreated at 650°C for 2 hours to remove impurities. Aluminum oxide, acid washed, was prepared for use by shaking with 10%, by weight, distilled water for partial deactivation.

After a sampling period, pesticide residues were extracted from the screens using the Soxhlet method of extraction for 4 hours with acetone-hexane (1+1) as the solvent. The extract was transferred to a Kuderna-Danish flask, attached to a rotary evaporator and the solvent mixture removed under vacuum at 37°C until the volume was reduced to approximately 2 milliliters. The sample extract was transferred quantitatively to an alumina column prepared as follows: 2.5 cm Na<sub>2</sub>SO<sub>4</sub> was added to a column (2 cm x 30 cm), the column was filled with hexane, and 20 grams of alumina was added. After the alumina settled to the bottom, 5 cm of Na<sub>2</sub>SO<sub>4</sub> was poured on top. The hexane was drained to 1 cm above the Na<sub>2</sub>SO<sub>4</sub> layer. The sample was placed quantitatively on the column and eluted with 75 milliliters of hexane. The eluate was collected in a Kuderna-Danish flask, placed on a rotary evaporator and reduced to approximately 1 milliliter at 37°C under vacuum. The sample was then analyzed on the gas chromatograph, frozen for later analysis, or further purified.

#### Air Sample Purification

This cleanup was found to be unnecessary by June 1979 because of the selectivity of the alkaline flame ionization

detector on the gas chromatograph.

The sample extracts were further purified using semi-micro (25 cm x 5 mm i.d.) chromatographic columns filled with 2 grams florisil (activated at 130°C for 12 hours) and topped with 1 cm anhydrous sodium sulfate. The column was then pre-rinsed with 15 milliliters of hexane.

The extract was transferred quantitatively to the florisil column and eluted from the column with 15 milliliters of 6% ether in hexane (v/v) into a 20 milliliter glass vial. The vial was changed and the column eluted again with 15 milliliters of 15% ethyl ether in hexane. The vial was changed once again and eluted with 15 milliliters of 50% ethyl ether in hexane. Eluates were treated separately by evaporating almost to dryness under a gentle stream of nitrogen and diluting to 1 milliliter. Samples were then analyzed immediately or stored by freezing until gas chromatographic analysis.

#### Gas Chromatographic Analysis

Analytical standards obtained from the Environmental Protection Agency<sup>1</sup> were used for making up stock standard solutions of 100 ppm phorate, counter, diazinon, methyl parathion, malathion, ethyl parathion, trithion, and guthion. Working mixed standards containing 1 ppm (1 ng/ $\mu$ l) in isooctane were prepared for use in analysis. All standards were kept frozen until use.

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<sup>1</sup>U.S.E.P.A., Research Triangle Park, North Carolina.

Organophosphates were identified by comparing retention times of sample peaks versus standards on the column being used (Fig. 6 and 7).

Amounts of pesticides were determined by measuring peak heights. A plot of total nanogram weight of the standard pesticides injected versus peak height was used to determine the amounts of pesticides present in the samples.

If a standard was not available for a sample peak, it was tentatively identified using published tables of relative retention times to ethyl parathion and quantified using relative response ratios (EPA 1979).

A Packard series 7300 gas chromatograph with a flame ionization detector modified to a phosphorus selective alkaline flame ionization detector (U.S. Department of Health, Education, and Welfare 1977) was used for analysis. The U-shaped glass column was 200 cm long x 4 mm i.d. and was packed with 1.5% OV-17/1.95% OV-210 on Gas Chrom Q (Fig. 6). The carrier gas was nitrogen with a flow of 60 cc/min. Temperatures were injector 210°C, column 195°C, and detector 250°C. The air flow was 200 cc/min and hydrogen flow was 10 cc/min. Electrometer attenuation was set at  $256 \times 10^{-11}$ .

Confirmatory identification on 14 samples was done using a column of 4% SE-30/6% OV-210 on Gas Chrom Q (Fig. 7). Relative retention data on the columns agreed with literature values (EPA 1979).

#### Bird Sample Collection and Analysis

There are a number of acceptable methods presently used

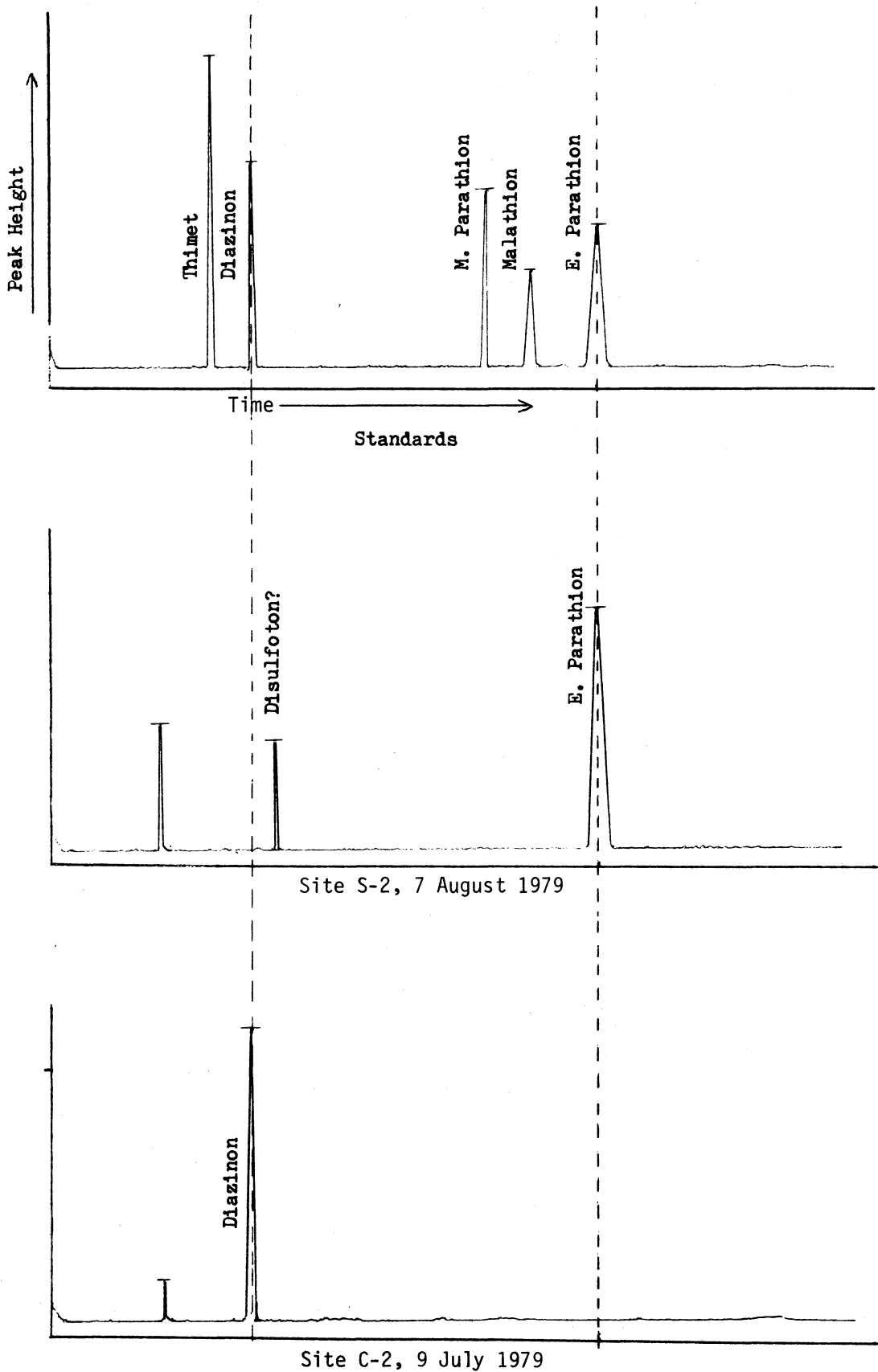


Figure 6. Gas chromatograph of standards compared with two samples on a 1.5% OV-17/1.95% OV-210 column.

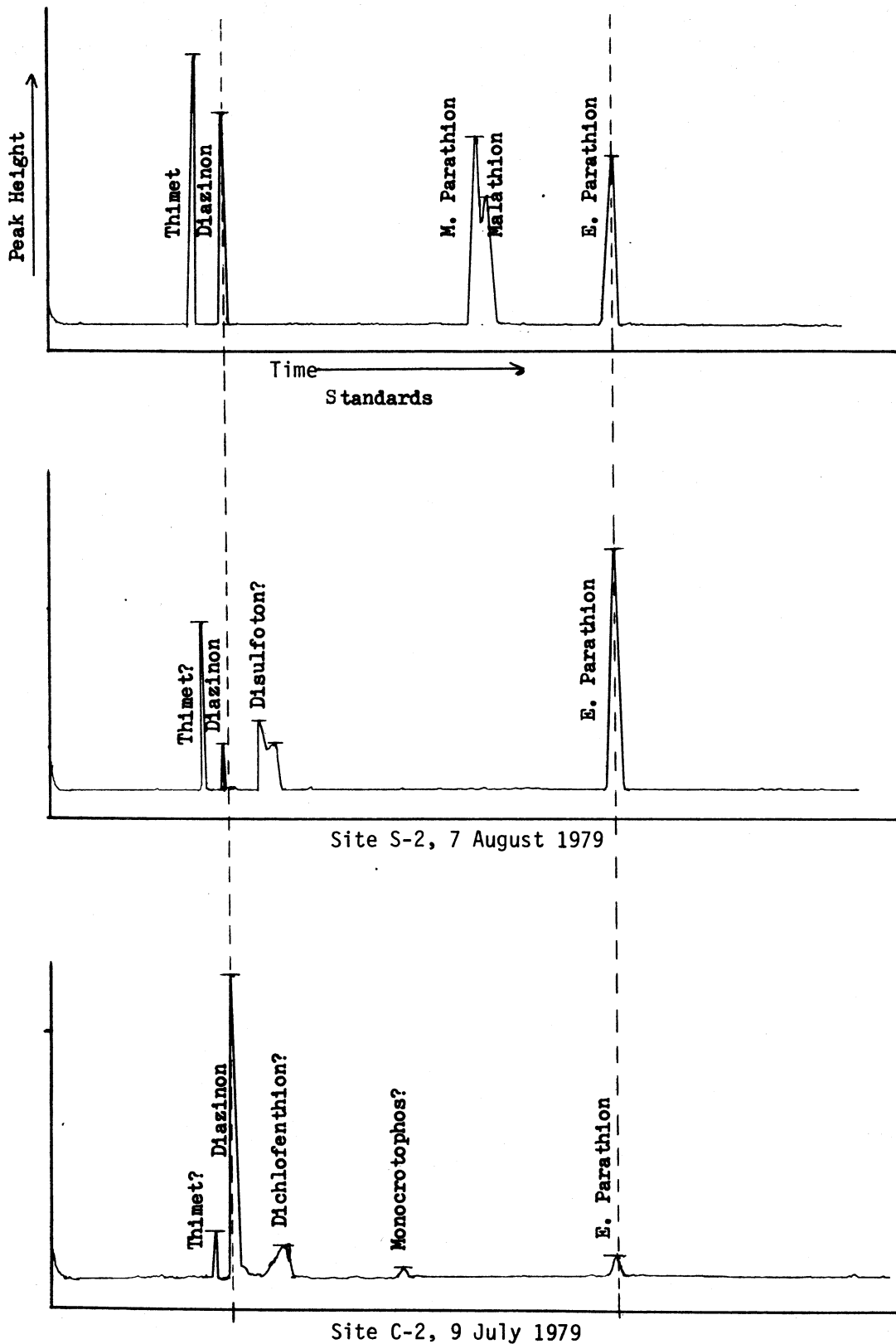


Figure 7. Gas chromatograph of standards compared with two samples on a 4% SE-30/6% OV-210 column. Confirmation of pesticides found in samples from figure 6.

for cholinesterase enzyme (ChE) analysis (Aldrich 1969). The colorimetric method of Ellman (1961) was chosen for use with the wild birds because it is relatively simple, quick, and inexpensive. Results are expressed as micromoles of substrate (acetylcholine iodide) hydrolyzed per minute per unit weight of brain tissue ( $\mu\text{moles}/\text{min}/\text{gram}$ ).

Federal and Wisconsin scientific collectors permits were obtained and birds were collected by shooting at or near the air sample sites. When a bird was collected it was put in an ice cooler. Specimens were taken to the laboratory within 4 hours for analysis. Data recorded included a code number, date, species and location. Analysis was then performed by the method of Ellman (1961) as recommended by Fleming (1979). The method is outlined in Appendix A.

#### Water Samples

Two ditches were preliminarily surveyed for organophosphate pesticides using the method of LaBel et al. (1979). Water samples were extracted by running the desired volume of water through XAD-2 resin and then eluted with acetone-hexane (15+85). The concentrated extract was analysed by gas chromatography.

## RESULTS AND DISCUSSION

### Air Samples

The detection limits for pesticide standards used in identifying and quantifying samples are presented in Table 1. These limits were determined by injecting pesticide standards at various concentrations and adjusting instrument parameters to obtain an optimum output.

Two recovery studies were undertaken. Before outside air sampling began a nylon screen was prepared, spiked with the standards and immediately extracted and analyzed. The same experimental design was repeated in a second recovery test except that the screen was exposed to the atmosphere for 3 days (16-19 July 1979) at a sample site. Recoveries dropped significantly (Table 2). Tessari and Spencer (1971) did not mention this as being a significant problem in their study. Their samplers may not have been directly exposed to sunlight and moisture as in this study. They did check for DDT "wash-of" with little loss, however organophosphates are likely to behave differently since they are more water soluble than DDT and easily degradable. Elliot (1980) recommended the use of OV-101 to replace the ethylene glycol because of less pesticide loss and easier cleanup. He has had success with this and it should be considered in future studies. After becoming aware of the degradation losses taking place, sampling times were reduced to 1-2 days as opposed to 3-7 days early in the summer of 1979. Elliot (1980) had experienced similar degradation problems during

TABLE 1. Detection limits for pesticide standards.

|  | <u>Pesticide</u> |         |                     |                    |           |
|--|------------------|---------|---------------------|--------------------|-----------|
|  | Diazinon         | Phorate | Methyl<br>Parathion | Ethyl<br>Parathion | Malathion |
| Detection<br>limit<br>( $\mu\text{g}/\text{m}^2$ ) | 0.25             | 0.25    | 0.50                | 0.50               | 0.50      |

TABLE 2. Recovery of pesticide standards from fortified cloth screen.

| Pesticide                            | Added<br>( $\mu\text{g}$ ) | Recovered<br>% |
|--------------------------------------|----------------------------|----------------|
| Immediate Extraction                 |                            |                |
| Diazinon                             | 1.0                        | 80             |
| Phorate                              | 1.0                        | 83             |
| Methyl Parathion                     | 1.0                        | 90             |
| Ethyl Parathion                      | 1.0                        | 80             |
| Malathion                            | 1.0                        | 75             |
| After 3 Days of Atmospheric Exposure |                            |                |
| Diazinon                             | 1.0                        | 5              |
| Phorate                              | 1.0                        | 0              |
| Methyl Parathion                     | 1.0                        | 0              |
| Ethyl Parathion                      | 1.0                        | 3              |
| Malathion                            | 1.0                        | 5              |

the summer of 1979 and had reduced sampling periods to one day or to hours in some cases. Pesticide levels obtained will be considered minimum values, because of the degradation which took place on the sample screens.

Other compounds which are listed, but not confirmed with standards, were identified using relative retention tables for the column being used according to EPA (1979) and were usually found only at trace levels. Standards injected agreed with these retention tables. Background information on all compounds identified can be found in Appendix B.

#### Air Sample Collection

In the spring and summer of 1979, 52 air samples from SA-S and SA-C were analyzed for selected organophosphorus pesticides. I was unable to analyze the air samples for all of the chemicals that were being applied during the growing season because of the complexity of analysis and an inability to obtain specific information about types and application times of chemicals being used in the area. Organophosphorus compounds were chosen for study on the basis of the analytical capabilities of the laboratory and observations of organophosphorus pesticides being used in the region. The method used did not include all organophosphorus pesticides in use. Major pesticides which were excluded are monitor and orthene because of their differing chemical properties.

#### Organophosphates in Air Samples

Diazinon, ethyl parathion, thimet, and malathion were

identified in air samples by comparison with standards. Disulfoton, dichlofenthion, monocrotophos, dimethoate, and fenthion have been tentatively identified from EPA relative retention data (EPA 1979). Retention times on the columns used were identical to those reported by the EPA (1979). The results of air sampling are presented in Appendix C.

The data indicates that diazinon and ethyl parathion occurred with greater frequency and quantity or they are more persistent on the samplers than the other compounds identified. A statistical summary for these two compounds is presented (Table 3).

A wide range of values was found for ethyl parathion, from a trace to 200 micrograms of compound/m<sup>2</sup> collection screen, and diazinon, from a trace to 24 micrograms/m<sup>2</sup>. These values are much higher than those found for ethyl parathion in the outdoor air of farm residences, up to 4.8 micrograms/m<sup>2</sup>, by Tessari and Spencer (1971). Their work was done in 1970 and their DDT values were high. The high levels of ethyl parathion and diazinon in this study may be a product of their increased use since the chlorinated pesticides have been phased out.

There was little or no drift of pesticides prior to the beginning of July 1979. The only organophosphorus pesticides expected to be in use during this period would be those applied as seed treatments or as granular applications at planting. This is a period when many fungicides are applied which are not organophosphates. Other pesticides may have

TABLE 3. Statistical summary of residues collected on sample screens.

| <u>Ethyl Parathion</u> |                 |                 |   |   |                                       |
|------------------------|-----------------|-----------------|---|---|---------------------------------------|
| Sample Site            | N <sup>1/</sup> | F <sup>2/</sup> | N $\bar{X}$ <sup>3/</sup><br>( $\mu\text{g}/\text{m}^2$ ) | F $\bar{X}$ <sup>4/</sup><br>( $\mu\text{g}/\text{m}^2$ ) | Range<br>( $\mu\text{g}/\text{m}^2$ ) |
| S-1                    | 13              | 8               | 3.32  | 5.40  | TR <sup>5/</sup> - 20.0               |
| S-2                    | 13              | 7               | 16.33   | 30.14   | TR - 200.00                           |
| S-3                    | 14              | 6               | 1.50  | 3.50  | TR - 12.0                             |
| C-1                    | 14              | 5               | 1.82  | 3.64  | TR - 12.5                             |
| C-2                    | 12              | 7               | 4.75  | 8.14  | TR - 40.0                             |

| <u>Diazinon</u> |    |   |   |   |                                       |
|-----------------|----|---|---|---|---------------------------------------|
| Sample Site     | N  | F | N $\bar{X}$<br>( $\mu\text{g}/\text{m}^2$ ) | F $\bar{X}$<br>( $\mu\text{g}/\text{m}^2$ ) | Range<br>( $\mu\text{g}/\text{m}^2$ ) |
| S-1             | 13 | 7 | 0.78  | 1.43  | TR - 4.0                              |
| S-2             | 13 | 6 | 0.69  | 1.50  | TR - 4.0                              |
| S-3             | 14 | 5 | 0.38  | 1.00  | TR - 1.0                              |
| C-1             | 14 | 7 | 0.50  | 1.00  | TR - 1.0                              |
| C-2             | 12 | 9 | 2.67  | 3.56  | TR - 24.0                             |

<sup>1/</sup> N = Number of samples taken.

<sup>2/</sup> F = Frequency of compound found out of N.

<sup>3/</sup> N $\bar{X}$  = Numerical mean (sample mean).

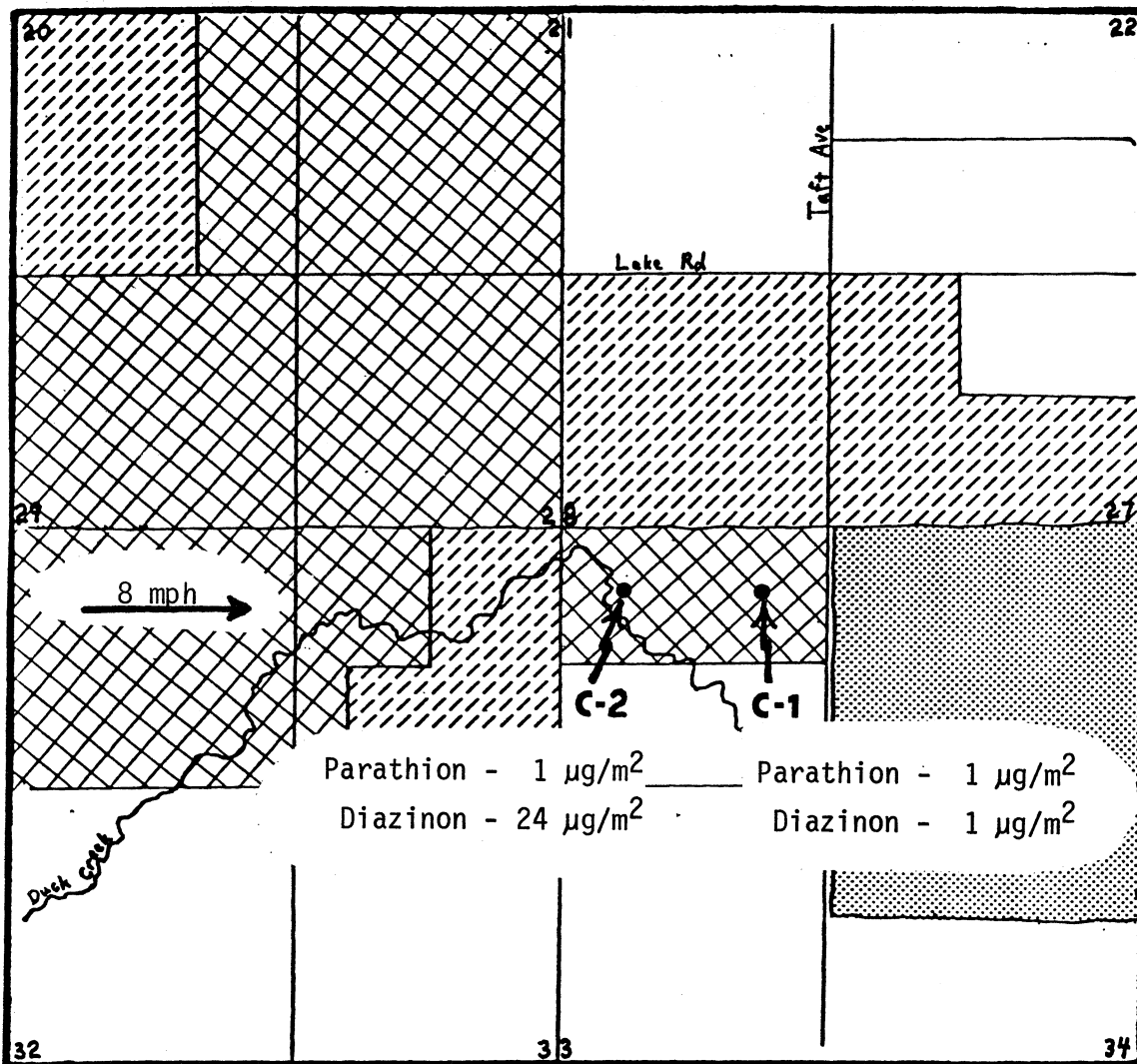
<sup>4/</sup> F $\bar{X}$  = Frequency mean (mean of samples containing pesticide).

<sup>5/</sup> TR = Trace (  $1\mu\text{g}/\text{m}^2$  )

been aerially drifting during this time but not analyzed.

A greater variety and larger quantities of pesticides were found in the air samples as the season progressed. Ethyl parathion usage was confirmed by noting the required posting of a warning sign on a snap bean field in the area. Warning signs are required by law when very toxic materials are being used and to warn individuals not to enter an area for a specified period of time. SA-C consistently had measurable pesticide residues even though no agricultural lands were present within 2.5 kilometers, except for one field immediately to the east. Possible interference from this field was checked from local weather data. On 9 July 1979 (Fig. 8) diazinon was found in SA-C at  $<1$  microgram/m<sup>2</sup> at C-1 and 24 microgram/m<sup>2</sup> at C-2. On 16 August 1979 (Fig. 9) ethyl parathion was found at 10 microgram/m<sup>2</sup> at C-1 and 40 microgram/m<sup>2</sup> at C-2. Winds at this time were westerly or southwesterly, thus precluding the possibility of contamination from the agricultural field to the east. Similarly on 7 August 1979 and 8 August 1979, compounds were found to be present on this area when winds were southwesterly or northwesterly. The finding of pesticide residues on SA-C indicates that applied pesticides may be drifting considerable distances ( $>2.5$  kilometers) and that with repeated spraying a continuous background level of pesticides may exist in the air of the Buena Vista Marsh during the spraying season.

Ethyl parathion was found in every sample from SA-S



1 mile




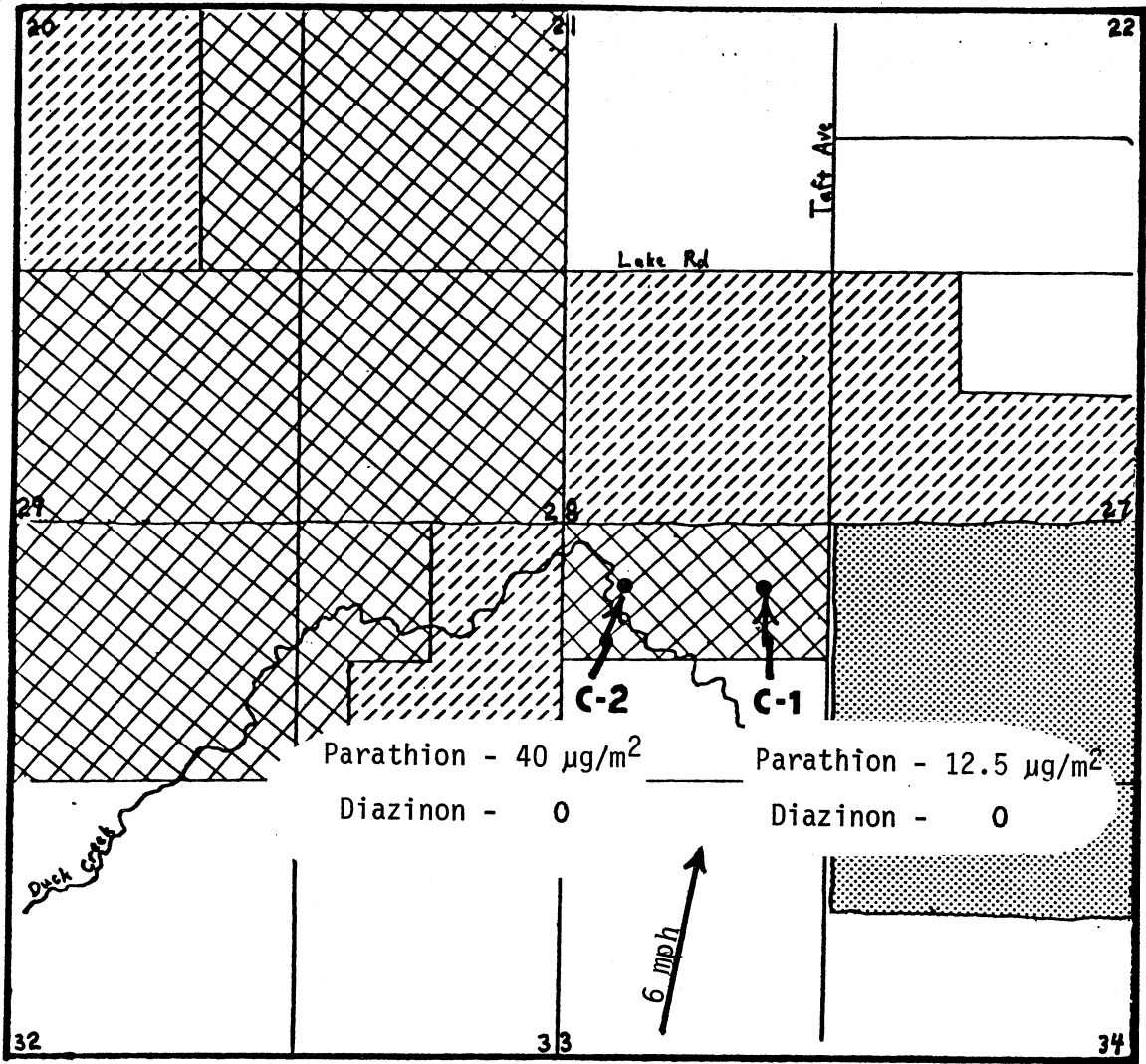
-  - SOCIETY TYMPANUCHUS CUPIDO PINNATUS
-  - PRAIRIE CHICKEN FOUNDATION
-  - ACTIVE CROPLAND

Figure 8. Pesticides found and wind conditions on SA-C, 9 July 1979.



- SOCIETY TYMPANUCHUS CUPIDO PINNATUS



- PRAIRIE CHICKEN FOUNDATION



- ACTIVE CROPLAND

Figure 9. Pesticides found and wind conditions on SA-C, 16 August 1979.

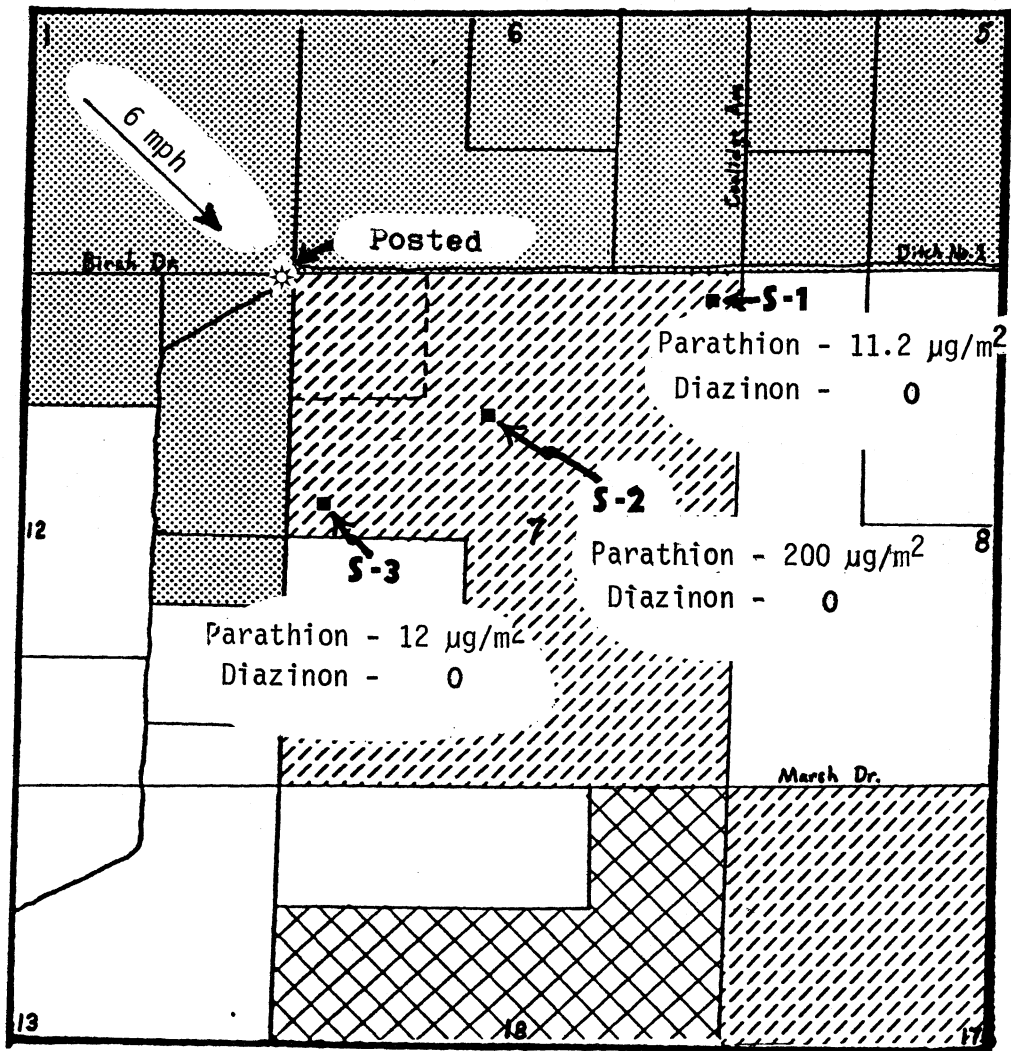
between 9 July 1979 and 16 August 1979. Adjacent fields were not always posted with a warning during this time. The pesticide may have come from more distant croplands.

On 8 August 1979 high levels of ethyl parathion were found on SA-S. Fields to the northwest of the study area were posted for spraying on this date; the wind was from the northwest at 5-10 mph (Fig. 10). Station S-2 was therefore directly in the path of any drift and subsequently received 200 micrograms/m<sup>2</sup> of ethyl parathion. S-2 is 0.5 kilometer from the agricultural fields; areas closer to the application site likely received higher doses. This level is approximately 1% of the recommended application rate on the target area. Station S-1 received 11.2 micrograms/m<sup>2</sup> and station S-3 received 12 micrograms/m<sup>2</sup>.

No measureable pesticide residues were found on 23 August 1979.

#### Cholinesterase Enzyme Assays

The relationship between brain cholinesterase activity and organophosphate exposure is good regardless of species, sex, and age (Ludke et al. 1975). There is general agreement that there is a direct relationship between brain cholinesterase inhibition and mortality, although there is a considerable difference of opinion among investigators regarding what degree of brain cholinesterase depression is diagnostic as to the cause of death. Mehrotra et al. (1967), working with house sparrows, generally considered 80% inhibition to be lethal, but observed brain cholinesterase inhibition of birds



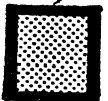
1 mile



- SOCIETY TYMPANUCHUS CUPIDO PINNATUS



- PRAIRIE CHICKEN FOUNDATION



- ACTIVE CROPLAND

Figure 10. Pesticides found and wind conditions on SA-S, 8 August 1979.

dying from malathion ranged from 31% to greater than 90%. Ludke et al. (1975) found mortality generally occurred in birds with brain cholinesterase below 40% of normal. He considered mortality accompanied by inhibition of brain cholinesterase greater than 50% suitable for diagnostic purposes and a 20% inhibition of brain cholinesterase activity indicative of exposure.

With this background and the ability to easily measure cholinesterase activity, its measurement can be used to determine if an animal has been exposed to organophosphate or carbamate pesticides. Any correlation between low cholinesterase levels and pesticide residues found in air samples might therefore indicate a direct effect of pesticide drift on wildlife in the Buena Vista Marsh.

Cholinesterase levels from 7 species of native birds were determined in the summer of 1979 (Tables 4 and 5).

Certain precautions were taken to assure that cholinesterase assay results were valid. The cholinesterase activity of birds suspected to have been exposed to cholinesterase inhibitors were compared with that of unexposed birds of the same species because of the great variation of activity between species (Stickel 1973). Storage of the enzyme containing tissue was such that deterioration of the enzyme between the time of death and the time of analysis was minimal. Ludke et al. (1975) found that there was a tendency toward reactivation of brain cholinesterase in suppressed bird samples when refrigerated (2°C, 7 days) or at ambient

TABLE 4. Cholinesterase levels of birds collected in the Buena Vista Marsh, SA-S, 1979.

| Sample No. | Date          | Species                | Site No. | ChE Level<br>( $\mu$ moles/min/gm) |
|------------|---------------|------------------------|----------|------------------------------------|
| S-5479-1   | 4 May 1979    | Red-winged             | S-3      | 15.73                              |
| S-5479-2   | "             | Blackbird              | S-3      | 22.55                              |
| S-5479-3   | "             | Cowbird                | S-3      | 17.60                              |
| S-5479-4   | "             | "                      | S-3      | 20.35                              |
| S-5479-5   | "             | "                      | S-3      | 18.70                              |
| S-5479-6   | "             | "                      | S-3      | 16.50                              |
| S-6479-1   | 4 June 1979   | Cowbird                | S-2      | 28.57                              |
| S-61179-1  | 11 June 1979  | Red-winged             | S-3      | 20.46                              |
| S-61179-2  | "             | Blackbird              | S-3      | 20.24                              |
| S-61179-3  | "             | Savannah               | S-2      | 19.80                              |
| S-61179-4  | "             | Sparrow                | S-3      | 22.88                              |
| S-61179-6  | "             | "                      | S-2      | 22.55                              |
| S-61179-5  | "             | Bobolink               | S-3      | 24.20                              |
| S-62179-3  | 21 June 1979  | Bobolink               | S-1      | 22.11                              |
| S-62179-1  | "             | Red-winged             | S-3      | 21.67                              |
| S-62179-2  | "             | Savannah               | S-2      | 23.54                              |
| S-62179-4  | "             | Sparrow                | S-1      | 20.35                              |
| S-62179-5  | "             | "                      | S-1      | 23.87                              |
| S-7279-1   | 2 July 1979   | Bobolink               | S-1      | 24.75                              |
| S-7279-2   | "             | Vesper Sparrow         | S-1      | 19.36                              |
| S-7279-3   | "             | Savannah               | S-2      | 19.80                              |
| S-7279-6   | "             | Sparrow                | S-1      | 22.33                              |
| S-7279-4   | "             | Bobolink               | S-3      | 25.30                              |
| S-7279-5   | "             | "                      | S-1      | 24.20                              |
| S-7979-1   | 9 July 1979   | Meadowlark             | S-1      | 16.72                              |
| S-7979-2   | "             | Savannah               | S-2      | 17.71                              |
| S-7979-3   | "             | Grasshopper<br>Sparrow | S-3      | 17.16                              |
| S-7979-4   | "             | Red-winged             | S-3      | 16.50                              |
| S-71679-1  | 16 July 1979  | Vesper Sparrow         | S-3      | 18.81                              |
| S-71679-2  | "             | Savannah               | S-2      | 22.99                              |
| S-71679-3  | "             | Sparrow                | S-2      | 19.14                              |
| S-71679-4  | "             | "                      | S-3      | 17.93                              |
| S-71679-5  | "             | "                      | S-1      | 17.71                              |
| S-71679-6  | "             | "                      | S-2      | 21.89                              |
| S-71679-7  | "             | "                      | S-1      | 18.81                              |
| S-8679-1   | 6 August 1979 | Savannah<br>Sparrow    | S-2      | 17.93                              |

TABLE 5. Cholinesterase levels of birds collected in the Buena Vista Marsh, SA-C, 1979.

| Sample No. | Date          | Species             | Site No. | ChE Level<br>( $\mu$ moles/min/gm) |
|------------|---------------|---------------------|----------|------------------------------------|
| C-6479-1   | 4 June 1979   | Bobolink            | C-2      | 19.97                              |
| C-6479-2   | "             | "                   | C-1      | 22.61                              |
| C-6479-3   | "             | "                   | C-2      | 19.47                              |
| C-61179-1  | 11 June 1979  | Bobolink            | C-2      | 21.34                              |
| C-61179-2  | "             | "                   | C-2      | 20.46                              |
| C-61179-3  | "             | Meadowlark          | C-2      | 17.71                              |
| C-62179-1  | 21 June 1979  | Bobolink            | C-1      | 23.10                              |
| C-7279-1   | 2 July 1979   | Savannah Sparrow    | C-1      | 17.82                              |
| C-7979-1   | 9 July 1979   | Grasshopper Sparrow | C-1      | 15.82                              |
| C-7979-2   | "             | Savannah Sparrow    | C-2      | 23.65                              |
| C-7979-3   | "             | Sparrow             | C-1      | 21.78                              |
| C-71679-1  | 16 July 1979  | "                   | C-1      | 18.37                              |
| C-8679-1   | 6 August 1979 | "                   | C-2      | 18.04                              |

temperatures (18-24°C, 2 days) but the increases were not significant. My procedure of placing the bird in an ice cooler and analyzing within 4 hours is well within these limits to prevent significant reactivation.

To obtain cholinesterase levels of unexposed birds, samples were collected in May and June 1979 prior to the spraying season. The early May 1979 values for red-winged blackbird (Agelaius phoeniceus) and brown-headed cowbird (Molothrus ater) however are low. These levels may be a product of the birds consuming treated seeds during the planting period. The effects of seed treatments on wildlife may in itself be significant. Wagner et al. (1965) reported the following effects could occur: reduced laying rate or temporary reproductive inhibition induced by seed treatments might alter the recruitment rate of a population by causing reduced clutch size, nest abandonment due to an insufficient number of eggs in the clutch to elicit incubation behavior, or a delay in nesting phenology. Any one or all of these alterations would be manifested in fall densities and age ratios.

Background cholinesterase activities are considered to be those obtained from bird species during the period 4 June 1979 to 2 July 1979 when pesticide levels in the air were found to be relatively low in the sample areas and organophosphates were not believed to be heavily applied (Table 6). Sufficient data was obtained for red-winged blackbird, bobolink (Dolichonyx oryzivorus), and savannah

TABLE 6. Background Cholinesterase Data (4 June-2 July 1979).

| Species              | Number of<br>Birds | Mean $\pm$ S.D.  |
|----------------------|--------------------|------------------|
| Red-winged blackbird | 3                  | 20.79 $\pm$ 0.62 |
| Brown-headed cowbird | 1                  | 28.57            |
| Bobolink             | 8                  | 22.83 $\pm$ 2.05 |
| Savannah sparrow     | 8                  | 21.89 $\pm$ 1.56 |
| Vesper sparrow       | 1                  | 19.36            |

sparrow (Passerculus sandwichensis). With all of the Bobolink data being used to obtain its background and no abnormally low values, only red-winged blackbird and savannah sparrow remain to be compared as possibly affected birds. No bobolinks were available later in the summer because they are one of the first birds to leave the area after nesting. Red-winged blackbirds and savannah sparrows remain longer but they became more difficult to obtain later in the summer as they began to flock and leave their nesting area (Kumlien and Hollister 1951).

One red-winged blackbird and 7 savannah sparrows had values which were at least 2 standard deviations below the mean (Tables 7 and 8). The red-winged blackbird's activity was 20.6% below the mean, while 5 savannah sparrow's activities were depressed nearly to that level (19.1%, 18.1%, 19.1%, 18.1% and 18.6%).

Using the most stringent criterion of 50% cholinesterase depression (Ludke et al. 1975), none of the collected birds were in danger of dying from direct poisoning due to cholinesterase inhibiting pesticides. However at least 6 of the birds had activities depressed about 20% below the mean of the species. This indicates exposure had occurred. Two additional birds had activities depressed greater than 2 standard deviations below the mean.

Little experimental work has been done concerning the effects of sublethal cholinesterase inhibition on birds. McEwen and Brown (1966) state that effects on individuals can

TABLE 7. Brain cholinesterase activities of birds collected on SA-S (9 July-6 August 1979).

| Species              | Background<br>( $\mu$ moles/min/gm) | Range<br>( $\mu$ moles/min/gm) | Date         | ( $\mu$ moles/min/gm) | %<br>Inhibition |
|----------------------|-------------------------------------|--------------------------------|--------------|-----------------------|-----------------|
| Red-winged blackbird | 20.79 $\pm$ 0.62                    | 20.24 - 21.67                  | 9 July 1979  | 16.50                 | 20.6            |
| Savannah sparrow     | 21.89 $\pm$ 1.56                    | 19.47 - 25.30                  | 9 July 1979  | 17.71                 | 19.1            |
|                      |                                     |                                | 16 July 1979 | 22.99                 |                 |
|                      |                                     |                                | 16 July 1979 | 19.14                 | 12.6            |
|                      |                                     |                                | 16 July 1979 | 17.93                 | 18.1            |
|                      |                                     |                                | 16 July 1979 | 17.71                 | 19.1            |
|                      |                                     |                                | 16 July 1979 | 21.89                 |                 |
|                      |                                     |                                | 16 July 1979 | 18.81                 | 14.1            |

TABLE 8. Brain cholinesterase activities of birds collected on SA-C (2 July-6 August 1979).

| Species          | Background<br>( $\mu$ moles/min/gm) | Range<br>( $\mu$ moles/min/gm) | Date          | ( $\mu$ moles/min/gm) | %<br>Inhibition |
|------------------|-------------------------------------|--------------------------------|---------------|-----------------------|-----------------|
| Savannah sparrow | 21.89 $\pm$ 1.56                    | 19.47 - 25.30                  | 2 July 1979   | 17.82                 | 18.6            |
|                  |                                     |                                | 9 July 1979   | 23.65                 |                 |
|                  |                                     |                                | 9 July 1979   | 21.78                 |                 |
|                  |                                     |                                | 16 July 1979  | 18.37                 | 16.1            |
|                  |                                     |                                | 2 August 1979 | 18.04                 | 17.6            |

range from undetectable to serious impairment of ability to survive and reproduce.

It is therefore possible that the levels found in this study might have increased susceptibility to predation or decreased ability to obtain food. Fifty-four percent (7 out of 13) of the red-winged blackbirds and savannah sparrows evaluated after 2 July 1979 had significantly decreased cholinesterase levels.

The unavailability of birds with lower cholinesterase levels due to mortality or behavioral changes is a possibility which could be overcome by using extensive ground searches.

#### Comparison of Air Samples and Cholinesterase Assays

Comparison of average cholinesterase levels (Tables 9 and 10) for red-winged blackbirds and savannah sparrows with pesticide residues found in air samples on SA-S (Fig. 11) and SA-C (Fig. 12) shows a possible relationship between low enzyme activity in the birds and high pesticide levels due to drift.

#### Water Samples

One of the drainage ditch samples (W-1) was obtained from Ditch-8 at Taft Road (Fig. 3) and contained 37 ppb of ethyl parathion. This ditch flows through some intensively farmed areas immediately upstream of this sampling point. The other ditch sample (W-2) was obtained from Ditch-2 (Fig. 3) on County F and did not contain any identifiable residues. This ditch runs through marsh land prior to the

TABLE 9. Average cholinesterase levels on SA-S, 1979.

| Species  | Brain Cholinesterase Level (umoles/min/gram) |              |               |               |              |              |               |              |
|--|--|--------------|---------------|---------------|--------------|--------------|---------------|--------------|
|  | <u>4 May</u>                                 | <u>4 Jun</u> | <u>11 Jun</u> | <u>21 Jun</u> | <u>2 Jul</u> | <u>9 Jul</u> | <u>16 Jul</u> | <u>6 Aug</u> |
| Red-Winged Blackbird<br>( <u>Agelaius phoeniceus</u> )   | 19.14<br>(2)*                                | -            | 20.35<br>(2)  | 21.67<br>(1)  | -            | 16.50<br>(1) | -             | -            |
| Brown-headed Cowbird<br>( <u>Molothrus ater</u> )        | 18.29<br>(4)                                 | 28.57<br>(1) | -             | -             | -            | -            | -             | -            |
| Bobolink<br>( <u>Dolichonyx oryzivorus</u> )             | -  | 20.68<br>(3) | 24.20<br>(1)  | 22.11<br>(1)  | 24.75<br>(3) | -            | -             | -            |
| Savannah Sparrow<br>( <u>Passerculus sandwichensis</u> ) | -  | -            | 21.74<br>(3)  | 22.59<br>(3)  | 21.06<br>(2) | 17.71<br>(1) | 19.74<br>(6)  | 17.93<br>(1) |
| Vesper Sparrow<br>( <u>Poocetes gramineus</u> )          | -  | -            | -             | -             | 19.36<br>(1) | -            | 18.81<br>(1)  | -            |
| Easter Meadowlark<br>( <u>Sturnella magna</u> )          | -  | -            | -             | -             | -            | 16.72<br>(1) | -             | -            |
| Grasshopper Sparrow<br>( <u>Ammodramus savannarum</u> )  | -  | -            | -             | -             | -            | 17.16<br>(1) | -             | -            |

(n)\*-Number of Birds

TABLE 10. Average cholinesterase levels on SA-C, 1979.

| Species  | Brain Cholinesterase Level (umoles/min/gram) |               |              |              |               |              |
|--|--|---------------|--------------|--------------|---------------|--------------|
|  | <u>11 Jun</u>                                | <u>21 Jun</u> | <u>2 Jul</u> | <u>9 Jul</u> | <u>16 Jul</u> | <u>6 Aug</u> |
| Bobolink<br>( <u>Dolichonyx</u><br><u>oryzivorus</u> )             | 20.90<br>(2)*                                | 23.10<br>(1)  | -            | -            | -             | -            |
| Savannah Sparrow<br>( <u>Passerculus</u><br><u>sandwichensis</u> ) | -  | -             | 17.82<br>(1) | 22.71<br>(2) | 18.37<br>(1)  | 18.04<br>(1) |
| Eastern Meadowlark<br>( <u>Sturnella magna</u> )                   | 17.71<br>(1)                                 | -             | -            | -            | -             | -            |
| Grasshopper Sparrow<br>( <u>Ammodramus</u><br><u>savannarum</u> )  | -  | -             | -            | 15.84<br>(1) | -             | -            |

(n)\*-Number of Birds

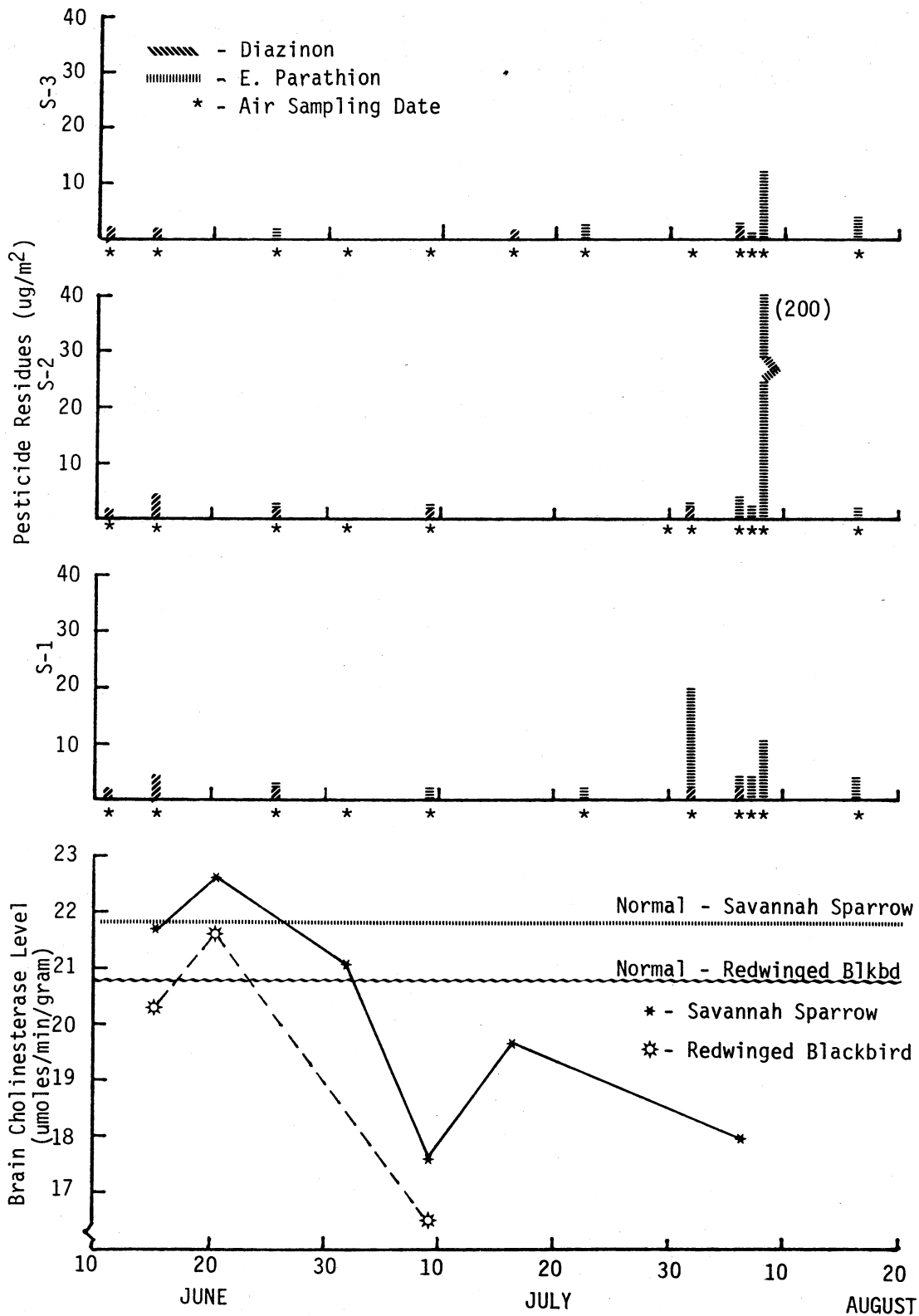


Figure 11. Comparison of air sample results and cholinesterase assays on SA-S, 1979.

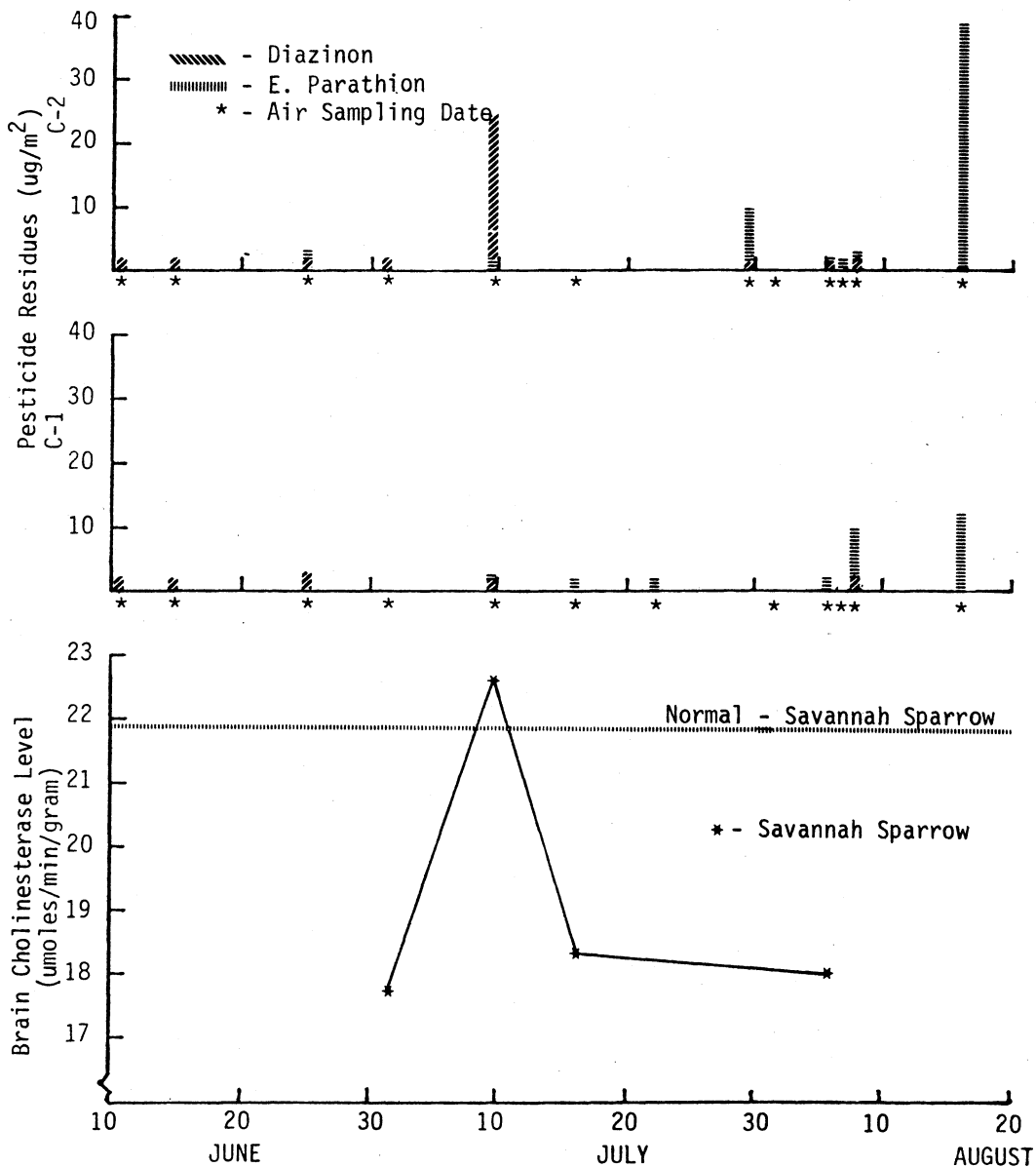


Figure 12. Comparison of air sample results and cholinesterase assays on SA-C, 1979.

sampling point. These samples were obtained on 23 August 1979 during a heavy rain and there may be a dilution factor, also levels may be higher at times when the pesticides are being applied.

This preliminary study indicates a need for additional studies on surface water pollution from agricultural chemicals in the Wisconsin Central Sands Region.

Only a small fraction of the total number of different compounds being used in the region were investigated. Any one of those which were not analyzed for are also potential pollutants. The possibility of harmful effects on wildlife or humans is justification for further study.

## SUMMARY AND CONCLUSIONS

This study was hampered by several factors. I was unable to obtain specific information about types and application times of chemicals being used in the area. Therefore, a broad spectrum of potential chemicals was searched over a broad period of time. Also some residues may have degraded during the sampling period. Hence, the levels of chemical residues found on the air samplers are minimal values of a small fraction of the total number of different compounds being used.

From the data obtained, the following conclusions are made:

1. A variety of organophosphorus pesticides are drifting (at least 2.5 kilometers) and contaminating the air over non-target areas of the Buena Vista Marsh.
2. Pesticides are suspected of producing an effect on the cholinesterase levels of native birdlife on the Buena Vista Marsh.
3. A relationship between aerial drift of pesticides and decreased cholinesterase activity in birds is possible.
4. Pesticides may be moving through the ecosystem via surface drainage water in the Buena Vista Marsh.

## RECOMMENDATIONS

This study is an indication of the potential hazards of pesticide movement in the Wisconsin Central Sands Region. The following areas require more study:

1. Further research should be done in measuring pesticide movement in air and water in the Central Wisconsin area.
2. Further research should be directed toward determining normal and exposure values of brain cholinesterase activity in a number of commonly occurring species in Central Wisconsin.

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APPENDIX A. Cholinesterase assay procedure and reagents.

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CHOLINESTERASE PROCEDURE

1. Homogenize whole brain in Tris buffer (Solution 3) at 1 gram of brain per 10 ml of buffer.
  - a. Preweigh a small beaker, add brain, weigh via subtraction, add buffer (if brain wt. = 500 mg, then add 5 ml buffer).
  - b. Pour into homogenizing tube and homogenize thoroughly.
  - c. May be assayed at this time or stored refrigerated (1-3 days) or frozen (indefinitely?) until assayed.
2. To a cuvette, add:
  - a. 2.9 ml chromagin reagent (Solution 1),
  - b. 20  $\mu$ l homoginate (to bottom and center of cuvette),  
(the mixture is stable at this point since the substrate has not yet been added)
  - c. 100  $\mu$ l acetylthiocholine iodide substrate (Solution 2)(if added on wall of cuvette, above the solution already contained in the cuvette, a new pipet tip will not be necessary for each sample),
  - d. and mix with plastic spatula or glass rod.
3. Read the change in absorbance on a spectrophotometer set at 405 nm.
4. Plasma may be assayed directly for plasma cholinesterase by omitting step 1 above and substituting plasma for homoginate in step 2b.
5. Calculations:  
$$\Delta\text{Absorbance}/\text{min} \times 110 \text{ (for brains)} = \mu\text{moles}/\text{min}/\text{gram of tissue}$$
$$\Delta\text{Absorbance}/\text{min} \times 11,353 \text{ (for plasma)} = \mu\text{moles}/\text{min}/\text{ml of plasma}$$

APPENDIX A. (continued)

CHOLINESTERASE ASSAY REAGENTS

SOLUTIONS NUMBER OF ASSAYS  
FOR STATED VOLUME

---

CHROMAGIN REAGENT .... Solution 1

DTNB (dithiobisnitrobenzene acid)  
0.00025M (0.099g/l) 9.9 mg  
Tris buffer, 0.05M, pH 7.4(25°C)  
Trizma HCl (6.61g/l) 661.0 mg  
Trizma base (0.97g/l) 97.0 mg  
Deionized water bring to 100 ml  
TOTAL = 100ml of solution

30-33

Refrigerate in dark bottle  
Shelf life = 1-2 months

---

SUBSTRATE .... Solution 2

Acetylthiocholine iodide, 0.156M  
(45.12g/l) 451.1 mg  
Deionized water bring to 10ml  
TOTAL = 10ml of solution

95-100

Refrigate in dark bottle  
Shelf life = 1 week

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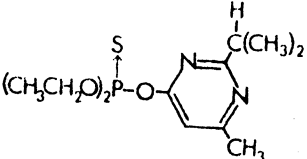
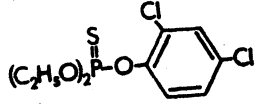
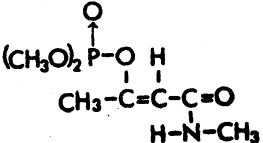
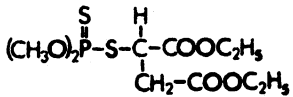
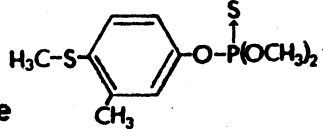
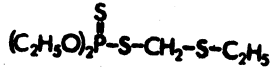
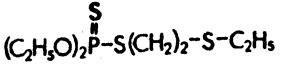
TRIS BUFFER .... Solution 3

0.05m, pH 8.0(25°C)  
Trizma HCL 4.44 gr  
Trizma base 2.65 gr  
Deionized water bring to 1 liter  
TOTAL = 1 liter of solution

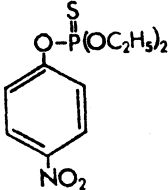
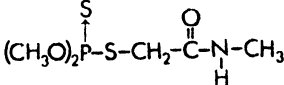
Store on shelf, no need to refrigate  
Shelf life is not critical

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APPENDIX B. General information on pesticides found in this study (EPA 1978).

| Name           | Synonyms                                       | Chemical Name   | Structure   | LD <sub>50</sub> (mg/kg) |
|----------------|--|---|---|--------------------------|
| Diazinon       | Basudin<br>Spectracide<br>Diazitol<br>Neocidol | O,O-diethyl O-(2-isopropyl-6-methyl-4-pyrimidinyl) phosphorothioate |    | 100 - 150                |
| Dichlofenthion | VC-13<br>Mobilawn                              | O,O-diethyl O-(2,4-dichloro-phenyl) phosphorothioate                |    | 270                      |
| Monocrotophos  | Azodrin<br>Nuvacron                            | 3-(dimethyl phosphinyloxy)-N-methylxicrotonamide                    |    | 21                       |
| Malathion      | Malathon<br>Emmaton<br>Cythion                 | O,O-dimethyl S-(1,2-di(ethoxycarbonyl)ethyl) phosphorodithioate     |    | 1000 - 1375              |
| Fenthion       | Baycid<br>Baytex<br>Entex                      | O,O-dimethyl O-(3-methyl-4-methylthio-phenyl) phosphorothioate      |   | 215                      |
| Phorate        | Thimet<br>Granutox                             | O,O-diethyl S-((ethylthio)-methyl) phosphorodithioate               |  | 1 - 5                    |
| Disulfoton     | DiSyston<br>Dithiosystox<br>Solvirex           | O,O-diethyl S-(2-(ethylthio)-ethyl) phosphorodithioate              |  | 12.5                     |

APPENDIX B. (continued)

| Name               | Synonyms                       | Chemical Name   | Structure   | LD <sub>50</sub> (mg/kg) |
|--------------------|--------------------------------|---|---|--------------------------|
| Ethyl<br>Parathion | Phoskil<br>Fosferno<br>Parawet | O,O-diethyl O-<br>(p-nitrophenyl)<br>phosphorothioate                     |  | 3 - 30                   |
| Dimethoate         | Cygon<br>De-fend               | O,O-dimethyl S-<br>(N-methyl-carbam-<br>oylmethyl)phos-<br>phorodithioate |  | 215                      |

APPENDIX C. Organophosphate pesticide residues in air samples on each sample date.

31 May 1979

|                        | <u>WIND SPEED</u> | <u>WIND DIRECTION</u> |
|------------------------|-------------------|-----------------------|
| <u>UWSP</u> (8 A.M.)   | -                 | -                     |
| <u>FIELD</u> (10 A.M.) | -                 | -                     |

Pesticide concentration for each site (ug pesticide/m<sup>2</sup> collection screen)

| PESTICIDE       | S-1 | S-2 | S-3 | C-1 | C-2 |
|-----------------|-----|-----|-----|-----|-----|
| DIAZINON        | TR* | TR  | TR  | TR  | TR  |
| ETHYL PARATHION | -   | -   | -   | -   | -   |
| MALATHION       | -   | -   | -   | -   | -   |
| PHORATE         | -   | -   | -   | -   | -   |
| MONOCROTOPHOS   | -   | -   | -   | -   | -   |
| DIMETHOATE      | -   | -   | -   | -   | -   |
| DISULFOTON      | -   | -   | -   | -   | -   |
| DICHLOFENTHION  | -   | -   | -   | -   | -   |
| FENTHION        | -   | -   | -   | -   | -   |

\*TR=Trace(<1ug/m<sup>2</sup>)

11 June 1979

|                        | <u>WIND SPEED</u> | <u>WIND DIRECTION</u> |
|------------------------|-------------------|-----------------------|
| <u>UWSP</u> (8 A.M.)   | 3-7 mph           | SW                    |
| <u>FIELD</u> (10 A.M.) | -                 | -                     |

Pesticide concentration for each site (ug pesticide/m<sup>2</sup> collection screen)

| PESTICIDE       | S-1 | S-2 | S-3 | C-1 | C-2 |
|-----------------|-----|-----|-----|-----|-----|
| DIAZINON        | TR* | TR  | 1   | TR  | TR  |
| ETHYL PARATHION | -   | -   | -   | -   | -   |
| MALATHION       | -   | -   | -   | -   | -   |
| PHORATE         | -   | -   | -   | -   | -   |
| MONOCROTOPHOS   | -   | -   | -   | -   | -   |
| DIMETHOATE      | -   | -   | -   | -   | -   |
| DISULFOTON      | -   | -   | -   | -   | -   |
| DICHLOFENTHION  | -   | -   | -   | -   | -   |
| FENTHION        | -   | -   | -   | -   | -   |

\*TR=Trace(<1ug/m<sup>2</sup>)

APPENDIX. (continued)

14 June 1979

|                        | <u>WIND SPEED</u> | <u>WIND DIRECTION</u> |
|------------------------|-------------------|-----------------------|
| <u>UWSP</u> (8 A.M.)   | 11-17 mph         | NE                    |
| <u>FIELD</u> (10 A.M.) | -                 | -                     |

Pesticide concentration for each site (ug pesticide/m<sup>2</sup> collection screen)

| PESTICIDE       | S-1 | S-2 | S-3 | C-1 | C-2 |
|-----------------|-----|-----|-----|-----|-----|
| DIAZINON        | 4   | 4   | TR* | TR  | TR  |
| ETHYL PARATHION | -   | -   | -   | -   | -   |
| MALATHION       | -   | -   | -   | -   | -   |
| PHORATE         | -   | -   | -   | -   | -   |
| MONOCROTOPHOS   | -   | -   | -   | -   | -   |
| DIMETHOATE      | -   | -   | -   | -   | -   |
| DISULFOTON      | -   | -   | -   | -   | -   |
| DICHLOFENTHION  | -   | -   | -   | -   | -   |
| FENTHION        | -   | -   | -   | -   | -   |

\*TR=Trace(<1ug/m<sup>2</sup>)

25 June 1979

|                        | <u>WIND SPEED</u> | <u>WIND DIRECTION</u> |
|------------------------|-------------------|-----------------------|
| <u>UWSP</u> (8 A.M.)   | 3-6 mph           | S                     |
| <u>FIELD</u> (10 A.M.) | 10-15 mph         | SW                    |

Pesticide concentration for each site (ug pesticide/m<sup>2</sup> collection screen)

| PESTICIDE       | S-1 | S-2 | S-3 | C-1 | C-2 |
|-----------------|-----|-----|-----|-----|-----|
| DIAZINON        | TR* | TR  | -   | 1   | TR  |
| ETHYL PARATHION | TR  | TR  | 1   | -   | TR  |
| MALATHION       | TR  | -   | -   | -   | -   |
| PHORATE         | -   | -   | -   | -   | -   |
| MONOCROTOPHOS   | -   | -   | -   | -   | -   |
| DIMETHOATE      | TR  | -   | -   | -   | TR  |
| DISULFOTON      | -   | -   | -   | -   | -   |
| DICHLOFENTHION  | -   | -   | -   | -   | -   |
| FENTHION        | -   | -   | -   | -   | -   |

\*TR=Trace(<1ug/m<sup>2</sup>)

APPENDIX C. (continued)

2 July 1979

|   | <u>WIND SPEED</u> | <u>WIND DIRECTION</u> |     |     |     |     |  |
|---|-------------------|-----------------------|-----|-----|-----|-----|--|
| <u>UWSP</u> (8 A.M.)  | 3-6 mph           | E                     |     |     |     |     |  |
| <u>FIELD</u> (10 A.M.)  | 5-10 mph          | SW                    |     |     |     |     |  |
| Pesticide concentration for each site (ug pesticide/m <sup>2</sup> collection screen) |                   |                       |     |     |     |     |  |
| PESTICIDE   | S-1               | S-2                   | S-3 | C-1 | C-2 |     |  |
| DIAZINON  | -                 | -                     | -   | -   | -   | TR* |  |
| ETHYL PARATHION   | -                 | -                     | -   | -   | -   | -   |  |
| MALATHION   | -                 | -                     | -   | -   | -   | -   |  |
| PHORATE   | -                 | -                     | -   | -   | -   | -   |  |
| MONOCROTOPHOS   | -                 | -                     | -   | -   | -   | -   |  |
| DIMETHOATE  | -                 | -                     | -   | -   | -   | -   |  |
| DISULFOTON  | -                 | -                     | -   | -   | -   | TR  |  |
| DICHLOFENTHION  | -                 | -                     | -   | -   | -   | TR  |  |
| FENTHION  | -                 | -                     | -   | -   | -   | -   |  |

\*TR=Trace(<1ug/m<sup>2</sup>)

9 July 1979

|   | <u>WIND SPEED</u> | <u>WIND DIRECTION</u> |     |     |     |    |  |
|---|-------------------|-----------------------|-----|-----|-----|----|--|
| <u>UWSP</u> (8 A.M.)  | -                 | -                     |     |     |     |    |  |
| <u>FIELD</u> (10 A.M.)  | 5-10 mph          | W                     |     |     |     |    |  |
| Pesticide concentration for each site (ug pesticide/m <sup>2</sup> collection screen) |                   |                       |     |     |     |    |  |
| PESTICIDE   | S-1               | S-2                   | S-3 | C-1 | C-2 |    |  |
| DIAZINON  | -                 | TR*                   | -   | TR  | -   | 24 |  |
| ETHYL PARATHION   | TR                | TR                    | -   | TR  | -   | TR |  |
| MALATHION   | -                 | -                     | -   | -   | -   | -  |  |
| PHORATE   | -                 | -                     | -   | -   | -   | TR |  |
| MONOCROTOPHOS   | TR                | TR                    | -   | TR  | -   | TR |  |
| DIMETHOATE  | -                 | -                     | -   | -   | -   | -  |  |
| DISULFOTON  | -                 | TR                    | -   | -   | -   | -  |  |
| DICHLOFENTHION  | -                 | -                     | -   | -   | -   | TR |  |
| FENTHION  | -                 | -                     | -   | -   | -   | -  |  |

\*TR=Trace(<1ug/m<sup>2</sup>)

APPENDIX C. (continued)

16 July 1979

|                 | WIND SPEED | WIND DIRECTION |
|-----------------|------------|----------------|
| UWSP (8 A.M.)   | 2-4 mph    | N              |
| FIELD (10 A.M.) | 15-20 mph  | N              |

Pesticide concentration for each site (ug pesticide/m<sup>2</sup> collection screen)

| PESTICIDE       | S-1  | S-2 | S-3 | C-1 | C-2 |
|-----------------|------|-----|-----|-----|-----|
| DIAZINON        | NS** | NS  | TR* | TR  | -   |
| ETHYL PARATHION | "    | "   | -   | -   | -   |
| MALATHION       | "    | "   | -   | -   | -   |
| PHORATE         | "    | "   | -   | -   | -   |
| MONOCROTOPHOS   | "    | "   | -   | -   | -   |
| DIMETHOATE      | "    | "   | -   | -   | -   |
| DISULFOTON      | "    | "   | -   | -   | -   |
| DICHLFENTHION   | "    | "   | -   | -   | -   |
| FENTHION        | "    | "   | -   | -   | -   |

\*TR=Trace(<1ug/m<sup>2</sup>)  
 \*\*NS=No sample

23 July 1979

|                 | WIND SPEED | WIND DIRECTION |
|-----------------|------------|----------------|
| UWSP (8 A.M.)   | 3-5 mph    | SSE            |
| FIELD (10 A.M.) | 5-10 mph   | W              |

Pesticide concentration for each site (ug pesticide/m<sup>2</sup> collection screen)

| PESTICIDE       | S-1 | S-2  | S-3 | C-1 | C-2 |
|-----------------|-----|------|-----|-----|-----|
| DIAZINON        | -   | NS** | -   | -   | NS  |
| ETHYL PARATHION | TR* | "    | 1   | TR  | "   |
| MALATHION       | -   | "    | -   | -   | "   |
| PHORATE         | -   | "    | -   | -   | "   |
| MONOCROTOPHOS   | -   | "    | -   | -   | "   |
| DIMETHOATE      | -   | "    | -   | -   | "   |
| DISULFOTON      | -   | "    | 1   | TR  | "   |
| DICHLFENTHION   | -   | "    | TR  | -   | "   |
| FENTHION        | -   | "    | -   | -   | "   |

\*TR=Trace(<1ug/m<sup>2</sup>)  
 \*\*NS=No sample

APPENDIX C. (continued)

31 July 1979

|                        | <u>WIND SPEED</u> | <u>WIND DIRECTION</u> |
|------------------------|-------------------|-----------------------|
| <u>UWSP</u> (8 A.M.)   | 6-11 mph          | N                     |
| <u>FIELD</u> (10 A.M.) | 10-15 mph         | NE                    |

Pesticide concentration for each site (ug pesticide/m<sup>2</sup> collection screen)

| PESTICIDE       | S-1  | S-2 | S-3 | C-1 | C-2 |
|-----------------|------|-----|-----|-----|-----|
| DIAZINON        | NS** | -   | NS  | NS  | TR* |
| ETHYL PARATHION | "    | -   | "   | "   | 10  |
| MALATHION       | "    | -   | "   | "   | -   |
| PHORATE         | "    | -   | "   | "   | -   |
| MONOCROTOPHOS   | "    | -   | "   | "   | -   |
| DIMETHOATE      | "    | -   | "   | "   | -   |
| DISULFOTON      | "    | -   | "   | "   | -   |
| DICHLOFENTHION  | "    | -   | "   | "   | -   |
| FENTHION        | "    | -   | "   | "   | -   |

\*TR=Trace(<1ug/m<sup>2</sup>)  
\*\*NS=No sample

2 August 1979

|                        | <u>WIND SPEED</u> | <u>WIND DIRECTION</u> |
|------------------------|-------------------|-----------------------|
| <u>UWSP</u> (8 A.M.)   | 2-4 mph           | SW                    |
| <u>FIELD</u> (10 A.M.) | 5-10 mph          | SW                    |

Pesticide concentration for each site (ug pesticide/m<sup>2</sup> collection screen)

| PESTICIDE       | S-1 | S-2 | S-3 | C-1 | C-2 |
|-----------------|-----|-----|-----|-----|-----|
| DIAZINON        | TR* | TR  | -   | -   | -   |
| ETHYL PARATHION | 20  | TR  | -   | -   | -   |
| MALATHION       | 4   | TR  | -   | -   | -   |
| PHORATE         | -   | -   | -   | -   | -   |
| MONOCROTOPHOS   | -   | -   | -   | -   | -   |
| DIMETHOATE      | -   | -   | -   | -   | -   |
| DISULFOTON      | TR  | TR  | -   | -   | -   |
| DICHLOFENTHION  | -   | -   | -   | -   | -   |
| FENTHION        | -   | -   | -   | -   | -   |

\*TR=Trace(<1ug/m<sup>2</sup>)

APPENDIX C. (continued)

6 August 1979

|                        | <u>WIND SPEED</u> | <u>WIND DIRECTION</u> |
|------------------------|-------------------|-----------------------|
| <u>UWSP</u> (8 A.M.)   | 8-11 mph          | E                     |
| <u>FIELD</u> (10 A.M.) | 10-15 mph         | SE                    |

Pesticide concentration for each site (ug pesticide/m<sup>2</sup> collection screen)

| PESTICIDE       | S-1 | S-2 | S-3 | C-1 | C-2 |
|-----------------|-----|-----|-----|-----|-----|
| DIAZINON        | TR* | -   | TR  | -   | TR  |
| ETHYL PARATHION | 4   | 4   | 2   | TR  | TR  |
| MALATHION       | -   | -   | -   | -   | -   |
| PHORATE         | -   | -   | -   | -   | TR  |
| MONOCROTOPHOS   | -   | -   | -   | TR  | TR  |
| DIMETHOATE      | -   | -   | -   | -   | -   |
| DISULFOTON      | -   | TR  | -   | -   | -   |
| DICHLOFENTHION  | -   | -   | -   | -   | TR  |
| FENTHION        | -   | -   | -   | -   | -   |

\*TR=Trace(<1ug/m<sup>2</sup>)

7 August 1979

|                        | <u>WIND SPEED</u> | <u>WIND DIRECTION</u> |
|------------------------|-------------------|-----------------------|
| <u>UWSP</u> (8 A.M.)   | 7-12 mph          | WSW                   |
| <u>FIELD</u> (10 A.M.) | 15-20 mph         | W                     |

Pesticide concentration for each site (ug pesticide/m<sup>2</sup> collection screen)

| PESTICIDE       | S-1 | S-2 | S-3 | C-1 | C-2 |
|-----------------|-----|-----|-----|-----|-----|
| DIAZINON        | 1   | -   | -   | -   | -   |
| ETHYL PARATHION | 4   | 2   | TR* | -   | TR  |
| MALATHION       | -   | -   | -   | -   | -   |
| PHORATE         | 1   | 1   | -   | -   | -   |
| MONOCROTOPHOS   | -   | -   | -   | -   | -   |
| DIMETHOATE      | -   | -   | -   | -   | -   |
| DISULFOTON      | TR  | 2   | TR  | -   | -   |
| DICHLOFENTHION  | 1   | -   | TR  | -   | TR  |
| FENTHION        | -   | -   | -   | -   | -   |

\*TR=Trace(<1ug/m<sup>2</sup>)

APPENDIX C. (continued)

8 August 1979

|                        | <u>WIND SPEED</u> | <u>WIND DIRECTION</u> |  |  |  |
|------------------------|-------------------|-----------------------|--|--|--|
| <u>UWSP</u> (8 A.M.)   | 3-5 mph           | N                     |  |  |  |
| <u>FIELD</u> (10 A.M.) | 5-10 mph          | NW                    |  |  |  |

| Pesticide concentration for each site (ug pesticide/m <sup>2</sup> collection screen) |      |       |      |      |     |
|---|------|-------|------|------|-----|
| PESTICIDE   | S-1  | S-2   | S-3  | C-1  | C-2 |
| DIAZINON  | -    | -     | -    | TR*  | TR  |
| ETHYL PARATHION   | 11.2 | 200.0 | 12.0 | 10.0 | 3.0 |
| MALATHION   | -    | -     | -    | -    | TR  |
| PHORATE   | -    | -     | -    | -    | -   |
| MONOCROTOPHOS   | -    | 1.0   | 2.0  | TR   | TR  |
| DIMETHOATE  | -    | -     | -    | -    | -   |
| DISULFOTON  | TR   | 4.0   | 1.0  | 1.0  | TR  |
| DICHLUFENTHION  | -    | -     | -    | TR   | -   |
| FENTHION  | -    | -     | -    | -    | -   |

\*TR=Trace(<1ug/m<sup>2</sup>)

16 August 1979

|                        | <u>WIND SPEED</u> | <u>WIND DIRECTION</u> |  |  |  |
|------------------------|-------------------|-----------------------|--|--|--|
| <u>UWSP</u> (8 A.M.)   | 2-4 mph           | S                     |  |  |  |
| <u>FIELD</u> (10 A.M.) | 5-10 mph          | W                     |  |  |  |

| Pesticide concentration for each site (ug pesticide/m <sup>2</sup> collection screen) |     |     |     |      |      |
|---|-----|-----|-----|------|------|
| PESTICIDE   | S-1 | S-2 | S-3 | C-1  | C-2  |
| DIAZINON  | -   | -   | -   | -    | -    |
| ETHYL PARATHION   | 4.0 | 2.0 | 4.0 | 12.5 | 40.0 |
| MALATHION   | -   | 2.0 | -   | -    | 4.0  |
| PHORATE   | -   | -   | 1.0 | TR*  | TR   |
| MONOCROTOPHOS   | -   | -   | -   | -    | -    |
| DIMETHOATE  | -   | -   | -   | -    | -    |
| DISULFOTON  | TR  | -   | 1.0 | -    | -    |
| DICHLUFENTHION  | -   | -   | -   | 1.0  | -    |
| FENTHION  | -   | -   | TR  | 1.0  | TR   |

\*TR=Trace(<1ug/m<sup>2</sup>)