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EFFECT OF PESTICIDES ON PHOTOSYNTHESIS

OF APPLE (*Malus sylvestris* Mill.)

DISSERTATION

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By

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* * * * *

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Parkash, D. 1971. Effect of Various Salt Concentrations on Different Grape Cultivars. Abstract only. Hort. Sci. 6(3):283.

FIELDS OF STUDY

Major Field: Pomology

Studies in Horticulture. Professor Fred O. Hartman.

Studies in Food Technology. Professors David C. Crean, Jean R. Geisman, and Wilbur A. Gould.

Studies in Pesticides and Pollution. Professors Leo E. Bendixen, William B. Collins, Paul E. Taiganidies and Acie C. Waldron.

Studies in Plant Physiology. Professors Michael L. Evans and Carroll A. Swanson.

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INTRODUCTION

Pesticides are an integral part of modern agriculture. Millions of tons of these chemicals are being used annually to protect our crops and homes from harmful insects, diseases, and unwanted weeds. Pesticides, in fact, have played a major role in increasing the yields of crops and in feeding the fast rising population of the world.

However, when used on commercial crops, these chemicals are evaluated only i) for their effectiveness against the target organism and ii) for visible symptoms of injury, if any, to the host plant. Their possible effect on the vital physiological processes, and subsequent influence on ultimate yields, is not emphasized. Potential damage to these processes may already have been inflicted even before visible signs of injury appear.

Photosynthesis, the ability of green plants to fix atmospheric CO_2 into carbohydrates by using energy from sunlight, is the principle process which directly influences plant productivity. Information about the pesticides which are effective in controlling plant pests without impairing the net photosynthate production can improve plant growth and increase crop yields. The present investigation was undertaken to study the influence of some commonly used pesticide chemicals on photosynthesis.

Primary objectives of the present investigations were:

1. To screen the various pesticide chemicals in use for their possible effect at the commercially recommended dosages on net CO_2 assimilation rate (NAR) of apple leaves.

2. To investigate the additive effect of repeated sprays of dicofol and dodine on the photosynthetic rate of Golden Delicious and Delicious cultivars.

3. To determine the influence of newly introduced combination sprays of benomyl and Superior Oil (70 sec. viscosity) on the NAR of apple trees.

4. To study the effect of four commonly used herbicides, diuron, monuron, simazine, and atrazine on the photosynthetic rate of apple leaves.

Separate experiments were conducted to meet the above objectives and are described in the text.

REVIEW OF LITERATURE

Pesticide chemicals are designed to either destroy harmful pests or to protect plants from pest induced injury. However, in the course of its action the pesticide can cause injury to the plant. Occasional reports (25, 36, 47) exist dealing with the overall influence of pesticides on the yield and growth of the host plant, but the information about their effect on photosynthesis or other physiological processes is limited. The influence of insecticides, fungicides, and miticides on fruit and vegetable physiology has been reviewed earlier by The National Academy of Sciences (5) and Harris (41).

In earlier studies, the net photosynthetic rate was measured by changes in leaf dry weight. It was not until the introduction of CO_2 absorption towers that the apparent photosynthesis of attached leaves could be measured (43). This method dominated until the late 1950's when the oxygen electrode and infra-red CO_2 analyser were introduced. The latter is a more sensitive and refined technique and measures the CO_2 absorbed by the plant (53).

Bordeaux mixture and other copper based inorganic fungicides have been in use for many years. Bain (10) as early as 1902 found an initial increase in the starch content of leaves treated with Bordeaux mixture and cuprous hydroxide, but subsequent sprays resulted first in injury and then death of the leaves. In more recent studies, 1% Bordeaux mixture and 0.5% copper oxychloride caused leaf scorch and premature leaf drop, lowered the fruit quality (97) and retarded shoot growth of apple (56, 97). Rather than damaging the leaves directly, Bordeaux mixture probably injures the tissue by adversely affecting some normal physiological processes (23, 91, 93, 94). Bordeaux mixture reduced photosynthesis and transpiration in apple (93, 94) and photosynthesis in sour cherry (73), but did not influence these processes in pecan foliage (69).

Of the sulfur-based inorganic fungicides, lime sulfur is most damaging to photosynthesis and yield of apple (2, 12, 13, 15, 44, 49, 91). It causes injury by inhibiting photosynthesis, increasing respiration, and by liberating heat on oxidation of sulfur to hydrogen sulfide (1, 13, 103, 104). High temperature, low humidity and continuous sunshine stimulated the inhibitory effect of sulfur compounds on photosynthesis (12, 44). However, from their work on sour cherry (<u>Prunus cerasus</u> L. var. Montmorency), Johnson, <u>et al</u>. (55) concluded that a spray program of lime sulfur resulted in better growth of the trees and subsequently better fruit yield. One percent colloidal sulfur also increased fruit weight, quality, and vitamin C content in apple (97).

Organic fungicides differ with regard to their influence on the host plant. McIntosh trees sprayed with zineb produced the highest yields followed in descending order by captan, dodine, dichlone, ferbam, glyodin and Bordeaux mixture (85). Zineb has increased photosynthesis in grapes (14), respiration, amount of chlorophyll, glucose and fructose levels in tomato leaves (95) and fruit weight, quality and vitamin C in apple (97). The favorable response with zineb was thought to be associated with an increase in the zinc content of the foliage (85). Captan, however, reduced photosynthesis in grapes (14) and dodine sprays injured 3% of McIntosh apples (82), but no significant differences were detected in the fruiting performance of Stayman Winesap trees treated with these chemicals (40). Ferbam, likewise, did not influence the photosynthetic activity of peach and apple foliage (79), but intensified the green color of limes (33). Of other fungicides, dichlone decreased the yield of McIntosh apples (84). Niacide-M and glyodin also suppressed the rate of CO_2 assimilation in apple leaves initially, but the rate increased later (91). Benomyl, a systemic fungicide, inhibited the Hill reaction in isolated chloroplasts and reduced the photosynthetic activity in the leaf tissues; but it did not affect respiration (59). Another systemic fungicide, Vitavax, is also known to inhibit photosynthesis (18).

The incorporation of petroleum oils in benomyl sprays had increased the efficiency and decreased the cost of controlling apple scab and powdery mildew. Though the dormant oils are highly phytotoxic, more refined oils (39, 74) having a narrow viscosity range and high unsulfonated residues (105) have been introduced which are comparatively less toxic. Naphthalinic type oil is known to be less phytotoxic than the paraffinic type (17) and 75-85 sec. to be the least phytotoxic viscosity range (16).

Schroeder (90) studied the effect of some summer oil sprays on the CO_2 absorption of apple leaves. First and second sprays of 1% oil (90 sec. viscosity and 85% sulfonation) at 14 day intervals reduced the CO_2 absorption rate by 17% and 50%, respectively. Two percent and 5% sprays of the same oil gave average reductions of 38.4% and 45%, respectively, in the first few days. Raising the viscosity and amount of oil in the spray decreased the CO_2 absorption rate. Spray oils are also known to inhibit photosynthesis in the foliage of bananas (80), oranges and lemons (108).

Oils influence other plant processes in addition to photosynthesis. They increased respiration in citrus leaves but decreased it in apples (76). Oils are also known to decrease the transpiration rate (21, 26).

A disproportionately small amount of information appears about the phytotoxic effects of inorganic insecticides because of their limited use. Lead arsenate is the most widely used inorganic compound for controlling insects, however, it has, at times, injured apple foliage (83). Damage does not always occur with use of lead arsenate. For example in cherries, lead arsenate with fixed copper gave higher soluble solids than ferbam plus parathion (100).

DDT has been studied more thoroughly than any other chlorinated-hydrocarbon insecticide. It depressed shoot growth (56), lowered fruit set, and decreased the yield (29) of apple. DDT reduced the photosynthetic rate of cucumber leaves, but had no significant effect on beans (42) and peaches (79). Moon and Harley (70) found no effect of DDT on the fruit enlargement rate of apple, thereby concluding that photosynthetic efficiency was not impaired. DDT sprays have been found to increase the respiration rate of younger peach leaves but reduced it in mature leaves (78).

As evidence of their inhibitory effect on photosynthesis, sprays of chlorinated hydrocarbons like sulphenone, aramite, dieldrin and toxaphene have also been shown to reduce dry weights of leaves in apple and pear (112). However, Kelthane did not reduce leaf growth or dry weight significantly (112). Lindane probably reduces photosynthesis by inhibiting photoxidation of water more than the steps of ATP synthesis coupled with it during non-cyclic photophosphorylation (96).

Helson (46) studied the effect of the insecticides, ryania, ryanodine and ryanodine-free-ryania (powdered wood) on the apparent photosythesis of attached apple leaves. He concluded that ryanodine itself had no marked chemical effect on apple leaves. But the deposits of either ryania or powdered wood inhibit photosynthesis either by shading or by interfering with the gas exchange of the leaves.

Reports differ with regard to the influence of organophosphates on plants. While parathion (0.12%) injured apple foliage and fruit in one study (35), it (0.1%) increased the growth of apple (56) and sour cherry (55) trees in other cases. Peach and apple foliage treated with parathion resulted in less photosynthate than Fermate- and thinner palisade tissue than chlordane-treated leaves (79). However, parathion did not affect the chlorophyll content of the fully expanded apple leaves (110). Of other organophosphates, malathion did not affect the fruit growth rate of Bartlett pear (112), but disulfoton increased yield of melons, tomatoes and green beans (27). Diazinone, ethion and azinophosmethyl reduced the rate of photosynthesis in apple foliage by 50%, 34% and 12%, respectively, over the control (45). It is believed that Methylnitrophos reduced photosynthesis by inhibiting ATP synthesis more than the steps of electron transport coupled with water photoxidation (96). On the other hand, Methylmercaptophos inhibited photoxidation of water more than the steps of ATP synthesis coupled with it during non-cyclic phosphorylation.

There is little information available on the effects of carbamate and dinitro groups of insecticides and miticides on photosynthesis or related plant processes. However, stimulation of apple shoot growth by 0.2% carbaryl (56) and an increase in the yield of melons, tomatoes and green beans by dinoseb (27) has been reported.

Of the commonly used herbicides, s-triazines (simazine and atrazine), anilides (monuron and diuron) and alkyl amides (Karsil) primarily act by inhibiting the Hill reaction of photosynthesis (31, 37). More specifically, these chemicals block non-cyclic photophosphorylation (4,8, 24, 107, 111). However, they may inhibit processes other than photosynthesis as well (22, 72). In addition to killing weeds through their effect on the above processes, the herbicides may also affect the photosynthetic process of the cultivated crop.

When applied to the soil, herbicides like atrazine, simazine, monuron and diuron are adsorbed on the cation exchange sites of the soil particles. The rate of adsorption is directly proportional to the cation exchange capacity (clay and/or organic matter content) of the soil (48, 98). However, atrazine and monuron are more mobile than diuron and simazine respectively (99, 48). After several applications, most of the simazine remained in the top 2-inches of soil while atrazine had spread to the 12-inch depth (98). Therefore, atrazine and monuron, through their more mobile nature in the root medium can influence photosynthesis and other plant processes more than simazine and diuron. Leaching was not believed to be an important factor in the disappearance of these chemicals from soils (48, 98).

Of the s-triazines, simazine and atrazine are commercially used in fruit plantations. Simazine has given satisfactory pre-emergence weed control in apples (62, 92), grapes (6), highbush blueberries (109) and pecans (75). Atrazine gave good weed control in apple orchards (92) and vineyards (3).

Application of simazine stimulated growth and cropping in apples (11, 58), citrus (38) and 'Pineapple' orange trees (86). However, Lord et al. (67) could not find any direct effect of this herbicide on the nutrient level, tree growth, or fruit color of 'McIntosh' apples. Annual applications of simazine in excess of 6 pounds per acre (recommended rate: 2-4 lbs./acre for selective weed control) caused injury to young apple trees (87). The leaves developed a pale green color and a more netted appearance than leaves of control trees, resulting in veinal chlorosis, necrosis and complete defoliation in acute cases.

The lowest concentration at which simazine adversely affected fresh weight increase of greenhouse grown 'McIntosh' apple trees was 1.6 ppm in sandy loam and fine sandy loam soils (81).

Dvorak (30) reported that in loamy soil with a low humus content (0.8% organic carbon), simazine at a concentration of 2.5 ppm reduced growth of shoots, roots and trunk diameter in 1-year-old scions of 5 apple cultivars on M9 rootstock. Lord et al. (68) also found simazine levels in excess of 3 ppm in soil to be detrimental for the growth of M7, M9 and MM 106 rootstocks.

It is known that atrazine (8, 9) and simazine (57, 77) lower chlorophyll and carotenoid contents because of direct interaction between the herbicide and pigment. Application of these chemicals reduced chlorophyll content in blue plum (60). Although, in citrus (38) and raspberries (34), atrazine decreased the chlorophyll content, simazine did not influence the level of this pigment. Krusteva (60) proposed that atrazine has a stronger phytotoxic effect than simazine.

Imbamba and Moss (50) fed atrazine through the base of excised leaves of atrazine-tolerant and -susceptible species. Atrazine suppressed CO_2 uptake in all species. This was shown to be due primarily to inhibition of photosynthetic reactions and only to a slight extent to the stomatal closure caused by atrazine. Simazine and atrazine also increased the amount of oxygen-uptake in blue plum (60). The effect of simazine is temperature dependent and is more pronounced on photosynthesis than respiration (102).

Of the ureas, diuron is used to control annual weeds and grasses in tree fruits, grapes, and blueberries (6, 75, 86, 109). Monuron gave a satisfactory weed control in vineyards (3), but it generally is used for complete weed kill.

Karnatz (58) reported stimulation of growth and cropping of newly planted apple trees by the use of diuron. However, Saidak and Rutherford (88) observed a faint veinal necrosis in a few leaves on trees which had received a total of 8 lbs. per acre of diuron (recommended rate: 4 lbs./acre). At higher rates (16 and 24 lbs./acre), the symptoms were more severe and there was a visible reduction in tree growth. In another trial, diuron was found consistently more toxic than simazine to young trees, but older trees showed the reverse relation (62).

Like other phenylureas, diuron (113) and monuron (71) strongly inhibit the Hill reaction. One molecule of monuron per 200 chlorophyll molecules causes 100% inhibition of the Hill reaction (13, 66). Diuron blocked photosynthesis in soybean (*Glycine max*, Merr.) (65) and doddar (*Cuscuta indecora*, chois.) (61). In bean (*Phaseolus vulgaris*, L.) plants, diuron inhibited photosynthesis to a lesser degree than simazine, simeton, or isocil (106).

Diuron and monuron are not known to affect respiration (89). Diuron had no influence on oxidative production of ATP, but it did inhibit photochemical ATP production in chlorella (54). The effect of diuron on enzyme activity, respiration intensity and growth was found to be more intense in sensitive species (*Amaranthus retroflexus*) than on comparatively resistant corn seedlings (51, 52). Lay and Ilnicki (65) concluded that diuron closes stomata in soybean (Glycine max, Merr.) due to an increase in intercellular CO_2 concentration as a consequence of reduced CO_2 assimilation. These authors proposed that diuron indirectly affects physiological processes other than photosynthesis in plants.

There are reports about the interaction of herbicides and insecticides in leaf tissue (19, 20). Synergistic effects of such groups of chemicals within the host plant and their influence on different plant processes is a very important, and so far, neglected field.

Leaf age also exercises a great influence on the photosynthetic rate. The concept of plastochron, period between initiation of successive leaves, has been used to judge the leaf age. Since the leaves in a constant environment are initiated at relatively uniform rates, Erickson and Michelini (32) proposed a plastochron index to relate morphogenetic measurements with anatomical, physiological, and biochemical events or processes. Larsen and Isebrands (64) concluded from their studies on cottonwood that the plastochron index could serve two useful purposes: (1) to adjust plants of different developmental stages to a standardized morphological time scale, and (2) to predict developmental processes or events from simple nondestructive measurements. Leaf plastochron index was also correlated with the rates of photosynthesis and respiration (28).

MATERIALS AND METHODS

The present investigation involved four separate studies:

A. Screening trials.

B. Multiple sprays: The effect of the repeated sprays of dicofol and dodine on NAR in Golden Delicious and Delicious.

C. Benomyl and oil: The influence of different levels of benomyl and oil on NAR of Golden Delicious foliage and their influence on the leaves of different ages.

D. The effect of diuron, monuron, simazine and atrazine on NAR.

The trees were grown in 8-inch plastic containers having a loam:peat:perlite (3:1:1) mixture. At the time of planting they were headed back to 4-6 buds from the bud union. Later, a single shoot was allowed to develop.

The trees were watered daily between 4 and 5 p.m. Approximately 0.5 liter of fertilizer solution (200 ppm each of N, P_2O_5, and K_2O) was applied once a week to each tree. Aphids, (*Aphis pomi* Degeer and *Anuraphis roseus* Baker), white flies (*Trialeurodes vaporariorum* Westwood) and mites (*Panonychus ulmi* Koch and *Bryobia arborea* Morgan and Anderson) when present were sprayed with parathion (15% W.P.; 0.9 $g.l^{-1}$) and Plictran (50% W.P.; 0.3 $g.l^{-1}$). These chemicals were selected because in preliminary screening trials they did not affect NAR of apple leaves.

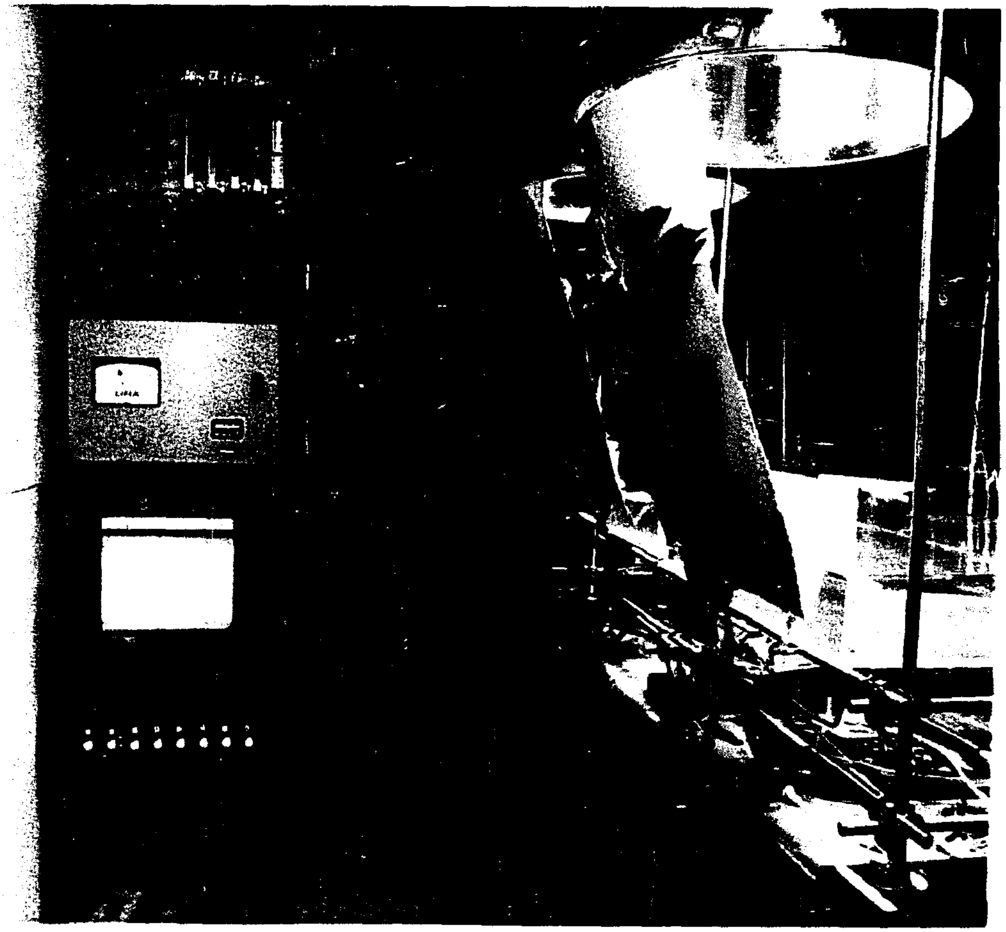


FIG. 1. APPARATUS USED FOR DETERMINING THE NET CO_2 ASSIMILATION RATE OF APPLE LEAVES
A: MSA INFRA-RED CO_2 GAS ANALYSER B: BRISTOL RECORDER C: LEAF CHAMBER WITH THE LEAF ENCLOSED IN IT D: RUNNING WATER BATH TO CONTROL TEMPERATURE IN THE LEAF CHAMBER E: ASBESTOS SHEET TO PREVENT EXCESSIVE EXPOSURE OF LIGHT AND HEAT TO THE TREE FOLIAGE F: SYLVANIA'S PHOSPHORUS COATED METAL-ARC LAMPS G: AIR FLOW METERS.

The trees were taken from the greenhouse (or cold frame from July 3 to October 1) to the growth chamber one day before taking measurements. Light intensity in the growth chamber was 2000 foot candles (0.0782 cal. $cm.^{-2}$ $min.^{-1}$ PAR). Day length was kept to 15 hours (21°C) and night to 9 hours (16°C).

The net CO_2 assimilation rate (NAR) was measured by an infra-red gas analyser (MSA Model 200 Lira) with a flowing reference cell (Fig. 1). The rate of CO_2 absorbed by the leaf was recorded on a Bristol Recorder. The intact leaves were placed in a plexiglass chamber (inside area $\approx$ 200 $cm.^3$) sealed by a closed pore rubber gasket. The leaf in the chamber was supplied with outside air (350-450 ppm CO_2) at the rate of 3 litres $min.^{-1}$ by a compressor with an air intake located 90 cms. above the roof of the building. Sylvania's phosphorus-coated metal-arc lamps provided 4000 foot candles (0.1720 cal. $cm.^{-2}$ $min.^{-1}$ PAR) of light intensity (spectral distribution in appendix 1) which is on the light saturation plateau for apple leaves (Appendix 2). A running water bath located between the light source and leaf chamber aided in maintaining the temperature at the leaf surface 25°C.

Since it required $\approx$ 30 minutes for a leaf to achieve a steady rate of CO_2 absorption, the two leaf chambers were used alternately (flow diagram in appendix 3). While the NAR in one chamber was being measured, the leaf in the other chamber was allowed to equilibrate.

A modification of the leaf plastochron index (32) was adopted in all but screening trials to check the physiological age of the leaves used for NAR measurements. For calculating leaf plastochron age, the youngest leaf with a blade length $\geqslant 3$ centimeters was employed as the index leaf.

A. Screening Trials

Several apple cultivars were used in the screening trial. In order to ensure the supply of enough plant material over the period, the trees were removed from the cold storage in groups and were grown in the greenhouse between the months of January and April, 1973. Natural light conditions and a minimum temperature of 21°C during day and 16°C during night were maintained in the greenhouse.

(a) Commercial formulations of one nematicide, 12 fungicides, 14 insecticides and 10 acaricides were tested for their effect on NAR in apple leaves (Appendix 4). The materials were applied at the commercially recommended rates. One leaf (8th, 12th or 15th from the base) was employed for each chemical. The leaf was sprayed with a hand atomizer until it was dripping wet. Uniform concentration of the pesticide in the spray solution was ensured by agitating it during application. The readings were taken before treatment and 1-, 6-, 24-, and 28-hrs. after spraying. The influence of the chemical was compared with the initial reading taken for the same leaf.

(b) In order to ascertain the time required for maximum noticeable response, the chemicals which had the greatest effect in (a) were tested at more frequent intervals. The observations were taken every half hour after treatment for 0-6 hours and 23-29 hours.

B. Multiple Spray Experiment

The acaricide dicofol (E.C. 42%) and the fungicide dodine (65% W.P.) had greatly depressed NAR in the screening trials. Therefore, an experiment was initiated for evaluating the effect of four sprays of these chemicals as applied to Delicious/M7 and Golden Delicious/M7 cultivars at 10-day intervals. Until planting on July 3, the trees for this study were kept in cold storage at 2°C. At the time of treatment, each tree had an average of 36 leaves. The trees were grouped according to their NAR values. Fifteenth leaf from the index leaf was employed for determining the net CO_2 assimilation rate. The same leaf was measured throughout the trial.

The treatments were applied in a randomized block arrangement. Dicofol (E.C. 42%, 1.25 $ml.l^{-1}$) and dodine (65% W.P.; 0.6 $g.l^{-1}$) were mixed in distilled water. The entire tree was sprayed with the hand atomizer until it was dripping wet. The control trees were sprayed with distilled water. Each treatment was replicated three times.

The first spray was applied on August 31, 1973 and subsequent sprays at 10-day intervals. Observations were taken 1-, 24-, 48-, 72-hours and 10 days after each spray. An additional reading was taken 20 days after the 4th or last spray.

C. Benomyl and Oil

Golden Delicious/M7 trees were planted on July 3. Until then they were kept in the cold storage (2^oC). At the time of treatment, the trees were grouped according to their shoot extension growth. Three levels of benomyl ($B_0 = 0$, $B_1 = 0.15\ g.l^{-1}$, $B_2 = 0.3\ g.l^{-1}$) and 70 sec. viscosity Superior Oil ($O_0 = 0$, $O_1 = 1.25\ ml.l^{-1}$, $O_2 = 2.5\ ml.l^{-1}$) were applied in a factorial arrangement with each treatment having three replications. Since the size of the experiment was large, each replication was treated individually at 12 day intervals (beginning from September 15). To ensure uniformity in its physiological age in all replications, the 22nd leaf from the index leaf was used for NAR determination.

Spraying was done in the same manner as in the previous experiment. Check trees were sprayed with distilled water. Observations were taken before and after 1-, 5-, 10-, 20-, 30-, 50-, 70-hrs, and 10- and 20-days of the treatment.

For the trial designed to investigate the effect of benomyl and oil sprays at different leaf positions, the trees (Delicious/M7) were also planted on July 3 but were kept in the cold storage (2^oC) until August 15 when they were moved to the cold frame. Oil (70 sec.; $2.5\ ml.l^{-1}$) and benomyl (50% W.P.; $0.3\ g.l^{-1}$) plus oil (70 sec.; $2.5\ ml.l^{-1}$) were applied to the whole tree. The check tree was sprayed with distilled water. Each treatment was replicated three times. Effects of these sprays on the NAR of 15th and 25th leaf were measured at 50-, 70-, and 90-hrs. after treatment.

D. Herbicide Experiment

Golden Delicious/M7 trees were grown in 8-inch plastic pots containing 6 kgs. of Crotan silt loam soil (O.M.: 3.2%, pH : 6.0). A 5-cm. layer of gravel was applied on the surface of the pot as mulch to ensure uniform moisture conditions. The trees were grouped into different replications according to their uniformity in NAR. Eight percent formulations of diuron, monuron, simazine and atrazine were applied at the rate of 4.5 kg. hectare^{-1}. In 50 grams of soil, 0.0146 gram of the herbicide was mixed and was spread evenly over the surface of the pot on September 5. The treatments were arranged in a randomized block design with three replications. Each tree was given 0.5 litres of water per day.

NAR and plastochron index were observed after 0.5-, 1.5-, 2.5-, 10-, 20-, 30-, 40-, and 50-days after the treatment. The fifteenth leaf from the index leaf was chosen for taking NAR measurements. The same leaf, on an average, had become 30th from the index leaf at the conclusion of the trial.

RESULTS

A. Screening Trials

Thirty-seven pesticide chemicals were screened for their possible effect on the photosynthesis of apple leaves. The formulations and concentrations of the different chemicals tested, and their effect on the NAR are given in appendix 4. Five pesticides increased and 20 decreased the net CO_2 assimilation rate by more than 5% (Fig. 2). The remainder of the chemicals influenced the NAR of leaves by less than 5% of their initial rates.

Superior 70 sec. oil, at the rate of 20 $ml.l^{-1}$ caused the greatest reduction in NAR (66%) (Fig. 2), followed by 0.6 $g.l^{-1}$ benomyl + 2.5 $ml.l^{-1}$ Superior Oil (41%), diazinone (26%) and dicofol (23%). Also causing a 10% or greater decrease in NAR were Omite (15%), dodine (14%), leptophos E.C. (14%), thiram (13%) and streptomycin (10%). However, ferbam, parathion, Plictran, tetradifon, and endosulfan treatments increased NAR by more than 5% of the initial rate.

The pesticides which either inhibited or stimulated NAR by more than 10% were studied in greater detail to determine how soon after treatment the reduction occurs. The chemicals chosen to control white flies, aphids, and mites during the period of trials, were also included in the

FIG. 2. EFFECT OF PESTICIDE SPRAYS ON NAR IN APPLE LEAVES. (EXPRESSED AS PERCENT REDUCTION IN NAR FROM THE INITIAL VALUE)

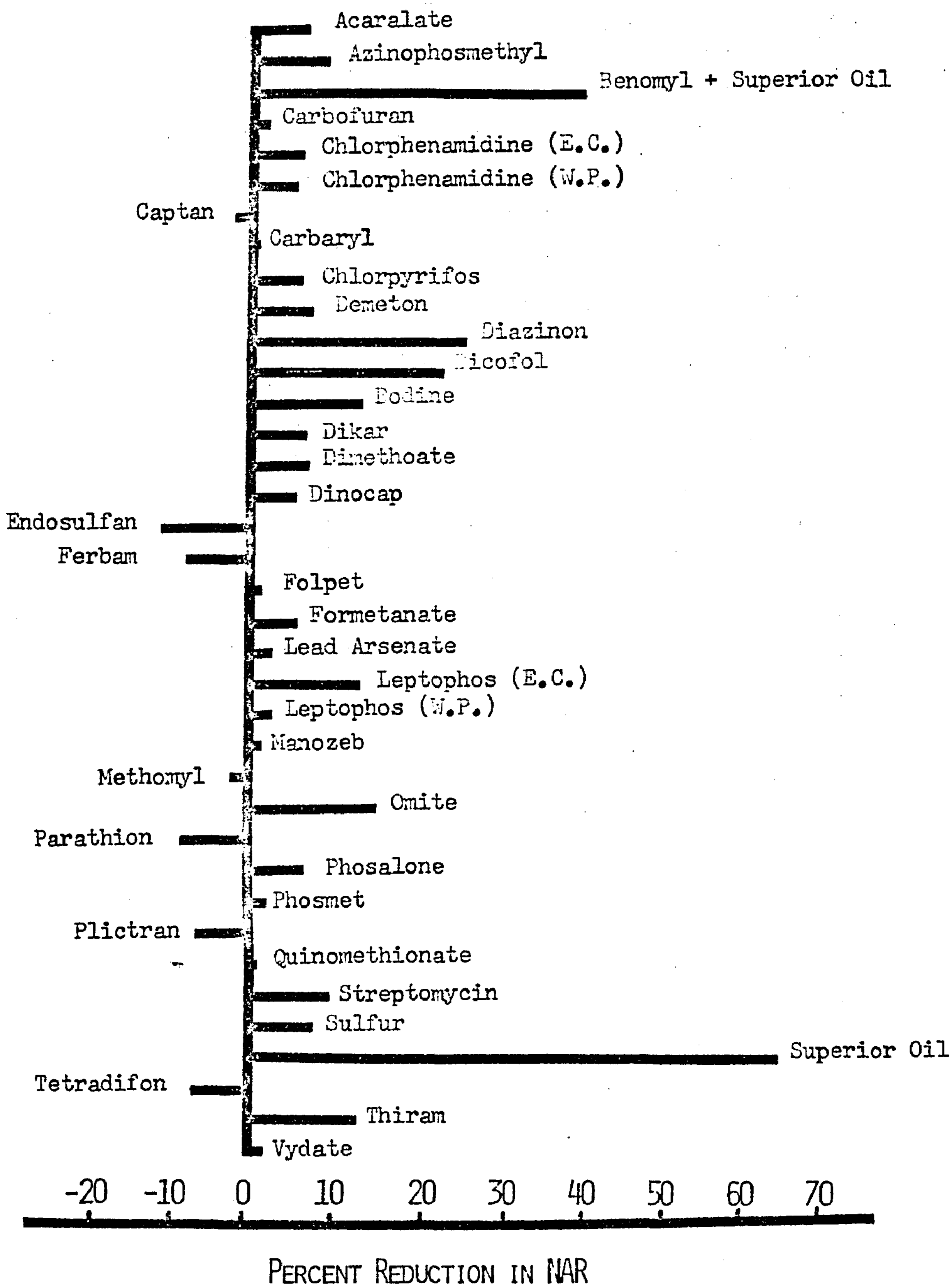


Table 1. Effect of Pesticides on the Net CO_2 Assimilation Rate[a] of Apple Leaves at Different Time Intervals after Spraying

Pesticide	Formulation	Concentration	Hours After Spray			
			0.5	2	5	25
Carbaryl	50 W.P.[b]	1.8 $g.l^{-1}$	98.0	96.0	91.6	105.6
Dicofol	42 E.C.[c]	1.3 $ml.l^{-1}$	75.9	82.3	81.4	70.5
Dimethoate	25 W.P.	1.3 $g.l^{-1}$	85.2	87.8	91.7	112.3
Dodine	65 W.P.	0.6 $g.l^{-1}$	52.8	65.9	75.6	88.3
Omite	30 W.P.	1.5 $g.l^{-1}$	47.4	53.9	61.0	62.4
Parathion	15 W.P.	1.8 $g.l^{-1}$	87.2	89.9	88.8	101.4
Plictran	50 W.P.	0.3 $g.l^{-1}$	93.5	92.2	90.0	82.5
Streptomycin	17 W.P.	100 ppm	97.4	98.4	96.9	104.9

[a]Expressed as percent of the reading taken before spray for the same leaf.

[b]W.P.: Wettable Powder

[c]E.C.: Emulsifiable Concentrate.

study. In most instances (Table 1), the pesticides reduced NAR within one half hour after treatment. The greatest reduction was caused by Omite (53%) followed, respectively, by dodine (47%), dicofol (14%), dimethoate (15%), parathion (13%), and Plictran (6%). The rates for dodine-, Omite-, and parathion-treated leaves showed recovery in the next 25 hours and increased for dimethoate-treated leaves in the same period. Leaves treated with dicofol and Plictran showed a further decrease in NAR values. The reduction in NAR caused by dicofol and Plictran 25 hours after the treatment was 27% and 17%, respectively. Carbaryl and streptomycin sprays had little influence on the NAR of apple leaves.

B. Multiple Spray Experiment

There was a sharp rise in the NAR of dodine-treated leaves 72 hours following the first spray (Fig. 3). In all cases, the rates dropped significantly 10 days after the spray. One hour after the second spray, the NAR declined further, recovering sharply in the next 72 hours, and dropping again 10 days later. This pattern was repeated in the periods following the third and fourth sprays. But the rise and drop in NAR was less pronounced in each successive spray. In all cases dicofol decreased NAR the most.

The average NAR values following 2nd, 3rd, and 4th sprays, though not differing significantly among each other, were significantly lower than the NAR values for the period following the first spray (Appendix 5).

Fig. 3. Effect of Four Sprays of Dicofol (1.3 ml.L^{-1}) and Dodine (0.6 g.L^{-1}) at 10 Day Intervals on NAR of Golden Delicious Leaves.

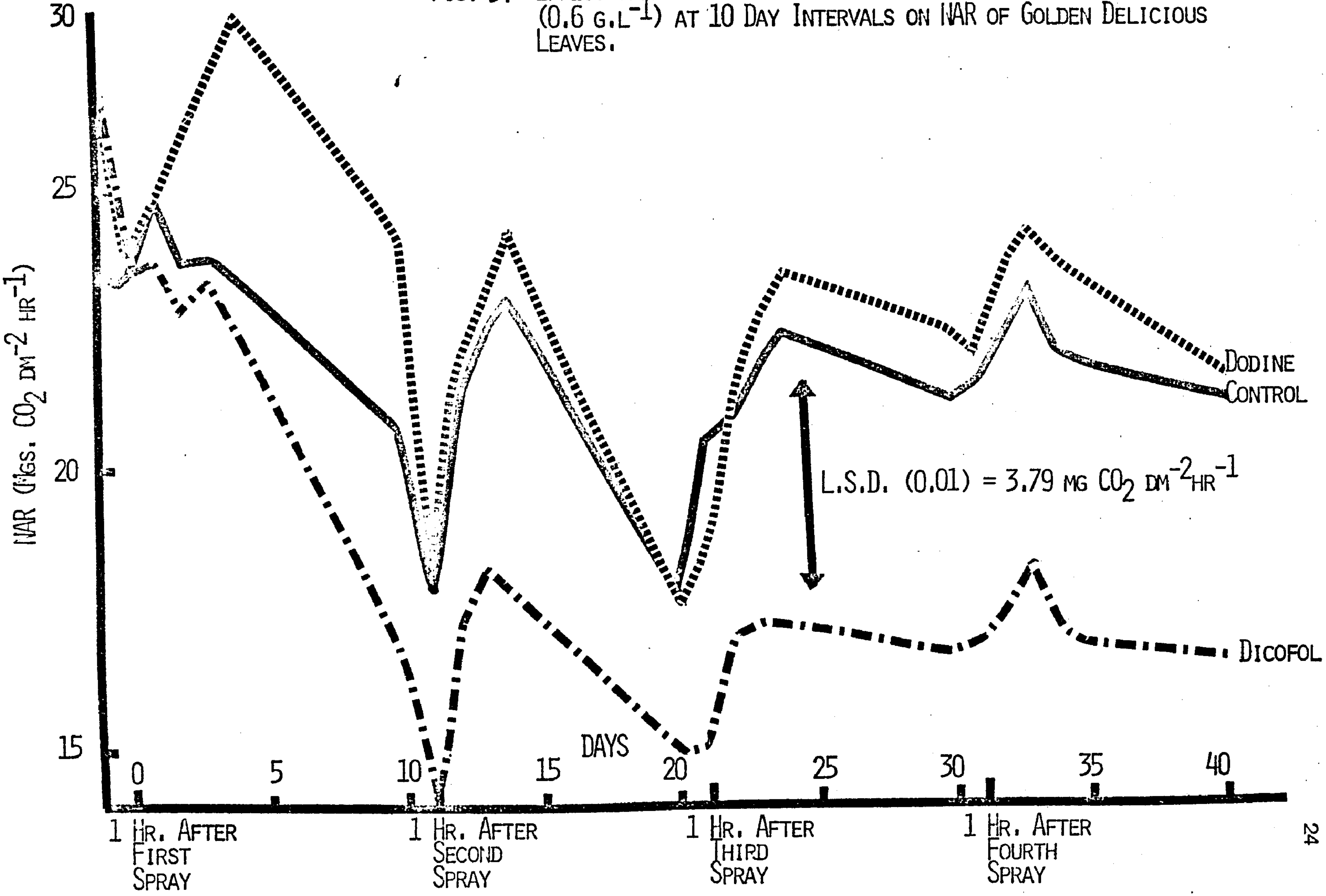
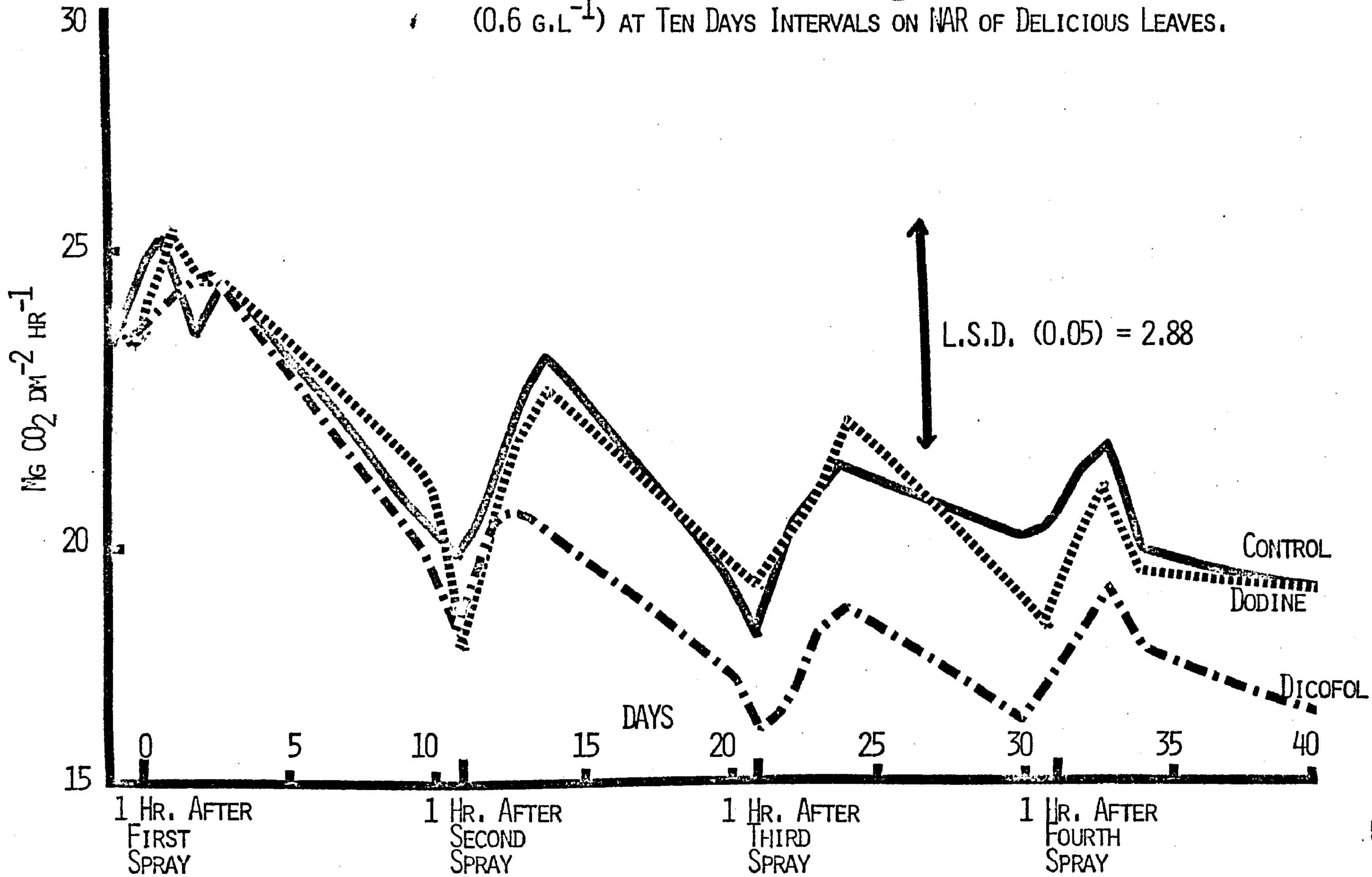


FIG. 4. EFFECT OF FOUR SPRAYS OF DICOFOL (1.3 ML.L^{-1}) AND DODINE (0.6 G.L^{-1}) AT TEN DAYS INTERVALS ON NAR OF DELICIOUS LEAVES.



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This indicates that the maximum decline in NAR occurred after the second spray and the subsequent sprays did not decrease NAR further. Also, the average NAR at 1 hour and 10 days after the treatment was significantly lower than the NAR values at 24-, 48-, and 72-hours (Appendix 6).

While the net CO_2 assimilation rates obtained for Delicious leaves (Fig. 4) were similar to Golden Delicious leaves (Fig. 3), the fluctuations in NAR values and the effects of the treatments were less pronounced in the Delicious leaves. Dicofol reduced NAR significantly in both cultivars, but the decrease was greater in Golden Delicious (Appendix 5). Dodine did not affect NAR of Delicious leaves significantly. The interactions between (cultivars x treatments x periods) and (cultivars x treatments x periods x time after spray) were not significant.

C. Benomyl and Oil Experiment

The effect of 3 levels each of benomyl (50% W.P.; $B_0 = 0$; $B_1 = 0.15$ $g.l^{-1}$ and $B_2 = 0.3$ $g.l^{-1}$) and Superior Oil (70 sec.; $O_0 = 0$; $O_1 = 1.25$ $ml.l^{-1}$; $O_2 = 2.5$ $ml.l^{-1}$) on the net CO_2 assimilation rate of Golden Delicious leaves is recorded in table 2. Both concentrations of oil produced a significant reduction in NAR. However, the decrease caused by the higher concentration (2.5 $ml.l^{-1}$) was not significantly different from 1.25 $ml.l^{-1}$-treatment. On the other hand, the lower level of benomyl (0.15 $g.l^{-1}$) significantly increased the net CO_2 assimilation rate. The decrease in NAR caused by the higher benomyl rate (0.3 $g.l^{-1}$) was not significant.

Table 2. Effect of Different Levels of Benomyl[a] and Oil[b] on the Net CO_2 Assimilation Rate of Golden Delicious Leaves (Mgs. CO_2 dm^{-2} hr^{-1})[c]

Oil / Benomyl	0	1.25 Ml 1^{-1}	2.5 ml 1^{-1}	Benomyl Mean
0	20.81^{B}	21.11^{BC}	18.78^{A}	$20.23^{A'}$
0.15 $g.1^{-1}$	21.92^{C}	20.44^{B}	20.23^{B}	$20.86^{B'}$
0.3 $g.1^{-1}$	20.58^{B}	19.29^{A}	20.58^{B}	$20.12^{A'}$
Oil Mean	$21.10^{B''}$	$20.25^{A''}$	$19.86^{B''}$	20.40

[a]Benomyl in 50% W.P. formulation.

[b]Oil used was 70-second viscosity superior oil.

[c]Figures with different letters differ significantly from each other. Figures having the same letters do not differ significantly from each other according to Duncan's multiple range test.

Table 3. Effect of Benomyl[a] and Oil[b] Sprays on Net CO_2 Assimilation Rate of Delicious Leaves of Two Different Ages (Mgs. CO_2 dm^{-2} hr^{-1})[c]

Treatment	Hours After Treatment								
	50	70	90	Mean	50	70	90	Mean	Mean
	15th Leaf				25th Leaf				
Control	20.49	20.27	20.70	20.49	20.81	20.06	20.45	20.44	20.46
Oil	20.48	20.34	20.25	20.35	21.73	21.80	22.21	21.91	21.13
Benomyl and Oil	19.88	19.49	19.01	19.46	22.48	22.00	22.00	22.16	20.81
Mean	20.28	20.03	19.99	20.10[A]	21.67	21.29	21.56	21.50[B]	20.80

[a]Benomyl: 50% W.P. @ 0.3 $g.l^{-1}$.

[b]Oil: 70-second viscosity superior oil @ 2.5 $ml.l^{-1}$.

[c]Figures bearing different letters are significantly different from each other according to Duncan's multiple range test.

The interaction between benomyl and oil levels was highly significant. The sprays of B_0O_2 (0 benomyl + 2.5 $ml.l^{-1}$ oil) B_2O_1 (0.6 $g.l^{-1}$ benomyl + 1.25 $ml.l^{-1}$ oil) reduced NAR significantly as compared to other treatments. The benomyl and oil treatments did not influence the total leaf count (Appendix 7).

In another study designed to determine the influence of benomyl and oil on the NAR of leaves differing in age, the treatment (2.5 $ml.l^{-1}$ oil; 0.6 $g.l^{-1}$ benomyl + 2.5 $ml.l^{-1}$ oil) effects were not significant (Table 3). However, the net CO_2 assimilation rate of the 25th leaf (from the index leaf) was significantly higher than the NAR of 15th leaf.

D. Herbicide Experiment

The effect of the surface soil applied herbicides diuron, monuron, simazine and atrazine on the NAR of Golden Delicious leaves is depicted in figures 5 and 6. All treatments resulted in a gradual decline with time in the rate of net CO_2 assimilation. Although monuron reduced the NAR from the beginning, the maximum treatment influence was evident 10 days after the application of the herbicides. By then, monuron had decreased NAR by 72%, atrazine by 64% and simazine by 22%. These effects persisted throughout the course of the trial. The NAR in monuron- and simazine-treated trees recovered slightly 30 days after the treatment. Monuron caused the greatest reduction in the net CO_2 assimilation rate (45%) followed by atrazine (37%) and simazine (12%). Diuron showed no

FIG. 5. EFFECT OF DIURON AND MONURON (80% W.P.; 4.5 KG. HECTARE^{-1}) AS APPLIED TO THE SURFACE SOIL ON THE NET CO_2 ASSIMILATION RATE OF GOLDEN DELICIOUS LEAVES.

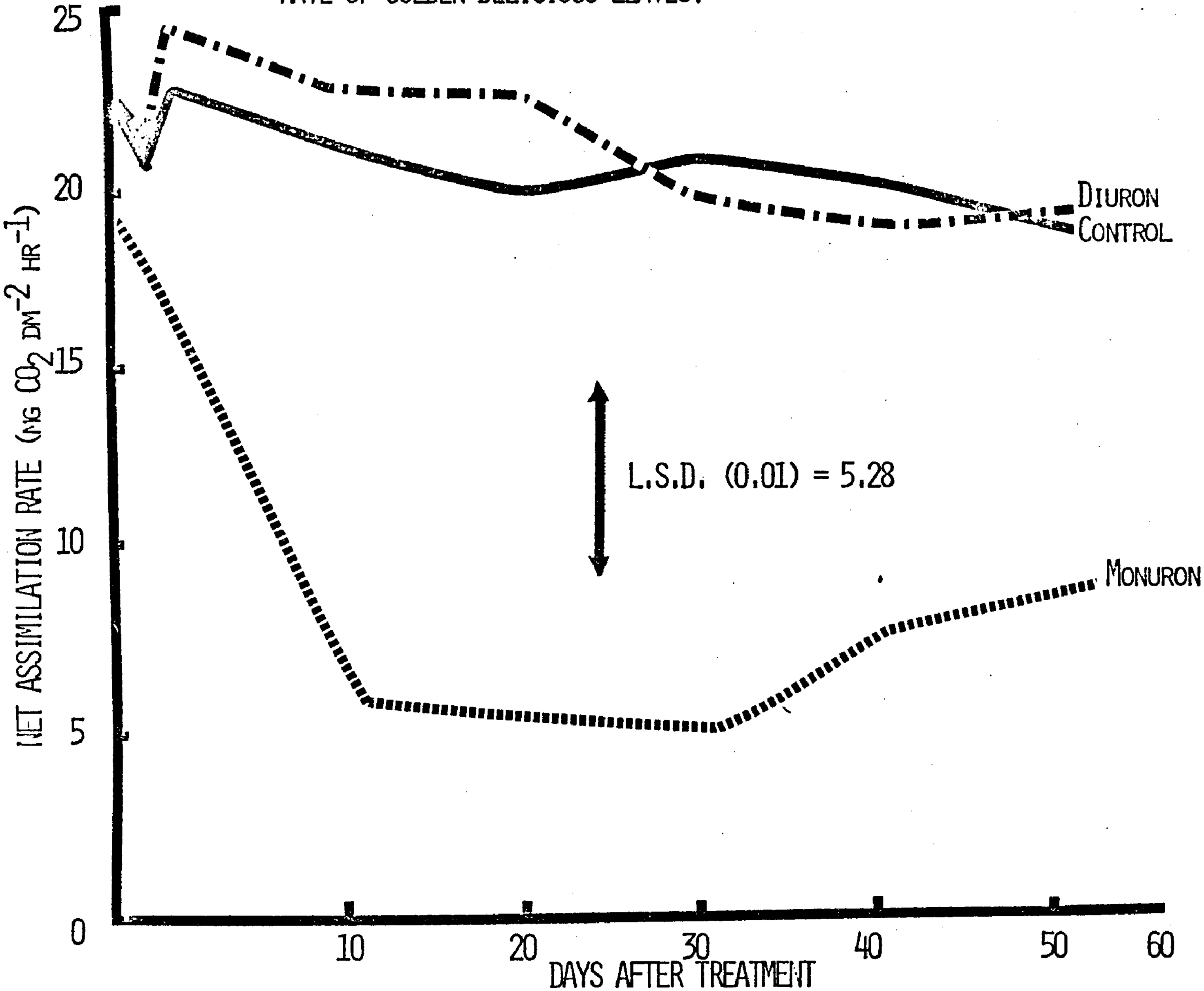


FIG. 6. EFFECT OF SIMAZINE AND ATRAZINE (80% W.P., 4.5 KG. HECTARE^{-1}) AS APPLIED TO THE SURFACE SOIL ON THE NET CO_2 ASSIMILATION RATE OF GOLDEN DELICIOUS LEAVES.

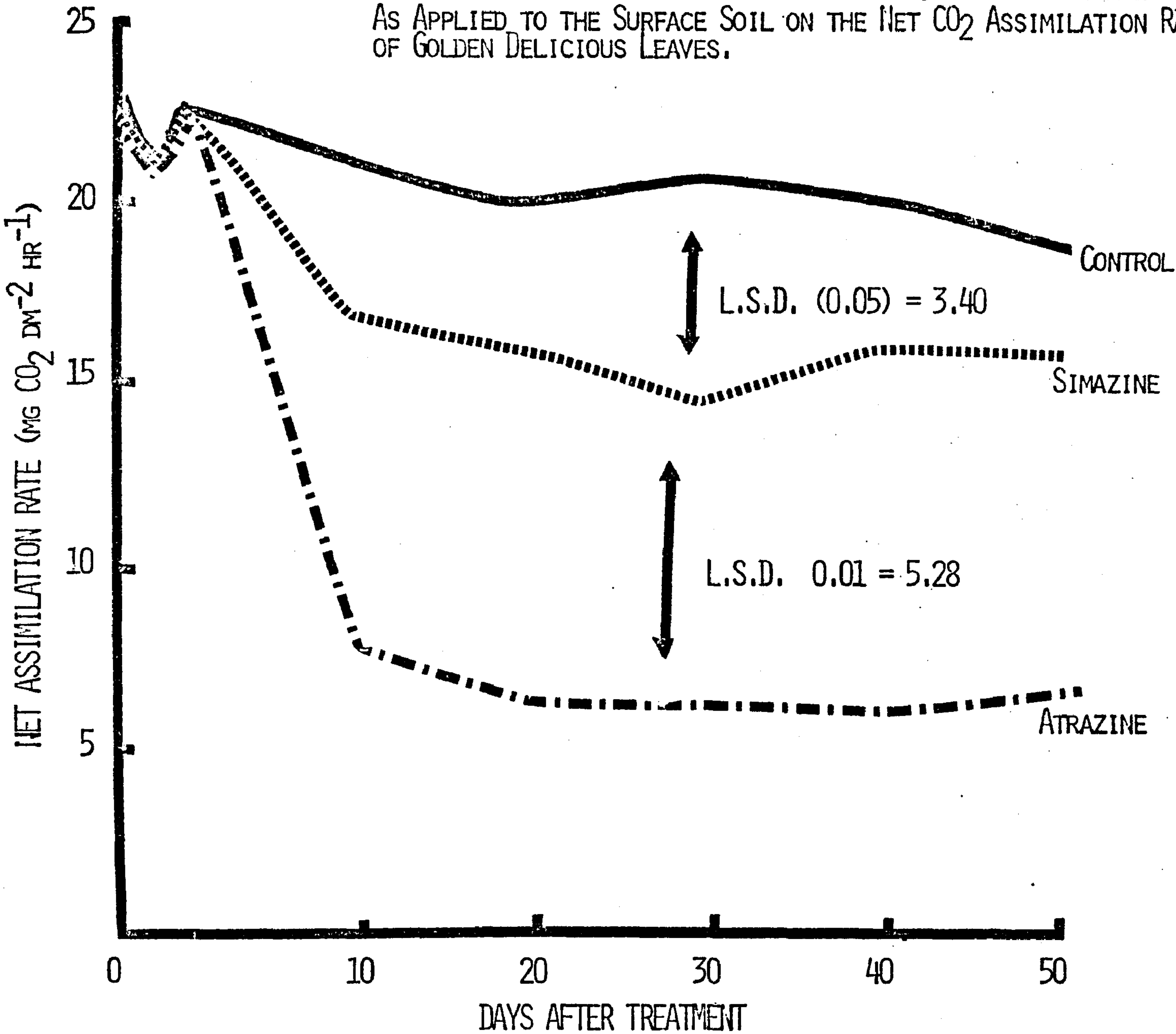
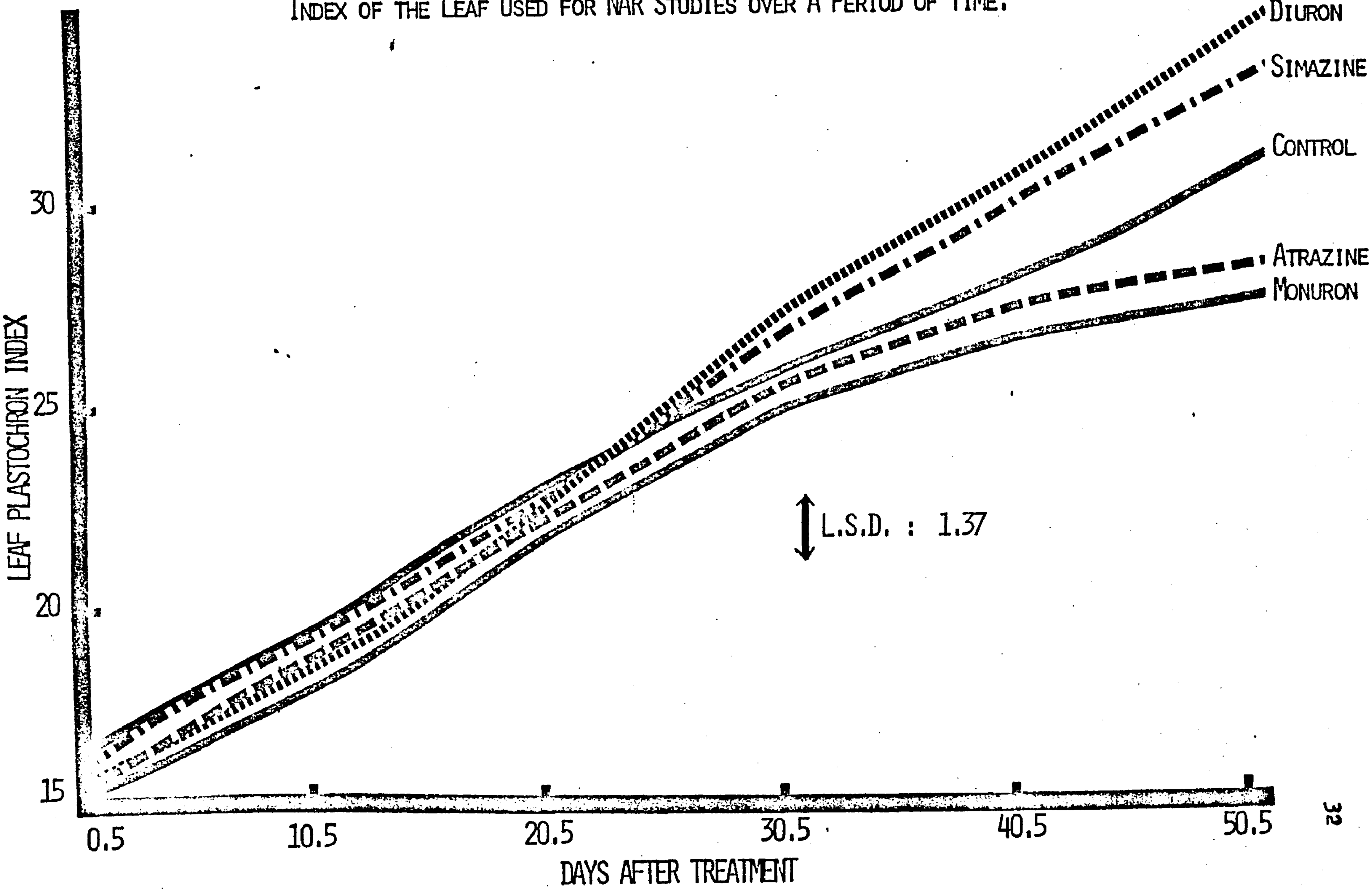


Fig. 7. Effect of Diuron, Monuron, Simazine and Atrazine on Plastochron Index of the Leaf Used for NAR Studies over A Period of Time.



significant effect on the net CO_2 assimilation rate. The NAR values for figures 5 and 6 are depicted in appendix 8.

The influence of herbicides on leaf plastochron index is depicted in figure 7. Near-linear curves started diverging 30 days after the treatment. Monuron caused the greatest depression in the leaf plastochron index, followed by atrazine, diuron, and simazine. At the conclusion of the trial, control trees had a lower leaf plastochron index than diuron- and simazine-treated trees. However, on an average, monuron- and atrazine-treated trees had a significantly lower leaf plastochron index than other treatments (Appendix 9).

DISCUSSION

Ferbam and parathion did not affect the chlorophyll content of apple leaves significantly (110). Therefore, the reported increase in the photosynthate levels of peach (79) and the stimulation in apple shoot growth by parathion (55), could be due to its direct influence on the net CO_2 assimilation rate. The screening trials (Fig. 2) in which endosulfan, ferbam, parathion, and tetradifon increased NAR by more than 6%, support the above view. Also, the decrease in NAR by diazinon and dicofol is in agreement with the earlier findings (44, 112).

The screening trials were also designed to determine the time required by a pesticide to influence the NAR. The NAR values dropped sharply in most cases 0.5 hours after the treatment (Table 1). However, the values gradually recovered in the next 25 hours for dodine- and parathion-treated leaves. The NAR of leaves sprayed with dimethoate increased on the second day after treatment. Sirois, et al. (91) report a similar case for Niacide-M and glyodin. These fungicides initially suppressed the rate of CO_2 assimilation in apple leaves, but the rates increased later. They suggested that the increased yields were brought about by the stimulatory effect of these chemicals on photosynthesis.

The results show that the chemicals used to control a specific group of pests may differ in their influence on NAR. The acaricides dicofol and Omite reduced NAR by more than 15% in the screening trials. However, tetradifon and endosulfan did not influence NAR in the present study. With the additional screening of different pesticides, it may be possible to select chemicals that are most efficient in pest control but have no deleterious effects on NAR.

The screening trials were designed to select chemicals most inhibiting to NAR in a short time so that they could be studied in a greater detail. Although the results obtained in the screening trials suggest a general trend, the results should not be over-emphasized as cultivar variability existed and only one leaf was used for each chemical tested.

In the multiple spray experiment, the treatments were assigned at random. However, the control trees of Golden Delicious initially had lower NAR. Therefore dicofol probably has a more retarding and dodine a less stimulating effect on NAR than appears in figure 3. The values obtained for the control-, dodine-, and dicofol-treated Golden Delicious leaves 10 days after the last spray were 90%, 82% and 62%, respectively, of their initial CO_2 absorption rates. Dicofol and dodine reduced NAR by 26% and 16%, respectively, in the screening trials also.

The reduction by dicofol of NAR of apple leaves and no appearance of phytotoxicity symptoms or influence on the tree growth supports the findings of others (101, 11[illegible]). However, the inhibition in NAR following the first and second spray ([illegible]% and 21%) and no significant recovery during the course of the trial indicates the lasting invisible damage dicofol may have on the trees (Appendix 5). Additional studies are needed to determine the influence of this chemical on fruit quality and yield that result from its effect on the NAR. Since dicofol reduced NAR more in Golden Delicious than in Delicious trees, trials comparing the influence of commonly used pesticides such as dicofol on different cultivars are needed. There are about 8 to 10 main cultivars of apple commonly grown in the United States, the number being much higher for apples sold at roadside stands. Evaluating the influence of commercially used pesticide chemicals on the photosynthetic efficiency can enable the apple industry to select pesticides which are least inhibiting to the NAR of a particular cultivar, thereby increasing the productivity. Variability amongst cultivars to a pesticide is a very complex problem and probably involves the interaction between several factors which need to be studied in greater detail. Depending on the constitution and thickness of the leaf cuticle and palisade tissue, and the climatic factors such as light intensity, temperature and moisture, the pesticides may influence the photosynthetic efficiency of various apple cultivars differentially.

The minimal values of NAR obtained at 1 hour and 10 days after the spray in all the treatments (Figs. 3 and 4) may be due to the way plant material was handled. The trees were grown in the greenhouse. The night before the spray treatment the trees were moved to the growth chamber (2000 ft. candles and 23°C). The next morning they were moved to the laboratory (23°C). For the next four days the trees were kept in the growth chamber until they were treated and readings taken 1-, 24-, 48-, and 72-hours following the spray. Low NAR values for all the treatments on 10th day suggest that one night was not sufficient for the plants to become adapted to the change in environment. Spraying the trees with distilled water caused a reduction in the NAR of leaves. Even in the screening trials (Table 1), there was a sudden drop in NAR 0.5 hours after the spray. This suggests that the readings taken on the 10th day, 1 hour and 24 hours after the spray were influenced by the environmental conditions as well as the pesticide chemical. Because the NAR values at 48 and 72 hours following the treatment did not differ significantly, it is believed that the trees were conditioned to the changed environment. Therefore, the readings taken at 48 and 72 hours after spraying are considered to be more valid expressions of the treatment effects on NAR. The decrease in the fluctuation of NAR values after each successive spray probably implies that the leaf was adapted to the changing conditions.

Oil sprays historically have been reported to inhibit photosynthesis. Schroeder (90) found a 17% decrease in the CO_2 absorbed by apple leaves on the first and third day following a 1% oil spray. Spray oils reduced photosynthesis in bananas (80), oranges and lemons (108) and parsnips (76). A 66% reduction in NAR in the screening trials by a 2% application of 70 second oil (Fig. 1) and 9% reduction obtained by 0.25% of the same oil (Table 2) in benomyl-and-oil experiment also support the earlier findings (90).

Because of the low costs involved, there is an increasing interest in the summer spray oils. Their use with lower rates of expensive fungicides like benomyl has been advocated to control plant diseases more economically. Addition of benomyl in general, appears to reduce the toxic effects of oil on NAR of Golden Delicious leaves (Table 2). However, the 9% reduction in NAR caused by oil (2.5 $ml.l^{-1}$) may not be of practical importance to discourage its inclusion in the spray programs.

In the benomyl and oil experiment designed to determine the effect of these chemicals on the leaves of different ages, the older, 25th leaf (from the index leaf) surprisingly had a higher NAR than the younger, 15th leaf (Table 3). The trees were started late in summer and they were growing rapidly in the greenhouse. Probably the 25th leaf had not aged enough to cause a reduction in its NAR (45). Insignificant differences by the treatments on NAR in this experiment may be because of the cultivar tolerance (Delicious was used rather than Golden Delicious in the earlier case). Also, the trees were conditioned at a lower light intensity (400 ft. candles) before being placed in the leaf chamber for the NAR determination. Lower light intensity, coupled with the lower temperature, might have lessened the effect of oil.

In the herbicide experiment, monuron (Fig. 5) decreased NAR of apple leaves the most (45% of control), followed by atrazine (37%) (Fig.6) and simazine (12%). The Diuron treatment did not affect NAR significantly (Fig. 5). The herbicide effects appeared 10 days after the application and persisted throughout the remainder of the trial.

All these herbicides are potential inhibitors of photosynthesis (36, 30). Because of their higher mobility, greater amounts of monuron (47) and atrazine (99) probably moved into the root zone and were absorbed thereby causing a greater reduction in NAR of apple trees than the less mobile diuron and simazine. Monuron and atrazine, through their influence on the net CO_2 assimilation rate, also reduced growth as indicated by reductions in total leaf count and leaf plastochron index (Fig. 7).

Simazine, on the other hand, reduced NAR significantly, but compared to the control, increased the leaf plastochron index in the later states of the experiment (Fig. 7). However, the average increase in the leaf plastochron index was not significant (Appendix 9). This increase is probably because of the lower leaf plastochron index values for the control treatment rather than a stimulatory influence of simazine. During the course of the experiment, the control treatment in one replication had produced only 6 leaves as compared to 15 by an average tree. However, there are reports of increased nitrogen and protein contents of leaves and stimulation in growth and cropping of apples by simazine treatments (11,57).

It is very difficult to correlate a complex phenomenon like growth directly to NAR. However, in the present studies 37% was the minimal reduction (Appendix 8) at which a significant decrease in the plastochron index was noted (Fig. 7). The nature of the pesticide chemical and its influence on other plant processes is a very important factor in determining the minimal change in NAR values required to influence the plant growth. In addition, it is reasonable to assume that the degree of inhibition in the NAR is inversely correlated with the time required for the appearance of visible reduction in plant growth. In the present studies, 37% inhibition in NAR by atrazine resulted in lower leaf plastochron index 30 days after the treatment. The lower rates of NAR inhibition will probably be reflected after longer periods of time in the apparent tree growth.

Larsen and Isebrands (64) indicated that leaf plastochron index could serve two useful purposes when applied to developmental studies of woody plants: (1) to adjust plants of different developmental stages to a standardized morphological time scale, and (2) to predict developmental processes and events from simple, non-destructive measurements. The leaf plastochron index served as a very useful tool in the benomyl-and-oil study where, because of the large size of the experiment, each replication was treated at intervals of 13 days. In each replication, regardless of the total number of leaves on the tree, the 22nd leaf from the index leaf was employed for NAR determination.

This, on an average, ensured less than 0.6 mg. CO_2 dm^{-2} hr^{-1} of deviation in NAR values in the three replications from their mean. The leaf plastochron index also served the useful purpose as an indicator or morphological growth. For more precise studies on photosynthesis, it is suggested that instead of using the time scale, the NAR of leaves having the same plastochron age be compared. Dickmann (28) also found a high correlation between the net CO_2 assimilation rate and the plastochron index.

Since the pesticides vary in their effect on the net CO_2 assimilation rate, there is a need for evaluating the influence of newly introduced chemicals on NAR and its relationship with growth, crop quality, and the yield. Knowledge about the chemicals which are effective in pest control without impairing the photosynthetic rate will certainly be beneficial to the fruit industry. The differential effects of the multiple sprays of dicofol and dodine obtained for the two apple cultivars in the present investigation, point out the need for additional information concerning cultivar response to specific pesticides and pesticide combinations. The synergistic effects of the different pesticide chemicals on NAR is an important and hitherto neglected area.

SUMMARY

Endosulfan increased while Superior oil, benomyl + Superior oil, dodine, diazinon, dicofol, Omite, leptophos (E.C.), and thiram decreased the net CO_2 assimilation rate of apple leaves by more than 10% in the screening trials. The remainder of the pesticides tested influenced this process by less than 10%. The major effects of the pesticides on NAR were evident 0.5 hour following the treatment.

In the multiple spray experiment, dicofol ($1.3\ ml.l^{-1}$) significantly reduced NAR in apple leaves. Golden Delicious was significantly affected more than Delicious. The first two sprays caused the maximum reduction in NAR. Successive sprays of dicofol did not depress the net CO_2 assimilation rate further. Dodine ($0.6\ g.l^{-1}$) did not influence NAR to an appreciable extent.

Increasing the concentration of Superior oil in the spray decreased the NAR of Golden Delicious leaves. It appeared that the addition of benomyl in the spray solution lessens the adverse effects of oil on NAR. There was no apparent difference caused by the treatments to the leaves of different ages (15th and 25th from the index leaf).

Of the four herbicides incorporated into the surface soil of the pots, monuron most severely inhibited NAR (45%), followed by atrazine (37%)

and simazine (12%). Diuron, however, appeared to have no effect on the net CO_2 assimilation rate. The herbicide inhibition of NAR resulted in decreased plant growth when measured as plastochron index. Effects of the herbicides on NAR were apparent 10 days after the treatment and persisted until the conclusion of the trial (50 days).

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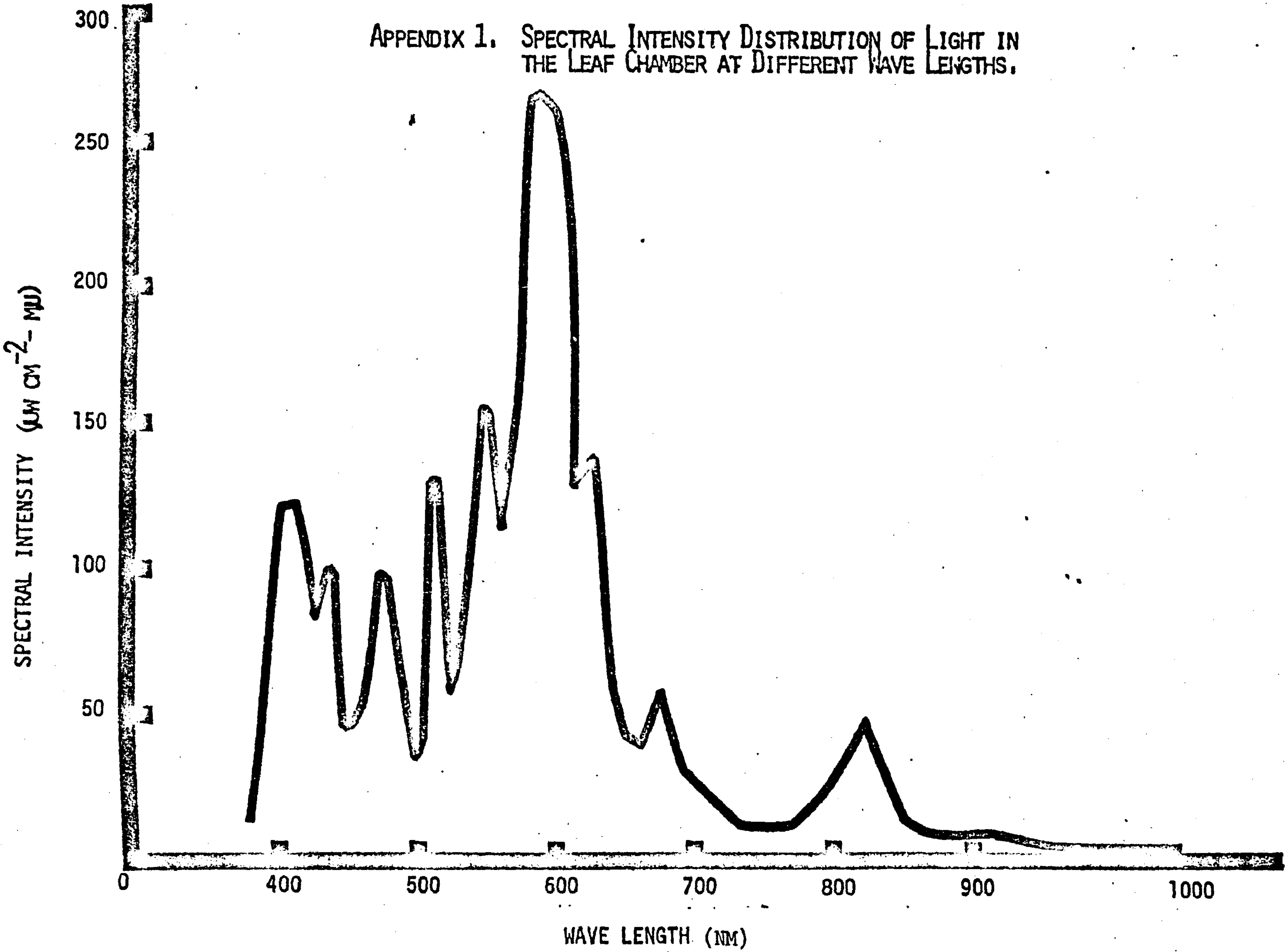
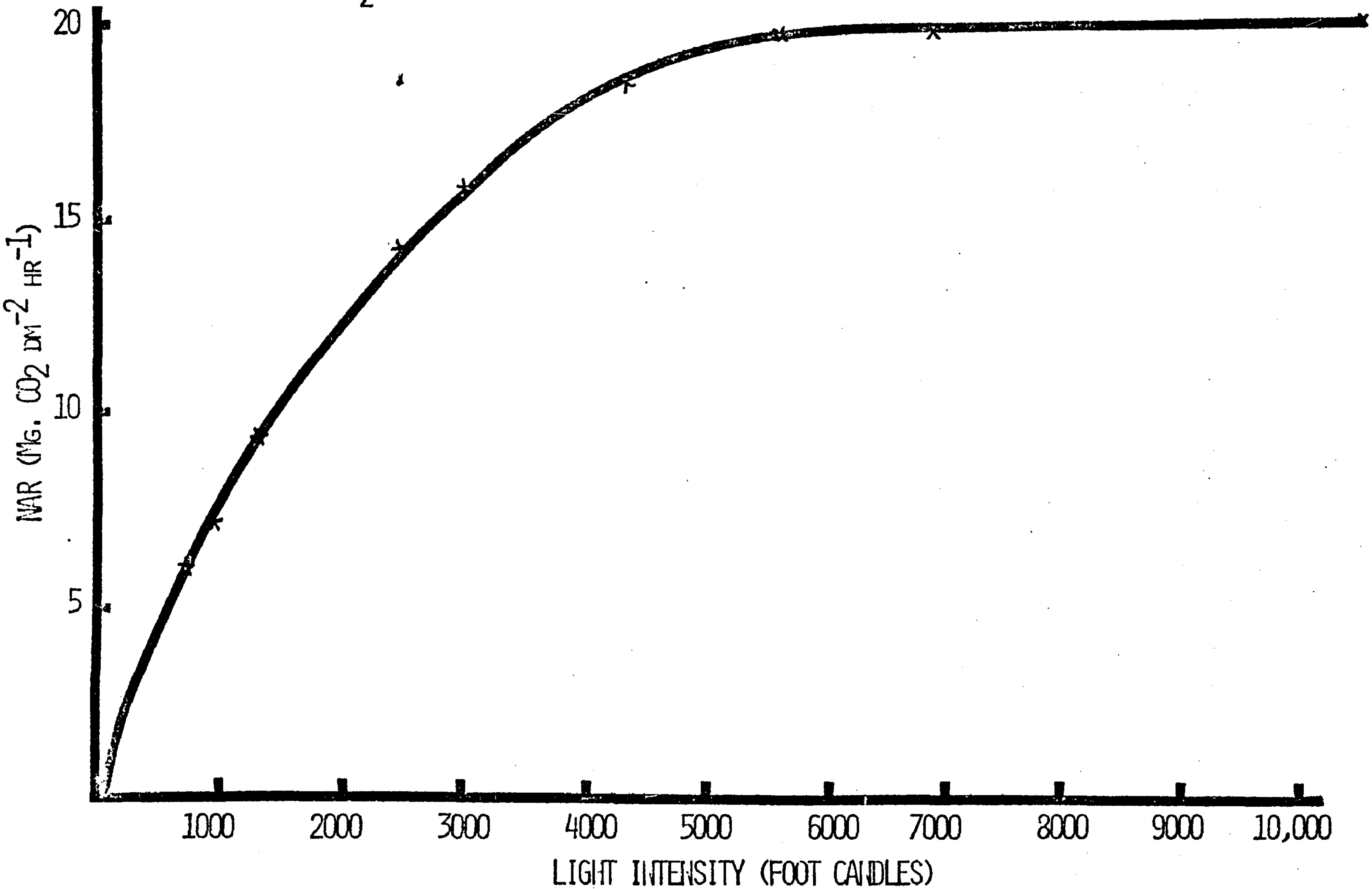
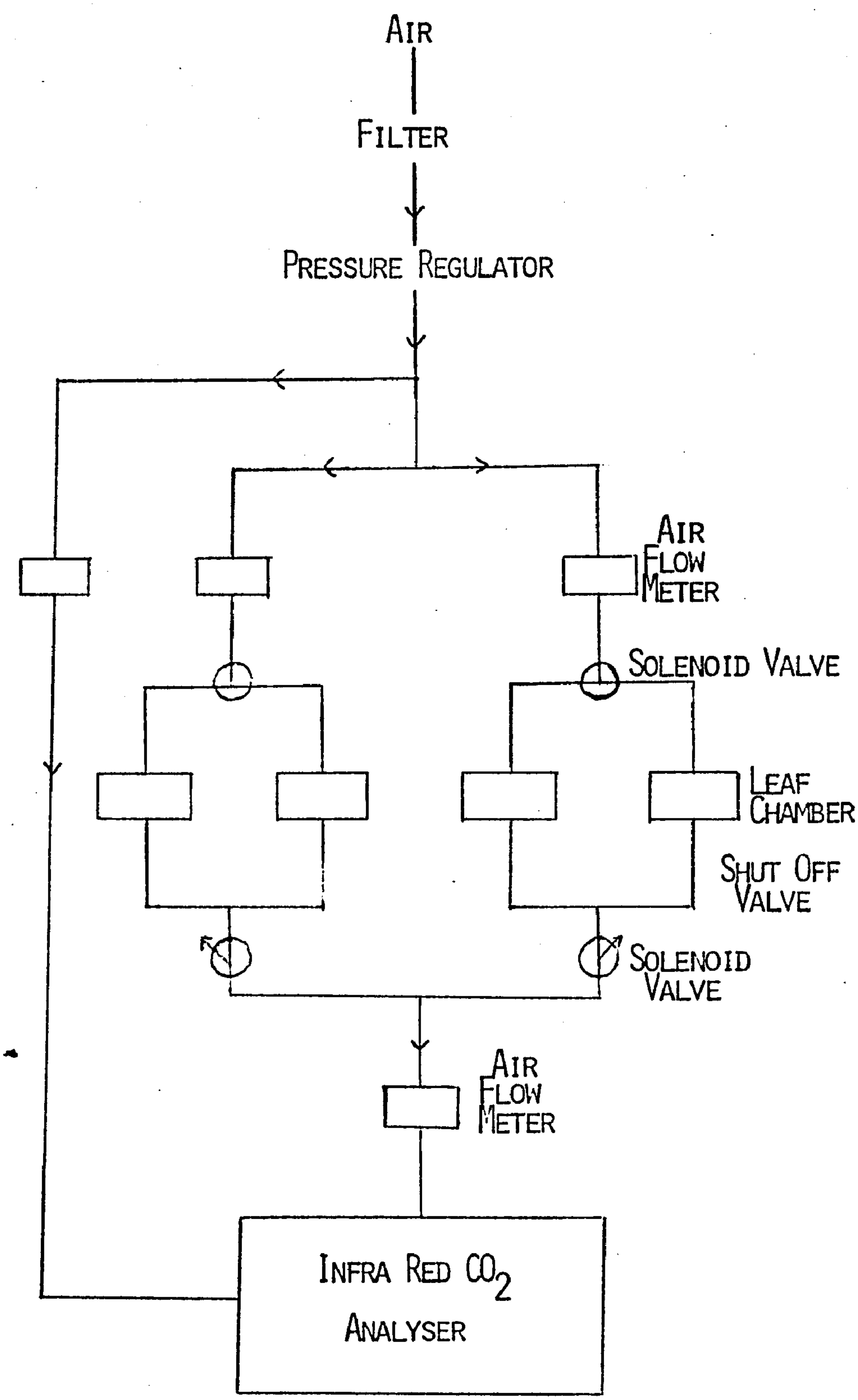
APPENDIX 1. SPECTRAL INTENSITY DISTRIBUTION OF LIGHT IN THE LEAF CHAMBER AT DIFFERENT WAVE LENGTHS.



APPENDIX 2. LIGHT RESPONSE CURVE: EFFECT OF LIGHT INTENSITY ON THE NET CO_2 ASSIMILATION RATE OF APPLE LEAVES.



Appendix 3. Flow Diagram for the Determination of Net CO_2 Assimilation Rate of Apple Leaves



Appendix 4. Effect of Pesticides[a] on the Net CO_2 Assimilation Rate[b] of Apple Leaves.

Pesticide				Percent Reduction in NAR				
				Hours After Treatment				
Common Name	Chemical Name	Formulation	Concentration Used	1	6	23	28	Average Reduction
‘Acaralate’	Isopropyl 4,4-dichloro-benzilate	2 E.C.[c]	2.5 ml.l^{-1}	7.69	7.69	4.62	5.78	6.45
Azinophos-methyl ‘Guthion’	0,0-Dimethyl-S[4-oxo-1,2,3-benzotriazin-3 (4H) -yl methyl] phosphorodithioate	50 W.P.[d]	0.6 g.l^{-1}	2.15	6.39	14.93	13.40	9.22
Benomyl and ‘Benlate’ Superior Oil	Methyl-1-(butylacarbamoyl) -2-benzimidazole-carbamate	50 W.P. +70 Sec.	0.6 g.l^{-1} +2.5 ml.l^{-1}	36.38	41.79	45.48	40.74	41.10
Captan	n-trichloro-methyl-thio-tetra-hydro-phthalimide	50 W.P.	2.4 g.l^{-1}	-1.61	9.68	-6.45	-6.45	-1.21
Carbaryl ‘Sevin’	1-Naphthyl methylcarbamate	50 W.P.	1.8 g.l^{-1}	0.00	0.00	4.41	-3.43	0.27
Carbofuran ‘Furadan’	2,3-dihydro-2,2-dimethyl-7-benzofuranyl methyl carbamate	42.4 E.C.	1.3 ml.l^{-1}	7.74	0.00	0.00	0.00	1.94
Chlorphenamidine ‘Fundalgalecron’	N'-(4-chloro-o-tolyl)-N,N-dimethylformamide	4 E.C.	1.3 m.l^{-1}	9.66	4.47	4.47	3.26	5.47

Appendix 4. (Continued)

Pesticide				Percent Reduction in NAR				
				Hours After Treatment				
Common Name	Chemical Name	Formulation	Concentration Used	1	6	23	28	Average Reduction
Chlorphenamidine `Fundalgalecron´	N'-(4-chloro-o-tolyl)-N,N-dimethylformamidine	97 W.P.	1.2 $g.l^{-1}$	3.07	0.64	21.19	0.00	6.23
Chlorpyrifos `Dursban´	0,0-Diethyl-0-(3,5,6-trichloro-2-byridyl) phosphorothioate	25 W.P.	1.2 $g.l^{-1}$	0.00	3.87	19.50	0.00	5.84
Demeton `Systox´	I-0,0-Diethyl-0-2-(ethylthio)ethylphosphorothioate	2 E.C.	0.3 $ml.l^{-1}$	10.37	8.63	6.89	3.48	7.34
	II-0,0-Diethyl-S-2-(ethylthio) ethylphosphorodithioate							
Diazinon	0,0-Diethyl-o-(2-isopropyl-4 methyl-6-primidiryl) phosphorothioate	50 W.P.	2.0 $g.l^{-1}$	26.17	26.17	27.14	27.14	26.66
Dicofol `Kelthane´	2,2,2-trichloro-1,1-di-(4-chlorophenyl) ethanol	42 E.C.	1.3 $g.l^{-1}$	26.93	22.16	22.68	22.68	23.61
`Dikar´	(Special formulation including active ingredients of Mancozeb and Dinocap)	72 W.P.	4.8 $g.l^{-1}$	0.00	2.37	11.96	11.87	6.55

Appendix 4. (Continued)

Pesticide				Percent Reduction in NAR				
				Hours After Treatment				
Common Name	Chemical Name	Formulation	Concentration Used	1	6	23	28	Average Reduction
Dimethoate 'Cygon'	0,0-Dimethyl-S-(N-methyl-carbamoylmethyl) phosphorodithioate	25 E.C.	1.3 ml.l^{-1}	11.10	5.55	8.37	2.78	6.95
Dinocap 'Karathane'	2-(1-methylheptyl)-4,6-dinitrophenyl crotonate and other nitrophenols and derivatives	25 W.P.	1.2 g.l^{-1}	4.70	5.92	5.55	5.20	5.34
Dodine 'Cyprex'	n-do-decyl-guanidine acetate	65 W.P.	0.6 g.l^{-1}	19.33	7.24	17.32	13.32	14.30
Endosulfan 'Thiodan'	6,7,8,9,10, 10-Hexachloro-1,5,5a,6,9, 9a-hexahydro-6, 9-methano2,4,3-benzodioxan-thiepin-3-oxide	50 W.P.	3.6 g.l^{-1}	-6.64	-3.35	-16.64	-15.02	-10.41
Ferbam	Ferric dimethyl-di-thio-carbamate	76 W.P.	2.4 g.l^{-1}	-7.9	-5.6	-10.7	-6.63	-7.71
Folpet	n-tri-chloro-methyl-thio-phthalimide	50 W.P.	2.4 g.l^{-1}	0.00	-3.77	4.12	4.10	1.11
Formetanate 'Carzol'	M-[[(Dimethyl amine) methyl-ene] amino] phenyl methyl-carbamate	92 W.P.	1.2 g.l^{-1}	3.89	3.89	12.87	0.89	5.39

Appendix 4. (Continued)

Pesticide				Percent Reduction in NAR				
				Hours After Treatment				
Common Name	Chemical Name	Formulation	Concentration Used	1	6	23	28	Average Reduction
Lead Arsenate			3.6 $g.l^{-1}$	5.91	5.91	-2.91	0.00	0.00
Leptophos 'Phosvel'	O-(2,5-Dichloro-4-bromophenyl)-O-methyl phenylthiophosphonate	30.6 E.C.	2.5 $ml.l^{-1}$	15.06	8.05	15.06	15.06	13.32
Leptophos 'Phosvel'		45 W.P.	1.2 $g.l^{-1}$	0.00	-4.48	6.67	6.67	2.24
Mancozeb 'Dithane'	Coordination product of zinc ion and manganese ethylene-bis-dithiocarbamate	80 W.P.	2.4 $g.l^{-1}$	0.00	0.00	3.87	0.00	0.97
Methomyl 'Lannate'	S-methyl N-[(methyl carbamoyl) oxy] thioacetimidate	25 W.P.	1.2 $g.l^{-1}$	3.95	-0.98	-7.36	-2.42	-1.72
'Omite'	2-(p-tert-butylphenoxy) cyclohexyl progargyl sulfite	30 W.P.	1.5 $g.l^{-1}$	13.47	12.49	17.61	17.61	15.32
Parathion	O,O-Diethyl-o-nitrophenyl phosphorothioate	15 W.P.	1.8 $g.l^{-1}$	-5.98	5.02	-16.64	-13.99	-8.12

Appendix 4. (Continued)

Pesticide		Formu-lation	Concen-tration Used	Percent Reduction in NAR				
				Hours After Treatment				Average Reduction
Common Name	Chemical Name			1	6	23	28	
Phosalone 'Zolone'	0,0-diethyl S-[(6-chloro-2-oxobenzoxolin-3-yl) methyl] phosphorodithioate	3 E.C.	1.3 ml.l^{-1}	10.05	10.05	7.49	2.47	6.77
Phosmet 'Imidan'	N-(mercaptomethyl) phthalimide S-(0,0-dimethyl phosphorodithioate)	50 W.P.	1.2 g.l^{-1}	-3.33	3.11	1.72	5.80	1.83
'Plictran'	Tricyclohexyltin	50 W.P.	0.3 g.l^{-1}	-10.44	-5.97	-4.52	-3.01	-5.99
Quinomethionate 'Morestan'	6-methyl-2,3-quinoxaline-dithiol cyclic carbonate	25 W.P.	1.2 g.l^{-1}	-7.41	13.46	-4.44	-0.85	0.19
Streptomycin	Streptomycin sulfate	17 W.P.	100 ppm	14.06	10.32	5.69	10.17	10.06
Sulfur		96 W.P.	7.2 g.l^{-1}	3.14	3.14	11.71	13.50	8.07
'Superior Oil'		70 Sec.	20 ml.l^{-1}	59.64	64.99	67.86	64.27	66.28
Tetradifon 'Tedion'	4-chlorophenyl 2,4,5-trichlorophenyl sulfone	25 W.P.	1.2 g.l^{-1}	0.00	0.41	-14.38	-10.21	-6.27

Appendix 4. (Continued)

Pesticide: Common Name	Pesticide: Chemical Name	Formulation	Concentration Used	Percent Reduction in NAR: Hours After Treatment: 1	6	23	28	Reduction
Thiram	Tetra-methyl-thiuram disulfide	75 W.P.	2.4 $g.l^{-1}$	8.04	8.04	30.14	7.28	13.38
‘Vydate’ 2 L	Methyl-N', N-Dimethyl-N [(Methylcarbamoyl) oxy] -1- Thiooxaminidate	2 E.C.	1.3 $g.l^{-1}$	2.62	2.15	0.85	0.00	1.41

[a] The pesticides are listed according to their official names. Trade name, if different from the official name, is included between inverted commas. Common name of a chemical which has not been assigned any official name, also appears within inverted commas.

[b] Expressed as percent reduction in NAR. Negative values show an increase in NAR following the treatment.

[c] E.C. = Emulsifiable Concentrate.

[d] W.P. = Wettable Powder.

Appendix 5. Effect of Multiple Applications of Dicofol[a] and Dodine[b] on the Net CO_2 Assimilation Rate of Golden Delicious and Delicious Apple Leaves (Mgs. CO_2 dm^{-2} hr^{-1}).

	Golden Delicious					Delicious				
	Period[c] Following Each Spray					Period[c] Following Each Spray				
Treatment	1	2	3	4	Mean	1	2	3	4	Mean
Control	23.21	20.39	21.29	22.04	21.73	23.56	21.11	20.27	20.49	21.36
Dodine	25.27	20.80	21.62	22.97	22.66	23.60	20.62	20.27	19.79	21.07
Dicofol	21.96	16.32	16.58	17.11	17.99	23.42	19.47	17.84	18.21	19.73
Mean	23.48	19.17	19.83	20.71	20.80	23.53	20.40	19.46	19.50	20.72

	Period	Tt. X Var.	Var. X Period	Tt. X Period
L.S.D. 0.05	0.527	0.627	0.745	0.913
L.S.D. 0.01	0.693	0.824	0.979	1.200

[a]Dicofol: 42% E.C. @ 1.25 $ml.1^{-1}$.

[b]Dodine: 65% W.P. @ 0.60 $g.1^{-1}$.

[c]Period: Mean of the readings taken 1 hr., 24 hrs., 48 hrs., 72 hrs. and 10 days following each spray.

Appendix 6. The Effect of Dicofol[a] and Dodine[b] on the Net Assimilation Rate (Mgs. CO_2 dm^{-2} hr^{-1}) of Golden Delicious Leaves at Different Time Intervals Following the Application of Spray

Spray Period[c]	Time After Spray 1 Hr.	24 Hr.	48 Hr.	72 Hr.	10 Days	Period Mean
1	23.50	24.66	24.05	24.92	20.38	23.50
2	17.64	20.28	21.38	21.76	17.86	19.78
3	18.11	19.38	20.35	20.93	19.45	19.64
4	19.41	20.65	21.28	19.99	19.17	20.10
Mean	19.67	21.25	21.77	21.90	19.22	20.76

L.S.D.: Time After Spray: (0.05) = 0.606
(0.01) = 0.796

Period X Time After Spray: (0.05) = 1.194
(0.01) = 1.569

[a]Dicofol: 42% E.C. @ 1.25 $ml.l^{-1}$.

[b]Dodine: 65% W.P. @ 0.60 $g.l^{-1}$.

[c]Period: Mean of the readings taken 1 hr., 24 hrs., 48 hrs., 72 hrs., and 10 days following each spray.

Appendix 7. Effect of Different Levels of Benomyl[a] and Oil[b] on the Average Number of Leaves Produced by Each Tree

Benomyl ($g.l^{-1}$) \ Oil ($ml.l^{-1}$)	0	1.25	2.5	Mean
0	8.3	9.7	9.0	9.0
0.15	9.7	9.3	9.0	9.3
0.3	8.0	9.7	9.3	9.0
Mean	8.7	9.6	9.1	9.1

[a]Benomyl: 50% W.P.

[b]Oil: 70 second viscosity superior oil.

Appendix 8. Effect of Diuron, Monuron, Simazine and Atrazine[a] on the Net CO_2 Assimilation Rate (Mgs. CO_2 dm^{-2} hr^{-1}) of Golden Delicious Leaves

Treatment	Days After Treatment									Mean
	0.5	1.5	2.5	3.5	10.5	20.5	30.5	40.5	50.5	
Control	22.69	21.19	20.96	22.84	21.13	19.90	20.76	20.15	18.94	20.95
Diuron	23.35	21.49	22.08	24.47	22.67	22.61	19.71	19.03	19.30	21.63
Monuron	19.17	18.64	17.46	16.88	5.80	5.71	4.98	7.61	8.57	11.64
Simazine	22.34	20.60	20.32	23.19	16.58	15.99	14.38	16.16	15.79	18.37
Atrazine	22.34	21.06	20.82	22.78	7.51	6.35	4.99	5.99	6.45	13.14
Mean	21.98	20.59	20.33	22.03	14.74	14.11	12.79	13.79	13.81	17.15

	Treatment	Days	Treatment X Days
L.S.D. 0.05:	1.331	1.787	3.400
L.S.D. 0.01:	1.759	2.362	5.284

[a]80% Formulations of the herbicides were used @ 4.5 kgs. $hectare^{-1}$.

Appendix 9. Effect of Diuron, Monuron, Simazine and Atrazine[a] on the Plastochron Index[b] of Golden Delicious Trees

Treatment	Days After Treatment									Mean
	0.5	1.5	2.5	3.5	10.5	20.5	30.5	40.5	50.5	
Control	16.29	16.69	17.08	17.32	19.47	23.08	25.75	27.97	30.68	21.59
Diuron	15.45	15.76	16.00	16.29	18.33	22.75	27.33	30.79	34.33	21.89
Monuron	15.76	15.97	16.28	16.54	18.15	21.89	25.09	26.83	27.68	20.47
Simazine	16.02	16.37	17.04	17.43	19.14	22.96	27.06	30.13	33.04	22.13
Atrazine	15.72	15.94	16.28	16.59	18.69	22.39	25.66	27.45	28.45	20.80
Mean	15.85	16.15	16.54	16.84	18.76	22.61	26.18	28.64	30.83	21.38

	Treatment	Days	Treatment X Days
L.S.D. 0.05:	0.90	1.21	2.71
L.S.D. 0.01:	1.20	1.61	3.59

[a] 80% Formulations of the herbicides were used @ 4.5 kgs. hectare^{-1}.

[b] Youngest leaf with $\geq$ 3 cm. blade length was used as index leaf for determining the plastochron index.