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Effects of Some Formulations of Methyl Parathion, Toxaphene and DDT on the Cotton Plant[1]

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M. E. MERKL observed in 1954 that differences in maturity of cotton plants were related to the kind of insecticide used for insect control but that observed differences in plant behavior were not related to differences in insect activity. This suggested that one or more of the insecticides had some direct action on the cotton plant. Similar reports were received from farmers, from other research and extension workers, and from representatives of the agricultural chemical industry. In 1959 and 1960 replicated field experiments were conducted to determine whether certain commercial formulations of methyl parathion (*O,O*-dimethyl *O-p*-nitrophenyl phosphorothioate), toxaphene (chlorinated camphene containing 67–69% chlorine), and DDT (1,1,1-trichloro-2,2-bis(*p*-chlorophenyl)ethane) have direct action on the metabolism of the cotton plant. Preliminary reports of this research have been given elsewhere.[3]

REVIEW OF LITERATURE

A number of chemical compounds related to methyl parathion have been reported to affect the growth and development of cotton. McIlrath (19) reported that ethyl parathion (*O,O*-diethyl *O-p*-nitrophenyl phosphorothioate) caused an increase in the number of lobes per leaf of cotton plants grown in the greenhouse. Fowler (11) observed there were more vegetative branches on cotton plants treated with ethyl parathion than on untreated plants and that there was a corresponding decrease in fruit production. From a study of carbon:nitrogen ratios, he attributed the increase in vegetative development to a direct physiological effect of the insecticide. There was an increase in nitrogen in treated plants which was negatively correlated with sugar content and he considered that this promoted vegetative growth until the C:N balance was restored. Hacskaylo and Ergle (12) found that cotton plants grown in nutrient cultures containing low concentrations of schradan (octamethyl pyrophosphoramide) had more vegetative growth than untreated control plants, and that bolls were borne on successively higher fruiting branches with increasing concentrations of schradan. Minton[4] reported a significant delay in the maturity of the cotton crop when zinophos (*O,O*-diethyl *O*-2-pyrazinyl phosphorothioate) was applied to the soil for wilt and nematode control 14 days prior to planting. Hacskaylo and Scales (13) found that cotton plants treated with 0.25 pound guthion (*O,O*-dimethyl *S*-(4-oxo-1,2,3-benzotriazin-3-(4*H*)-ylmethyl) phosphorodithioate) per acre under insect-free conditions in the greenhouse produced more flowers than untreated plants.

Materials related to methyl parathion are able to participate in plant metabolism under certain conditions. Jansen et al. (15) found that tetraethyl pyrophosphate and other organic phosphate insecticides inhibited acetyl esterase from citrus fruits. Hall (14) treated plants of carnation and tomato with tetraethyl pyrophosphate. High concentrations caused epinasty, chlorosis and necrosis of the growing point with resultant loss of apical dominance. Lower concentrations caused stem elongation, earlier flowering, increased uptake of molecular oxygen and increased the accumulation of inorganic phosphate. Hall postulated that these effects of tetraethyl pyrophosphate at low concentrations were due to enzymatic action, possibly by stimulating the natural synthesis of high energy phosphate. Lord (16) found that the hydrolysis of phenyl acetate by enzymes of bean and mangold leaves was inhibited by spraying plants with ethyl parathion and of bean leaves by spraying with systox (*O,O*-diethyl *S*(and *O*)-2-(ethylthio)ethyl phosphorothioate).

Methyl parathion was not known to have direct action on plant metabolism before the preliminary reports of our own work. Since then, Brown et al. (5) have reported two field experiments in which the percent of open bolls in October was less in cotton treated with methyl parathion than in untreated check plots. Adkisson et al. (1) reported an experiment in which there were no significant differences in rate of flowering, number of flowers, percentage of boll retention, boll weight, boll maturation period, earliness or yield of cotton treated with (a) methyl parathion, (b) methyl parathion + DDT, (c) guthion, (d) guthion + DDT, (e) dieldrin (1,2,3,4,10,10-hexachloro-6,7-epoxy-1,4,4a,5,6,7,8,8a-octahydro-1,4-*endo-exo*-5,8-dimethanonaphthalene) + DDT or (f) DDT. They also reported other experiments in which they found that neither methyl parathion + DDT, nor toxaphene + methyl parathion + DDT, nor guthion + DDT, nor toxaphene + DDT, nor sevin (1-naphthyl *N*-methylcarbamate) + DDT affected treated plants. Beckham and Morgan (3) compared the growth, development, and maturity of cotton plants treated with (a) toxaphene + DDT, (b) methyl parathion + DDT and (c) toxaphene + methyl parathion + DDT. Cotton treated with methyl parathion + DDT matured later than cotton treated with toxaphene + DDT but this difference was not consistent and they considered that it was not pronounced or of any major importance under their conditions.

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[3] Pfrimmer, T. R., and Roark, B. Some effects of chlorinated hydrocarbon and phosphate insecticides on cotton production. Paper presented at the Annual Meeting of the Southeastern Branch, Entomol. Soc. Am., Savannah, Ga. 1960.

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[4] Minton, E. B. Report of cotton nematodes and soil fumigation committee. Proc. 21st Meeting Cotton Disease Council. Greenville, S. C. January 9–10, 1961.

Table 1—Components of insecticide formulations and rates of application for each treatment.

Component	Pound/acre/application*				
	Methyl parathion formulation	Toxaphene formulation	DDT formulation	Toxaphene + DDT formulation	Methyl parathion + DDT§
80% methyl parathion (technical grade)†	0.21 (0.17)	-		-	0.21 (0.17)
Toxaphene (technical grade)	-	2.00	-	1.33	-
DDT (technical grade)	-	-	0.66	0.66	0.66
Emulsifiers‡	0.03	0.13	0.08	0.14	0.11
Spreader-Sticker	-	-	-	0.27	-
Aromatic naptha solvent	0.46	-	-	-	0.46
Petrol. distillate solvent	-	1.20	-	-	-
Xylene type solvent	-	-	1.94	0.97	1.94
Epichlorohydrin	-	0.01	-	0.01	-

* Rates increased 50% after July 6 in 1959, and July 8 in 1960. † 80% 0,0-dimethyl 0-p-nitrophenyl phosphorothioate + 20% xylene. Active ingredient in parenthesis.
‡ The same emulsifiers were used in all formulations. § Methyl parathion + DDT emulsion was prepared by mixing methyl parathion formulation and DDT formulation.

Table 2—Growth and production of flower buds by cotton plants 42 days after planting in 1960 (means for 40 hills). Treatments begun when plants were in seedling stage; arranged in order of bud production.

Treatment	Height/ plant, cm.	Plants/ hill	Plant wt./ hill, g.†	Buds/ hill	Bud wt./ hill, mg.†
Toxaphene + DDT	33	5.8	27	17a‡	211a‡
DDT	31	6.0	26	17a	210a
Toxaphene	33	5.9	26	15a	181a
No insecticide	33	5.9	28	14a	163a
Methyl parathion	34	6.2	29	6b	26b
Methyl parathion + DDT	32	5.5	23	3b	12b
Anal. of variance	ns	ns	ns	***	***
Coeff. of variability	6	9	14	21	33

† Oven dry weight basis. ‡ Averages followed by the same letter do not differ significantly by Duncan's (6) multiple range test at the 5% level. Averages followed by different letters differ at the 0.1% level. ns - Not significant at 5% level.
*** Significant at 0.1% level.

Lumsden and Smith (17), Pickett (26), Allen and Casida (2) and many others have reported that under certain conditions DDT and its analogues have auxin-like effects on plants. R. C. Roark (27) reviewed the early work with toxaphene, including effects on plants. In general, toxaphene has little effect on most species of plants but it is highly toxic to a few, including curcurbits and potatoes. Brown et al. (4) compared cotton treated with toxaphene + DDT with cotton treated with calcium arsenate and with untreated control plants. They found no differences between treatments in square and boll shedding nor in earliness and yield. Weekly applications of toxaphene + DDT resulted in the production of a significantly greater number of bolls, but the increase in boll number was offset by a reduction in boll size so that yields were not affected. They attributed this effect of toxaphene + DDT to direct action of the insecticide. Hacskaylo and Scales (13) studied the direct action of certain insecticides on cotton plants grown in the greenhouse under insect-free conditions. Plants treated with guthion + DDT had less dry weight and yield than comparable plants treated only with guthion.

MATERIALS AND METHODS

About 5 acres of sandy loam were planted to American upland cotton (*Gossypium hirsutum*, L., cultivar, Deltapine 15) on April 30, 1959, and on May 13 and 14, 1960. One hundred pounds of nitrogen per acre were applied in the seed bed before planting using commercial anhydrous ammonia. Rows were 40 inches apart and hills were spaced at 19-inch intervals along each row. The final stand averaged from about 3 to 5 plants per hill in 1959 and from 5 to 6 plants per hill in 1960. A healthy, vigorous stand of cotton free of weeds was maintained throughout the growing season in each year.

The experimental area was divided into 24 similar plots of ⅕ acre each. There were 6 experimental treatments and 4 replications of each treatment arranged in a randomized complete block experimental design. The treatments were: (a) methyl parathion, (b) toxaphene, (c) DDT, (d) toxaphene + DDT, (e) methyl parathion + DDT and (f) no insecticides. All insecticide formulations were water-emulsifiable concentrates obtained from a commercial source. The components of each formulation and their rate of application are given in Table 1. The spray emulsion used for the methyl parathion + DDT treatment was obtained by mixing proper proportions of methyl parathion concentrate and DDT concentrate with water immediately before application. Insecticide treatments were started on May 8 in 1959 and on May 23 in 1960, when the cotyledons were fully expanded. They were repeated at 4- or 5-day intervals as weather conditions permitted. Treatments were applied with an 8-row high-clearance spray machine using 2 No. X-4 nozzles/row and applying 4 gallons per acre of spray emulsion. Commencing with the 13th application (July 6) in 1959 and with the 11th application (July 8) in 1960, the rate/acre was increased 50% by using 3 No. X-4 nozzles/row and applying 6 gallons per acre of spray emulsion. Altogether, 25 applications were made in 1959 and 24 applications in 1960.

Table 3—Distribution of vegetative and fruiting branches on main axis and boll production per plant for cotton plants which received different treatments (means for 30 plants). Evaluated Oct. 24 to Nov. 1, 1960. Cotyledonary node was counted as No. 1. Treatments in order of number of vegetative branches produced.

Treatment	Nodes on main axis	Vegetative branches			Fruiting branches	
		Lowest node on main axis	No. produced	No. bolls produced†	Lowest node on main axis	No. bolls produced
Methyl parathion	27	5.8	4.1a‡	0.8	10.0a‡	10.3
Methyl parathion + DDT	27	6.1	3.7a	0.4	9.7a	11.3
Toxaphene + DDT	28	5.7	1.8b	0.2	7.4b	12.1
No insecticide	29	5.5	1.7b	0.2	7.4b	9.9
Toxaphene	28	5.5	1.5b	0.1	7.0b	12.1
DDT	27	5.7	1.3b	0.1	7.0b	11.2
Anal. of variance	ns	ns	***	ns	***	ns
Coeff. of variability	4	5	12	66	2	14

† Primary vegetative branches sometimes have secondary branches which bear bolls.
‡ Averages followed by the same letter do not differ significantly by Duncan's (6) multiple range test at the 0.1% level. ns -Not significant at 5% level.
*** Significant at 0.1% level.

Each plot was 24 rows wide. The eight central rows were used to determine the yield of seed cotton (cotton seed and lint before ginning). They were picked by hand 4 times in 1959 and 2 times in 1960. All other data were obtained from the 6 rows of cotton adjacent to each side of the 8 central rows. Methods for obtaining data on thrips and tarnished plant bugs have been described previously (24, 25).

RESULTS AND DISCUSSION

The treatments had little or no effect on plant survival or early growth. In both 1959 and 1960 there were fewer early flower buds (squares) on plants treated with methyl parathion or methyl parathion + DDT than on plants treated with toxaphene or DDT or toxaphene + DDT or no insecticide. Table 2 shows the height and dry weight of the aerial portion of the plants and the number of squares for each treatment 42 days after planting (1960 data). At this date the plots had received eight applications of insecticide. The plants were removed from the field and carefully dissected in the laboratory so that the smallest macroscopic squares could be counted. No empty fruiting positions were found.

In both 1959 and 1960 the first fruiting branch developed higher on plants treated with methyl parathion or methyl parathion + DDT than on plants treated with toxaphene or DDT or toxaphene + DDT or no insecticide. Data for 1960 is shown in Table 3.

Evidence of delayed reproductive development (Table 2) was obtained 42 days after planting on plants which were sprayed with a formulation containing methyl parathion. There were few insects on the treated plots before this date. Of those insects which damage cotton, only thrips and tarnished plant bugs were present in detectable numbers (Tables 4 and 5). Insect data for 1959 were in agreement with data presented for 1960. Plants treated with methyl parathion or methyl parathion + DDT belonged to a different population than plants treated with toxaphene or DDT or toxaphene + DDT with respect to number of squares (Table 2) and position of first fruiting branch (Table 3) but there was no similar division of insecticide treatments for thrips (Table 4) and tarnished plant bug (Table 5) populations. Injury to seedling cotton by uncontrolled infestations of thrips sometimes results in delayed maturity of the crop (22) but in this experiment thrips and tarnished plant bug infestations were low and little injury occurred even in unsprayed plots. Unsprayed plants had the most thrips and plant bugs but they resembled plants sprayed with DDT, toxaphene, and toxaphene + DDT in reproductive development rather than plants sprayed with methyl parathion and methyl parathion + DDT. Thus, the delayed initiation of the reproductive stage which was observed when seedling cotton plants were sprayed with the methyl parathion formulation seems unrelated to infestations of insects. This suggests that the effect was due to some component of the formulation which was active in plant metabolism. Heavy aromatic naphthas of the type used to formulate insecticides have been shown (23) to have no visible effects on cotton plants at 4 times the concentration used in our experiment. Therefore, it seems most probable that the observed effects were due to the active ingredient, methyl parathion. The observed plant response may have involved a metabolite of methyl parathion since certain related phosphorus esters are known to be translocated readily in the plant (28) and to be metabolized in the plant to other active biochemicals (20, 21).

Eaton and Rigler (9) and Eaton (7, 8) point out that the position of the first fruiting branch is sensitive to many factors. These include genotype, day length, light intensity, temperature, humidity, spacing, mutilation, and applications of growth regulators. Mauney and Ball (18) have provided evidence that the deciding factor in determining whether a bud develops into a vegetative or a fruiting branch is associated with physiological forces impinging upon it rather than to fundamental morphological differences in the buds. Their reports are consistent with our conclusion that the morphological nature of a branch may be altered biochemically by applications of methyl parathion. Reports (see "Review of Literature") of closely related compounds which have affected cotton and other plants also support this conclusion.

The delay in initiation of the reproductive stage shown in Tables 2 and 3 was reflected in the yields (Tables 6 and 7). Plots sprayed with methyl parathion or methyl parathion + DDT had less seed cotton at first picking (Table 7) than plots sprayed with toxaphene or DDT or toxaphene + DDT. It can be calculated from the data of Table 6 that plots sprayed with methyl parathion or methyl parathion + DDT required about 10 days longer than comparable plots sprayed with toxaphene or toxaphene + DDT to produce a given quantity of September seed cotton. There were no significant differences in total yield of seed cotton for any treatments.

Besides time of initiation of the first fruiting form, the rate of seed cotton production is also determined by the net rate of production and shedding of forms, by boll period and by seed cotton per boll (10). However, careful measurements carried out during the course of these experiments did not reveal any differences between treatments in the rate of accumulation of bolls after square initiation, nor in boll period, nor in boll size. Thus, the difference in time of initiation of the first fruiting branches and the resultant delay in square and boll production seems to account for nearly all of the observed differences in early seed cotton between plants treated with methyl parathion and plants not treated with methyl parathion. Brown et al. (5) reported that yields were reduced when cotton plants were sprayed with methyl parathion. Their first application of methyl parathion was made at the commencement of flowering in one experiment and at the appearance of the first squares in the other. Thus, their results seem to be at

Table 4—Number of thrips (*Frankliniella* spp., (primarily *fusca* Hinds)) per 80 plants from plots which received different treatments in 1960. Treatments in order of seasonal averages.

Treatment	May 27	May 31	June 6	June 10	Aver..
No insecticide	7	59a†	39a	58a	41a
Methyl parathion	6	21c	12b	10b	12b
DDT	3	28b	8c	8b	12b
Toxaphene	8	21c	6d	8b	11b
Methyl parathion + DDT	7	20c	10bc	4c	10b
Toxaphene + DDT	2	7d	3e	2d	4c
Anal. of variance	ns	**	***	***	***
Coeff. of variability	29	30	26	30	37

† Averages in the same column followed by the same letter do not differ significantly by Duncan's (6) multiple range test at the 5% level. ns - Not significant at 5% level.
, * Significant at 1% and 0.1% levels.

Table 5—Number of tarnished plant bugs (*Lygus lineolaris* (Palisot de Beauvois)) per 400 sweeps from plots which received different treatments in 1960. Treatments in order of seasonal averages.

Treatment	June 14	June 17	June 23	Aver.
No insecticide	7	13a†	32a	17a
DDT	3	4b	12b	6b
Toxaphene	6	5b	4b	5bc
Methyl parathion	2	3b	8b	4bc
Toxaphene + DDT	1	2b	6b	3bc
Methyl parathion + DDT	0	2b	3b	2c
Anal. of variance	ns	*	**	***
Coeff. of variability	43	31	32	35

† Averages in the same column followed by the same letter do not differ significantly by Duncan's (6) multiple range test at the 5% level. ns - Not significant at 5% level.
*, **, *** - Significant at 5%, 1%, and 0.1% levels.

Table 7—Precent of total yield of seed cotton picked on given dates after different treatments. Treatments in order of 2-year averages.

Treatment	Sept. 29, 1959	Sept. 21, 1960	1959-60 average
DDT	66a†	59a	63a
Toxaphene + DDT	60a	53ab	57a
Toxaphene	52b	45b	49b
No insecticide	42bc	47b	45bc
Methyl parathion + DDT	46bc	31c	39c
Methyl parathion	38c	31c	35c
Anal. of variance	**	***	***
Coeff. of variability	16	17	17

† Averages in the same column followed by the same letter do not differ significantly by Duncan's (6) multiple range test at the 5% level. **, *** Significant at the 1.0% and 0.1% levels.

Table 6—Pounds per acre of seed cotton picked on given dates after different treatments.

Treatment	1959					1960			Aver. total yield
	Sept. 9	Sept. 29	Oct. 27	Nov. 19	Total	Sept. 21	Nov. 14	Total	
Methyl parathion	317a	769	1265c	463b	2814	866ab	1866d	2732	2773
Toxaphene	762abc	845	1194bc	304ab	3105	1239cd	1535bcd	2774	2940
DDT	1010c	909	771a	231a	2920	1470d	1045a	2515	2718
Toxaphene + DDT	857bc	909	933ab	233a	2931	1190bcd	1179ab	2369	2650
Methyl parathion + DDT	444ab	907	1191bc	375ab	2916	782a	1802cd	2584	2750
No insecticide	416ab	754	1308c	269a	2746	1090abc	1379abc	2469	2608
Anal. of variance	*	ns	**	*	ns	**	**	ns	ns
Coeff. of variability	48	16	16	34	12	19	18	12	12

† Averages in the same column followed by the same letter do not differ significantly by Duncan's (6) multiple range test at the 5% level. ns - Not significant at 5% level.
*, ** Significant at 5% and 1% levels.

variance with our conclusion that the main effect of methyl parathion on maturity of cotton operates through its influence on the production of fruiting branches in response to applications made in the pre-square stage of growth. They did not find differences between treatments in percent of bolls and squares shed nor in lint percent, seed index, seeds per boll nor boll weight. The morphological nature of the delayed maturity of the plants they treated with methyl parathion is not clear from their report. Adkisson et al. (1) reported that methyl parathion had little or no effect on the maturity of cotton plants. However, their first application of insecticides was made after the cotton had begun to develop squares and their results are consistent with the hypothesis that the main effect of methyl parathion on the maturity of the cotton plant takes place at the pre-square stage of plant development.

No epinasty, leaf malformations or premature chlorosis were observed on the plants in these experiments at any time. However, certain other differences in plant response were noted between the treatments. The leaves of plants sprayed with methyl parathion or methyl parathion + DDT were greener in late July, August, and September than corresponding leaves of plants treated with toxaphene, DDT, or toxaphene + DDT. Plants sprayed with methyl parathion or methyl parathion + DDT were also taller at the end of the season than plants of other treatments. Apparently, differences in plant height were related to differences in internode length since there were no differences between treatments in the number of internodes on the main axis (Table 3). These differences are most simply interpreted as secondary effects associated with the delayed initiation of the reproductive stage when plants were sprayed methyl parathion.

Finally, consistent differences in growth or morphological characteristics or yield due to direct action of DDT or toxaphene or toxaphene + DDT on the cotton plant were not detected. Plants treated with these insecticides bore their first fruiting branch on the same node and had the same number of floral buds six weeks after planting as control plants which received no insecticides. It is apparent therefore, that under the conditions of these experiments the formulations of DDT and toxaphene had considerably less direct action on the cotton plant than the methyl parathion formulation.

SUMMARY

There was a delay in the initiation of fruiting branches and a corresponding delay in the production of floral buds when cotton plants were sprayed 8 times during their first 6 weeks of growth with a commercial formulation of methyl parathion. This response was not related to infestations of insects and it is suggested that it was due to direct action of methyl parathion on the metabolism of the cotton plant. Subsequently, after a total of 24 or 25 applications of methyl parathion, these same plants yielded fewer open bolls at first harvest. There were no differences between treatments in the total yield of seed cotton at final harvest. There were no obvious effects on plant metabolism from treatment with formulations of toxaphene, DDT, or toxaphene + DDT.

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