

### **III RESULTS AND DISCUSSION**

---

---

## A. Germination, Root Shoot Length and Phytotoxicity :

### 1. Germination

The effect of monocrotophos and monocrotophos in combination with bavistin on germination percentage root-shoot length and phytotoxicity studied in soybean variety MACS-13 is given in Table 1. The response of seed germination in both the cases of pesticidal treatments was inhibitory at higher concentrations while it was stimulatory at lower concentrations. The germination percentage of the seeds treated with monocrotophos + bavistin was comparatively less than that of non treated control seeds. However, the situation is altogether different in the seeds treated with monocrotophos alone which showed stimulation in germination percentage as compared with control at recommended and below recommended dose of monocrotophos, while it was greatly affected at above recommended dose of monocrotophos. ←

### 2. Root-shoot length :

Root length was largely affected as compared with control with increasing concentration of pesticides (Plate-19). Similarly shoot length was also hampered owing to pesticidal treatment. The maximum inhibition of shoot length being observed in the seeds treated with monocrotophos + bavistin. Among the shoot and root length, shoot length was much affected.

Table 1 : Effect of monocrotophos and monocrotophos in combination with bavistin on germination, root, shoot length and phytotoxicity in soybean.

Treatment	Germination *	Shot length <sup>@</sup> cm	Root length <sup>@</sup> cm	Root/shoot ratio	Phytotoxicity %
Control	80.00 ± 1.2	5.54 ± 1.2	5.54 ± 0.5	0.98	-
<u>Monocrotophos</u> M <sub>1</sub> (0.075% V/V)	81.00 ± 1.5	3.0 ± 0.5	2.90 ± 0.75	0.96	46.59 ± 5.1
M <sub>2</sub> (0.15% V/V)	83.20 ± 3.5	1.50 ± 0.2	1.70 ± 0.35	1.13	68.69 ± 3.9
M <sub>3</sub> (0.3% V/V)	25.00 ± 4.5	0.60 ± 0.03	1.40 ± 0.25	2.3	74.21 ± 2.87
<u>Monocrotophos<sup>#</sup> + Bavistin</u> M <sub>1</sub> +B <sub>1</sub> (0.034% W/W)	78.00 ± 2.00	2.10 ± 0.32	2.70 ± 0.70	1.28	50.27 ± 3.1
M <sub>2</sub> + B <sub>2</sub> (0.068% W/W)	75.00 ± 1.5	0.82 ± 0.05	1.66 ± 0.25	2.02	69.42 ± 3.0
M <sub>3</sub> +B <sub>3</sub> (0.136% W/W)	20.00 ± 2.5	0.50 ± 0.01	1.20 ± 0.20	2.4	77.90 ± 2.1

\* = After 48 h.      @ = after 120 h.      # = Concentrations are as above      ± = S.D.

Plate - 19 Effect of monocrotophos (a) and monocrotophos in combination with bavistin on root and shoot length in soybean.

C = Control

1 - Monocrotophos (0.075% v/v)

2 - Monocrotophos (0.15% v/v)

3 - Monocrotophos (0.30 % v/v)

4 - Monocrotophos + Bavistin (0.034% w/v)

5 - Monocrotophos + Bavistin (0.068% w/v)

6 - Monocrotophos + Bavistin (0.136% w/v)



a



b

PLATE - 19

Root-shoot ratio was found more in seeds treated with monocrotophos + bavistin than that of monocrotophos alone. This is because shoot length is much affected. The percent phytotoxicity studied in relation with radicle length clearly indicated that monocrotophos + bavistin treatment was more toxic than monocrotophos alone. This can clearly be seen from the Table 1.

The use of chemicals is in vogue since long as it is one of the potent tools in ensuring the eradication of the microflora associated within or outside the seed surface. The chemicals no doubt combat with the pathogenic and non pathogenic flora of the seed, but deviation from the concentration recommended or reactions of plant to particular chemicals some times lead to ill effect, which are pronounced either at the time of germination or at seedling stage. The ill effects observed during the present investigation are confined to the higher concentration of pesticides. The inhibition of germination due to pesticides such as Antracol and Kitazin in Brassica nigra. (Krishnamurthy and Rao, 1980), Kitazin in Dolichos biflorus (Reddy and Vidyavati, 1983), Endosulfan in Vigna radiata (Gupta et al. 1983), Carbaryl in Vigna sinensis (Sengupta et al. 1988), Thiodan-35 in Pea (Agrawal and Soam, 1988), and Dithane-M 45 in crop plants like Jowar, green gram, Bajra, Pea, Maize, Sunflower, Ragi etc. (Somashekhar and Sreenath, 1986) have been reported. Contrary to this the insecticide Sevin has found to be beneficial in promoting the root, shoot length in Vigna radiata with increasing concentrations (Pathak and Mukherji, 1986). However, high concentrations inhibited the growth of root and shoot. The treatments of Metasystox (a systemic insecticide) on seed germination of Vigna mungo

reported to be inhibitory by Prasad and Mathur (1983), while Phosphamidon (an organophosphorus insecticide) found to be stimulatory in action on seed germination of Hordeum vulgare (Singh et al. 1979).

According to Singh et al. (1979) generally insecticide treatments did not reduce germination but did reduce seedling height. Monocrotophos and monocrotophos in combination with bavistin did exhibited similar effect on root-shoot length in soybean of present investigation. Pardeshi et al. (1989). Studied effects of different fungicide on seedling vigour and seed viability in soybean. According to them the fungicide captan was found most effective in all varieties of soybean tested followed by sulphur, thiram and bavistin. Khare (1976) reported that fungicide treatment with bavistin and thiram increased germination and vigour in soybean. Saxena and Beg (1989) reported that low concentration of lindane did not exhibit any effect on the process of differentiation while higher concentration did exhibit inhibition of growth in Vigna unguiculata. Though insecticide and fungicide have been reported to be stimulatory in action at lower concentration as far as germination percentage and root-shoot ratio is concerned (Pardeshi et al. 1989, Patil and Shirashyad 1989), monocrotophos and monocrotophos in combination with bavistin inhibited root length which may be due to direct contact with pesticide. The reduction in shoot length may be as a result of transpiration of more pesticides to the meristematic zone of shoot apex through root system. However, to support this statement needs further investigation.

#### 4. Leaf area :

62

The leaf area expansion in response to foliar application of monocrotophos and monocrotophos in combination with bavistin is given in Table 2 and Plate 20. It is very clear from Table and Plate that recommended concentration (0.15%) of monocrotophos and monocrotophos in combination with bavistin (0.15% + 0.068%) favoured the leaf area expansion and as the concentration increased it showed reduction. Among these two treatments monocrotophos alone exhibited better results. Similar results were observed by Kulkarni (1989) in okra, guar and tomato under the influence of foliarly applied methyl-parathion and phosphamidon.

#### 5. Growth :

The shoot and root length of monocrotophos and monocrotophos in combination with bavistin treated plants is depicted in Table 3. It is vividly clear from the Table that lower concentrations of monocrotophos are stimulatory in action while all the concentrations of monocrotophos and monocrotophos in combination with bavistin were found inhibitory in action. Root length is also largely affected by monocrotophos and monocrotophos in combination with bavistin as compared with monocrotophos alone.

Somsekhar and Sreenath (1986) studied effects of carbamate and dithane M-45 on crop plants. Their results indicated that the germination percentage remain dose dependent while root and shoot growth adversely affected at higher dose but lower dose did not influence the growth of seedlings. The reduction in growth as a result of treatment was more in roots compared with the shoot. Further they reported that reduction in



Table 2 : Effect of foliar application of monocrotophos and monocrotophos in combination with bavistin on leaf area expansion in soybean (50 days old)

Treatment	Leaf area* (cm <sup>2</sup> )
Control	128.92 ±10.3
<u>Monocrotophos</u>	
M <sub>1</sub> (0.075% VN)	90.07 ± 8.2
M <sub>2</sub> (0.15% VN)	176.35 8.2
M <sub>3</sub> (0.3% VN)	87.19 ± 7.5
<u>Monocrotophos @ + Bavistin</u>	
M <sub>1</sub> +B <sub>1</sub> (0.034% WN)	115.23 ±3.2
M <sub>2</sub> +B <sub>2</sub> (0.068% WN)	164.45 ±3.9
M <sub>3</sub> +B <sub>3</sub> (0.136 w/v)	76.45 ± 2.5

@ = Concentrations are as above \* = Area of single 3rd and trifoliate leaf from the base. ± = S.D.

Plate - 20 Effect of foliar application of monocrotophos (a) and monocrotophos in combination with bavistin (b) on leaf area expansion in soybean (50 days old).

C = Control

1 - Monocrotophos (0.075% v/v)

2 - Monocrotophos (0.15% v/v)

3 - Monocrotophos (0.30 % v/v)

4 - Monocrotophos + Bavistin (0.034% w/v)

5 - Monocrotophos + Bavistin (0.068% w/v)

6 - Monocrotophos + Bavistin (0.136% w/v)

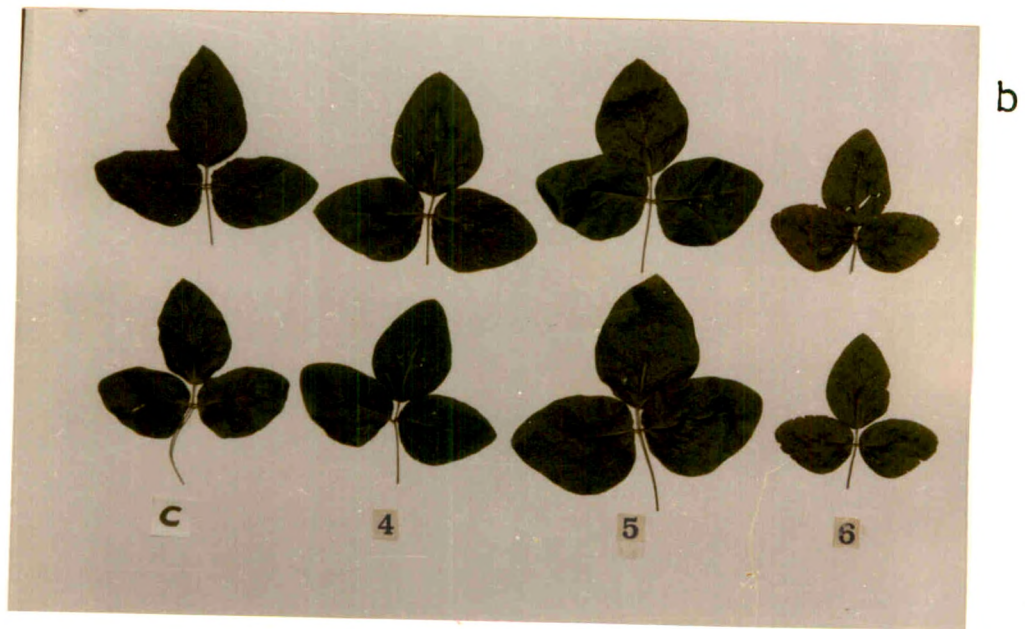
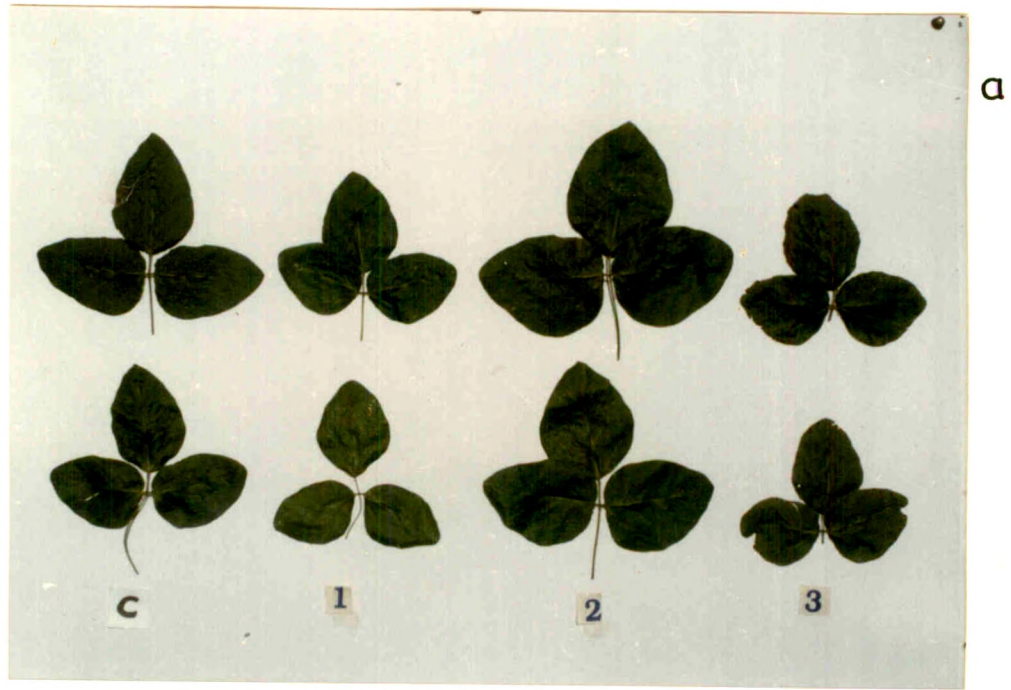


PLATE - 20

Table 3: Effect of foliar application of monocrotophos and monocrotophos in combination with bavistin on overall growth of soybean (50 days old).

Treatment	Shoot length cm	Root length cm	Root/shoot ratio	Root nodule/plant	Nodule diameter mm	Leaves/Plant	Pods/plant
Control	43.42 ± 2.16	18.44 ± 1.18	0.42	28.2 ± 2.78	2.14 ± 0.33	5.2 ± 1.44	14.6 ± 5.95
<u>Monocrotophos</u>							
M <sub>1</sub> (0.075% VN)	52.28 ± 1.58	15.85 ± 0.73	0.30	26.8 ± 2.31	2.32 ± 0.34	7.2 ± 0.97	31.00 ± 3.09
M <sub>2</sub> (0.15% VN)	54.30 ± 2.69	20.26 ± 1.08	0.37	37.2 ± 5.26	2.46 ± 0.18	9.8 ± 0.97	36.6 ± 3.32
M <sub>3</sub> (0.3% VN)	44.02 ± 1.25	11.48 ± 0.65	0.26	24.6 ± 2.72	1.92 ± 0.42	8.0 ± 1.09	21.4 ± 8.3
<u>Monocrotophos * + Bavistin</u>							
M <sub>1</sub> +B <sub>1</sub> (0.034% W/W)	34.00 ± 1.78	12.17 ± 3.26	0.36	5.4 ± 3.26	0.34 ± 0.048	6.8 ± 1.6	3.50 ± 0.60
M <sub>2</sub> +B <sub>2</sub> (0.068% W/W)	31.01 ± 2.00	6.8 ± 0.79	0.22	4.0 ± 1.4	0.36 ± 0.12	5.0 ± 0.66	3.75 ± 0.82
M <sub>3</sub> +B <sub>3</sub> (0.136 w/v)	30.00 ± 1.58	9.25 ± 0.76	0.31	1.8 ± 0.83	0.22 ± 0.04	4.6 ± 0.48	1.75 ± 0.43

\* = Concentration are as above. ± S.D.

epicotyl and hypocotyl growth can be attributed to an anomalous behaviour of plumle. This clearly suggests that the response of pesticide varies with plant to plant.

Nodule formation, nodule diameter, leaves per plant and pods per plant were greatly influenced by the foliar application of 0.15% monocrotophos. The values of these parameters are comparatively more than that of control values. Contrary to this the monocrotophos and monocrotophos in combination with bavistin exhibited inhibitory action Singh et al. (1991) studied effects of insecticide on nodulation in soybean cv. Gaurav. They noted that nodule number at 60 days was highest after treatment with 20 g carbosulfon/kg seeds and lowest after seed treatment with monocrotophos. In the present investigation the nodule number was increased in the soybean cv MACS-13 when monocrotophos (0.15% v/v) was applied foliarly. From the data it appears that foliar application of monocrotophos did favour nodulation in soybean than soil application.

#### 6. Biomass :

Biomass in terms of percentage dry weight in leaf, stem and root of soybean was increased over control due to lower concentration of monocrotophos, while the plants sprayed with monocrotophos in combination with bavistin did not exhibit much increase in biomass over control as compared with monocrotophos treatment (Table 4). However, the higher concentrations of both the treatments showed detrimental effect.

The foregoing discussion lead us to surmise that the monocrotophos at below recommended and recommended dose favour the overall growth

Table 4 : Effect of foliar application of monocrotophos in combination with bavistin on dry weight in soybean (50 days old)

Treatment	BIOMASS (g) (% dry wt.)		
	Leaf	Stem	Root
Control	33.00	32.00	31.00
<u>Monocrotophos</u> M <sub>1</sub> (0.075% v/v)	57.00	61.00	62.00
M <sub>2</sub> (0.15% v/v)	52.00	49.00	51.00
M <sub>3</sub> (0.3% v/v)	47.00	32.00	41.00
<u>Monocrotophos * + Bavistin</u> M <sub>1</sub> +B <sub>1</sub> (0.034% w/v)	41.00	38.00	36.00
M <sub>2</sub> + B <sub>2</sub> (0.068% w/v)	42.00	29.00	31.00
M <sub>3</sub> +B <sub>3</sub> (0.136 w/v)	37.00	22.00	27.00

\* Concentrations are as above

The values are mean of three determinations.

Dry weight is difference between 1 gm fresh wt. kept in oven at 60°C for 48 h.

of soybean but its combination with bavistin even at recommended and below recommended dose did not favour to influence the growth in soybean and hence the compatibility is not suitable as far as soybean crop is concerned.

#### B. Stomatal regulation :

Large number of pesticides are used in agriculture for the protection of crop plants against diverse pests. The residues left over the sprayed surfaces of crop plants have become a matter of concern with respect to health hazards to men and animals and environmental pollution (Finiayson and Mc Carthy, 1973). The residual effects of these pesticides has been known to remain in the environment for a long time (Sharma and Chopra 1970, Deshpande and Swamy 1987); residue level of any pesticide may affect the plant growth, stomatal regulation and metabolism. Since stomata play pivotal role in gaseous diffusion process, the effect of these insecticides as a spray on stomatal regulation in soybean was studied.

The response of diffusive resistance and diffusive conductance for water vapour and CO<sub>2</sub> and transpiration rate in the leaves of soybean sprayed with different concentration of monocrotophos and monocrotophos in combination with bavistin with respect to days after foliar application is shown in Table 5 and 6. It is very clear from Tables that the stomata are present on both the surfaces, the maximum being on lower surface which is the indicative of the fact that the soybean leaves are amphistomatous in nature and transpire maximum through lower surface. The effect of monocrotophos on stomatal regulation was not changed much when the plants were sprayed with 0.075% and 0.15% (v/v).

Table : 5. Effect of foliar application of monocrotophos on stomatal regulation in soybean.

*under Repression*

Treatment	Parameters	Days after foliar application								
		I	II	III	IV	V	VI	VII	VIII	
Control	DR.H <sub>2</sub> O	L	0.96	0.94	0.96	0.96	0.96	1.08	1.14	1.14
		U	1.78	1.5	2.07	2.07	1.59	1.96	1.22	2.31
	DC.H <sub>2</sub> O	L	1.04	1.06	1.04	1.04	1.04	0.92	0.87	0.90
		U	0.56	0.66	0.48	0.48	0.63	0.51	0.81	0.43
	TR.H <sub>2</sub> O	L	17.44	17.63	16.87	16.87	13.49	15.67	19.60	12.51
		U	9.99	11.83	8.75	8.75	8.71	9.13	12.88	6.66
DR.CO <sub>2</sub>		0.47	0.25	0.20	0.20	0.24	0.24	0.23	0.18	
	DO.CO <sub>2</sub>	2.13	3.93	4.87	4.87	4.09	4.88	4.81	5.55	
Monocrotophos M <sub>1</sub> (0.075% V/V)	DR.H <sub>2</sub> O	L	0.9	0.94	1.02	1.02	0.95	1.04	1.05	1.99
		U	1.29	1.37	1.60	1.60	1.48	1.44	1.67	3.66
	DC.H <sub>2</sub> O	L	1.11	1.06	0.98	0.98	1.05	0.96	0.95	0.50
		U	0.78	0.72	9.62	9.62	0.67	0.69	0.59	0.27
	TR.H <sub>2</sub> O	L	13.58	18.42	16.83	16.83	14.40	16.53	20.71	7.37
		U	12.98	13.33	11.47	11.47	9.46	12.41	13.50	4.26
DR.CO <sub>2</sub>		0.18	0.34	0.20	0.20	0.25	0.25	0.22	0.12	
	DO.CO <sub>2</sub>	2.11	2.90	4.20	4.20	3.90	3.98	4.39	9.14	



Table : 5 (contd...)

Treatment	Parameters	Days after foliar application							
		I	II	III	IV	V	VI	VII	VIII
M <sub>2</sub> (0.15 % V/V)	DR.H <sub>2</sub> O	1.17	1.33	0.97	1.12	1.01	1.09	1.04	1.79
		2.63	1.18	1.61	2.39	1.81	1.9	2.49	3.00
	DC.H <sub>2</sub> O	0.85	0.75	1.03	0.89	0.99	0.91	0.96	0.55
		0.38	0.46	0.61	0.41	0.55	0.52	0.40	0.33
	TR.H <sub>2</sub> O	13.37	12.98	18.31	15.94	13.46	15.95	20.36	8.28
		6.37	8.35	11.78	8.10	8.03	9.66	9.44	5.07
M <sub>3</sub> (0.3 % V/V)	DR.CO <sub>2</sub>	0.16	0.17	0.23	0.17	0.22	0.20	0.17	0.12
	DO.CO <sub>2</sub>	6.09	5.61	4.18	5.63	4.52	4.83	5.68	7.70
	DR.H <sub>2</sub> O	1.85	0.94	1.02	1.18	1.1	1.08	1.12	1.96
		4.60	2.15	2.03	1.84	2.3	2.19	2.39	2.19
	DC.H <sub>2</sub> O	0.54	1.06	0.98	0.84	0.91	0.92	0.89	0.51
		0.21	0.46	0.49	0.54	0.43	0.45	0.41	0.45
M <sub>3</sub> (0.3 % V/V)	TR.H <sub>2</sub> O	8.98	17.80	18.18	15.22	12.80	16.17	19.4	7.69
		3.82	8.48	9.84	10.29	6.43	8.57	9.92	6.78
M <sub>3</sub> (0.3 % V/V)	DR.CO <sub>2</sub>	0.94	0.20	0.20	0.17	0.18	0.18	0.17	0.14
	DO.CO <sub>2</sub>	10.6	4.99	4.91	5.84	5.45	5.34	5.81	6.70

DR : Diffusive resistance ( s cm<sup>-1</sup>) for water, DC : Diffusive conductance ( c ms<sup>-1</sup>) for water, TR : Transpiration rate (µg cm<sup>-2</sup>s<sup>-1</sup>)  
 Experimental conditions : Average light intensity 1200.00 µE m<sup>-2</sup>s<sup>-1</sup>, Average relative humidity : 48.00 %  
 Leaf area exposed : 1 cm<sup>2</sup>, The values are mean of three determinations.

Table : 6. Effect of foliar application of monocrotophos in combination with bavistin on stomatal regulation in soybean.

Treatment	Parameters	Days after foliar application							
		I	II	III	IV	V	VI	VII	
Control	DR.H <sub>2</sub> O	L	0.96	0.94	0.96	0.96	0.96	1.08	1.14
		U	1.78	1.5	2.07	2.07	1.59	1.96	1.22
	DC.H <sub>2</sub> O	L	1.04	1.06	1.04	1.04	1.04	0.92	0.87
		U	0.56	0.66	0.48	0.48	0.63	0.51	0.81
	TR.H <sub>2</sub> O	L	13.70	17.44	17.63	16.87	13.49	15.67	16.90
		U	6.30	9.99	11.83	8.75	8.71	9.13	12.88
Monocrotophos + Bavistin M <sub>1</sub> +B <sub>1</sub> (0.034 w/v)	DR.CO <sub>2</sub>	L	0.47	0.23	0.25	0.20	0.24	0.24	0.23
		U	2.13	4.39	3.93	4.87	4.09	4.88	4.81
	DR.H <sub>2</sub> O	L	1.90	1.95	3.41	2.99	3.23	2.08	7.11
		U	6.66	7.13	19.60	44.40	16.10	12.60	10.22
	DC.H <sub>2</sub> O	L	0.52	0.51	0.29	0.22	0.30	0.48	0.140
		U	0.15	0.14	0.068	0.022	0.002	0.079	0.098
TR.H <sub>2</sub> O	L	11.42	7.8	4.97	5.87	5.37	7.93	2.79	
	U	3.44	2.38	0.94	0.449	1.17	1.39	2.03	
DR.CO <sub>2</sub>	L	0.072	0.068	0.034	0.012	0.032	0.042	0.035	
	U	13.77	14.60	29.11	77.81	31.21	23.64	27.83	

Table : 6 (contd..)

Treatment	Parameters	Days after foliar application						
		I	II	III	IV	V	VI	VII
M <sub>2</sub> + B <sub>2</sub> (0.068 w/v)	DR.H <sub>2</sub> O	2.07 10.10	1.68 7.98	5.07 17.60	3.34 55.00	2.44 15.40	2.02 12.90	1.97 13.20
	DC.H <sub>2</sub> O	0.53 0.99	0.48 0.12	0.19 0.056	0.29 0.018	0.40 0.064	0.49 0.077	0.50 0.075
	TR.H <sub>2</sub> O	8.64 1.72	10.67 2.94	3.62 1.10	5.7 0.38	7.52 1.27	6.38 1.38	10.07 1.60
	DR.CO <sub>2</sub>	0.0520	0.060	0.026	0.010	0.058	0.041	0.040
	DO.CO <sub>2</sub>	19.22	16.65	37.09	94.67	17.22	24.10	24.60
	DR.H <sub>2</sub> O	2.3 5.94	2.66 8.47	5.0 10.80	2.6 25.0	2.89 18.20	1.68 5.87	2.64 1.4
M <sub>3</sub> + B <sub>3</sub> (0.136 w/v)	DC.H <sub>2</sub> O	0.43 0.16	0.37 0.11	0.2 0.092	0.38 0.04	0.34 0.084	0.59 0.170	0.37 0.71
	TR.H <sub>2</sub> O	9.73 3.98	6.45 2.10	3.87 1.90	7.82 0.89	6.77 1.15	10.07 3.14	7.66 1.56
	DR.CO <sub>2</sub>	0.072	0.052	0.102	0.022	0.029	0.082	0.15
	DO.CO <sub>2</sub>	13.75	18.92	9.76	44.34	34.42	12.14	6.58

DR : Diffusive resistance ( s cm<sup>-1</sup>) for water, DC : Diffusive conductance ( c ms<sup>-1</sup>) for water, TR : Transpiration rate ( µg cm<sup>-2</sup>s<sup>-1</sup>)  
 Experimental conditions : Average light intensity 1200.00 µE m<sup>-2</sup>s<sup>-1</sup>, Average relative humidity : 48.00 %  
 Leaf area exposed : 1 cm<sup>2</sup>; The values are mean of three determinations.

However, at higher concentration of monocrotophos (0.3% V/V) the stomatal regulation was considerably affected. The diffusive resistance for water vapour was increased as compared with the values of control and plants sprayed with lower concentration of monocrotophos. Contrary to this the stomatal regulation was greatly affected followed by foliar application of different concentration of monocrotophos in combination with bavistin which can be clearly seen from Table 6. It is interesting to note here that the diffusive resistance for water vapour was tremendously increased at all the concentration of the pesticide sprayed as compared with control. The increase in diffusive resistance checked the transpiration rate.

The graphical representation of diffusive resistance for water vapour and transpiration rate studied in both the surfaces of soybean leaves with respect to different concentrations of monocrotophos is shown in Fig.1. It is very clear from the figure that as the concentration increases the diffusive resistance increases and transpiration rate gets declined (Fig.1a). The above situation was more or less the same in case of lower surface of the leaves. The only difference is that the lower surface transpire more by keeping the diffusive resistance value at lower ebb as compared with the upper surface (Fig. 1b). Contrary to this the diffusive resistance and transpiration rates were adversely affected due to foliar application of monocrotophos in combination with bavistin (Fig. 2a).

The diffusive conductance and resistance for CO<sub>2</sub> monitored after foliar application of pesticide is graphically represented in Fig.3a and b. In case of monocrotophos sprayed plants the CO<sub>2</sub> resistance was decreased with increasing concentration of monocrotophos while CO<sub>2</sub> conductance

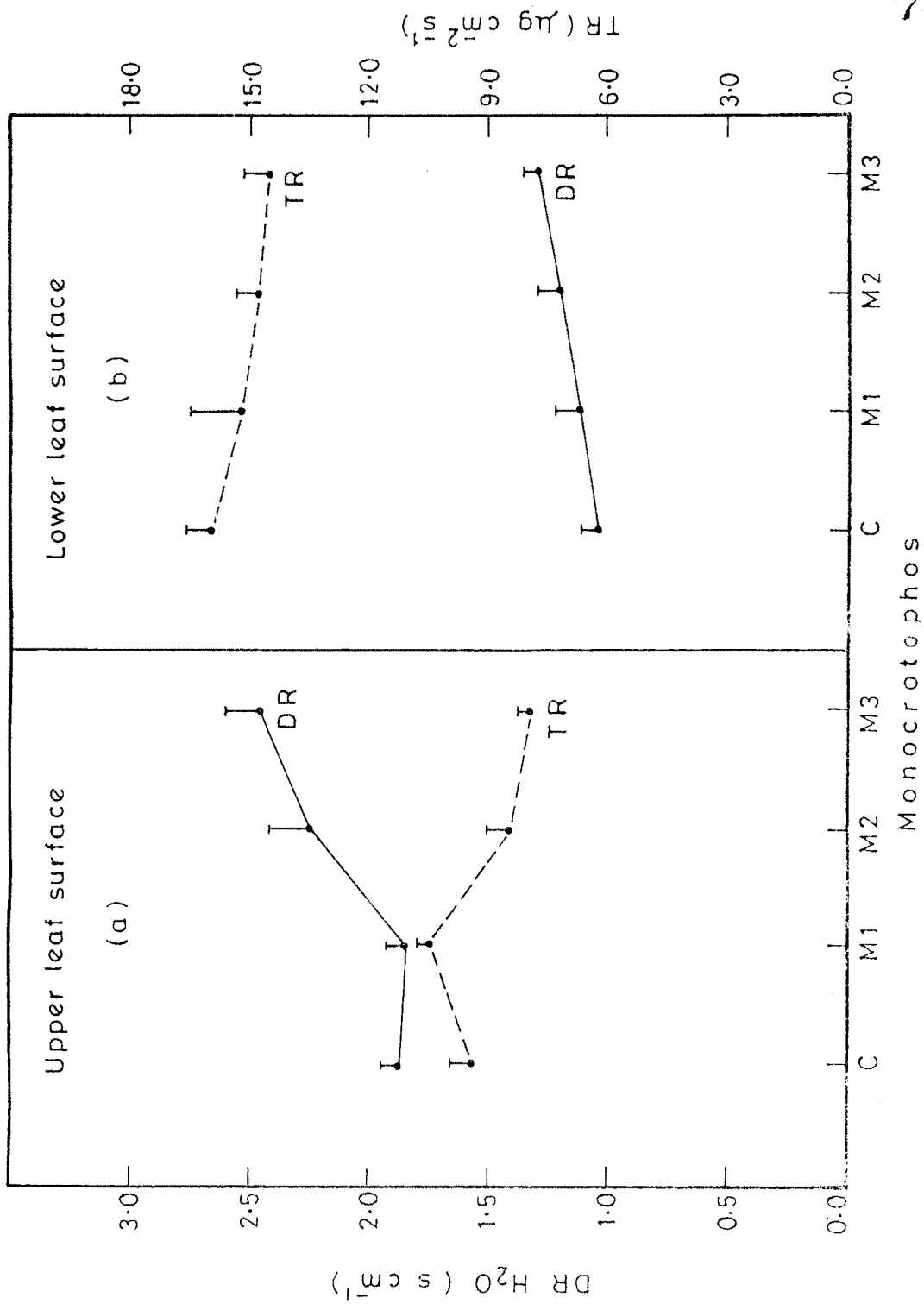


Fig. 1

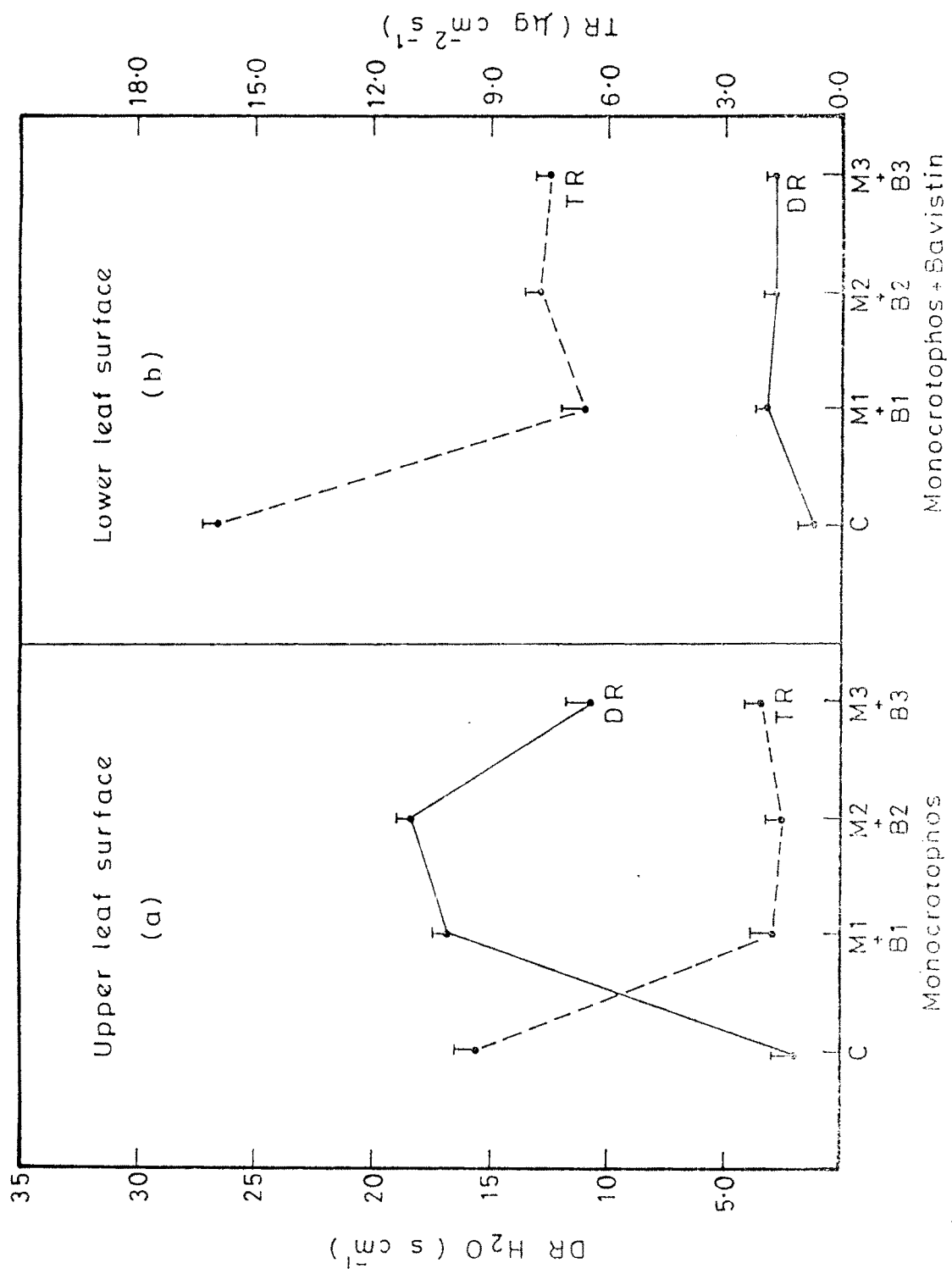


Fig. 2

was increased (Fig.3a). Whereas resistance for CO<sub>2</sub> increased tremendously at higher concentration in the plants sprayed with monocrotophos in combination with bavistin and conductance for CO<sub>2</sub> was declined (Fig.3b). However, both diffusive resistance and conductance for CO<sub>2</sub> showed immediate response to pesticide that is the resistance for CO<sub>2</sub> sharply declined followed by foliar application of monocrotophos in combination with bavistin, diffusive conductance for CO<sub>2</sub> was sharply increased, both these parameters remained more or less the same at recommended dose of pesticide application and at above recommended dose, resistance was increased while conductance was declined. Highest value for CO<sub>2</sub> resistance at high dose of pesticide clearly indicates that the concentrations above recommended doses affect the stomatal regulation and possibly develop constraint in the gaseous diffusion process. This can be correlated with residual level left over the sprayed surfaces. As such the residual content of organophosphorus pesticides are known to remain in the environment for a long time (Attri and Rattan 1974, Rajukkannu *et al.* 1976, Deshpande and Swamy 1987, Patil and Kulkarni 1989).

Based on the information obtained from studies on stomatal physiology different chemicals were used to regulate the water requirement of plants. The closing of stomata reduces the water requirement of plants while enhance stomatal opening on the other hand leads to increased transpiration and desiccation (Das 1977). Paraquat and 2,4,5-T are some examples of herbicides which kill the plants by enhancing transpirational water loss by forced opening of the stomata (Rao *et al.* 1977, Patil *et al.* 1991). Stomatal behaviour also formed the basis of sensitivity of weeds to herbicides. Thiocarbamates (EPTC) and mollinate inhibit transpiration in C<sub>4</sub>

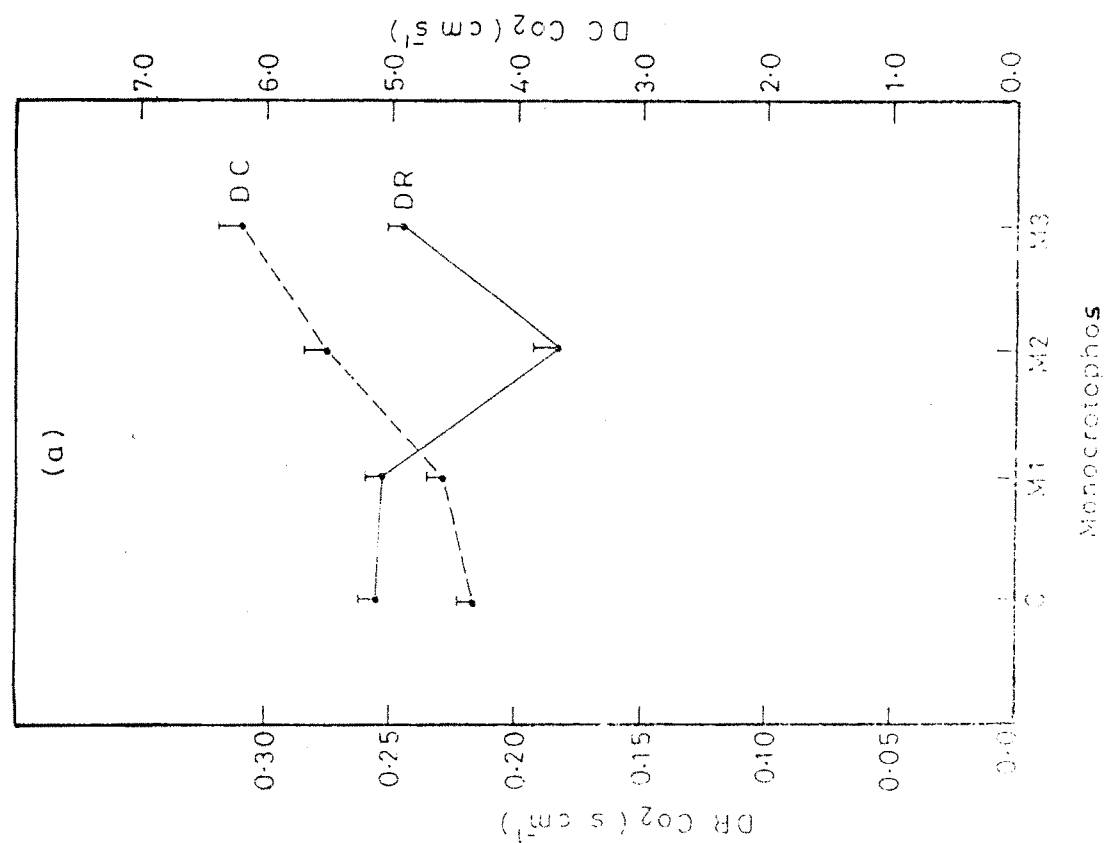
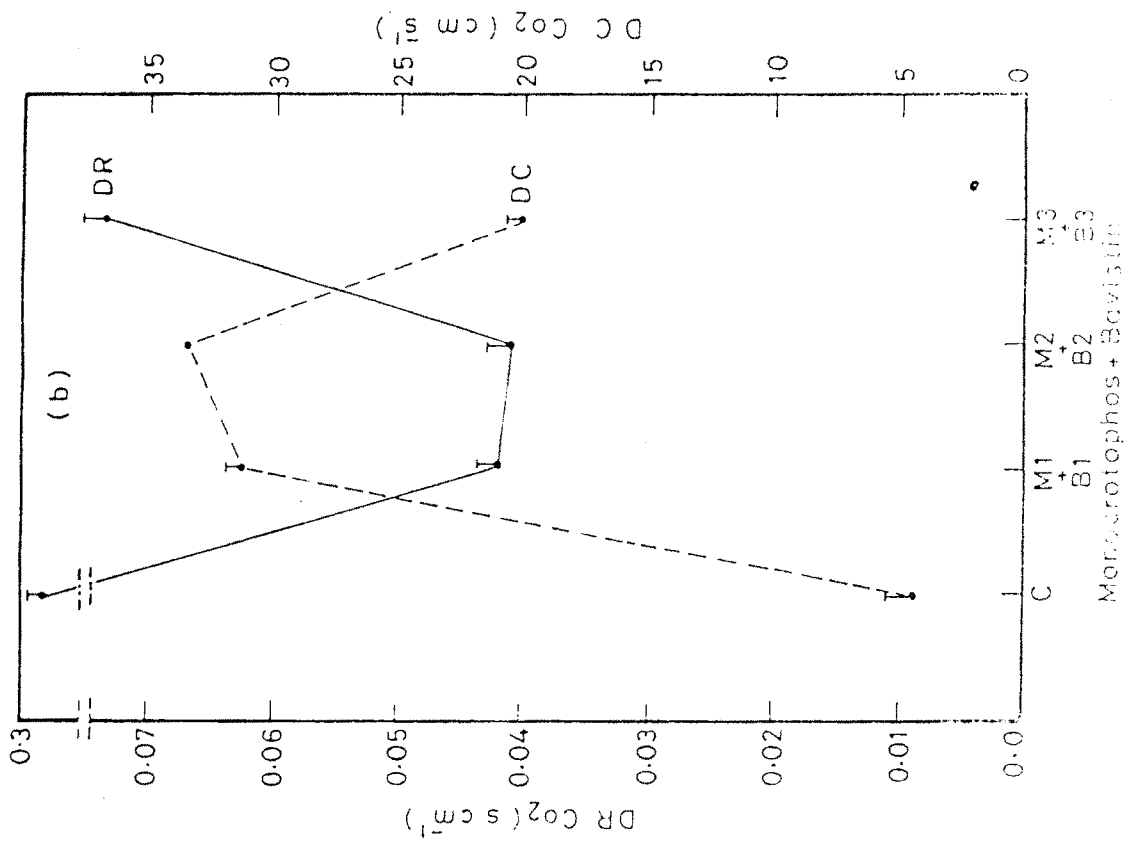


Fig. 3



crop plants and results in their better water management. The transpiration in  $C_3$  weeds was found to be enhanced by thiocarbamates leading to their desiccation and death (Das and Santakumari, 1975). Thus under crop weed association, thiocarbamate compounds are extremely useful in promoting growth of plants alone.

The compounds which reduce transpiration and improve water efficiency of plants are known as antitranspirants (Das and Raghavendra 1979, Patil 1995). Several herbicides such as alachlor, butachlor reduce transpiration by restricting stomatal opening. Alachlor even improved the yield of maize plants inspite of reduction in transpiration (Santakumari et al. 1977). In soybean stomata remain more widely open in the morning and more closely shut at mid day in the fusicoccin sprayed plants than in the control plants (Stoinova and Lilovd 1991). All these above observations hold good for insecticidal spray, which leads stomatal closure. Apart from this the maximum increase in diffusive resistance was noticed at higher concentration of insecticidal sprays, because of which transpiration rate was arrested. However, at lower concentration, though the transpiration rate was arrested initially, it was corrected later on by the plants and the overall growth under lower concentration of monocrotophos was found good. None of the concentrations of monocrotophos in combination with bavistin showed good growth as compared with the control plants which indicates that this compatibility does not suit for the soybean.

Thus from agronomic point of view the data convey that the monocrotophos should always be used at, or below recommended concentrations. Little ignorance, may seriously affect the plant metabolism and would cause environmental pollution by persisting residual problem.

## C. Organic constituents :

### 1. Chlorophylls :

The data on chlorophyll content of soybean foliage under the influence of foliar application of monocrotophos and monocrotophos in combination with bavistin is given in Table-7. and represented histographically in Fig.4. The data clearly indicates that lower concentration of monocrotophos stimulates chlorophyll synthesis while inhibits at higher concentration. On the other hand the soybean plants sprayed with monocrotophos in combination with bavistin showed drastic reduction at all the concentrations employed. This is very clear from Fig.4 in which the level of total chlorophylls represented histographically with respect to different concentrations of pesticides employed in soybean for foliar application.

### 2. Carotenoids :

The carotenoids are located in the chloroplasts and the chromatophores as water insoluble protein complexes. The specific orientation of the carotenoids in relation to the chlorophylls within lamellar system of chloroplasts must be an important aspect of photosynthetic process (Devlin and Witham 1986). The carotenoids act as accessory light harvesting pigments, absorbing light and passing the excitation energy on to the antenna chlorophylls. They also protect the chlorophylls against photooxidation. In order to know the fate of carotenoid content under the influence of pesticidal application, it was studied in the present investigation and the values of carotenoids are incorporated in Table-7. It is

Table 7 : Effect of foliar application of monocrotophos and monocrotophos in combination with bavistin on chlorophyll and carotenoid content in the leaves of soybean (50 days old).

Treatment	Chlorophyll (mg 100 <sup>-1</sup> g fresh tissue)			Carotenoids
	Chl. a	Chl. b	Total Chl.	
Control	338.00 ± 10.2	98.8 ± 5.2	436.9 ± 7.0	252.0 ± 6.5
<u>Monocrotophos</u>				
M <sub>1</sub> (0.075% v/v)	332.50 ± 11.2	112.2 ± 5.0	444.7 ± 6.0	262.00 ± 2.2
M <sub>2</sub> (0.15% v/v)	360.1 ± 7.9	122.1 ± 5.0	482.2 ± 5.3	258.00 ± 3.5
M <sub>3</sub> (0.3% v/v)	182.1 ± 10.20	125.7 ± 4.3	300.66 ± 5.2	153.00 ± 1.2
<u>Monocrotophos * + Bavistin</u>				
M <sub>1</sub> +B <sub>1</sub> (0.034% w/v)	183.4 ± 6.5	125.32 ± 2.5	308.46 ± 5.4	160.00 ± 3.5
M <sub>2</sub> +B <sub>2</sub> (0.068% w/v)	171.5 ± 4.5	123.00 ± 4.5	294.4 ± 5.4	156.00 ± 1.9
M <sub>3</sub> +B <sub>3</sub> (0.136 w/v)	131.0 ± 7.5	105.1 ± 10.6	236.00 ± 3.2	120.00 ± 6.0

\* = Concentrations are as above. ± = S.D.

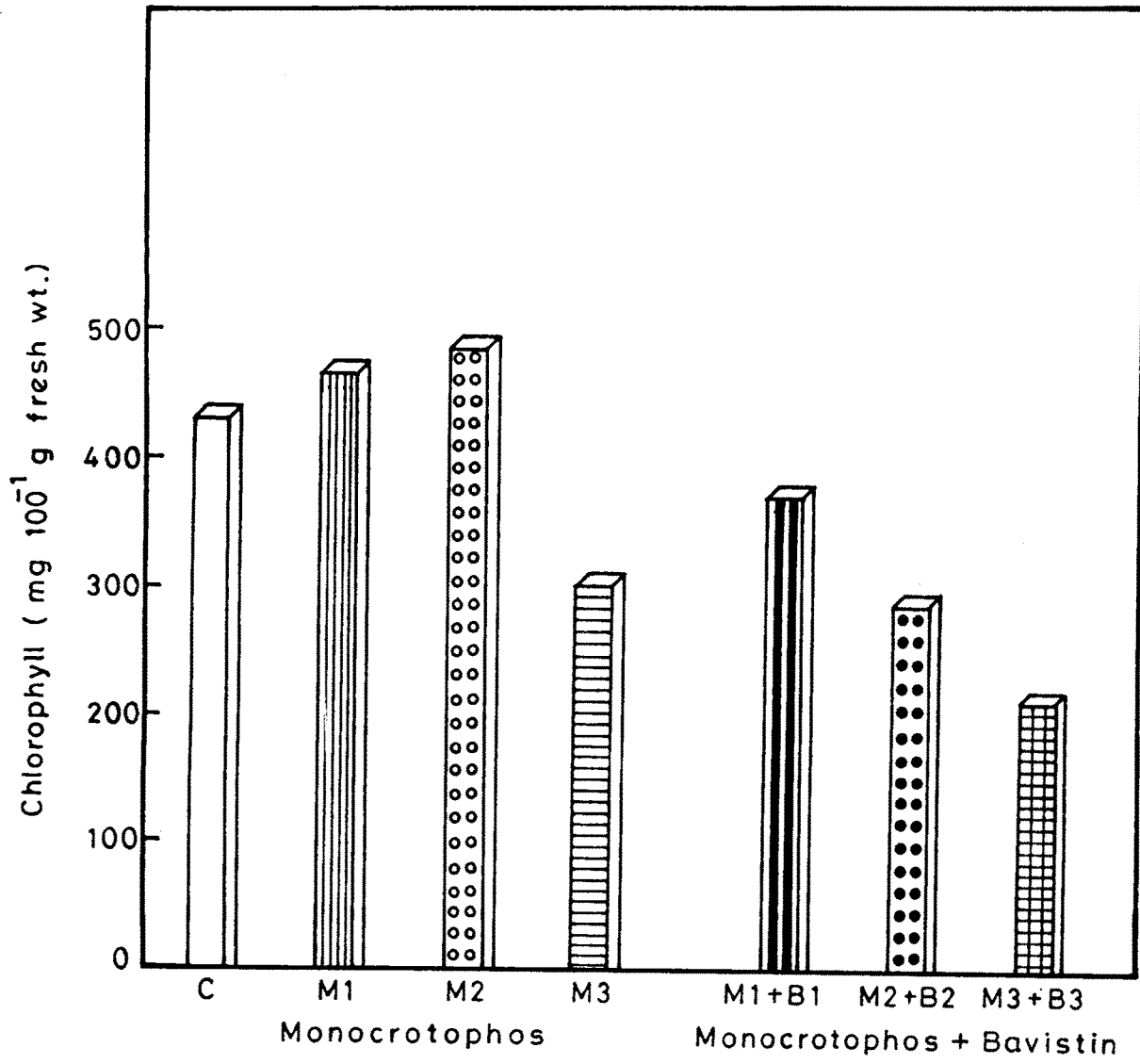


FIG.4

vividly clear from the Table that, the carotenoid content not much affected at lower concentration of monocrotophos but it did affected at higher concentration by about 39% than that of control. However, the carotenoid level was largely affected at all the concentration of monocrotophos in combination with bavistin. None of the concentration was favoured the stimulation of carotenoid. As high as more than 50% reduction in carotenoid content was noticed at above recommended dose employed foliarly on soybean.

The organophosphorus pesticides namely metacid-50 and dimecron when applied foliarly on tomato, okra and guar leaves exhibited increase in chlorophyll content at recommended and below recommended doses (Kulkarni 1989). Sukul and Handa (1989) also reported increase in total chlorophyll content in chickpea leaves followed by foliar application of synthetic pyrethroids. Most of the herbicides when applied foliarly, exhibited inhibition of chlorophyll synthesis (Nandihalli and Bhowmik 1992, EL Sheekh et al. 1994, Patil et al. 1991).

The universal function of carotenoids is the protection of chlorophyll molecules against photodynamic damage caused by singlet oxygen, armed by the transfer of energy from an excited photosensitizer such as chlorophyll. Carotenoids also act as component of photosystem I and II in the light reaction of photosynthesis. Possibly the reduction in chlorophyll content in case of soybean treated with monocrotophos in combination with bavistin is due to inhibition of carotenoid biosynthesis.

From the above results one thing is clear that lower concentrations of monocrotophos stimulate chlorophyll synthesis while higher

concentration inhibit it. The compatibility of monocrotophos with bavistin appear to hinder metabolic activities by entering into tissue system symplasticly or apoplasticly.

### 3. Polyphenols :

Polyphenols are the secondary metabolites which play an important role in disease resistance. The polyphenol content studied in soybean leaves under the influence of pesticidal application depicted ~~histographically~~ in Fig 5. It is very clear from the figure that the polyphenol level increased by 19.54 and 22.55% over control due to foliar application of monocrotophos at 0.075% and 0.15% respectively, while declined by 35.33% at higher concentration. On the other hand the application of monocrotophos in combination with bavistin did not show any increase in polyphenol level over control.

hist. graph  
there is  
no level  
up  
No increase

Early work by Reilly and Klarman (1972) demonstrated that several fungicides can induce production of the phytoalexin hydroxyphaseollin in soybeans. They found manab, benomyl and nabam to stimulate relatively high level of this phenolic phytoalexin. Amine decomposition products of these fungicides were also stimulatory. This study did not, however, establish a link between these effects and the mechanism of action of these fungicides. In the present investigation, however, bavistin being a fungicide did not favoured stimulation of polyphenol in soybean. Perhaps its compatibility with monocrotophos might have affected the phenol biosynthetic pathway.

Similarly little is known of indirect effect of insecticides on plants. No doubt, this is partly because of a lack of interest or expertise among

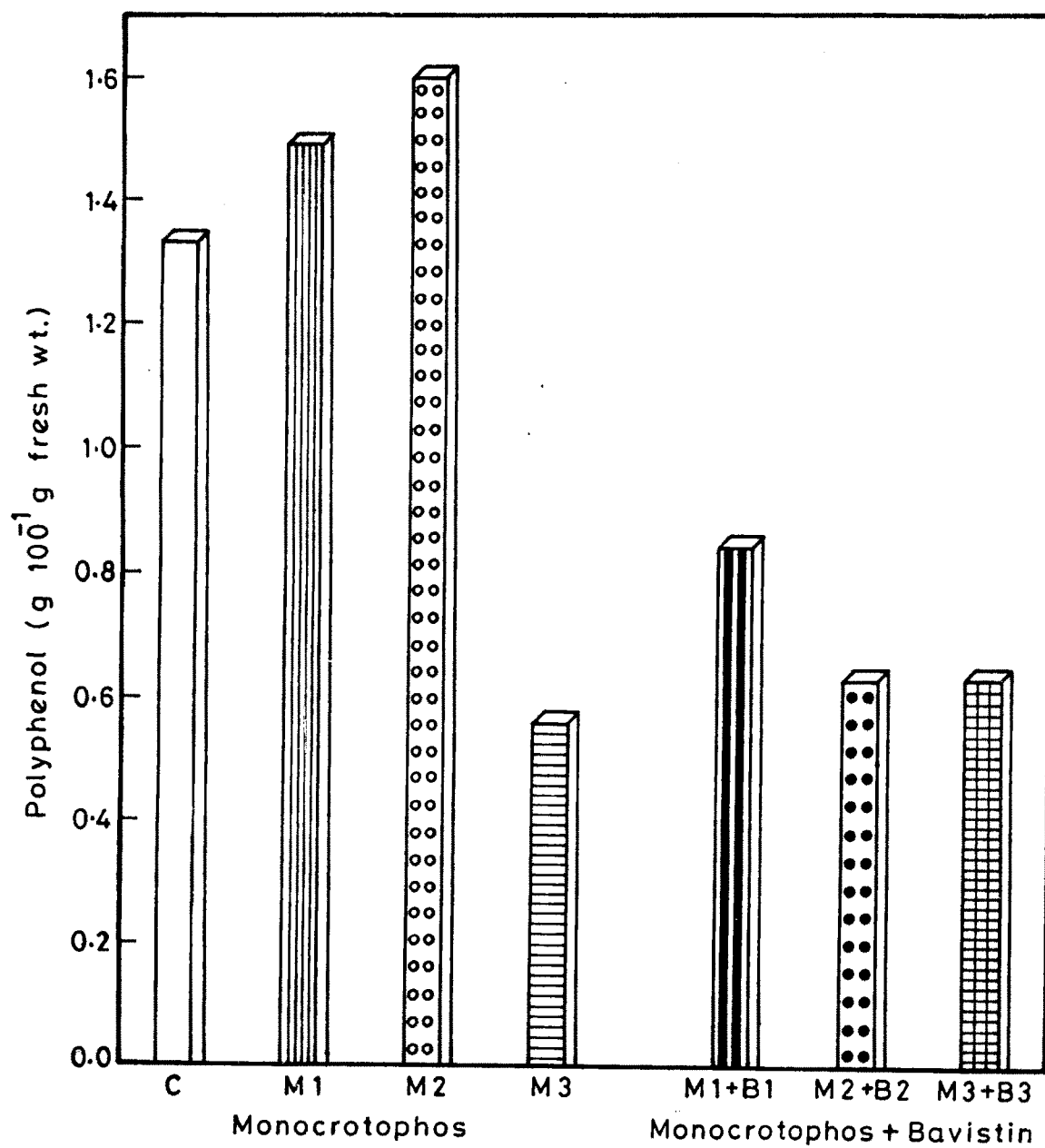


FIG. 5

entomologist in plant physiology and vice versa (Lydon and Duke, 1989). Although one might not expect insecticides to be phytotoxic, at least one pesticide, chlorinated camphor, has been registered for use as both a herbicide and a insecticide (Sherman et al. 1983, Green et al. 1987). However, the mechanism of action of this mixture of compounds on plants was never determined. The only report of an effect of an insecticide on secondary metabolism that has been found was that of methomyl on the phenolic compounds of cotton (Parrott et al. 1983). It caused almost three-fold increase in cyanidin-3-glycoside and a 50% increase in the content of tannin in mature leaves. In the present investigation foliar application of monocrotophos on soybean also exhibited increase in polyphenol content.

Clearly, an understanding of the effects of pesticides on the secondary metabolism of the plants is important. Alteration of secondary metabolism can dramatically change plant responses to biotic stress viz. pathogens, insects, nematodes and other plants and to stress imposed by the physical environment. Kulkarni (1989) observed that methylparathion and phosphamidon used for spray on tomato, okara and guar favoured polyphenol synthesis. More over lower concentrations of both the insecticides worked good in case of tomato plant in stimulating polyphenol content. Stimulation of polyphenols by lower concentrations due to the insecticides may minimise fungal attack too or in other words the plant become resistant to pathogen attack (Wang 1961). However, it needs further study to elucidate plant pathogen and polyphenol relationship under the influence of insecticidal spray. At present our knowledge of such effects is extremely limited. But a greatly expanding literature on the mechanism of action of pesticides and on the biological and physiological



roles of secondary products will provide a better understanding of these relationships.

#### 4. Nitrogen, Protein and Amino acids :

##### i) Nitrogen and protein :

Nitrogen plays an important role in crop plants for it is linked with the productivity. All the breeding programmes of the recent years are oriented to the nitrogen geared productivity. Pesticides affect the nitrogen metabolism in plants through effects on nitrogen fixation, either by effects on the plant or on the bacterium (Pozuelo et al., 1989). In most cases, the evidence indicates that reductions in nodulation and/or fixation by existing nodules are due to effects on the plant rather than on the bacterium (Goring and Laskowski, 1982). In some cases, herbicides have been reported to increase nitrogen fixation. For example, cyanazine, simazine have been shown to increase acetylene reduction activity (=nitrogenase) in Lupinus albus. Similarly, in the last few years, it has been witnessed unprecedented progress in the area of pesticide mode of action. Nowhere has the progress been as rapid as with herbicides that inhibit branched chain amino acid biosynthesis (Stidham, 1991). Since nitrogen plays a key role in metabolism, growth, reproduction, heredity and takes part in protein and amino acid synthesis, the effect of monocrotophos and monocrotophos in combination with bavistin on nitrogen protein and amino acids in the soybean was studied.

Nitrogen content and the protein (obtained by multiplying the nitrogen value by the factor 5.71 suggested by Sadasivam and Manikam, 1992 for soybean) content in the leaf, stem and root of soybean is given in

Table 8 and represented histographically in Fig. 6 and 7. Both nitrogen and protein content was enhanced by the monocrotophos. As high as 11% and 27.11% increase in nitrogen content over control was observed in leaf tissue due to 0.075% and 0.15% foliar application of monocrotophos respectively. Similarly, the protein content was also increased in insecticide sprayed leaves of soybean. However, at higher concentrations of monocrotophos (0.3%) both nitrogen and protein content was inhibited. In case of stem and root the nitrogen and protein content was found more in the plants sprayed with 0.15% and 0.075% monocrotophos respectively. However, the increase in protein and nitrogen content in stem at 0.15% monocrotophos was less than control but more than the values obtained at other concentrations of monocrotophos. The soybean plant sprayed with monocrotophos in combination with bavistin exhibited gradual reduction in root protein and nitrogen content with increasing concentrations while in leaf and stem except one concentration ( $M_2+B_2$ ), all the concentrations showed decrease in protein and nitrogen content over control.

ii) Amino acids :

Effect of monocrotophos and monocrotophos in combination with bavistin on amino acid composition was studied in soybean. The qualitative analysis of amino acids along with the Rf values is depicted in Table 9 and Plate 21. Most amino acids were effected by monocrotophos treatment even at low concentration 0.075%. The effects were more pronounced at higher concentrations. It is very clear from table and plate that there are 10 different amino acids visualised on chromatogram. These are viz. ornithine, histidine, glycine, serine, threonine, alanine, tyrosine, tryptophan, phenyl alanine and leucine. It is also clear from the table that their intensity varies

Table 8 : Effect of foliar application of monocrotophos and monocrotophos in combination with bavistin on nitrogen and protein content in leaf, stem and root of soybean (50 days old).

Treatment	Nitrogen/Protein (g 100 <sup>-1</sup> g dry wt.)		
	Leaf	Stem	Root
Control	(2.36) 13.50	(1.40) 8.02	(2.21) 12.61
<u>Monocrotophos</u>			
M <sub>1</sub> (0.075% v/v)	(2.62) 14.99	(0.62) 3.54	(2.29) 13.10
M <sub>2</sub> (0.15% v/v)	(3.00) 17.13	(0.90) 5.08	(1.32) 07.53
M <sub>3</sub> (0.3% v/v)	(1.14) 6.56	(0.37) 2.11	(0.84) 04.79
<u>Monocrotophos * + Bavistin</u>			
M <sub>1</sub> +B <sub>1</sub> (0.034% w/v)	(0.8) 04.56	(0.8) 04.56	(1.53) 8.73
M <sub>2</sub> +B <sub>2</sub> (0.068% w/v)	(2.89) 16.55	(0.9) 05.08	(1.05) 05.99
M <sub>3</sub> +B <sub>3</sub> (0.136 w/v)	(1.06) 6.05	(0.17) 00.97	(0.6) 03.42

\* = Concentrations are as above, ( ) = Values in parenthesis are of nitrogen content  
All the values are mean of three determinations.

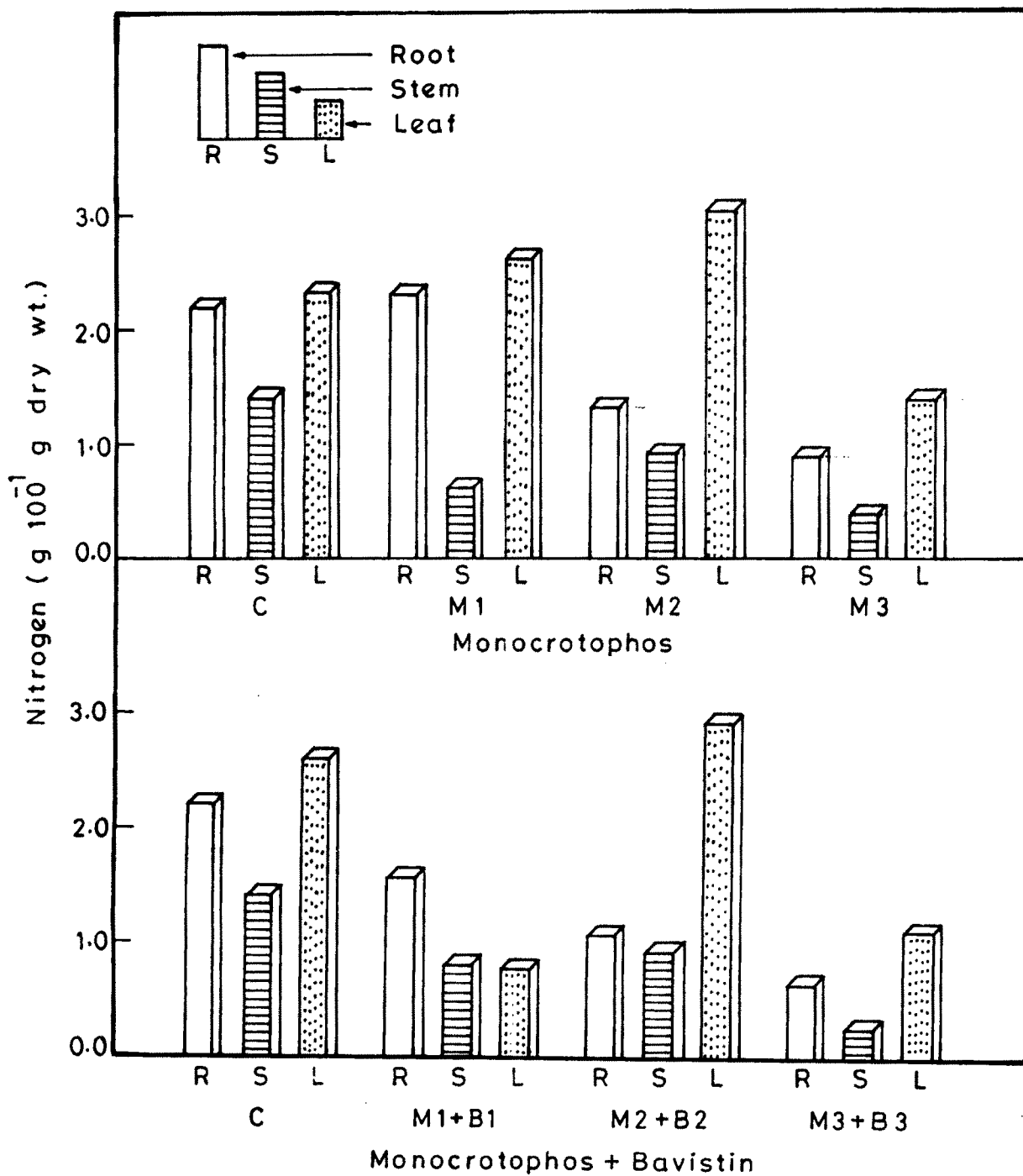


FIG. 6

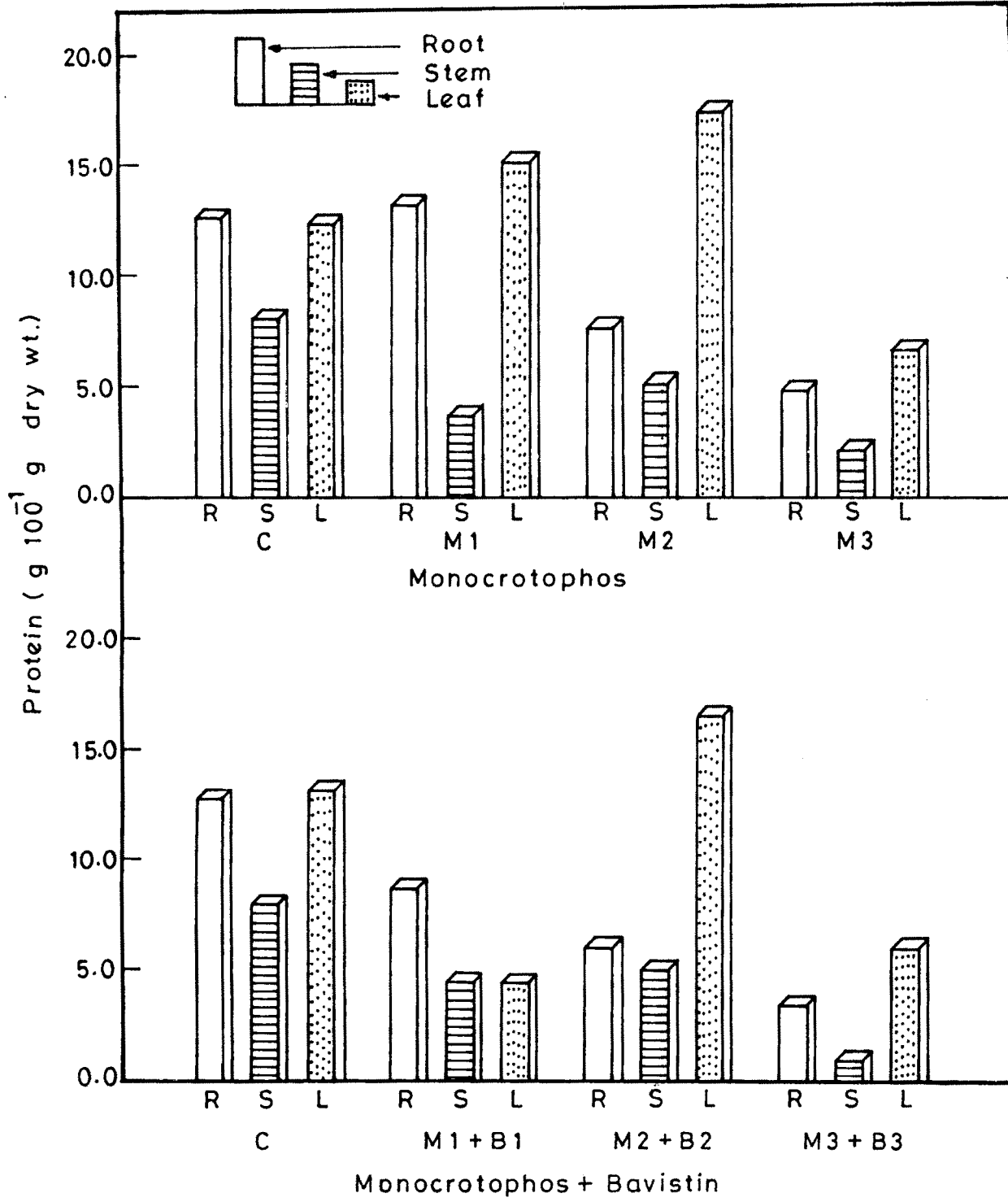


FIG. 7

Table 9 : Effect of foliar application of monocrotophos and monocrotophos with bavistin on amino acid composition in soybean leaves.

Spot No.	Rfx	Probable identification	Band Intensity								
			Control	M <sub>1</sub>	M <sub>2</sub>	M <sub>3</sub>	M <sub>1</sub> +B <sub>1</sub>	M <sub>2</sub> +B <sub>2</sub>	M <sub>3</sub> +B <sub>3</sub>		
1	10.84	Ornithine monohydrochloride	++	Trace	+	++	-	-	-	-	
2	14.54	Histidine monohydrochloride	++++	+++	++++	++++	++++	++++	++++	+++	
3	18.46	Glycine	++++	+++	++++	+++	++++	++++	++++	+++	
4	22.98	DL-Serine	++++	+++	++++	++	++++	+++	+++	+++	
5	27.10	DL-Threonine	++	++	+++	++	++++	++	++	+++	
6	31.06	DL-Alanine	Trace	-	++	+	++	++	+	+	
7	42.52	L-Tyrosine	+++	+	++++	++	++++	+++	+++	+++	
8	52.61	DL-Tryptophan	++++	+++	++++	++++	++++	++++	++++	+++	
9	61.52	DL-Phenylalanine	+++	++	++++	++++	++++	++++	++++	+++	
10	68.05	DL-nor-Leucine	+	+	+++	-	++++	++	++	+++	

- not detectable + detectable ++ Less +++ Moderate +++++ Optimum ++++++ Maximum

Plate 21. Chromatogram showing amino acid composition of soybean leaves under the influence of foliar application of monocrotophos (a) and monocrotophos in combination with bavistin (b).

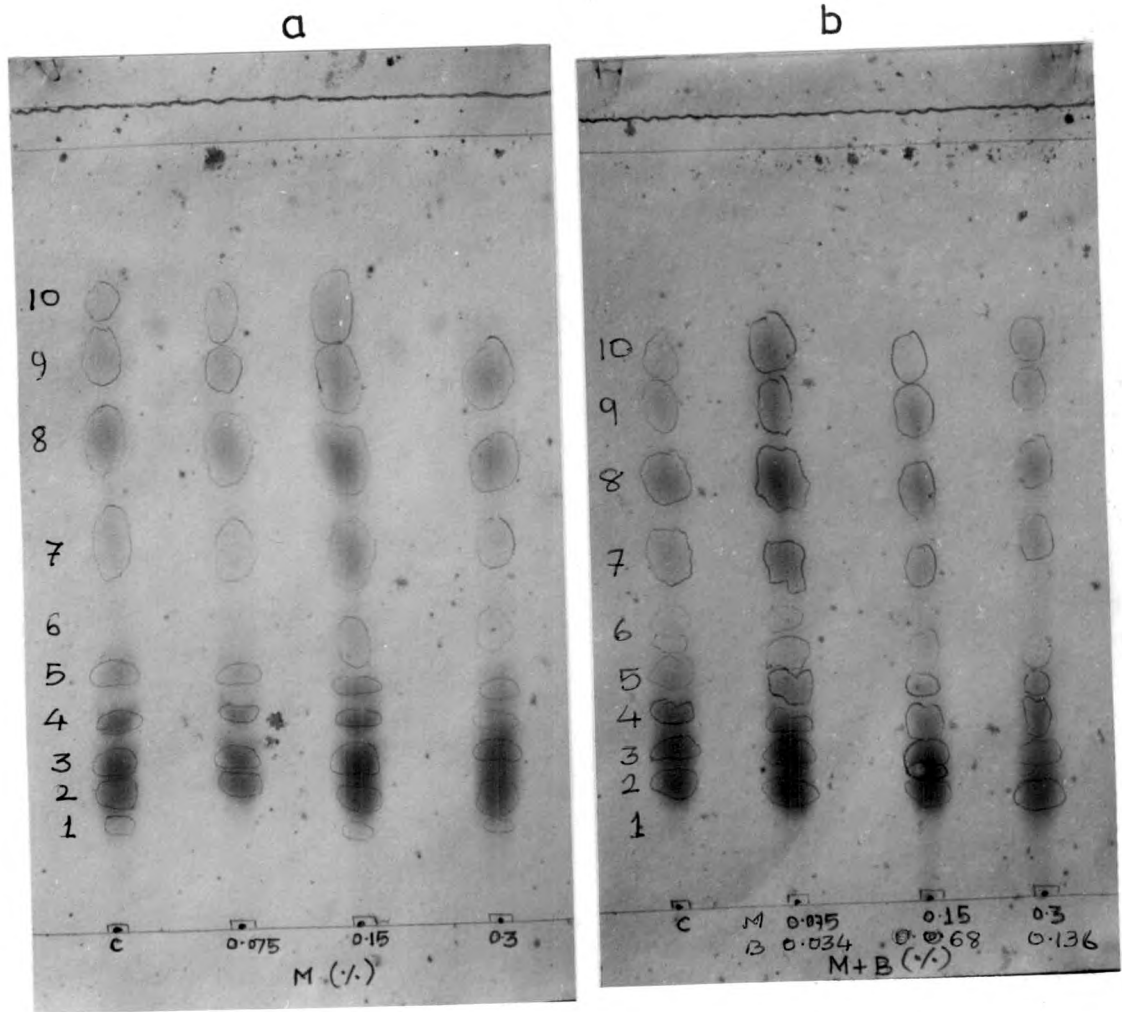


PLATE - 21



with increasing concentration of insecticidal application. The amino acids such as glycine, threonine, alanine, tyrosine, phenyl alanine and leucine showed increase in band intensity with increasing concentration of monocrotophos and monocrotophos in combination with bavistin. It is interesting to note here that the alanine which was present in trace in control was appeared in the plants sprayed with pesticide. Similarly the ornithine was completely disappeared from the plants sprayed with monocrotophos in combination with bavistin.

Many workers have studied the effect of different pesticides on aminoacid composition and biosynthesis in different plants. In duck weed (Lemna gibba) glyphosate caused reduction in the levels of aromatic amino acids needed for normal protein synthesis and thus the growth was reduced (Jaworski, 1972). The effect of various herbicides on glutathione levels in barley, tobacco, soybean and corn was examined by Smith (1985) and reported that the plants fail to increase the total amount of glutathione. Singh and Salunkhe (1970) reported significant increase in protein and amino acids due to foliar application of herbicides of S-triazine group in leaves of beans, peas and corn grown under green house condition.

Zwoliniska (1984) studied the effect of sencor on potted potatoes and reported that higher doses of herbicide treatment increase isoleucine, tyrosine and phenyl-alanine, whereas the lower dose raised threonine, valine, isoleucine and lysine in addition to above amino acids. Homeyer et al. (1985) reported that formation of alanine is directly proportional to increasing concentrations of herbicide chlorsulfuron. Thirumarna and Xavier (1987) reported decrease in amino acid content in black gram especially

with 0.01% methyl-parathion. Cooley and Foy (1992) studied the effect SC-0224 and glyphosate on free amino acids, soluble proteins and protein synthesis in Lemna gibba and reported increase in quantity of arginine, alanine, valine iso-leucine and tyrosine as compared with the control. They have attributed this with possible inhibition of protein synthesis. Paul et al. (1995) studied monocrotophos induced changes in amino acid contents of tobacco leaves. According to them amino acids were effected by monocrotophos showing decrease in alanine, lycine, leucine and tryptophan.

The data of the present investigation depicted in Table 9 and Plate 21 clearly indicates that the insecticides viz. monocrotophos and monocrotophos in combination with bavistin are responsible for increasing the level of nitrogen and protein content at lower concentration and certain amino acids at all the concentrations. This increased amino acid level at higher concentration can be attributed to the inhibition of the protein synthesis or increased protein hydrolysis or decreased use of amino acids as respiratory carbon sources and/or increased amino acid biosynthesis. The reduction in nitrogen, protein and nodule number in the soybean plant due to the foliar application of monocrotophos in combination with bavistin can be correlated with bavistin inhibited bacterial growth. As such thiram + bavistin mixtures were inhibitory to the isolates of rhizobium and cause complete growth inhibition at 20 ppm and reduction in nodule number in mung bean (Chakraborty et al. 1985). In soybean 75% defoliation also resulted decrease in nodule activity (Bullock, 1990).

It is noteworthy to mention here that the changes in composition of sugars and aminoacids could be important in pathogenesis of foliar

pathogens. Such changes induced by insecticides may result in development of iatrogenic diseases since the changed chemical environment on the phylloplane may affect the growth of pathogens directly or indirectly altering the activity of antagonists in the phylloplane.

For instance D.D.T. application converted a rust resistant wheat to a susceptible one due to accumulation of sugar in leaf tissue. Maleic hydrazide reduced Alternaria infection but increased Fusarium wilt in tomato. 2,4-D decreased sugar content of leaf tissue thereby favouring Heliminthosporium infection of maize. Zineb spray used to control Plasmopara on grape worsened powdery mildew on vine and Botrytis on fruits. Similarly insecticidal formulations were found to increase incidence of Peronospora parasitica on Broccoli (Paul et al., 1995).

In this direction such studies concerned with the effects of growth regulators, fungicides and herbicides on physiology and biochemistry of plants were made but very little information is available on insecticides commonly used in present day agriculture. The present investigation is a part of it, which needs further investigation to relate precisely the role of insecticide in plant growth and metabolism. ←

#### D. Nitrate and Nitrite Reductase :

The three major inputs viz. water, carbondi-oxide (CO<sub>2</sub>) and nitrogen governs agricultural productivity. Nitrogen is mainly taken up as nitrate, which is reduced to ammonia before being assimilated into amino acids. Reduction of nitrate to ammonia is catalyzed by two enzymes viz. nitrate reductase (NR) and nitrite reductase (NIR). In leaves, these enzymes differ in cellular location, electron donor, and energy generation system. NR is

donor

located in the cytoplasm (Ritenour et al. 1967), uses NADH as its electron donor (Beevers and Hageman, 1969), and derives its source of NADH, primarily from the oxidation of 3-P-glyceraldehyde (Klepper et al. 1971), or malate (Neyra and Hageman, 1976). NIR is located in the chloroplast, utilizes reduced ferredoxin as its electron donor and derives its reductive from photosynthetic electron flow.

Since nitrate and nitrite reductase enzymes are associated with chloroplast and cytoplasm and pesticide affect the pigment system, the effect of monocrotophos and monocrotophos in combination with bavistin on NR and NIR was studied in soybean. The activities of these enzymes in response to foliar application of monocrotophos and monocrotophos in combination with bavistin assayed in the leaves of soybean is given in Table 10. In case of leaf and stem at lower concentration of monocrotophos the activity of NR reduced, there after increased at 0.15% monocrotophos and then again declined at higher concentration. However, this reduction was not below the level of control. On the other hand there was gradual reduction in the leaf NR activity as compared with the control, with increase in concentration of M+B, while in stem the NR activity was increased beyond control and in root it was inhibited considerably. Both these pesticides exhibited marked inhibition of NR activity in the root as compared with the control. Though M+B application raised the NR activity slightly over the value observed at lower concentration, it was below the level of control.

The NIR activity in leaf and root was gradually inhibited with increasing concentration of monocrotophos, while it was increased in case of stem. The NIR activity in the plants sprayed with monocrotophos in

Table 10: Effect of foliar application of monocrotophos and monocrotophos in combination with bavistin on nitrate and nitrite reductase activity in soybean (50 days old).

Treatment	Nitrate reductase $\mu$ mole $\text{NO}_2$ formed $\text{l}^{-1} \text{g}^{-1}$ fresh wt.			Nitrite reductase $\mu$ mole $\text{NO}_2$ reduced $\text{h}^{-1} \text{g}^{-1}$ fresh wt.		
	Leaf	Stem	Root	Leaf	Stem	Root
Control	382.33	19.73	616.66	382.33	19.73	616.66
<u>Monocrotophos</u> M <sub>1</sub> (0.075% v/v)	246.66	12.33	488.99	307.33	54.2	616.66
M <sub>2</sub> (0.15% v/v)	505.66	43.16	622.00	295.99	69.06	381.00
M <sub>3</sub> (0.3% v/v)	394.66	37.00	123.66	287.9	59.19	164.00
<u>Monocrotophos + Bavistin</u> M <sub>1</sub> +B <sub>1</sub> (0.034% w/v)	217.77	48.1	137.00	233.3	36.90	419.3
M <sub>2</sub> + B <sub>2</sub> (0.068% w/v)	126.66	70.3	170.00	271.32	38.7	414.64
M <sub>3</sub> +B <sub>3</sub> (0.136 w/v)	115.92	88.8	292.5	244.00	29.5	153.3

\* = Concentrations are as above

The values are mean of three determinations.

combination with bavistin exhibited increase in the leaf NIR activity with increase in concentration but, certainly all the values were below the level of control. In case of stem the NIR activity was found to be increased over control, no much change was exhibited by the root NIR except it was greatly affected at higher concentration of M + B.

From the above results one thing is clear that monocrotophos at M<sub>2</sub> concentration triggers the NR activity in leaf, root and stem but inhibits NIR activity both in leaf and root; and stimulates in stem.

Many workers have studied the effect of various herbicides on the activity of NR and NIR. Effect of thiocarbamate (Klepper, 1975), glyphosate (Hoagland, 1985), fluchloralin and benthocarb (Reddy and Rao, 1985), had inhibitory effect on NR and NIR. Contrary to this stimulatory response of these enzymes due to herbicidal application such as 2,4-D (Beevers *et al.* 1963) S-triazine (Singh and Salunkhe 1970, Mohandas *et al.* 1978) thiobencarb (Reddy *et al.* 1983) have also been reported.

Thombre *et al.* (1989) studied the effect of fungicidal seed treatment and reported no adverse effect on rhizobial symbiosis or seed yield in soybean. Singh *et al.* (1991) while studying effect of monocrotophos seed treatment in soybean reported decrease in nodule number at 60 days of plant growth while increase in nodule number after treatment with 20 g carbosulfan/kg seed. Reddy *et al.* (1986) studied the effect of pesticides on plant growth, nodulation, nitrogen fixation and protein content in pea and reported inhibition of nodulation and symbiotic nitrogen fixation. Significant increase in leghaemoglobin content of nodules at lower concentration of herbicide fluchloralin in chickpea has also been reported by

Palwa and Jaiprakash (1992). Further Misra and Gupta (1985) studied effect of organophosphorus insecticides on nitrogen metabolism during germination of mung and cowpea. The results indicated that the insecticides viz. phorate, disulfoton, monocrotophos and fensulfothion at 25, 50 and 100 ppm did not cause any major interference in nitrogen metabolism in these legumes.

Thus from the above foregoing literature it is very clear that the pesticide treatment either enhance or inhibit NR and NIR activity. From the present investigation one thing is very clear that the higher concentration of monocrotophos inhibits the NR and NIR activity. The chloroplast enzyme nitrate reductase is dependent on photosynthetic electron flow to ferredoxin, which is used by the enzyme as reductant. Inhibitors of photosynthetic electron transport therefore stop or reduce the reduction of nitrite to ammonia ion. This is very clear from the Table 10, where nitrite reductase is affected due to monocrotophos and monocrotophos in combination with bavistin. An increase in NR with a concomitant decrease in NIR in thiobencarb treated leaves of tropical weeds was reported by Reddy et al. (1983). However, such observation was not noticed in soybean when sprayed with monocrotophos and monocrotophos in combination with bavistin. There are reports that if nitrate continues to be reduced to nitrite in the cytoplasm and if NIR gets inhibited, toxic levels of nitrite and nitrous acid accumulate rapidly which cause injury and death of plants (Klepper, 1988). Possibly this may be the reason of induced phytotoxicity at higher concentration of monocrotophos and monocrotophos in combination with bavistin. The inhibition of NR and NIR activity can also be

correlated with decrease in chlorophyll content at higher concentration of pesticidal treatment (Table 7 ) in soybean.

#### E. Mineral Nutrient Status

The minerals are important constituents of living systems at all levels and by and large the plant nutrition is concerned with nutrients as well as nutrients uptake and their distribution in plants. The essential nutrients required by higher plants are exclusively of inorganic nature. The supply and absorption of these inorganic constituents is needed for growth and metabolism of crop plants. It is well known that fundamental processes of plant physiology and biochemistry such as photosynthesis and respiration are affected or regulated by mineral nutrients. More over, many a times plants have to face some unfavourable conditions such as water and salt stress, pollution stress, etc. during which the possibility of disturbances in mineral nutrition may occur. Besides, in order to overcome pest and disease attack, plants are oftenly subjected to pesticidal treatment. Some times the pesticides are used either frequently or in a higher doses without knowing their lurking danger. Hence it is thought worthwhile to study the nutrient status in root, stem and leaf of soybean plant under the influence of foliar application of monocrotophos (Table 11) and monocrotophos in combination with bavistin (Table 12).

Monocrotophos and monocrotophos in combination with bavistin altered levels of macro and micro elements of leaf, stem and root at all the concentrations. The changes caused by pesticidal application are not of greater magnitude. Sodium is generally not required by green plants but the work carried out in the last decade reports that sodium contributes to



Table 11: Effect of foliar application of monocrotophos on mineral content in soybean (50 days old)

Treatment	Plant Part	Mineral constituents ( g 100 <sup>-1</sup> g dry wt. )									
		Na	K	Ca	Mg	Mn	Fe	Zn	Co	P	
Control	Leaf	0.028	2.74	4.86	1.22	0.01	0.54	0.03	0.011	2.36	
<u>Monocrotophos</u> M <sub>1</sub> (0.075% v/v)		0.038	2.84	6.64	1.46	0.0138	0.66	0.05	0.0012	1.11	
M <sub>2</sub> (0.15% v/v)		0.022	2.94	4.06	1.36	0.138	0.64	0.04	0.0014	2.9	
M <sub>3</sub> (0.3% v/v)		0.028	2.16	4.90	1.16	0.0104	0.44	0.18	0.010	1.06	
Control	Stem	0.08	2.50	3.22	1.86	0.0034	0.42	0.07	0.009	1.41	
<u>Monocrotophos</u> M <sub>1</sub> (0.075% v/v)		0.042	2.84	4.98	1.22	0.004	0.96	0.01	0.0028	0.8	
M <sub>2</sub> (0.15% v/v)		0.024	2.78	2.28	0.70	0.0032	0.26	0.06	0.001	0.89	
M <sub>3</sub> (0.3% v/v)		0.036	2.92	2.10	0.50	0.0036	0.18	0.02	0.0016	0.75	
Control	Root	0.048	1.58	7.12	1.32	0.0166	1.98	0.12	0.001	2.21	
<u>Monocrotophos</u> M <sub>1</sub> (0.075% v/v)		0.036	2.34	2.78	0.66	0.0146	1.42	0.08	0.0014	1.53	
M <sub>2</sub> (0.15% v/v)		0.046	2.08	2.46	0.56	0.0108	0.82	0.06	0.0018	1.05	
M <sub>3</sub> (0.3% v/v)		0.046	2.32	2.4	0.82	0.013	0.20	0.06	0.0016	0.6	

The values are mean of three determinations.

Table 12 : Effect of foliar application of monocrotophos in combination with bavistin on mineral content in soybean (50 days old)

Treatment	Plant Part	Mineral constituents ( g 100 <sup>-1</sup> g dry wt. )									
		Na	K	Ca	Mg	Mn	Fe	Zn	Co	P	
Control	Leaf	0.028	2.74	4.86	1.22	0.01	0.54	0.03	0.011	2.36	
<u>Monocrotophos + Bavistin</u>											
M <sub>1</sub> +B <sub>1</sub> (0.034% w/v)		0.032	2.88	3.60	1.72	0.0118	0.58	0.032	0.0012	2.18	
M <sub>2</sub> +B <sub>2</sub> (0.068% w/v)		0.026	3.9	5.2	1.08	0.0114	0.54	0.035	0.0014	3.00	
M <sub>3</sub> +B <sub>3</sub> (0.136 w/v)		0.026	2.66	3.00	0.80	0.011	0.18	0.020	0.0010	1.15	
Control	Stem	0.08	2.50	3.22	1.86	0.0034	0.42	0.07	0.009	1.405	
<u>Monocrotophos + Bavistin</u>											
M <sub>1</sub> +B <sub>1</sub> (0.034% w/v)		0.042	2.60	2.24	0.62	0.0044	0.42	0.06	0.0014	0.62	
M <sub>2</sub> +B <sub>2</sub> (0.068% w/v)		0.044	2.54	1.54	0.3	0.0219	0.16	0.020	0.002	0.89	
M <sub>3</sub> +B <sub>3</sub> (0.136 w/v)		0.044	3.66	2.50	0.82	0.0036	0.12	0.020	0.0016	0.37	
Control	Root	0.048	1.58	7.12	1.32	0.0166	1.90	0.12	0.001	2.21	
<u>Monocrotophos + Bavistin</u>											
M <sub>1</sub> +B <sub>1</sub> (0.034% w/v)		0.048	2.4	3.5	0.58	0.0034	1.24	0.04	0.001	1.11	
M <sub>2</sub> +B <sub>2</sub> (0.068% w/v)		0.044	2.52	1.38	0.5	0.0104	0.72	0.06	0.0010	1.32	
M <sub>3</sub> +B <sub>3</sub> (0.136 w/v)		0.044	2.52	2.18	0.44	0.022	0.06	0.02	0.0012	0.84	

The values are mean of three determinations.

maintain the osmotic potential of the cell and thus has a positive effect on the water regime of the plants (Mengel and Krikby, 1982). The values of sodium reported in table reveals that it stimulates at lower concentration in case of leaf, and inhibit in stem and root as compared with control (Table 11) This situation was more or less same in the plants sprayed with monocrotophos in combination with bavistin (Table 12).

Potassium plays a major role in plant metabolism and exhibits its higher concentration wherever high energy transformation takes place and also helps in maintaining ionic balance under stress condition and govern stomatal mechanism. The content of potassium was increased in leaf tissue at lower concentrations and at all the concentrations in stem and root as compared with control (Table 11). The same trend was found in the plants sprayed with monocrotophos in combination with bavistin (Table 12).

Calcium a polyvalent cation which governs membrane permeability is useful structural component of the cell is greatly affected by the pesticides in case of stem and root while in leaf the calcium content enhanced as compared with the control (Table 11 & 12). For continuous uptake of calcium needs effective operation of transpiration stream (Waisel *et al.*, 1966). The decreased calcium content at higher concentration of pesticidal treatment can be correlated with decreased transpiration rate in soybean.

Magnesium is generally taken up by plants in lower quantities than calcium and potassium and act as a co-factor in almost all enzymes activating phosphorylation process. It's concentration was found to be triggered in leaf tissue at lower concentration; however, in case of stem and root none of the concentrations favoured magnesium content.

Manganese is also equally important mineral element which play an important role in activation of decarboxylases and dehydrogenases of T.C.A. cycle and bring about oxidation of IAA by activating IAA oxidase (Mumford *et al.* 1962, Tayler *et al.* 1968). It also involved in oxidation-reduction process in photosynthetic electron transport system (Bishop, 1971). The manganese content was found more in leaf tissue at all the concentrations whereas in case of root, none of the concentration helped in stimulating its content, in stem surprisingly lower and higher concentrations were stimulatory in action (Table 11). The situation is remained same in leaf and stem tissue as far as response of manganese to monocrotophos in combination with bavistin is concerned. Whereas in case of root the higher concentration caused more accumulation of manganese as compared with the control (Table 12).

Iron is an indispensable trace element because it functions both as structural element and as a cofactor for several enzymatic reactions and chlorophyll synthesis. The fate of this element studied under the influence of pesticides indicated stimulatory action at lower concentrations in case of leaf and stem and in case of root only at higher concentration of monocrotophos. While in case of monocrotophos in combination with bavistin treated plants no significant change in iron content was noticed at lower concentrations, but it was greatly reduced at higher concentration. In stem and root the decrease in iron level with increase in concentration of pesticide was noticed.

Zinc is closely involved in nitrogen metabolism of plants (Mengel and Krikby, 1982). Zinc deficiency shows sharp decrease in level of RNA and the ribosome content of the cells (Price *et al.* 1972). It is also required in the

synthesis of tryptophan (Tusi, 1948) and tryptophan which act as a precursor of I.A.A., which may be influenced indirectly by Zinc. This element was found to be stimulated at all the concentration of insecticide in leaf tissue, maximum being at higher concentration. However, in root and stem none of the concentration favoured its stimulation (Table 11). On the other hand the zinc content was found slightly more than control at lower concentrations and reduced below the level of control at higher concentration in the plants sprayed with monocrotophos in combination with bavistin. The level of zinc in root and stem was however, not favoured by pesticidal application (Table 12).

Cobalt is found in plants as cyanocobalmin (vit.B<sub>12</sub>) and is associated with group transfer reactions. It is required by the micro-organisms in their symbiotic association with legume roots for nitrogen fixation. It is interesting to note here that the cobalt content was hampered at all the concentrations in the stem, increased at all the concentrations in root and in leaf it was increased at lower concentrations, but declined at higher concentration (Table 11). This situation and the content of cobalt was almost same in case of leaf, inhibited at all the concentration of stem while no significant change was observed in root due to pesticidal application (Table 12).

Phosphorus is a constituent of biomembranes in the form of phospholipids and also act as carrier of genetic information in DNA and RNA molecule and responsible for strong acidic nature of nucleic acid. Besides, inorganic phosphate takes part in many enzyme reactions, increases respiration, regulates photosynthesis and carbohydrate metabolism. The phosphorus content studied in soybean under the

influence of monocrotophos showed increasing tendency with increasing concentration in leaf while it was considerably reduced in root and stem. (Table 11). While in the plants sprayed with monocrotophos in combination with bavistin resulted increase in phosphorus content both in leaf and stem and decreased in root (Table 12).

The mineral content in soybean plant has been studied by many workers (Khan 1988, Ye *et al.* 1991, Prapharsi *et al.* 1992, Slipcevic *et al.* 1992, Durand and Lucan 1994). However, very little is known regarding the effect of insecticide on mineral nutrient status in soybean. Gupta and Beg (1987) studied influence of endosulfan spray on mineral status in mungbean and reported alteration in the levels of macro and micro elements. Similarly influence of systemic insecticides on the contents of nitrogen, phosphorus and potassium in the aerial parts of tomato has been investigated by Oya *et al.* (1994). Their study indicated increased phosphorus and potassium content but nitrogen content was not affected due to insecticide application.

It is very clear from the present investigation and the literature survey that such type of exploratory studies point out the need for detailed and indepth experiments with pesticides to clarify the mechanism involved in growth response and influence of pesticides on mineral uptake, particularly under the field conditions. Our knowledge at the moment, does not extend much beyond the simple observations of changes in nutrient levels in various plant parts that seem to have been brought about by the presence of pesticide. Hence it is not possible to decide whether they are the result of more or less efficient uptake and utilization or whether they indicate changes in requirement.