

CHANGES IN CHLOROPLAST PIGMENTS AND SEEDLING GROWTH ATTRIBUTES OF MUNGBEAN AS INDUCED BY PESTICIDAL SEED TREATMENTS

G. Panduranga Murthy* and Vinay B. Raghavendra

P. G. Department of Studies and Research in Botany
University of Mysore, Manasagangotri, Mysore - 06 (India)

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ABSTRACT

Seed treatment with Gaucho (an insecticide) and Krilaxyl (a fungicide) at the concentrations of recommended dosages, below recommended and above recommended and in combination dosages were done on Mungbean cv. PS-116 at 12, 24 and 48 hours exposure periods. Wet treatment of both the pesticides exhibited non significant variations on germination and they varied in respect of root length and shoot length at recommended and higher dosages of longer exposure periods. The calculated results of vigour index, tolerance index were decreased and the percent phytotoxicity was increased along with the increase in concentrations over control. The content of chloroplast pigments, (Chlorophyll 'a', 'b', total chlorophyll and carotenoid) were also reduced drastically at higher and in combination dosages of both the pesticides at all the exposure periods. Lower concentrations of lower exposure periods did not induce much change on germination and other parameters

Keywords: Germination, early seedling growth, pesticide seed treatments, chloroplast pigments, fresh weight, dry weight and mungbean.

INTRODUCTION

Pulses occupy a unique position in Indian diet, since they form the bulk of plant source of high protein. In the face of increasing dependence of our world population on relatively some crop plants, the shape of our future will depend on our understanding of these plants and overcoming the limitations of their productivity. One of the major cause for the loss of food in terms of quantity and quality due to incidence of many pests and diseases. A number of pesticides have been used widely nowadays for early pests control but, most of these pesticides are not highly selective. Applications of pesticides are not only affect the pests but also toxic to most of the crop plants (Prasad and Mathur, 1983). Various workers have been reported adverse effects of pesticides on seed germination early growth and have concluded that, seed treatment, seed dressing in the form of emulsifiable concentrations were phytotoxic to majority of the crops (Patil and Shirashyad, 1989; Panduranga Murthy and Leelavathi, 2002). Hence,

it was decided to investigate whether, the individual and in combination treatment of an insecticide (Guacho) and fungicide (Krilaxyl) would have an additive or synergistic effects on germination, early growth and pigment content of Mung bean var PS-116.

MATERIAL AND METHODS

The experimental material 'Mungbean' is a member of family Papilionaceae, Genetically pure seeds of above pulse crop var PS-115 were procured from G.K.V.K. University of Agricultural Sciences, Bangalore, Guacho (insecticide) and Krilaxyl (fungicide) were purchased from authorised dealers, Bangalore.

The seeds were surface sterilized by immersing the seeds in $HgCl_2$ for 2 minutes and repeatedly washed with sterile distilled water. For each treatment 25 gms of seeds were taken and thoroughly mixed with the slurry of different concentrations of above pesticides viz.,

recommended dosage (Gaucho-10 gms/kg, Krilaxyl- 6 gms/kg of seeds). 25% of the below recommended. 25% of the above recommended and in combination of both the pesticides (50% among recommended). After treatment the seeds were kept for different exposure periods of 12, 24 and 48 hours. Untreated seeds in distilled water served as control.

Then, they were subjected for germination in germinator at $28\pm 2^{\circ}\text{C}$ using between paper method and replicated thrice. On termination day (ISTA 1995) percent germination, seedling growth, fresh weight (on 4th day) and dry weight were recorded at all the exposure periods. Chlorophyll (Arnon 1949) and Carotenoid (Kirk and Allen, 1965) were estimated. Vigour index, tolerance index and percent phytotoxicity were calculated using standard formula prescribed by Abdul Baki and Anderson (1973), Turner and Marshal (1972) Chiou and Mullar (1972) respectively.

All the tabulated data were analysed using arithmetic mean and standard deviation of three determinants (Keul, 1959).

RESULTS AND DISCUSSION

Germination percentage:

The tabulated data reveals that, the Gaucho and Krilaxyl are strong inhibitors of the root and shoot growth of Mungbean. The higher and in combination dosages of two pesticides gave severe reduction on germination and was greatly reduced and delayed by long-term treatment (Table - 1).

This decrease of germination in the treated series is attributed to the inhibitory action of Gaucho and Krilaxyl on the metabolic activities (Krishna Murthy and Rao, 1980; Pradhan and Basu, 1980; Reddy and Vidyavathi, 1984; and Chakravarthi, 1986).

Seedling growth:

Root and shoot growth (Table -1) was found to be affected while, increased and in combination treatments of longer exposure periods (Kamble and Sabale, 1999, Panduranga Murthy and Leelavathi, 2002). This inhibition of seedling growth indicates that, the effect on cell division of root and shoot meristem and cell elongation (Somashekar and Sreenath, 1987).

The ratio of root and shoot was increased as the dose of both the pesticides increased suggesting an inhibition of root development. This may be due to the higher concentrations of the Gaucho and Krilaxyl accelerated the shoot growth while affecting the growth of root adversely (Gaikwad and Pawar, 1979; Benjamin 1986).

Chlorophyll and Carotenoids:

Chlorophyll 'a' and Chlorophyll 'b' (Table - 2) were reduced significantly at higher concentrations and in combination of both the chemicals during longer exposure periods (Ramulu & Rao, 1987).

This reduction of Chlorophyll content may be due to formation of enzyme i.e., Chlorophyllase, which is responsible for Chlorophyll degradation (Majumdar *et al.*, 1991). At in combination dosage of longer exposure period, the ratio of chlorophyll a/b was reduced at greater extent.

In addition to these, carotenoids also decreases significantly with increased in concentrations of all the exposure periods (Table - 2). The data also clearly indicated that, the calculated results of vigour index and tolerance index were decreased significantly and the percent toxicity was marginally increased, while increase and in combination of long term treatment by both the pesticides are represented in Table -2. (Dhakshina moorthy and Sivaprakasam, 1989).

Similarly, the fresh weight and dry weight (Table - 2) were greatly reduced at higher concentrations of individual and in combination of longer exposure periods as compared to control (Ramadoss and Sivaprakasam, 1994).

In the present study, a inhibitory effects of both the pesticides on germination, early growth, pigment content etc., at higher dosages of longer exposure periods have been recorded. But, the lower exposure periods of lower concentrations did not induce much variation on germination and other above parameters. In a study of combined treatment of insecticide and fungicide have also gave drastic reduction on germination and early growth at longer exposure periods (Krurup & Kivera, 1982). It is concluded that, both the pesticides treated seedlings of non-target crop plant, Green gram, which shows phytotoxic effects even at recommended and higher dosages of longer exposure periods under laboratory conditions.

Table - 1

CROP TREATMENTS	GERM(% EP	MRL(cms)		MSL (cms)		R/S	Mungbean var. PS - 116		P.P(%) AM \pm SD
		AM \pm SD	AM \pm SD	AM \pm SD	AM \pm SD		VI	T.I	
BR	12 H	89.66 \pm 2.07	8.0141 \pm 0.09	14.1600 \pm 0.03	0.5661 \pm 0.01	1988.12 \pm 94.60	85.56 \pm 0.73	14.43 \pm 0.03	
	24 H	83.11 \pm 4.91	6.2776 \pm 0.16	12.3818 \pm 1.69	0.5127 \pm 0.04	1549.75 \pm 172.49	81.00 \pm 6.29	16.65 \pm 3.59	
	48 H	71.66 \pm 6.23	4.5770 \pm 0.07	10.9042 \pm 0.94	0.4229 \pm 0.03	1112.10 \pm 137.10	70.75 \pm 1.51	29.23 \pm 1.51	
Gaucho	12 H	74.00 \pm 1.04	7.3330 \pm 0.27	11.9109 \pm 0.39	0.5415 \pm 0.01	1350.05 \pm 46.14	78.29 \pm 3.94	21.70 \pm 3.40	
	24 H	65.55 \pm 0.78	5.6633 \pm 0.49	10.6556 \pm 0.15	0.5381 \pm 0.08	1069.43 \pm 14.5	75.28 \pm 6.73	24.71 \pm 6.73	
	48 H	51.60 \pm 4.71	4.1070 \pm 0.81	8.5484 \pm 0.60	0.4814 \pm 0.10	650.32 \pm 50.47	63.56 \pm 12.977	36.42 \pm 12.90	
Dosages	12 H	63.33 \pm 2.5	5.3010 \pm 0.10	10.0101 \pm 0.28	0.4955 \pm 0.03	948.11 \pm 17.12	57.66 \pm 2.82	42.33 \pm 0.60	
	24 H	58.66 \pm 1.88	4.2161 \pm 0.44	9.8412 \pm 0.19	0.4297 \pm 0.04	823.67 \pm 17.69	55.97 \pm 5.20	44.01 \pm 5.20	
	48 H	41.36 \pm 4.71	3.5497 \pm 0.69	7.4094 \pm 0.13	0.4798 \pm 0.09	459.31 \pm 74.35	54.75 \pm 10.14	45.24 \pm 10.14	
BR	12 H	79.20 \pm 1.08	6.7140 \pm 0.42	13.6105 \pm 0.33	0.4936 \pm 0.03	1609.70 \pm 39.82	71.68 \pm 3.90	28.31 \pm 1.31	
	24 H	72.70 \pm 2.06	5.1483 \pm 0.47	11.2382 \pm 0.59	0.4571 \pm 0.01	1189.88 \pm 61.34	68.30 \pm 4.86	31.68 \pm 4.86	
	48 H	50.00 \pm 8.16	3.9454 \pm 0.48	8.0690 \pm 2.04	0.5101 \pm 0.11	620.23 \pm 213.62	60.96 \pm 7.89	38.96 \pm 7.89	
Krilaxy/ Dosages	12 H	64.66 \pm 2.72	5.9600 \pm 0.06	9.7770 \pm 2.29	0.5944 \pm 0.01	1007.90 \pm 23.60	63.63 \pm 2.25	36.36 \pm 0.64	
	24 H	50.80 \pm 2.06	4.7311 \pm 0.35	7.2258 \pm 0.32	0.6502 \pm 0.05	607.76 \pm 33.79	62.96 \pm 5.83	37.02 \pm 5.83	
	48 H	37.88 \pm 2.14	3.5643 \pm 0.24	4.6927 \pm 0.45	0.7666 \pm 0.08	312.58 \pm 23.61	55.28 \pm 4.87	44.80 \pm 4.93	
Gaucho + Krilaxy	12 H	48.66 \pm 1.13	5.0401 \pm 0.10	8.8106 \pm 0.01	0.5686 \pm 0.02	673.97 \pm 2.62	53.38 \pm 2.02	44.18 \pm 2.02	
	24 H	38.33 \pm 1.49	3.7931 \pm 0.21	6.7267 \pm 0.64	0.5649 \pm 0.04	406.58 \pm 28.37	50.39 \pm 2.55	49.60 \pm 2.15	
	48 H	23.33 \pm 2.40	2.8506 \pm 0.02	5.1822 \pm 0.20	0.5503 \pm 0.02	187.40 \pm 18.68	43.77 \pm 1.86	55.89 \pm 2.26	
CONTROL	12 H	41.70 \pm 0.33	4.6401 \pm 0.12	7.8991 \pm 0.43	0.5494 \pm 0.01	510.37 \pm 25.20	49.54 \pm 1.50	46.60 \pm 1.75	
	24 H	30.77 \pm 1.09	3.7661 \pm 0.09	7.5482 \pm 0.04	0.4988 \pm 0.00	348.17 \pm 21.58	46.94 \pm 0.06	49.95 \pm 0.19	
	48 H	25.99 \pm 1.03	2.9082 \pm 0.25	6.3427 \pm 0.10	0.4592 \pm 0.04	240.40 \pm 4.76	44.89 \pm 3.18	55.10 \pm 3.18	
CONTROL	12 H	100.00 \pm 1.56	9.3660 \pm 0.27	19.4016 \pm 0.43	0.4867 \pm 0.03	2876.76 \pm 89.20	-	-	
	24 H	97.88 \pm 2.06	7.5255 \pm 0.16	16.9270 \pm 0.49	0.4451 \pm 0.02	2394.27 \pm 85.24	-	-	
	48 H	91.66 \pm 4.71	6.4600 \pm 0.10	13.4870 \pm 0.84	0.4802 \pm 0.01	1831.15 \pm 135.5	-	-	

Germination Percentage based on normal seedlings only

Germ: Germination, MRL: Mean Root Length, MSL: Mean Shoot Length, R/S: Root & Shoot Ratio, VI: Vigour Index, TI: Tolerance index, PP: Percent Phytotoxicity, EP: Exposure period, H: Hour, AM: Arithmetic mean, SD: Standard deviation, BR: Below recommended, RD: Recommended dosage, AR: Above recommended, I+F: Insecticide + Fungicide.
Values are represented as Arithmetic mean and standard deviation of three determinants.

Table - 2

CROP TREATMENTS	EP	CHL 'a' (mg.g ⁻¹)			CHL 'b' (mg.g ⁻¹)			CHL a/b ratio			Mungbean var. PS - 116			Fresh wt. (gm)			Dry wt. (gms)		
		AM	±	SD	AM	±	SD	AM	±	SD	AM	±	SD	AM	±	SD	AM	±	SD
Gauchó Dosages	BR	12 H	0.0830	±0.00	0.0860	±0.00	0.9651	±0.00	0.1690	±0.00	0.0471	±0.00	7.65	±0.28	0.38	±0.02			
		24 H	0.0699	±0.00	0.0689	±0.00	1.0146	±0.02	0.1387	±0.02	0.0491	±0.00	5.22	±0.05	0.25	±0.04			
		48 H	0.0453	±0.00	0.0471	±0.00	0.9618	±0.00	0.0925	±0.00	0.0594	±0.00	4.07	±0.05	0.16	±0.00			
Gauchó Dosages	RD	12 H	0.0710	±0.00	0.0755	±0.00	0.9403	±0.03	0.1469	±0.00	0.0466	±0.00	6.66	±0.02	0.26	±0.02			
		24 H	0.0673	±0.00	0.0664	±0.00	1.0140	±0.02	0.1339	±0.00	0.0482	±0.00	4.88	±0.01	0.25	±0.00			
		48 H	0.0403	±0.00	0.0426	±0.00	0.9467	±0.00	0.0829	±0.00	0.0544	±0.00	3.52	±0.06	0.13	±0.00			
Gauchó Dosages	AR	12 H	0.0665	±0.00	0.0640	±0.00	1.0390	±0.00	0.1304	±0.00	0.0424	±0.00	5.65	±0.35	0.20	±0.01			
		24 H	0.0609	±0.00	0.0563	±0.00	1.0818	±0.00	0.1172	±0.00	0.0440	±0.00	3.62	±0.10	0.13	±0.00			
		48 H	0.0338	±0.00	0.0315	±0.00	1.0734	±0.00	0.0653	±0.00	0.0566	±0.00	3.09	±0.01	0.11	±0.00			
Krilaxyl Dosages	BR	12 H	0.0780	±0.00	0.0344	±0.00	2.2674	±0.06	0.1124	±0.00	0.0496	±0.00	4.14	±0.21	0.19	±0.04			
		24 H	0.0679	±0.00	0.0393	±0.00	1.7339	±0.09	0.1072	±0.00	0.0511	±0.00	3.39	±0.02	0.16	±0.00			
		48 H	0.0423	±0.00	0.0452	±0.00	0.9373	±0.02	0.0876	±0.00	0.0540	±0.00	3.13	±0.01	0.12	±0.00			
Krilaxyl Dosages	RD	12 H	0.0660	±0.00	0.0425	±0.00	1.5529	±0.08	0.1081	±0.00	0.0444	±0.00	2.86	±0.02	0.15	±0.02			
		24 H	0.0617	±0.00	0.0248	±0.00	2.5069	±0.20	0.0846	±0.00	0.0489	±0.00	2.27	±0.02	0.12	±0.00			
		48 H	0.0350	±0.03	0.0373	±0.00	0.9393	±0.00	0.0724	±0.00	0.0502	±0.00	2.06	±0.03	0.10	±0.00			
Gauchó + Krilaxyl	AR	12 H	0.0410	±0.00	0.0388	±0.00	1.0567	±0.09	0.0798	±0.00	0.0242	±0.00	2.10	±0.15	0.10	±0.01			
		24 H	0.0360	±0.00	0.0255	±0.00	1.4123	±0.06	0.0615	±0.00	0.0263	±0.00	1.88	±0.06	0.08	±0.01			
		48 H	0.0290	±0.00	0.0329	±0.00	0.8813	±0.02	0.0590	±0.00	0.0314	±0.00	1.73	±0.05	0.06	±0.00			
CONTROL	I+F	12 H	0.0345	±0.00	0.0260	±0.00	1.3269	±0.04	0.0607	±0.00	0.0189	±0.00	2.41	±0.17	0.11	±0.01			
		24 H	0.0249	±0.00	0.0205	±0.00	1.2183	±0.11	0.0455	±0.00	0.0202	±0.00	2.24	±0.07	0.09	±0.00			
		48 H	0.0155	±0.00	0.0180	±0.00	0.8621	±0.02	0.0335	±0.00	0.0236	±0.00	0.09	±0.00	0.03	±0.00			
CONTROL	I+F	12 H	0.0925	±0.00	0.0975	±0.00	0.9487	±0.02	0.1901	±0.00	0.0404	±0.00	9.06	±2.29	0.91	±0.01			
		24 H	0.0712	±0.00	0.0767	±0.00	0.9294	±0.03	0.1486	±0.00	0.0492	±0.00	7.34	±0.41	0.48	±0.03			
		48 H	0.0475	±0.02	0.0486	±0.00	0.9777	±0.02	0.0963	±0.00	0.0563	±0.00	6.21	±0.02	0.23	±0.00			

CHL: Chlorophyll, Tot CHL: Total Chlorophyll, CAR: Carotenoids, WT: Weight, AM: Arithmetic mean, SD: Standard deviation, BR: Below recommended, RD: Recommended dosage, AR: Above recommended, I+F: Insecticide + Fungicide.

Values are represented as Arithmetic mean and standard deviation of three determinants.

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REFERENCES

1. Abdul Baki, A.A. and Anderson, J.D. Vigour determination of Soybean seed by multiple criteria. *Crop. Sci.*, **3**, 630-633 (1973)
2. Anonymous, International rules for seed testing. *Seed Sci. & Technology*, **13**, 421-463 (1995)
3. Arnon, D.I. Copper enzymes in isolated Chloroplast, polyphenoloxidase in *Beta vulgaris*, *Plant Physio.*, **24**, 1-15 (1949)
4. Benjamin, L. Effect of Carbofuran on the germination rate and initial development of seedlings. *Phyto Parasitica*, **14**, 219-230 (1986)
5. Chakravarti, S.K. Effect of Bavistin 25 Ds on germination and seedling vigour of wheat. *Pesticides*, **3**, 23-26 (1986)
6. Chiou, C.H. and Muller, C.H. Allelopathic Mechanism of *Archostaphylos glandulosa* variety *Zazaerisis*, *Am. Mid. Nat.*, **88**, 324-347 (1972)
7. Gaikwad, S.K. and Pawar, V.M., Effect of systemic insecticide on the germination and seedling development of Okra (*Abelmoschus esculentus* (L.) Moench.) *Seed Research*, **7(1)**, 28-33 (1979)
8. Gupta, R.C., Beg, M.U. and Chandel, P.S. Effect of endosulfan on the seed germination and seedling growth of *Vigna radiata* Linn. *Pestology*, **7**, 25-28 (1983)
9. Kamble, A.B. and Sabale, A.B. Influence of Bavistin and Monocrotophos on seed germination and seedling growth of *Trigonella foenum-graecum*, *Poll. Res.*, **18(1)**, 61-65 (1999)
10. Keul, M. The use of studentised range in connection with an analysis of variance. *Euphytica*, 1-112 (1952)
11. Kirk, J.T.O. and Allen, R.I. Dependence of Chloroplast pigment synthesis on protein synthesis, effect of actidione. *Biochemical and Bio-physical research communications*, **21(6)**, 523-530 (1965)
12. Krarup, H.A. and Kivera, S.C. Effect of fungicides and insecticides on the germination and emergence of Peas (*Pisum sativum* L.), *Agro. Sur.*, **10**, 75-78 (1982)
13. Majumdar, S., Ghosh, S., Gliek, B.R. and Dum broff, E.B. Activities of Chlorophyllase, phosphoenolpyruvate carboxylase and ribulose-1,5-bi phosphate carboxylase in the primary leaves of Soybean during senescence and drought. *Physiol. Plant.* **81**, 473-480 (1991)
14. Panduranga Murthy, G. and Leelavathi, S. Effects of insecticidal and fungicidal seed treatments on seedling vigour and chlorophyll content in Red gram and Sunflower during seed germination. *Asian Jr. of Microbiol. Biotech. Env. Sc.*, **4(2)**, 271-275 (2002)
15. Panduranga Murthy, G. and Leelavathi, S. Effects of Xenobiotics on early seedling growth and chlorophyll content in Sunflower cv. 'Morden', During seed germination. *Eco. Env. & Cons.*, **8(2 & 3)**, 133-135 (2002)
16. Patil, T.M. and Shirashyad, V.S. Effect of methylparathion and Phosphamidon on seed germination and on the activity of peroxidase and α -amylase in some vegetable seeds. *Geobios.*, **16(2-3)**, 57-60 (1989)

17. Ramadoss, S. and Sivaprakasam, K. Effect of Cowpea seed treatment with fungicides and insecticides on the seedling vigour. *Madras Agric. Jr.*, **81**, 297-299 (1994)
 18. Ramulu, C.A. and Rao, D. Effect of Monocrotophos on seed germination, growth and chlorophyll content of 'Custard bean', *Comp. Physiol. Ecol.*, **12**, 102-105 (1987)
 19. Reddy, J.M.K. and Vidyavathi, P. Effect of fungicide on the growth and seedling metabolism of *Dolichos biflorus*. *Geobios.*, **10**, 174-178 (1984)
 20. Rodriguez, M.T., Gozatez, M.P. and Linares, J.M. Degradation of chlorophyll and chlorophyllase in senescing barley leaves. *J. Plant. Physiol.*, **129**, 369-374 (1987)
 21. Sabater, B. and Rodriguez, M.T. Control of chlorophyll degradation in detached leaves of Barley and Oat through effect of Kinetin on chlorophyllase leaves. *Physiol. Plant.*, **43**, 274-276 (1978)
 22. Sreenivasa Rao, A. and Rammohana Rao, P. Study on pesticide residues in vegetables. *Poll. Res.*, **19(4)**, 661-664 (2000)
 23. Turner, L.G. and Marshal, C. Accumulation of Zinc by sub cellular fraction of root of *Agrostis tennisi* in relation to Zinc tolerance. *New Phytol.*, **71**, 671-675 (1972)
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