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Research Paper



Effect of Carbendazim on Morphological and **Biochemical parameters of Sorghum bicolor**

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ABSTRACT

Hybrid Sorghum (G-204) seeds were treated with a systemic fungicide Carbendazim in varied concentrations. Morphological and biochemical parameters were analyzed. Multiple sets were prepared for comparision with untreated sets. All parameters were analyzed using standard methods and the results were subjected to 2 tailed "t" test, Pearson's Correlation Matrix and Bray- Curtis similarity index. The data was analyzed to know the effect of Carbendazim on Sorghum seeds. A strong correlation exists between Vigor index and nonreducing sugar, vigor index and total carbohydrates and nonreducing sugar and total carbohydrates in the samples treated with the fungicides. 96% similarity is seen between shoot lengths to root length when Bray-Curtis similarity index was applied.

Keywords : Sorghum bicolor, Carbendazim, Vigor index, biochemical parameters

INTRODUCTION

Sorghum (S. bicolor) is the most important millet crop occupying largest area among the cereals next to rice. It is mainly grown for its grain and fodder. Alternative uses of sorghum include commercial utilization of grain in food industry and utilization of stalk for the production of value-added products like ethanol, syrup and jaggery and bioenriched bagasse as a fodder and as a base material for cogeneration. Sorghum is mostly grown for grain as well as dual purpose for food and fodder. It is extensively cultivated in Mysore district where the excess use of insecticides and pesticides has reduced its yield considerably There is very little information available with regard to the effect of fungicide and other pesticides on pulses and cereals in general and Sorghum in particular. Some of the important studies using fungicide on various plants and that need mention here are (Garcia, 2004) on the "Effect of fungicide Carbendazim on Phenolic metabolism of Tobacco plants"; (Mahfouz, 2011) on "Effect of using Vitavax fungicide on vegetative characteristics"; Reddy and Vidyavathi (1983) on the "Effect of a fungicide on the growth and seedling metabolism of Dolichos biflorus".

Carbendazim is a widely used broad-spectrum benzimidazole fungicide and a metabolite of benomyl.

The fungicide is used to control plant diseases in cereals and fruit, including citrus, bananas, strawberries, pineapples, and pome. (Wight, 2009) It is also controversially used in Queensland, Australia on macadamia plantations. A 4.7% solution of carbendazim hydrochloride is sold as Eertavas and marketed as a treatment for Dutch elm disease. Studies have found that high doses of carbendazim cause infertility and destroy the testicles of laboratory animals.

Maximum pesticide residue limits (MRLs) have reduced since discovering its harmful effects. The MRLs for fresh produce in the EU are now between 0.1 and 0.7 mg/kg with the exception of loquat, which is 2 mg/kg. The limits for more commonly consumed citrus and pomme fruits are between 0.1 and 0.2 mg/kg.

With a view to understand the effect of fungicide Carbendazim on the morphological and biochemical parameters of Sor-

ghum, the present work was undertaken.

MATERIALS AND METHODS.

The seed samples of Hybrid Sorghum bicolor were obtained from Zuari Seeds Pvt Ltd, India sold in local agricultural agency in Mysore district, Karnataka, India. The seeds were treated with thiram to store the seeds for longer time. The seeds were soaked in fungicide at concentrations of 0.05%, 0.1%, 0.2% and 0.3% for 24 hours. Carbendazim 50% W.P a broad spectrum systemic fungicide for cereal crops was purchased from Green crop international Pvt Ltd, Maharastra, sold in local agricultural seeds shop in Mysore. The seeds were allowed to germinate for 10 days on top of the paper method (Nene and Thapliyal, 1993.). The seeds were then analyzed for varied biochemical and morphological parameters. Three sets in each concentration were maintained along with the control for comparison.

Seedling vigor index (SVI) was calculated following modified formula of Abdul-Baki and Anderson (1973); Conductivity test was used to quantify the leakage of electrolytes from the seed coat. (Hendricks, and Taylorson, 1976); Chlorophyll was extracted in 80% acetone and determined by Arnon method. (Arnon, 1949); Reducing sugars were estimated by Dinitrosalicylic acid method (Miller, 1972) ; Total carbohydrates and Starch estimation was followed by the Anthrone method (Hedge, 1962); Total Proteins were estimated using Lowry's method (Lowry et al 1951). The data obtained were subjected to Statistical analysis for conclusive determinations.

RESULTS AND DISCUSSION.

The results of the treatments along with a paired't' test are presented in Table. 1. for the morphological and biochemical parameters. The root length and fresh weight were significantly influenced in 0.3% concentration. The shoot length and root length significantly varied in all the concentrations of fungicide treatments. Fresh Weight of the sample increased considerably in all the concentrations of the treatment. Electrical Conductivity was highly influenced in 0.1% concentration, while in other concentrations it had increased but there was no significant increase in the conductivity of seed leachates.

Carbohydrate content in the class of biomolecules were significantly influenced by the treatment of fungicide and showed positive increase in 0.1% and 0.3%, whereas in the 0.2% concentration the carbohydrate content decreased considerably.

Starch content and the protein content were significantly influenced in all the treatments of fungicide. Chlorophyll content in all the treatments varied slightly and there was no significant increase in the content with varied concentrations.

In order to understand the relations between all the parameters; the data in Table. 1 was subjected to Pearson's correlation matrix. As per the Pearson's correlation matrix a very high significance at 0.01% level is observed between vigor index and nonreducing sugar; vigor index to total carbohy6drates and nonreducing sugars to total carbohydrates. All these parameters were positively correlated to each other. However, correlation at 0.05% level also exists. Percent germination to shoot length; percent germination to root weight; shoot length to root weight; root length to fresh weight; chlorophyll a to total chlorophyll; chlorophyll b to total chlorophyll. All these are positively correlated. These observations indicate that clusters of 2 parameters were predominant in the present study.

In order to understand similarities between the various parameters the data in Table.1 was also subjected to Bray-Curtis similarity index. A "fixed Stopping rule" at 90% similarity was applied. Shoot length and root length were at closest distance (Fig 1). However, 90% and above similarity was represented by leaf weight and total protein, root weight and seed weight, Chlorophyll a and Chlorophyll b. These results pointout that Shoot length and root length along with leaf weight and protein content were the prominent parameters influenced by Carbendazim in Sorghum bicolor (G-204).

According to Siddiqui (2004) an increase in germination values compared to control may be specific response of Z. mays to the treatment of fungicide. The fungicide effect of carbendazim showed slight reduction in the germination rates compared to control seeds.

A study on Hibiscus esculantus and Capsicum annum by Ahmed & Siddiqui, (1995) and Siddiqui et al., (1997) have reported an increase in chlorophyll, protein and carbohydrate contents, but increase in the germination rate has not been reported previously in the seeds treated with benlate fungicide. The present study also shows an increase in the content of protein and carbohydrate and slight decrease in the rate of germination in the Sorghum seeds treated with Carbendazim. The phenomenon also underscores the need for a comparative study of germination behavior of different species in response to a particular fungicide. Increase in fresh weight, seed weight and root length in all the concentrations of Sorghum seeds implies threshold limits of fungicide that can be tolerated by the plants. Use of benlate has also been found to cause an increase in fresh and dry weights of Sesbania ses-

ban at 0.25g/l concentration (Siddiqui et al., 1997). Siddiqui et al., 1999 also reported the inhibition of seed germination and seedling growth in Penesetum americanum L. due to the application of organophosphate insecticides. Shive & Hugh (1976) showed that a fungicide triarimol not only prevented the gibberellin synthesis in Phaseolus vulgaris but also checked the dry weight increase over fresh weights at higher concentrations. The study suggests that a secondary toxicity mechanism non-operative at lesser concentration may become operative at higher concentration to cause a reduction in growth parameters like fresh and dry weight and root and shoot length. Windham & Windham (2004) have reported that systemic fungicides which are based on SBI (sterol biosynthesis inhibitor) are closely related to plant growth regulators the use of which at higher than labeled rates shorten the internodes which may lead to slow shoot growth. Use of high rates of systemic fungicides as seed treatments may also reduce the growth of small grains such as barley and wheat.

According to Pandey (1988), leakage as measured by conductivity is related to membrane disorganization. The lower the membrane integrity, the greater the electrolyte leakage in the steep water, thus the greater the conductivity measurement (Powell et al., 1984). Woodstock et al. (1985) found relationships between weathering deterioration, germination respiratory metabolism, and leaching in cotton seeds. The deterioration of membranes due to weathering was confirmed by electron microscopy of cotyledon's lipids and proteins bodies and correlated well with conductivity measurements.

The percent of germination and seedling vigor index decreased gradually with the advancement in storage of seeds in both the seeds treated and untreated with fungicides (Bapurayagouda et al 2010). Similar results were seen in the present study where the seeds were stored for a month. It may be generalized that treatments with fungicides have a deleterious effect on seeds of various crops.

CONCLUSION

Treatment of Sorghum seeds with Carbendazim influences Root length and fresh weight, Vigor index, and DNA significantly. The influence is more pronounced between the biochemical parameters like chlorophyll a and chlorophyll b and morphological parameters like shoot length and fresh weight. A major effect of the treatment has an impact on root length which inturn may have an influence on their morphological and biochemical parameters.

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Table1: Effect of Carbenda	azim on metabolism	of S	Sorghum	seeds
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Morph	ological Parameters.						
SI No	Parameters	Control	0.05%	0.1%	0.2%	0.3%	
1	Vigor index	3053	3044	3184	2938	3508*	
2	Percent germination	100	93.33	96.67	93.33	96.67	
3	Shoot length (Cms)	18.4	15.6**	17.3*	16.4*	18.0	
4	Root length (Cms)	14.3	17.2*	15.5**	15.3	18.2***	
5	Fresh Weight (g)	1.339	1.508	1.53*	1.458*	1.754***	
6	Leaf weight (g)	0.845	1.062	0.838	1.012	1.068*	
7	Root weight (g)	0.367	0.253	0.316	0.307	0.322*	
8	Seed weight (g)	0.173	0.309**	0.352***	0.339*		
Bioche	emical parameters						
9	Electrical Conductivity mS/cm	0.020	0.020	0.025***	0.030	0.040	
10	Chlorophyll a mg/g	0.006	0.005	0.010*	0.003*	0.005	
11	Chlorophyll b mg/g	0.006	0.008	0.010*	0.002	0.003*	
12	Total Chlorophyll mg/g	0.012	0.012	0.017	0.004**	0.009*	
13	Reducing Sugar mg/g	0.05	0.09**	0.17**	0.10***	0.15**	
14	Nonreducing sugar mg/g	0.14	0.05**	0.16***	0.06**	0.45***	
15	Total Carbohydrates mg/g	0.20	0.15*	0.34***	0.18*	0.60***	

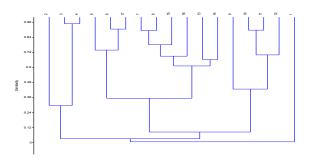
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16	Total Starch mg/g	0.25	0.15**	0.08**	0.17*	0.21**	
17	Total Proteins mg/g	0.53	0.62*	0.85***	1.04**	1.02*	

*= Values are Significant **= Values are very significant ***= values are extremely significant.

Fig 1: Bray- Curtis similarity index for morphological and biochemical parameters of Sorghum seeds treated with carbendazim.



(1. Vigor index 2. Percent germination 3. Shoot length 4. Root length 5. Fresh Weight 6. Leaf weight 7. Root weight 8. Seed weight 9. Electrical Conductivity 10. Chlorophyll a 11. Chlorophyll b 12. Total Chlorophyll 13. Reducing sugar 14. Non-reducing Sugar 15. Total Carbohydrates 16. Total Starch 17. Total Proteins.)

Table 2: Pearson's Correlation Matrix for Morphological and Biochemical parameters of Sorghum seeds treated with Carbe	ndazim.

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
1	1																
2	.294	1															
3	.504	.922*	1														
4	.692	399	230	1													
5	.869	215	.056	.892*	1												
6	.236	687	506	.801	.582	1											
7	.159	.880*	.925*	552	265	643	1										
8	.239	760	515	.562	.656	.546	607	1									
9	.727	107	.276	.590	.827	.493	.099	.580	1								
10	.226	.439	.298	197	.001	714	.192	093	266	1							
11	104	.161	134	157	217	528	203	177	652	.831	1						
12	.182	.427	.189	097	059	603	.046	258	458	.928*	.938*	1					
13	.622	173	.030	.469	.744	.058	183	.739	.577	.505	.189	.296	1				
14	.967**	.372	.626	.591	.804	.226	.329	.159	.804	.078	311	.001	.507	1			
15	.973**	.264	.537	.614	.870	.206	.236	.333	.837	.189	227	.063	.688	.974**	1		
16	.100	.474	.514	069	148	.136	.536	625	.130	569	603	456	665	.289	.072	1	
17	.369	411	041	.364	.632	.401	082	.817	.858	229	572	513	.654	.428	.545	233	1

**.Correlation is significant at the 0.01 level (2-tailed). *.Correlation is significant at the 0.05 Level (2-tailed).

(1. Vigor Index 2. Percent Germination 3. Shoot length 4. Root Length 5. Fresh weight 6. Leaf weight 7. Root Weight 8. Seed Weight 9. Electrical Conductivity 10. Chlorophyll a 11. Chlorophyll b 12. Total Chlorophyll 13. Reducing Sugar 14. Nonreducing Sugar

15. Total Carbohydrates 16. Total Starch 17. Total Proteins.)

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