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Effect of pre harvest phytosanitation spray on seed quality of soybean (*Glycine max* L.)

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Abstract
The present investigation was undertaken with main objective to determine the effect of pre harvest phytosanitation spray on seed quality of soybean. The foundation seeds of soybean variety of Dsb-21 were used for of crop rising. The crop was grown and pre harvest phytosanitation spray was carried at 15 days before harvesting in Seed Technological Research (STR) experimental site of Seed Unit and laboratory experiments were conducted NSP and Department of seed Science and Technology, University of Agricultural Sciences Dharwad. The five different pesticides along with one control [P_1: Hexaconazole 5%SC (2ml/L), P_2: Carbendazim 50%WP (0.5g/L), P_3: Nativo 75% WG {Tebuconazole 50% + Trifloxystrobin 50%} (0.5g/L), P_4: Streptocycline (0.15g/L) + COC 50%WP (2.5g/L), P_5: Spinosad 45%SC (0.2ml/L), P_6: Control] were used as treatments. Among different pesticides, spraying with nativo showed highest germination percentage (89.92%) and highest seedling vigour indices {SVI-I=3302 & SVI-II=7609}. The lowest disease incidence in blotter paper method (15.33%) and agar plate method (3.33%) was found in nativo and streptocycline spray. Lowest insect damaged (3.08%) seeds were observed in nativo and spinosad pre harvest spray respectively.

Keywords: Soybean, pre harvest, phytosanitation, spray

1. Introduction

Soybean (*Glycine max* L.) with chromosome number 2n=40, of the family Fabaceae, is a common legume plant cultivated for more than 3000 years, present day soybean has evolved from *Glycine ussuriensis* a wild legume native to Northern China. In India, annual yield losses due to various diseases are estimated to be 12% of total production. Over a hundred pathogens are known to affect soybean, of which 66 fungi, six bacteria and eight viruses have been reported to be associated with soybean seeds (Sinclair, 1978) [20]. By definition, Seedborne pathogens are any infectious agent associated with seeds that have the potential to cause seed, seedling, and plant diseases (Agarwal and Sinclair, 1996) [2]. Plant pathogenic bacteria, fungi, nematodes, and viruses occur with seed either as contaminants adhering to the seed surface, loosely mixed with seed or as infection inside the seed tissues (Neergaard, 1979) [14].

Seed borne infection or infected seed is a very important discouraging factor, which possesses a serious problem in seed certification. The infection of seed caused by various fungal pathogens *viz*. purple seed stain (*Cercospora kikuchii, C. sojina*), anthracnose (*Colletotrichum truncatum*), pod and stem blight (*Phomopsis* spp.) and *Fusarium* seed infection on soybean seeds, once used for diagnostic purposes, is now being used as indicative of quality. Seedborne pathogens affect seed quality and cause diseases that significantly impact the yield and marketability of seed lots (Machado *et al.*, 2002; Mathur and Kongsdal, 2003) [9, 12].

In pulses, the main pests and diseases of stored seeds are field carry over pests as they are infest or infect in the field before harvest and get manifested during storage and causing pronounced loss. Diseases like purple seed strain, anthracnose, soybean mosaic, pod blight, fusarium wilt and insect pests like lesser grain borer, rice weevil, bruchids and psylids are carried to storage from field infestation. As per the ancient adage, "Prevention is better than cure", controlling these pests and diseases in the field prevents them from entering godowns and spreading further to healthy seeds. Pre-harvest phytosanitary spray is a method to arrest or remove these pathogens or insects in the field itself thereby delimiting the damage during storage. It involves the spraying of fungicides and/or insecticides during the seed formation and development at needy concentrations.

2. Material and Methods

The seed production was carried out at plot No. 201, H-Block of Seed Technological Research

(STR) experimental site of Seed Unit during *rabi summer* of 2020-21. Laboratory experiments were conducted at Seed Testing Laboratory (STL) of National Seed Project (NSP) and Department of Seed Science and Technology, College of Agriculture, University of Agricultural Sciences, Dharwad. The foundation seeds of soybean variety of Dsb-21 were used for of crop rising. The crop was grown and pre harvest phytosanitation spray was carried at 15 days before harvesting.

2.2 The following pesticides were used as the treatments for pre harvest phytosanitation spray along with control *i.e,* no pre harvest spray

- P_1: Hexaconazole 5% SC (2ml/L)
- P_2: Carbendazim 50%WP (0.5g/L)
- P_3: Nativo 75% WG {Tebuconazole 50% + Trifloxystrobin 25%} (0.5g/L)
- P_4: Streptocycline (0.15g/L) + COC 50%WP (2.5g/L)
- P_5: Spinosad 45%SC (0.2ml/L)
- P_6: Control

The selected chemicals as per treatments were sprayed at 80 DAS *i.e.* when the crop was harvested 15 days after spraying of chemicals. The chemicals were sprayed according to dosage as per the treatments. The harvesting was done at maturity by uprooting the whole plant. The plants from each net plot were collected, threshed and were processed manually for extraction of seeds.

Standard germination test was conducted as per the procedure given by International Seed Testing Association (Annon., 2018) [3]. Further observation on seed quality parameters such as seedling shoot length (cm), root length (cm), seedling dry weight (mg/seedling), seedling vigour index values were measured and analysed statistically. The seedling vigour index was calculated as the formula suggested by (Abdul-Baki and Anderson, 1973) [1] by multiplying the germination per cent with seedling shoot length and root length (cm). The speed of germination was calculated by using the formula as suggested by (Maguire, 1962) [10]. Electrical conductivity of seed leachate was determined by using five grams of seeds, which were surface sterilized with acetone for half a minute and thoroughly washed in distilled water three times. 25 ml of distilled water was added to seeds and kept in the incubator maintained at 25 °C ± 1°C temperature for twelve hours. The electrical conductivity of the seed leachate was measured in the digital conductivity bridge (ELICO) with a cell constant 1.0 and the mean values will be expressed in Deci Simons per meter (dSm^{-1}) (Milosevic *et al.*, 2010) [13].

The seeds were selected randomly and observed for the disease and insect pest incidence. The number of seeds infected or infested from the total number of seeds observed is recorded, and then expressed in percentage.

$$\text{Diseased or pest damaged seeds (\%)} = \frac{\text{Number of diseased or pest damaged seeds}}{\text{Total number of seeds observed}} \times 100$$

Disease pathogens – Standard blotter paper method and agar plate method.

Insect pests – Visual examination

The RCBD Design was followed with 6 treatments and 6 replications (Gomez, 1984) [6] method of variance was applied for the analysis and interpretation of the experimental data of present investigation.

3. Results

3.1 Seed quality parameters

There was a significant difference in germination percentage, root length, shoot length, seedling dry weight and seedling vigour indices. And the results are in favour with the pre harvest sanitation spray compared to control. Speed of germination, EC, Seed volume and seed moisture didn't had any effect from the pre harvest spray. The highest germination percentage (89.92%) was found with nativo (P_3) pre harvest spray and all other pre harvest spray treatments were on par with P_3 and control (P_6) had lowest germination percentage (83.4%). The longest root length (20.85 cm) and shoot length (16.66 cm) was found in streptocycline + COC (P_4) and carbendazim (P_2) pre harvest phytosanitary spray respectively, and other treatments were on par with P_4 and P_2 respectively, and short root length (18.52 cm) and shoot (14.80cm) was found in control. The high (84.68 mg) seedling weight was found in nativo treatment (P_3) which was on par with other treatments and lowest (75.87 mg) was found in control (P_6). In the seedling vigour indices, the highest SVI-I (3302) and SVI-II (7609) was found in nativo (P_3), and lowest SVI-I (2782) and SVI-II (6327) was found in control (P_6).

Table 1: Influence of pre harvest phytosanitary spray on germination percentage, speed of germination and electrical conductivity.

	Treatments	Germination %	Speed of germination	Electrical conductivity(dS/m)
P_1	Hexaconazole 5%SC (2ml/L)	87.75 (69.90)	0.32	18.52
P_2	Carbendazim 50%WP (0.5g/L)	89.00 (70.89)	0.32	18.40
P_3	Nativo 75% WG (0.5g/L)	89.92 (71.88)	0.31	18.97
P_4	Streptocycline (0.15g/L) + COC 50%WP (2.5g/L)	88.00 (69.96)	0.31	18.30
P_5	Spinosad 45%SC (0.2ml/L)	86.17 (68.37)	0.31	18.17
P_6	Control	83.42 (66.12)	0.34	17.76
	S.Em ±	0.98	0.01	0.46
	C.D at 1%	4.03	N.S	N.S
	C.V	3.47	4.90	6.17

*Figures in the parentheses indicate arcsine root transformed values.

Table 2: Influence of pre harvest phytosanitary spray on average root length (cm), shoot length (cm), average seedling dry weight (mg), seedling vigour index-I (SVI-I) and seedling vigour index-II (SVI-II.

	Treatments	Root length (cm)	Shoot length (cm)	Seedling dry weight (mg)	SVI - I	SVI - II
P_1	Hexaconazole 5%SC (2ml/L)	19.98	16.42	79.68	7000	79.68
P_2	Carbendazim 50%WP (0.5g/L)	20.10	16.66	83.18	7403	83.18
P_3	Nativo 75% WG (0.5g/L)	20.43	16.21	84.68	7609	84.68

P_4	Streptocycline (0.15g/L) + COC 50%WP (2.5g/L)	20.85	16.58	82.75	7303	82.75
P_5	Spinosad 45% SC (0.2ml/L)	19.89	16.20	79.70	6893	79.70
P_6	Control	18.52	14.80	75.87	6327	75.87
	S.Em ±	0.37	0.32	1.36	136	1.36
	C.D at 1%	1.52	1.33	5.57	557	5.57
	C.V	4.54	4.93	4.11	3.34	4.70

3.2 Seed borne disease and insect pest incidence

The disease incidence was observed by two methods *i.e.*, standard blotter paper method and agar plate method. The lowest disease incidence in blotter paper method (15.33%) and agar plate method (3.33%) was found in nativo (P_3) and streptocycline + COC (P_4) respectively, while highest in blotter paper method (27.67%) and agar plate method (17.08%) was found in control (P_6) treatment. And the results from the pre harvest spray of other treatments. From the results the standard blotter method showed superior results than agar plate method.

The insect pest incidence was lower (3.08%) in spinosad treatment (P_5) and highest (5.00%) was found in control. This may because the pre harvest insecticidal spray had controlled the occurrence of insect pests such as bruchids. This result is in support with the findings of (Ranganatha, 2011; Malarkodi and Srimathi, 2007) [11, 16] in cowpea and green gram.

Table 3: Influence of seed rate and pre harvest phytosanitary spray on seed borne diseases (Blotter paper method and Agar plate method) and seed infestation.

	Treatments	% Diseased seeds		% Seed damaged or infested by insect pests.
		Blotter paper method	Agar plate method	
P_1	Hexaconazole 5%SC (2ml/L)	18.00 (27.26)	8.75 (17.16)	4.42 (12.11)
P_2	Carbendazim 50%WP (0.5g/L)	16.67 (23.81)	8.75 (17.16)	4.17 (11.77)
P_3	Nativo 75% WG (0.5g/L)	15.33 (21.93)	8.33 (16.74)	4.08 (11.63)
P_4	Streptocycline (0.15g/L) + COC 50%WP (2.5g/L)	17.00 (30.62)	3.33 (10.37)	4.00 (11.52)
P_5	Spinosad 45%SC (0.2ml/L)	23.67 (31.27)	12.50 (20.70)	3.08 (10.05)
P_6	Control	27.67 (36.46)	17.08 (24.37)	5.00 (12.91)
	S.Em ±	0.78	0.47	0.28
	C.D at 1%	3.18	1.94	1.15
	C.V	6.65	6.53	5.91

*Figures in the parentheses indicate arcsine root transformed value.

4. Discussion

4.1 Seed quality parameters

The reason for low performance of seeds in control might be due to presence of high disease or may be due to high insect damage. The disease pathogens affect seeds through degrading stored food materials and rotting or degrading the internal and embryonic tissues or immediately after germination the seedlings affected with disease rot or die. Insect pests damage seeds through feeding on the seeds. The pre harvest phytosanitary spray controlled some of the seed borne diseases and insect pests that are carried from the field. The obtained results are in support with, (Prasanth and Patil, 2012; Begum *et al*, 2008; Divyashree, 2007; and Kumari *et.al*, 2002) [4, 5, 8, 15] in soybean and green gram.

4.2 Seed borne disease and insect pest incidence

The reduction in diseases in pre harvest sprayed treatments might be due to fungi toxic and antibiotic effect of fungicides and bactericide. The results can be supported with (Kumar, 2019; Shaha and Choudary, 2016; Rao *et.al*, 2015; Prasanth and Patil, 2012; and Satya, 2004) [7, 18, 19] in soybean and pigeon pea.

The reduction in pest damaged seeds is because of the pre harvest insecticidal spray had controlled the occurrence of insect pests such as pod borers. This result is in support with the findings of (Ranganatha, 2011; Malarkodi and Srimathi, 2007) [11, 16] in cowpea and green gram.

The disease and pest incidence in seeds was lower in pre harvest sprayed treatments compared to control because of protective and curative action of chemicals.

4.3 The fungicides act as

- **Respiration inhibitors:** (Succinate dehydrogenase inhibitors): prevents the germination of fungal spores. (E.g. Carbendazim)
- **Quinone inhibitors (Qols):** Prevent ATP production in germinating spores. (E.g. trifloxystrobin).

The quinone inhibitors (Qols) along with controlling pathogens, they also help in plants by triggering physiological responses like lowering the ethanol production which is a stress hormone, increasing the chlorophyll content and increasing lignifications of cell walls. (Russel, 2009) [17].

- **Sterol biosynthesis inhibitors / demethylation inhibitors:** Disrupt or rupture membrane of fungi cells, organelles after spore germination. (E.g. hexaconazole, tebuconazole)
- **Bactericide:** (Streptocycline): It works by blocking the ability of 30S ribosomal subunits to make proteins, which results in bacterial death.

Spinosad: alter the function of nicotinic and GABA-gated ion channels, causing rapid excitation of the insect nervous system, leading to involuntary muscle contractions, tremors, paralysis and death.

5. Conclusion

The preharvest phytosanitary spray didn't had any effect on plant growth parameters since the spraying was done at 15 days before harvesting, but influenced seed quality parameters significantly compared to control. The highest germination percentage (89.92%), seedling vigour indices (SVI-I=3302, SVI-II=7609), lower disease incidence in seeds (15.33% - blotter paper method) was observed with nativo spray and less insect pest damaged seeds (3.08%) with spinosad spray.

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