Australian Journal of Crop Science

AJCS

AJCS 16(04):539-544 (2022) ISSN:1835-2707
doi: 10.21475/ajcs.22.16.04.p3563

Industrial treatment of soybean seeds subjected to high volumes of fungicides, insecticides, biostimulants and micronutrients during storage

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Abstract

The objective of this study was to examine the performance of soybean seeds at different storage periods as well as their physiological quality after the application of spray mixtures of insecticides, fungicides, polymers, micronutrients and biostimulants. The experiment was laid out in a completely randomized design with four replicates. Treatments were as follows: T1 - untreated seeds (control); T2 - insecticide and fungicide + polymer + drying powder; T3 - insecticide and fungicide + polymer + drying powder + micronutrient; and T4 - insecticide and fungicide + polymer + drying powder + micronutrient + biostimulant. For each industrial seed treatment (IST), the specific spray volumes tested were 0, 400, 600 or 1100 mL 100 kg seeds^{-1}. Soybean seeds were stored for 0, 15, 30, 45, 60 and 90 days and subsequently evaluated for physiological potential. At each storage period, the following tests were conducted: germination, first count, accelerated aging, emergence speed index, final emergence in sand substrate, whole-seedling length, shoot length and root length. Industrial seed treatment reduces seed physiological quality. The seed treatment corresponding to the highest spray volume provided the lowest means in all tested treatments. For all analyzed variables, the ST4 treatment, to which biostimulant was added, presented the lowest averages, indicating that the greater the volume of spray, the greater the damage caused to the physiological quality of the seeds, both before and after storage.

Keywords: Deterioration; *Glycine max* (L) Merr.; Germination; Pesticides, Physiological quality.
Abbreviations: IST_industrial seed treatment; SGT_standard germination test; FC_first count, ESI_emergence speed index; FE_final emergence in sand substrate; AA_accelerated aging; SP_storage period; TT_time of treatment; DF_degrees of freedom; CV_coefficient of variation; RL_root length; SL_shoot length; WSL_whole-seedling length; ST1_treatment 1; ST2_treatment 2; ST3_treatment 3; ST4_treatment 4.

Introduction

Grown in several regions of Brazil, the soybean crop stands out as the most economically important agricultural commodity in the agribusiness sector. Today, Brazil is considered the world's largest grain producer. The 2020/2021 harvest yield is estimated to have reached values close to 135.91 million tons, which represents an 8.9% increase compared with the previous harvest, in addition to exports corresponding to approximately 83.61 million tons (CONAB, 2021).

Industrial seed treatment (IST) has become a routine and essential practice, since sowing in appropriate soil-climatic conditions hardly takes place. In this respect, it is essential to highlight the importance of using chemical products as well as seeds of high physiological and sanitary quality, since vigorous seeds free of pathogens and pests stand uniformly, and exhibit superior seedling performance and high yields (Henning et al., 2005 and Henning et al., 2010).

The use of products such as pesticides, micronutrients, biostimulants, polymers and inoculants in combinations in IST provides nutrients and acts on the physiology and protection of seedlings (Ludwig et al., 2011; Castro et al., 2008; Binsfeld et al., 2014; Dan et al., 2012; França-Neto et al., 2015). In this scenario, Sfredo and Oliveira (2010) observed numerous benefits from the incorporation of micronutrients, as the insertion of cobalt and molybdenum into IST favors the symbiosis between soybean seeds and bacteria of the genus *Bradyrhizobium* spp., resulting in successful biological nitrogen fixation.

In addition to chemical products for protection and nutrient supply, the inclusion of biostimulants during IST offers advantages such as increasing the amounts of oils and proteins in the seeds, as well as greater effectiveness under unfavorable weather conditions in the field. Pereira et al. (2020) found that insecticides and fungicides contribute to deterioration and loss of vigor of seeds, when associated with storage.

Seed deterioration is considered a continuous and irreversible process that begins with physiological maturation (Delouche and Baskin, 1973). In this scenario, IST plays an important role. After treatment, seeds are subjected to storage to maintain the physiological quality of the lots until the time of sale (Carvalho and Nakagawa, 2012). Despite being considered a process inherent to maturation, metabolic changes are triggered according to factors such as temperature, relative humidity and the presence of insects and microorganisms (Marcos-Filho, 2015). Among the main factors that affect physiological potential, storage coupled with the composition and volume of the spray mixture, can be considered decisive factors for the maintenance of physiological potential (Pereira et al., 2021).

The hypothesis of this study tested association of high spray volumes of different chemical products with detrimental effects on seed physiological quality, especially during storage. Therefore, the present study proposed to investigate the performance of soybean seeds BMX Alvo RR at different storage periods as well as their physiological quality after the application of sprays based on insecticides, fungicides, polymers, micronutrients and biostimulants.

Results and discussion

The Shapiro-Wilk and Bartlett tests were initially applied and confirmed the hypotheses of normality and homogeneity of variances (p-value > 0.05). Table 2 shows the results of the F test for the analysis of variance. All variables under study showed significance (p-value < 0.05) for both the main effects and the double and triple interactions. In other words, it was necessary to decompose the analyzed factors to identify where statistically significant differences were present.

Considering each storage period, both before and after IST, significant differences were detected between the seed treatments for SGT and FC, with ST4 providing the lowest means at all evaluated periods (Table 2). In this respect, it is essential to highlight that IST with high spray volumes (1100 mL 100 kg seeds^{-1}) compromises the maintenance of seed quality, especially during storage. However, the results obtained in this study disagree with those described by Abati et al. (2020), Segalin et al. (2013) and Santos et al. (2018), who found that spray volumes below 1200 and 1400 mL 100 kg seeds^{-1} did not affect the quality of soybean seeds.

In the comparison between times of treatment (Table 3) within each storage period, significant differences were detected for the treatments. In addition, longer storage periods resulted in lower means for the variables under analysis, both before and after IST. After IST, the means of the analyzed variables were higher as compared with the values measured before IST.

For each storage period, before and after industrial seed treatment, emergence speed index, final emergence in sand substrate and accelerated aging differed significantly between the treatments, whereas ST4 recorded the lowest means in most periods (Table 4). These results corroborate with Lemes et al. (2019), who observed a decrease in emergence as well as an increase in abnormal seedlings after the accelerated aging test in soybean seeds subjected to IST.

Statistically significant differences were observed between the times of treatment, considering the seed treatments at each storage period. Finally, longer storage periods resulted in lower means for the analyzed variables, both before and after IST. After IST, the means of the variables increased, as compared with the condition prior to treatment. Marcos Filho and Souza (1983) found discordant results, whereas the treatment of soybean seeds with fungicides favored the maintenance of vigor before storage. Conversely, Da Silva et al. (2014) stated that seed performance after 90 days of storage was considered superior and more efficient.

According to the F-test of analysis of variance, there were significant differences between the treatments (p-value < 0.05), due to both the main effect and the double and triple interaction effects. In other words, it was necessary to decompose the analyzed factors to identify, where the statistically significant differences occurred.

Analysis of each storage period showed significant differences before and after IST on root length, shoot length and whole-seedling length (Table 6). Treatment ST4 recorded the lowest means of the respective variables. Brzezinski et al. (2017), reported that high spray volumes negatively affect physiological potential, especially in low-vigor seeds, since the deleterious effect is maximized under such conditions. Regarding seedling length, results by Abati et al. (2020) pointed out that increases in spray volume reduce the physiological quality of soybean seeds, when associated with the application of drying powder in IST.

Considering each storage period, significant differences were observed between the tested treatments in times of treatment. Longer storage periods were found to result in lower means for the studied variables, both before and after seed treatment. Krohn and Malavasi (2004) suggested that after seed treatment, physiological performance is superior, and the quality reduction is only observed after four months, due to the phytotoxicity of the applied products.

Materials and Methods

Plant materials

For the establishment of the trials, 2.5 kg of unconventional seeds with transgenic technology were used. Seed treatment was carried out in an industrial unit belong to a private company.

Conduction of the study

For the treatment, the seeds were coated in a continuous seed coating machine and subsequently packed in Kraft paper bags that were kept in environmental conditions in the laboratory, at a temperature of 25 °C and 65% relative humidity. The experiment was laid out in a completely randomized design, in a 2 × 4 × 6 factorial arrangement with four replicates, totaling 48 treatments. Treatments were defined as shown in Table 1.

Evaluated characteristics

Physiological quality was assessed by the following tests: germination test (Brasil, 2009), first germination count (Brasil, 2009), accelerated aging test (Marcos-Filho, 2020), emergence speed index (Maguire, 1962) final emergence in sand substrate (Nakagawa, 1999), whole-seedling length (Abati et al., 2014), shoot length (Abati et al., 2014) and root length (Abati et al., 2014). The evaluations were performed at the storage periods of 0, 15, 30, 45, 60 and 90 days.

Statistical analysis

The obtained data were analyzed using R software version 4.0.2 (R Core Team, 2020). The assumptions of normality and homogeneity of variances of the variables were checked by the Shapiro-Wilk and Bartlett tests. The F test of analysis of variance was applied to detect differences between treatments, times of treatment and storage periods.

Table 1. Products and spray volumes used in industrial seed treatment.

Treatment	Chemical product	Concentration	Spray volume (mL 100 kg^{-1})
ST1	-	-	Control (no treatment)
ST2	Insecticide + Fungicide[1] Polymer[2] Drying powder[3]	200 mL100 kg^{-1} seeds 200 mL100 kg^{-11} seeds 150 mL100 kg^{-11} seeds	400
ST3	Insecticide + Fungicide[1] Polymer[2] Drying powder[3] Micronutrient[4]	200 mL100 kg^{-11} seeds 200 mL100 kg^{-11} seeds 150 mL100 kg^{-11} seeds 200 mL 100kg^{-11} seeds	600
ST4	Insecticide + Fungicide[1] Polymer[2] Drying powder[3] Micronutrient[4] Bioestimulant[5]	200 mL100 kg^{-11} seeds 200 mL100 kg^{-11} seeds 150 mL100 kg^{-11} seeds 200 mL 100kg^{-11} seeds 0.5 L 100 kg^{-11} seeds	1100

[1]Inseticidee + Fungicide = pyraclostrobin + thiophanate-methyl; [2]Polymer = film formulation; [3]Drying powder = film formulation; [4]Micronutrient = cobalt + molybdenum; [5]Bioestimulant = kinetin + gibberellic acid + 4-indole-3-ylbutyric acid.

Table 2. Analysis of variance for the variables of standard germination test (SGT), first count (FC), emergence speed index (ESI), final emergence in sand substrate (FE) and accelerated aging (AA).

Source of variation	DF	Mean square				
		SGT (%)	FC (%)	ESI	FE (%)	AA (%)
TT	1	216*	276*	100.16*	252*	1541*
ST	3	3506*	3823*	24.43*	3322*	7553*
SP	5	6350*	4662*	253.50*	22790*	16706*
TT × ST	3	58*	59*	4.70*	777*	297*
TT × SP	5	60*	47*	2.04*	216*	88*
ST × SP	15	90*	268*	8.47*	722*	320*
TT × ST × SP	15	23*	15*	1.55*	308*	29*
Residual	144	13	15	0.34	42	17
CV (%)	-	7.97	4.68	8.12	8.44	11.36
Overall mean	-	49.61	83.28	7.17	76.89	35.83

*Considered significant if p-value < 0.05 by the F test; SP: storage period; TT: time of treatment; ST: seed treatment; DF: degrees of freedom and CV: coefficient of variation (%).

Table 3. Mean values of percentage of normal seedlings in the standard germination test (SGT) and first count (FC) of germination of soybean seeds as a function of time of treatment, decomposed within the storage periods (SP) and seed treatments (ST)*.

SP (days)	ST	SGT (%)		FC (%)	
		Before IST	After IST	Before IST	After IST
0	ST1	76.50 aA	76.50 aA	100.00 aA	100.00 aA
0	ST2	67.50 bAB	74.00 aA	93.50 bA	98.50 aA
0	ST3	62.50 bBC	70.00 aA	92.50 bA	97.50 aA
0	ST4	55.00 aC	56.50 aB	90.00 aB	91.00 aB
15	ST1	66.50 aA	66.50 aA	98.00 aA	98.00 aA
15	ST2	61.00 bAB	67.00 aA	93.00 bB	97.50 aA
15	ST3	57.00 aB	57.50 aB	87.00 bC	94.50 aA
15	ST4	48.00 aC	49.50 aC	80.50 bD	87.00 aB
30	ST1	56.50 aB	56.50 aB	93.00 aA	93.00 aA
30	ST2	57.50 aA	60.50 aA	90.00 bB	93.50 aA
30	ST3	44.00 bC	53.00 aC	84.00 bC	90.00 aA
30	ST4	43.50 bD	46.50 aD	76.50 bD	81.50 aB
45	ST1	51.00 aB	51.00 aB	89.50 aA	89.50 aA
45	ST2	55.50 aA	55.50 aA	84.50 bB	90.00 aA
45	ST3	49.50 aB	49.50 aB	82.00 aB	85.50 aA
45	ST4	36.50 bC	45.00 aC	68.00 bC	77.50 aB
60	ST1	41.00 aB	41.00 aB	86.50 aA	86.50 aA
60	ST2	50.50 aA	49.50 aA	82.50 aAB	85.00 aA
60	ST3	35.00 bB	43.50 aAB	78.50 aB	79.50 aA
60	ST4	27.50 aC	21.50 bC	58.50 bC	68.00 aB
90	ST1	33.00 aB	33.00 aA	78.50 aA	78.50 aA
90	ST2	45.50 aA	34.50 bA	77.50 aAB	68.00 bA
90	ST3	31.00 aB	28.50 aA	61.00 bB	67.00 aA
90	ST4	13.00 aC	7.50 bB	34.00 aC	33.50 aB

*Means followed by different lowercase letters in the columns and uppercase letters in the rows differ from each other by Tukey's test (p-value < 0.05).

Table 4. Mean values of emergence speed index (ESI), final emergence in sand substrate (FE) and accelerated aging (AA) of soybean seeds as a function of time of treatment, decomposed within storage periods (SP) and seed treatments (ST)*.

SP (days)	ST	ESI		FE (%)		AA (%)	
		Before IST	After IST	Before IST	After IST	Before ST	After IST
0	ST1	10.33 aB	10.33 aB	100.00 aA	100.00 aA	79.50 aA	79.50 aA
0	ST2	11.48 aA	10.98 aA	100.00 aA	100.00 aA	77.50 aA	78.00 aAB
0	ST3	10.55 aAB	10.50 aB	100.00 aA	99.00 aA	66.00 aB	70.50 aB
0	ST4	9.92 aB	9.26 bC	94.00 bB	97.00 aB	40.00 aC	44.00 aC
15	ST1	9.00 aA	9.00 aAB	94.00 aA	95.00 aA	54.00 aA	54.00 aC
15	ST2	9.08 bA	9.52 aAB	98.00 aA	96.00 aA	63.00 bA	72.00 aA
15	ST3	9.30 bA	9.88 aA	99.00 aA	96.00 bA	57.50 bA	66.00 aB
15	ST4	8.49 bB	8.79 aB	93.00 aB	93.00 aB	26.50 aB	28.50 aD
30	ST1	6.67 aB	8.46 aA	88.00 aAB	92.00 aA	45.00 aA	45.00 aB
30	ST2	8.64 aA	8.57 aA	96.00 aA	95.00 aA	45.00 bA	64.50 aA
30	ST3	8.63 bA	9.16 aA	95.00 aA	91.00 bA	48.00 bA	58.00 aA
30	ST4	8.03 aA	6.67 bB	82.00 aB	82.00 aB	19.00 aB	23.50 aC
45	ST1	6.63 aB	6.63 aA	79.00 bB	88.00 aA	35.00 aA	35.00 aA
45	ST2	8.27 aA	8.21 aA	92.00 aA	90.00 aA	29.50 bA	47.50 aA
45	ST3	7.79 bA	7.92 aA	92.00 aA	89.00 bA	27.00 bA	39.00 aA
45	ST4	5.12 aC	5.12 aB	59.00 aC	59.00 aB	5.50 bB	19.00 aB
60	ST1	5.03 aB	6.48 aAB	58.00 bB	77.00 aA	26.00 aA	26.00 aA
60	ST2	7.50 aA	7.61 aA	87.00 aA	82.00 bA	14.50 bB	27.50 aA
60	ST3	6.83 bA	5.74 aB	85.00 aA	65.00 bA	12.50 aB	17.00 aB
60	ST4	3.83 aB	3.83 aC	41.00 aC	41.00 aB	2.50 abC	1.00 aC
90	ST1	2.36 aB	2.36 aA	23.00 aB	23.00 aAB	4.00 aB	4.00 aBC
90	ST2	6.62 aA	2.70 bA	78.00 aA	24.00 bAB	11.00 bA	20.50 aA
90	ST3	1.55 bB	1.96 aAB	21.00 bB	29.00 aA	3.00 bB	8.00 aB
90	ST4	1.10 aB	0.70 bB	15.00 aB	10.00 bB	0.50 aB	0.00 aC

*Means followed by different lowercase letters in the columns and uppercase letters in the rows differ from each other by Tukey's test (p-value < 0.05).

Table 5. Analysis of variance for root length (RL), shoot length (SL) and whole-seedling length (WSL).

Source of variation	DF	Mean square		
		RL	SL	WSL
TT	1	32.20*	0.85*	43.30*
ST	3	166.70*	4.36*	220.20*
SP	5	391.80*	67.45*	676.20*
TT × ST	3	6.90*	2.59*	8.20*
TT × SP	5	4.10*	1.53*	3.60*
ST × SP	15	1.80*	0.75*	13.70*
TT × ST × SP	15	0.70*	0.33*	7.20*
Residual	192	0.50	0.12	1.10
CV (%)	-	5.48	5.34	5.53
Overall mean	-	12.24	6.38	18.62

*Considered significant if p-value < 0.05 by the F test; SP: storage period; TT: time of treatment; ST: seed treatment; DF: degrees of freedom; and CV: coefficient of variation (%).

Table 6. Mean values of root length (RL), shoot length (SL) and whole-seedling length (WSL) as a function of time of treatment, decomposed within storage periods (SP) and seed treatments (ST).

SP (days)	ST	RL		SL		WSL	
		Before IST	After IST	Before IST	After IST	Before IST	After IST
0	ST1	16.60 aA	16.60 aAB	8.34 bA	8.64 aAB	24.12 aA	24.12 aB
0	ST2	14.98 bA	17.72 aA	7.66 bA	9.44 aA	24.14 bA	27.04 aA
0	ST3	16.36 aA	16.40 aAB	7.82 bA	8.78 aA	23.76 aA	25.06 aAB
0	ST4	14.36 aB	15.36 aB	7.54 aB	7.66 aB	21.96 aB	23.98 aB
15	ST1	15.10 aB	15.10 aAB	7.16 aAB	7.16 aB	22.06 aAB	22.06 aB
15	ST2	15.68 aA	16.08 aA	7.24 bA	8.28 aA	22.88 bA	24.12 aA
15	ST3	15.02 aB	14.66 aB	7.22 bA	7.64 aB	21.88 aB	22.14 aB
15	ST4	9.68 bC	12.64 aC	6.96 aB	7.06 aB	18.68 aC	19.46 aC
30	ST1	13.86 aB	13.86 aB	6.56 aAB	6.56 aB	20.02 aB	20.02 aB
30	ST2	14.94 aA	15.02 aA	6.90 bA	7.34 aA	18.96 bA	22.10 aA
30	ST3	13.96 aB	11.36 bB	6.48 aB	6.38 aB	18.62 aB	19.50 aB
30	ST4	10.00 aC	8.36 bC	6.47 aB	6.16 bB	16.84 aC	17.62 aC

45	ST1	12.10 aA	12.10 aB	6.10 aB	6.10 aA	18.70 aA	18.70 aB
45	ST2	11.32 bA	13.92 aA	6.92 aA	6.14 bA	19.80 aA	20.38 aA
45	ST3	12.48 aA	12.38 aB	6.16 aB	6.00 aA	14.30 bB	18.56 aB
45	ST4	7.52 bB	9.76 aC	5.74 aC	5.42 aB	13.50 aB	15.32 aC
60	ST1	10.16 aA	10.16 aC	5.72 aA	5.72 aA	15.98 aB	15.98 aB
60	ST2	10.20 bA	12.78 aA	5.44 bB	5.92 aA	15.88 bB	19.08 aA
60	ST3	9.52 bA	11.14 aB	5.70 aB	5.54 aA	20.70 aA	16.88 bB
60	ST4	5.78 bB	8.58 aD	5.24 aB	4.24 bB	11.22 aC	12.84 aC
90	ST1	8.68 aA	8.68 aA	4.74 aA	4.74 aA	13.54 aAB	13.54 aA
90	ST2	7.60 bA	9.68 aA	4.72 aA	4.80 aA	12.42 bB	15.60 aA
90	ST3	7.98 bA	9.50 aA	4.70 aA	4.94 aA	14.94 aA	14.52 aA
90	ST4	4.14 bB	5.76 aB	4.10 aB	3.78 bB	8.90 aC	9.56 aB

*Means followed by different lowercase letters in the columns and uppercase letters in the rows differ from each other by Tukey's test (p-value < 0.05).

Tukey's test was applied to compare the means, when significance was observed in the F test of analysis of variance. The significance level of 5% was considered in all tests.

Conclusions

Industrial seed treatment reduces the physiological quality of soybean seeds. Regardless of the chemical products used in IST. The seed treatment corresponding to the largest spray volume provided the lowest means in all tested treatments.

Acknowledgments

The authors thank the Coordination for the Improvement of Higher Education Personnel (CAPES) for the fellowships granted to the first, fourth and sixth authors; and the State University of Maringá for supporting the research.

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