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Interaction of Mechanical Damage and Chemical Treatment and Its Effects on Soybean Seed Physiological Quality

Carla Coppo[1], Alessandro L. Braccini[1], Fernando A. Henning[2], Renata C. Pereira[1], Breno G. Silva[3], Silas M. Oliveira[1], Rayssa F. Santos[1], Géssica G. Bastiani[1], Luana C. Catelan[4], Helen Mariana Cock Protzek[1] & Yana M. Borges[5]

[1] State University of Maringá, Maringá, Paraná, Brazil

[2] Embrapa Soybean, Brazilian Agricultural Research Corporation, Londrina, Paraná, Brazil

[3] Luiz de Queiroz College of Agriculture, University of São Paulo, São Paulo, Brazil

[4] Julio de Mesquita Filho, University of São Paulo, São Paulo, Brazil

[5] Federal Institute of Eduction, Science and Technology of Amazonas, Manaus, Amazonas, Brazil

Correspondence: Carla Coppo, State University of Maringá, Maringá, Paraná, Brazil. Tel: 55-44-98447-4089. E-mail: engagrocarlacoppo@gmail.com

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Abstract

The objective was to evaluate the effects of chemical treatment and levels of mechanical damage and the lignin content of seed coat on soybean seed physiological quality. Two soybean cultivars were used: BMX Lança (58I60 RSF IPRO) and BMX Zeus (55I57 RSF IPRO), with different levels of mechanical injury identified by the tetrazolium test. The chemical treatments used were: control; Carbendazim + Thiram; Carbendazim + Thiram + Dry Powder; Imidacloprid + Thiodicarb; Imidacloprid + Thiodicarb + Dry Powder. A completely randomized design was used, in an 8 × 5 factorial scheme (Types of Samples × Seed Treatment). Physiological quality was evaluated by germination, primary root length, seedling dry mass, accelerated aging and seedling emergence tests. Also, the lignin content in seed coat, one-thousand-seed weight and uniformity test were performed. Data were submitted to analysis of variance (F test) and the mean comparison by the Tukey's test ($p < 0.05$). Cultivars showed differences in the tegument lignin content. The treatment with Imidacloprid + Thiodicarb + dry powder promoted greater reduction in seed physiological potential, intensifying in seeds with more severe damage levels. The lignin content in soybeans seed coat influences the occurrence of mechanical injuries. Seeds with greater intensities of mechanical damage are more susceptible to phytotoxic effects promoted by chemical treatment, since such effects are intensified with the incorporation of dry powder in the seed treatment.

Keywords: physiological potential, injury, seed treatment, lignin content

1. Introduction

The rise of soybean production in the world was achieved due to the success of scientific research and plant breeding; both made possible to adapt soybean cultivars to soil and climate for crop production throughout the Brazilian country, as well as the development of technologies that successfully contributed to improve productivity, agronomic desirable agronomic characteristics of cultivars as well as the industrial value of soybean products (Sediyama et al., 2013).

In the production process, mechanical harvesting is one of the main causes of mechanical injury to soybean seeds, which can cause micro fissures and cracks when performed improperly, mainly during the threshing operation. This is due to the thin seed coat, making vital parts of the embryonic axis, such as the radicle, hypocotyl and plumule very exposed or little protected to mechanical damage (Carvalho & Nakagawa, 2012).

Chemical seed treatment is a frequently practice during soybean seed processing, with the objective of seed protection to exploit the maximum genetic potential (Parisi & Medina, 2013). The presence of fissures or cracks in the seed coat caused by mechanical damage may contributes to increase the phytotoxic effect on seeds and, consequently reduce the physiological potential (Oliveira et al., 2021).

The seed coat is essential to preserve seed structure and also has many important functions (Matilla, 2019). Lignin, present in leaves, stems and seed coat, is characterized as a complex phenolic polymer, important for mechanical support, water transport and plant protection (Campbell & Sederoff, 1996). In this context, Huth et al. (2016), and Alvavez et al. (1997), showed that soybean varieties with lower lignin contents are more susceptible to mechanical damage. However, under these conditions it is essential to highlight the impact of oxidative stress due to the production of reactive oxygen species. Similarly results were observed by Capeleti et al. (2005), the level of mechanical damage is directly correlated with the lignin content of the soybean seed coat.

Therefore, the objective of this research was to evaluate the effect of chemical treatment, relating them to different levels of mechanical injury and to the lignin content of the seed coat and to verify if such factors affect the physiological quality of the seeds.

2. Method

Seeds of two soybean cultivars, BMX Lança (58I60 RSF IPRO) and BMX Zeus (55I57 RSF IPRO), produced in the 2019/2020 from 'Sementes Mutuca' seed company, in Arapoti-PR, Brazil, were used. To determine the levels of mechanical damage, four samples were collected and subjected to the tetrazolium test as described by França-Neto and Krzyzanowski (2018). Results of this the test, allowed the classification of different damage levels as follows: 65—'Lança' seeds with severe damage (between 7 and 10% injuries); 66—'Lança' with severe damage; 67—'Lança' without restriction (less than 6%); 68—'Lança' without restriction; 341—hp. 'Zeus' seeds with severe damage (above than 10% injuries); 347—'Zeus' with severe damage; 357—'Zeus' with severe damage; 369—'Zeus' with severe damage.

Seed treatment, physiological quality assessment and biochemical analysis of lignin content were carried out at the Main Campus of the State University of Maringá, Maringá-Paraná, Brazil.

To perform the seed treatment, the material representing each cultivar was homogenized, divided into samples of 180 g, and subjected to the following chemical treatments: TS1—control (no treatment); TS2—Carbendazim + Thiram (Derosal Plus®; 200 mL per 100 kg^{-1} of seed); TS3—Carbendazim + Thiram + dry powder (Derosal Plus®; 200 mL per 100 kg^{-1} of seeds and Talkum Gloss®; 200 g 100 kg^{-1} of dry powder); TS4—Imidacloprid + Thiodicarb (Cropstar®; 300 mL 100 kg^{-1} of seeds); TS5—Imidacloprid + Thiodicarb + dry powder (Cropstar®; 300 mL 100 kg^{-1} of seeds and Talkum Gloss®; 200 g 100 kg^{-1} of dry powder). The "on-farm" treatment was carried out by using Kraft paper bags as container, a process characterized by intense agitation to promote the homogenization of the product evenly on the seed coat. Seed quality was evaluated using the following tests:

(1) Germination: performed according to MAPA (2009) with four replications each of 50 seeds per treatment. Seed samples were distributed in paper towel, type paper moistened with water equivalent to 2.5 times the weight of dry paper and rolls were prepared. In sequence, samples were kept in a germinator (Mangelsdorf model) regulated at 25 °C constant. The evaluation was performed (germination first count, on the fifth day) and the mean percentage of normal seedlings was recorded (eighth day after sowing).

(2) Length of Primary Root: it was followed the procedure described by Nakagawa (1999), with four replications each of 10 seeds distributed in the upper third of the previously moistened paper towel; the prepared rolls were kept vertically in the germinator at 25 °C under a daily photoperiod of 12 hours for seven days. The primary root length was evaluated with the aid of a graduated ruler and the results were expressed in cm.seedling^{-1}, per treatment.

(3) Root Dry Mass: determined using the primary roots collected from the previous test, which were later placed in paper bags and kept in an oven with forced air circulation at 65 °C for 48 h. The results were obtained by dividing the sample mass by the number of normal seedlings and expressed in grams/seedling per treatment (Nakagawa, 1999).

(4) Seedling Emergence in Sand Substrate: determined through four replications of 50 seeds each, distributed on the surface of 5 cm of sand, in plastic containers (38 cm × 28 cm × 10 cm). The seeds were covered with a 3 cm layer of sand. Water was added to reach 70% of sand water holding capacity.

The containers were kept at room temperature, and the normal seedling count was made when the first pair of open leaves were observed, as performed by Nakagawa (1999). Results were expressed by the means of percentage of seedlings emerged per treatment.

(5) Accelerated Aging: the seeds were deposited superficially, over a wire mesh screen suspended inside a plastic box (11.0 cm × 11.0 cm × 3.5 cm), in a single layer, containing about 42 g. In sequence, 40 ml of distilled water were added to the bottom of each accelerated aging plastic box, which was then transferred to an accelerated aging chamber, kept for 48 h at 41 °C (Marcos-Filho, 1999). After this period, the seeds were tested for

germination, as described above, and the results were expressed as mean percentage of normal seedlings for each treatment.

(6) Seed Water Content: The seeds of all samples were tested in a portable water content meter (Al Agrologic Precision Technology AL-101).

(7) Thousand-Seed Weight (TSW): An analytical balance (0.001 g) was used. Thousand-seed weight was determined by weighing eight subsamples of 100 seeds for each replication, calculating the results and correcting the water content to 13% on a fresh weight basis (MAPA, 2009). The means of the repetitions was multiplied by 10.

(8) Uniformity Test: also known as sieve retention, this test aims to determine the size and diameter of seeds. The methodology is based on arranging the sieves in order of size and using two repetitions of 100 g of seeds and measuring the average of the sieve percentages between the top and bottom that the seeds were contained (MAPA, 2009).

(9) Lignin Content of Seed Coat: quantification was carried out in two stages. The first is the removal of cell wall proteins. Initially, the resulting biomass was weighed (60 mg) and homogenized in a vortex mixer, successively; then 5 times with the addition of 1.5 ml of sodium phosphate buffer (50 mM, pH 7.0); 5 times with the addition of 1.5 ml of 1% Triton® (v/v) in phosphate buffer (50 Mm, pH 7.0); 10 times with the addition of 1.5 of 1M NaCl in phosphate buffer (50 Mm, pH 7.0); 10 times with the addition of 1.5 ml of distilled water and finally 2 times with the addition of 1 ml of acetone. Reagent exchanges were marked by centrifugation (1,400xg) of the precipitate for 5 minutes. At the end of the process, the precipitate was dried in an oven (60 °C) for 24 hours and the dry mass obtained was defined as the protein-free cell wall fraction (PFCW) (Adapted from Moreira-Vilar et al., 2014). The second stage was characterized by the quantification of lignin by the acetyl bromide method, carried out in glass tubes with thread, in which the PFCW fractions were weighed (20 mg) and conditioned. Then, 0.5 ml of 25% acetyl bromide reagent, prepared in cooled acetic acid, was added. The flasks were placed in a water bath (70 °C) every 30 seconds. After this interval, the flasks were removed from the bath, respecting the order in which they were initially added, and cooled in an ice bath. The reaction was stopped by adding 0.9 ml of 2M NaOH and 0.1 ml of hydroxylamine-HCl. For the lignin dilution, 4 ml of acetic acid were added and the samples were centrifuged (1,400xg, 5 min.). After centrifugation, the diluted supernatant was used for readings in a spectrophotometer at 280 nm. Lignin concentration was determined according to a standard curve and expressed in mg of lignin g^{-1} of PFCW (Adapted from Moreira-Vilar et al., 2014).

Data were analyzed using R software version 4.0.2 (R Core Team, 2020). The hypothesis of normality and homogeneity of variances of the variables was verified using the Shapiro-Wilk and Bartlett tests, respectively. The analysis of variance F test was applied to identify differences between treatments and samples (genotypes). The Tukey's test was used to mean comparisons of the treatments. A significance level of 5% was considered in all tests ($p < 0.05$).

3. Results and Discussion

It was verified through the Shapiro-Wilk and Bartlett tests that the hypotheses of normality and homogeneity of variances were accepted. The results of the F test of the analysis of variance, noting that there was significance both for the main effects and for the double interaction types of samples × seed treatment, being necessary to carry out the unfolding between the factors analyzed to identify where the statistically significant differences are found.

For the variable germination first count, it was found that, in general, there was a significant difference among the treatments (Table 1), since the TS5 showed the lowest mean values for this parameter. As for the comparison between the types of samples, significant differences were observed, *i.e.*, the samples of 'Lança', with the lowest severity of mechanical injury, showed the highest averages, highlighting the sample of 'Lança 68', in which the germination first count showed the highest vigor. Costa et al. (2005) showed that as the intensity of mechanical injury increases in soybean seeds, there is a reduction in the percentage of germination and seed vigor.

Table 1. Germination first count (%) from soybean seeds, as a result of seed treatment and types of samples with different levels of mechanical injury

Samples *		Seed treatment *				
		TS1	TS2	TS3	TS4	TS5
BMX Lança (58I60 RFS IPRO)	65-SD	93.50 aA	92.00 aA	92.00 aA	93.50 aA	78.50 bA
	66-SD	86.50 aA	92.00 aA	87.00 aAB	85.50 aB	73.50 bA
	67-NR	90.50 abA	90.00 abA	96.00 aA	85.00 bcB	78.00 cA
	68-NR	94.00 aA	93.00 abA	94.50 aA	87.50 bcAB	82.00 cA
BMX Zeus (55I57 RSF IPRO)	341-VSD	51.00 bC	64.00 aB	67.50 aC	46.00 bE	37.50 cD
	347-SD	68.00 bB	87.50 aA	85.50 aAB	69.50 bC	56.00 bB
	357-VSD	66.50 abB	63.50 bcB	78.00 aBC	52.50 cdE	44.00 dCD
	369-VSD	70.00 bcB	84.50 aA	77.00 abBC	61.50 cdD	53.50 dBC

Note. TS1: control (no treatment); TS2: Carbendazim + Thiram; TS3: Carbendazim + Thiram + dry powder; TS4: Imidacloprid + Thiodicarb; and TS5: Imidacloprid + Thiodicarb + dry powder; Severe damage: SD; Very severe damage: VSD; No restriction: NR. *Means followed by the same lowercase letter in the row and uppercase in the column (p-value < 0.05) do not differ from each other by Tukey's test.

For the germination variable (Table 2) and root length (Table 3) it was observed that TS5 showed the lowest averages. When the types of samples, were compared, significant differences were observed; the samples of 'Lança' type presented the highest averages in the germination and root length variables, in comparison with the samples of the cultivar Zeus, highlighting the sample of 'Lança 65'.

The presence of cracks in the seed coat, caused by mechanical injury, contributes to emphasize possible phytotoxic effect of chemical products on seeds, thus causing reduction of seed physiological potential (Oliveira et al., 2021).

In this context, it is important to stress that Abati et al. (2018), found different responses in relation to germination, depending on soybean cultivar. These authors considered the use of dry powder resulted in lower speed of germination.

Table 2. Germination (%) as a function of seed treatment and types of samples with different levels of mechanical injury

Samples *		Seed treatment				
		TS1	TS2	TS3	TS4	TS5
BMX Lança (58I60 RFS IPRO)	65-SD	96.00 aA	95.00 aAB	94.00 aAB	96.00 aA	88.00 bA
	66-SD	92.50 bA	99.00 aA	92.50 bAB	91.50 bAB	86.00 cA
	67-NR	96.00 aA	96.50 aAB	98.00 aA	90.00 bAB	83.00 cAB
	68-NR	97.00 aA	95.00 abAB	97.00 aA	91.00 bcAB	87.00 cA
BMX Zeus (55I57 RSF IPRO)	341-VSD	74.50 bcC	81.00 abC	82.00 aC	70.00 cD	61.50 dE
	347-SD	82.50 abB	92.00 aABC	90.00 aABC	88.00 abBC	77.50 bBC
	357-VSD	96.00 aA	87.00 abBC	84.50 abBC	82.50 bcC	71.00 cCD
	369-VSD	77.50 bBC	87.50 aBC	86.00 aBC	71.00 bcD	68.00 cDE

Note. TS1: control (no treatment); TS2: Carbendazim + Thiram; TS3: Carbendazim + Thiram + dry powder; TS4: Imidacloprid + Thiodicarb; and TS5: Imidacloprid + Thiodicarb + dry powder; Severe damage: SD; Very severe damage: VSD; No restriction: NR. *Means followed by the same lowercase letter in the row and uppercase in the column (p-value < 0.05) do not differ from each other by Tukey's test.

As explained by Carvalho et al. (2020) in research on soybean seeds treated at different times before storage or sowing, the first symptoms of phytotoxicity promoted by seed treatment in seedlings are detected by the evaluation of primary root growth during germination. Rocha et al. (2020) found that soybean seeds treated with insecticides show signs of phytotoxicity in seedlings more intensely than when compared with fungicides. In the same perspective, studies with the use of insecticides, especially neonicotinoids, indicated the occurrence of irreversible damages to the physiological quality of the seeds. Since the translocation by way the tegument leads to a loss of vigor, results are accentuated with the increase of doses (Pereira et al., 2020).

In the development of new molecules for commercialization and use in agriculture, systemic ones are the most desired by researchers (Zhang et al., 2018). Systemic ingredients must have greater water solubility and thus promote greater penetration into seeds (Garcia, 1999). The penetration of products used in seed treatment is desirable, but depending on the active ingredient and the rate of application used, it can result in phytotoxicity in seedlings. To avoid phytotoxicity, França Neto et al. (2016) it is recommend to verify the necessity to use chemicals, followed by the efficiency and compatibility between them. Also check the spray volume, as well as the high volume can remove the seed coat during treatment, from mechanically damaged seeds, and also reduce their physiological potential.

Table 3. Root length (cm) as a function of seed treatment and types of samples with different levels of mechanical injury

Samples *		Seed treatment				
		TS1	TS2	TS3	TS4	TS5
BMX Lança (58I60 RFS IPRO)	65-SD	17.86 aA	18.82 aA	18.60 aA	17.68 aA	16.77 aA
	66-SD	17.28 aA	17.61 aA	19.69 aA	11.73 bB	7.99 cBC
	67-NR	12.44 bB	18.44 aA	19.52 aA	12.40 bAB	9.48 bBC
	68-NR	17.85 aA	17.56 aA	19.36 aA	12.82 aAB	12.64 aAB
BMX Zeus (55I57 RSF IPRO)	341-VSD	16.43 aA	18.43 aA	18.54 aA	13.36 aAB	4.90 bC
	347-SD	15.02 abAB	18.33 aA	17.99 abA	13.19 bAB	5.82 cC
	357-VSD	15.92 abAB	17.77 aA	17.28 abA	14.39 bAB	9.65 cBC
	369-VSD	15.81 bAB	16.89 abA	17.84 aA	14.15 cAB	9.83 dBC

Note. TS1: control (no treatment); TS2: Carbendazim + Thiram; TS3: Carbendazim + Thiram + dry powder; TS4: Imidacloprid + Thiodicarb; and TS5: Imidacloprid + Thiodicarb + dry powder; Severe damage: SD; Very severe damage: VSD; No restriction: NR. *Means followed by the same lowercase letter in the row and uppercase in the column (p-value < 0.05) do not differ from each other by Tukey's test.

Checking the root dry mass variable (Table 4), with the exception of 'Lança 65' and 'Zeus 369' samples, was observed significant differences among the treatments, where TS5 showed the lowest averages. With espect to types of samples, significant differences were observed only for TS1 and TS2; the samples of 'Lança' showed the highest averages, respectively, mainly for 'Lança 68'.

Taylor and Salanenka (2012) reported that some chemical products are prone to promote phytotoxicity effects on soybean seedlings, and the primary symptoms are slow germination and emergence, low percentage of seedling emergence and thickening, in addition to hypocotyl shortening. In the present study it was found that the lower means from the treatment with TS5 insecticide were probably due to the severity of the mechanical injury associated with the treatment used. However, it is essential to point out that, under the current conditions, chemical products used in seed treatment are able to penetrate the seed fissures and also translocate via seed coat, promoting irreversible damage to vital parts of the embryo, such as plumule and radicle (Taylor & Salanenka, 2012; Pereira et al., 2021). This translocation effect can happen, but it depends on the active ingredient and dosage, thus causing phytotoxicity effect in the first vegetative stages of the plant development.

The use of dry powder in seed treatment usually accelerates the natural drying process of the product but can result in future imbibition damage in seeds (Abati et al., 2018). These difficulties may occur during the imbibition process and is caused by a negative action of the fast water uptake on cell membranes, thus resulting in problems during germination (Toledo et al., 2010).

Table 4. Dry Root Mass (g) as a function of seed treatment and types of samples with different levels of mechanical damage

Samples *		Seed treatment				
		TS1	TS2	TS3	TS4	TS5
BMX Lança (58I60 RFS IPRO)	65-SD	0.11 bAB	0.11 bBC	0.27 aA	0.14 abA	0.10 bA
	66-SD	0.10 aAB	0.09 aC	0.13 aA	0.15 aA	0.14 aA
	67-NR	0.13 bA	0.11 bBC	0.35 aA	0.09 bA	0.12 bA
	68-NR	0.11 bAB	0.14 abABC	0.75 aA	0.10 bA	0.11 bA
BMX Zeus (55I57 RSF IPRO)	341-VSD	0.15 abA	0.20 abA	0.38 aA	0.14 abA	0.07 bA
	347-SD	0.06 bcB	0.13 aABC	0.14 aA	0.12 abA	0.06 cA
	357-VSD	0.12 abcAB	0.14 abABC	0.16 aA	0.10 bcA	0.08 cA
	369-VSD	0.16 aA	0.16 aAB	0.21 aA	0.21 aA	0.09 aA

Note. TS1: control (no treatment); TS2: Carbendazim + Thiram; TS3: Carbendazim + Thiram + dry powder; TS4: Imidacloprid + Thiodicarb; and TS5: Imidacloprid + Thiodicarb + dry powder; Severe damage: SD; Very severe damage: VSD; No restriction: NR. *Means followed by the same lowercase letter in the row and uppercase in the column (p-value < 0.05) do not differ from each other by Tukey's test.

The emergence of seedlings in sand (Table 5) showed significant differences and the lower mean values were from TS5. At the same time, significant differences were observed and the samples of the cultivar 'Lança 66' and 'Lança 68' showed the higher values.

Lopes et al. (2011) reported that the damage caused by mechanical harvesting was more severe, since it had a negative effect on seed viability, vigor, and field seedling emergence.

Table 5. Seedling emergence in sand (%) as a function of seed treatment and types of samples with different levels of mechanical damage

Samples *		Seed treatment				
		TS1	TS2	TS3	TS4	TS5
BMX Lança (58I60 RFS IPRO)	65-SD	100.00 aA	100.00 aA	96.00 abAB	93.00 bAB	85.00 cA
	66-SD	99.00 aA	93.00 abB	97.00 abAB	90.00 bBC	72.00 cB
	67-NR	100.00 aA	98.00 aAB	99.00 aA	93.00 bAB	89.00 bA
	68-NR	100.00 aA	100.00 aA	100.00 aA	99.00 aA	92.00 bA
BMX Zeus (55I57 RSF IPRO)	341-VSD	82.00 cC	96.00 aAB	94.00 abB	86.00 bcBC	63.00 dC
	347-SD	92.00 bB	94.00 abB	100.00 aA	85.00 cBC	72.00 dB
	357-VSD	89.00 bB	96.00 aAB	99.00 aA	82.00 cC	68.00 dBC
	369-VSD	90.00 bcB	97.00 abAB	99.00 aA	84.00 cC	64.00 dBC

Note. TS1: control (no treatment); TS2: Carbendazim + Thiram; TS3: Carbendazim + Thiram + dry powder; TS4: Imidacloprid + Thiodicarb; and TS5: Imidacloprid + Thiodicarb + dry powder; Severe damage: SD; Very severe damage: VSD; No restriction: NR. *Means followed by the same lowercase letter in the row and uppercase in the column (p-value < 0.05) do not differ from each other by Tukey's test.

For the accelerated aging variable (Table 6), it was observed the occurrence of significant differences among the treatments, where TS5 showed the lowest averages. Regarding the comparison among the types of samples significant differences demonstrated that the samples of 'Lança' were responsible for the highest averages in most treatments.

The water content during seed aging process favors the increase in seed temperature, due to the activation of the respiratory process (Carvalho & Nakagawa, 2012). The highest sensitivity of larger seeds to mechanical injury is a fact, but the higher physiological potential of small seeds is not always true.

Table 6. Accelerated aging (%) as a function of seed treatment and types of samples with different levels of mechanical damage

Samples *		Seed treatment				
		TS1	TS2	TS3	TS4	TS5
BMX Lança (58I60 RFS IPRO)	65-SD	51.00 cB	61.50 bC	76.00 aBC	48.00 cB	58.00 dB
	66-SD	78.00 aA	84.50 aA	85.50 aAB	38.00 bB	15.00 cCD
	67-NR	79.00 bcA	83.00 abA	88.00 aA	72.00 cA	59.00 dA
	68-NR	78.00 abA	80.50 abAB	83.50 aAB	75.00 bA	66.50 cA
BMX Zeus (55I57 RSF IPRO)	341-VSD	11.50 cE	33.00 aD	33.00 aD	22.00 bC	12.00 cCD
	347-SD	34.50 bC	31.50 bcD	64.50 aC	17.00 cC	24.50 bcC
	357-VSD	20.00 dD	57.50 bC	87.50 aAB	46.00 cB	11.00 dD
	369-VSD	9.50 dE	73.50 aB	41.00 bD	21.50 cC	8.50 dD

Note. TS1: control (no treatment); TS2: Carbendazim + Thiram; TS3: Carbendazim + Thiram + dry powder; TS4: Imidacloprid + Thiodicarb; and TS5: Imidacloprid + Thiodicarb + dry powder; Severe damage: SD; Very severe damage: VSD; No restriction: NR. *Means followed by the same lowercase letter in the row and uppercase in the column (p-value < 0.05) do not differ from each other by Tukey's test.

Seed water content showed values of 10.3% for 'Lança' and 10.4% in 'Zeus". The thousand-seed weight, determined according to the methodology described by MAPA (2009) and corrected for 13% water content, resulted in weights of 155.4 and 221.3 g, for the cultivars Lança and Zeus, respectively.

In addition, significant differences among treatments for PFCW (protein free cell wall) were observed (Table 7); the samples of 'Lança' type exhibited the highest averages, with emphasis for Lança 68; in contrast, the 'Zeus 341' sample had the lowest average in PFCW variable.

Table 7. Protein-free cell wall (PFCW in mg g^{-1}) as a function of sample types

Samples *		PFCW (mg g^{-1})
BMX Lança (58I60 RFS IPRO)	65-SD	65.34 bc
	66-SD	67.25 ab
	67-NR	68.07 ab
	68-NR	70.24 a
BMX Zeus (55I57 RSF IPRO)	341-VSD	59.44 e
	347-SD	63.07 cd
	357-VSD	62.44 cde
	369-VSD	61.62 de

Note. Severe damage: SD; Very severe damage: VSD; No restriction: NR.*Means followed by distinct lowercase letters in the column (p-value < 0.05) differ from each other by Tukey's test.

According to the above, it can be noted that samples of Lança cultivar show absence or lighter levels of mechanical injury, and this was attribute to their higher lignin content in the seed coat. This result confirms Capeleti et al. (2005) observations, in research with soybean cultivars, demonstrating that that the lignin contents in the seed coat is directly related with seed tolerance to mechanical injuries. The lignification of the tegument favors lower sensitivity to damage and this fact was explained by Mertz-Henning et al. (2015), who observed superior level in soybean seeds, with a higher concentration of lignin in the seed coat.

4. Conclusions

The lignin content in the soybean seed coat influences the occurrence of mechanical damage. Seeds with greater intensities of mechanical damage are more susceptible to phytotoxic effects, such effects are intensified with the incorporation of dry powder to the seed treatment. Drying powder use causes harm when used in conjunction with certain insecticides.

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