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Physiological quality of soybean seeds treated with imidacloprid before and after storage

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Abstract: This study aimed to evaluate the effect of different slurry volume of imidacloprid insecticide on the physiological potential during the storage of seeds of three soybean cultivars. A completely randomised experimental design was adopted, in a 6 × 3 × 2 factorial scheme, which treatments were six slurry volumes with imidacloprid insecticide (0, 200, 500, 800, 1 100 and 1 400 mL/100 kg of seeds), 3 soybean cultivars (SYN15630, M5947, and NS5959) and 2 storage periods (0 and 30 days after seed treatment), with four replications. The treated seeds were stored in kraft paper bags with controlled temperature and humidity. The physiological potential of the seeds was evaluated by standard germination test, electrical conductivity, accelerated aging, seedling emergence in the sand seedbed, and the field. The germination and vigor of soybean seeds were reduced during storage, especially with the increase in the volumes of the insecticide solution.

Keywords: *Glycine max* (L.) Merrill; neonicotinoid; deleterious effect; imbibition; deterioration

The seed treatment is considered an efficient practice to ensure adequate plant populations, a projection for the year 2020 foresees an 8.57% increase in the use of insecticides *via* seeds (França-Neto et al. 2010, Zhang 2018). Nonetheless, sowing is often carried out in adverse conditions to germination and emergency, resulting in failures and the need for reseeding. Therefore, the treatment of seeds with phytosanitary products, in addition to being a fundamental practice to guarantee an adequate stand, enables the reduction of costs resulting from the incidence of pathogens (Henning et al. 1997).

Subsequently, the physiological maturation starts the deterioration process, which is marked by changes that culminate in the vigor loss and quality of the seeds. Such modifications are initiated due to changes related to temperature, humidity, and the presence of microorganisms in the seed (Marcos-Filho 2015b).

Beyond storage, another relevant point to be considered is the volume of solution since it directly affects the maintenance of seed vigor throughout storage (Braccini et al. 2015). However, there are few studies relating to the slurry volume with the storage of seeds. Brzezinski et al. (2017) and Suzukawa et al. (2019) pointed out that the high slurry volume is detrimental to the physiological quality of the seeds, with more expressive results being found, especially in low vigor lots. In this context, the established hypothesis is that the high slurry volume of the imidacloprid insecticide associated with the storage period provides a deleterious effect on the seeds. Under these conditions, it is important to note that an increase in the slurry volume can potentiate such damage caused in corn and soybean seeds (Dias et al. 2014, Yang et al. 2018).

Given the above, this study aimed to evaluate the effect of different slurry volumes of imidacloprid on

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the physiological potential of three soybean cultivars during storage.

MATERIAL AND METHODS

The seeds of each cultivar were treated separately with different slurry volumes of the imidacloprid insecticide, obtained by diluting the product in water and subjected to different storage periods. Thus, the assay was conducted with a completely randomised design, with 36 treatments, composed by the combination of the standard dose of insecticide (250 mL/100 kg of seeds) diluted between six volumes of insecticide solution (0, 200, 500, 800, 1 100 and 1 400 mL of seeds), three soybean cultivars (SYN15630 IPRO, M5947 IPRO, and NS5959 IPRO) and two storage periods (0 and 30 days), in a 6 × 3 × 2 factorial scheme, with four replications.

The seeds of each cultivar were treated separately in plastic bags, according to each volume of pre-established solution and packed in Kraft paper bags. To simulate conventional storage, the seeds were kept under controlled conditions of humidity (65%) and temperature (25 °C). The evaluation of the physiological potential of the seeds was carried out in each of the storage periods (0 and 30 days), through standard germination test (Brasil 2009), electrical conductivity (AOSA 2002), accelerated aging (Marcos-Filho 1999), seedling emergency in a sand seedbed and seedling emergency in the field, which were performed according to Nakagawa (1999).

The data obtained for each response variable were initially tested for normality (Shapiro-Wilk) and homogeneity of variances (Levene). The physiological potential parameters were subjected to ANOVA analysis of variance ($P \leq 0.05$). For the cultivar factor, the comparison of averages was by *t*-test *LSD* (least significant difference) ($P \leq 0.05$), while for storage periods, it was compared by the *P* values of the analysis of variance. Finally, for the slurry volumes, as well as their interactions, regression analysis was applied. All statistical analyses were performed using the Sisvar software (Ferreira 2011).

RESULTS AND DISCUSSION

Concerning germination, the treatment of seeds, on the average of the three cultivars evaluated, with an increase in the volume of imidacloprid manure, provided a decrease in the germination of soybean seeds up to the high level of the seeds treatments (Figure 1). In these conditions, Dias et al. (2014) and Yang et al. (2018) suggest that the occurrence of diffusion of non-ionic compounds throughout the pericarp is capable of damaging the endosperm, inhibiting the germination process and the formation of the radicle. However, it is important to note that, even in the presence of the cutinised membrane, the active ingredients can pass the endoderm during the process of seed soaking (Baldini et al. 2018).

When unfolding the first-order interaction between storage periods and the cultivars for the germination variable, it was observed that for cv. M5947, the storage periods did not influence this response variable. On the other hand, cv. NS5959 showed a higher percentage of germination in the first storage period (0 days), whereas the cv. SYN15630 obtained better results in the second storage period (30 days).

Also, the average germination values as a function of the cultivars inside of the storage periods, cv. NS5959 showed superior germination, compared to the others, for both periods. However, for the 30 days, this cultivar did not differ from cv. SYN15630 for that attribute ($P \leq 0.05$) by *F*-test and *t*-test (*LSD*), respectively. There was also a difference between the cultivars, in which cv. NS5959 seeds showed a higher germinative percentage in the two storage periods ($P \leq 0.05$).

Elaborating the third-order interaction (insecticide (I) × storage periods (S) × cultivars (C)) for electrical conductivity (EC), different behaviors were observed regarding regression, which were quadratic, cubic, and decreasing for cv. M5947 at

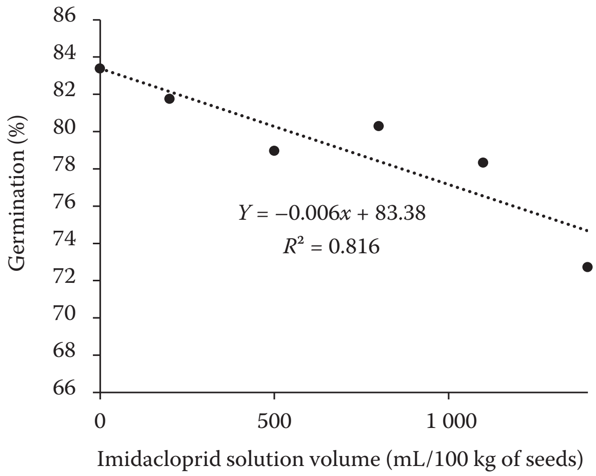

Figure 1. Percentage of soybean seeds germination, in response to soybean seed treatment with different imidacloprid slurry volumes, on the average of the three cultivars evaluated

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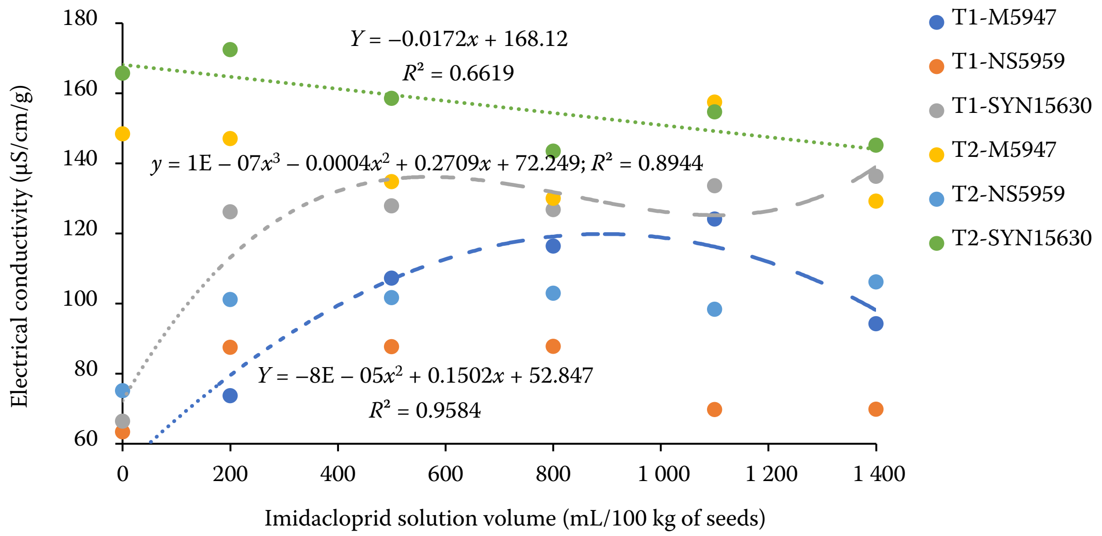

Figure 2. Electrical conductivity of soybean seeds, in the function of the seed treatment with different imidacloprid slurry volumes, unfolded inside of the cultivars (M5947, NS5959, and SYN15630) and seed storage periods (T1 – before; T2 – after) combination

the beginning of storage 0 days. And for seeds of cv. SYN15630 in the absence of storage and stored for 30 days, respectively, and the other combinations between cultivars and storage periods did not adjust significantly (Figure 2). França-Neto et al. (2015) and Santos et al. (2018) emphasise that the range of products applied in seed treatment, together with the high volume of solution, have a deleterious effect on the physiological quality and seed germination.

The augmentation of the effects of seed storage periods, inside of the levels of insecticide and the cultivars for EC, it was found that, in general, seed storage for 30 days provided an increase of 35.09% for the response variable (Table 1). However, the seed storage did not provide difference to the electrical conductivity of seeds for cv. NS5959 when seed treatment was carried out with 200 mL/100 kg of seeds, for cv. M5947 in the slurry volume of 500 and 800 mL/100 kg of seeds and cv. SYN15630 in the largest imidacloprid slurry volumes (800, 1 100 and 1 400 mL/100 kg of seeds) (Table 1).

On the other hand, in addition to the effects of the cultivars, inside of insecticide levels and seed storage periods, on the EC, it was found that cv. SYN15630 showed higher values for the response variable concerning cv. NS5959 (Table 1). In turn, the EC averages of the seeds of cv. SYN15630 were higher than those of cv. M5947, only when combined 200 mL/100 kg of seeds and zero days of storage,

Table 1. Electrical conductivity (µS/cm/g) in soybean seeds, in the function of imidacloprid solution volumes and the cultivars

Slurry volume (mL 100/kg of seeds)	Cv. M5947		Cv. NS5959		Cv. SYN15630	
	0	30	0	30	0	30
0	57.18^{Bc}	148.40^{Aa}	63.40^{Ba}	75.10^{Ab}	66.43^{Bb}	165.48^{Aa}
200	73.65^{Bbc}	147.01^{Aa}	87.47^{Aa}	101.07^{Aab}	126.13^{Ba}	172.36^{Aa}
500	107.24^{Aab}	134.72^{Aa}	87.67^{Ba}	101.68^{Aab}	127.82^{Ba}	158.47^{Aa}
800	116.31^{Aab}	129.96^{Aa}	87.73^{Aa}	102.93^{Aab}	126.73^{Aa}	143.39^{Ab}
1 100	124.09^{Ba}	157.39^{Aa}	69.69^{Ba}	98.34^{Aab}	133.59^{Aa}	154.68^{Aa}
1 400	94.22^{Bb}	129.14^{Aa}	69.81^{Ba}	106.10^{Aa}	136.24^{Aa}	145.15^{Aa}
Significant minimum difference			28.6447			

Averages followed by distinct upper case letters in the lines and lower case letters in the columns differ from each other. ($P \leq 0.05$) by the F-test

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500 mL/100 kg of seeds and 30 days of storage and 1 400 mL/100 kg of seeds and zero days of seed storage, statistically equaling in other situations (Table 1).

When advancing the second-order interaction (I × C) observed for the seedling emergency in sand seedbed (SES), an estimated linear decrease of 1.32% and 1.61% was found for cvs. NS5959 and M5947, respectively, for each 100 mL/100 kg of seeds (Figure 3A). In turn, the cv. SYN15630 showed quadratic behavior in the regression analysis, which the minimum SES point, provided by the level of insecticide of 862.50 mL/100 kg of seeds, was 77.34% (Figure 3A).

Concerning seedling emergency in the field (SEF), seed treatment with increasing levels of slurry volume provided an estimated decrease of 1.29% to the response variable, for each 100 mL of the insecticide per 100 kg of seeds, on the average of the three cultivars evaluated (Figure 3B). It is notorious that nonionic compounds with lipophilic characteristics can cross the cutinised membrane of the seeds during imbibition. However, because of the high solubility in water, the diffusion of these systemic insecticides has a deleterious effect on seeds. This occurs because of the lipophilicity of the active ingredients, in other words, the affinity of these compounds for the plasma membrane, waxes, and cutin of low vigor seeds submitted to the storage (Dias et al. 2014, Yang et al. 2018).

Developing the effect of the soybean cultivars, inside of the imidacloprid slurry volumes, for the seedling emergency in the sand (SES), were not found significant differences ($P > 0.05$) (when using the lowest volumes of insecticide solutions (0 and 200 mL/100 kg of seeds). Differently, cv. SYN15630 showed a lower percentage of seedling emergency, about the others, when the seeds were treated with the volumes of 500 and 800 mL/100 kg of seeds. In these circumstances, Brzezinski et al. (2017) suggest that the increase in slurry volume reduces the length of roots and the seedling emergency in the sand, especially in low vigor seed lots.

To the slurry volume of 1 100 mL/100 kg of seeds, cv. NS5959 showed a higher average of SES, compared to the cv. M5947, however, in the slurry volume of 1 400 mL/100 kg of seeds, cv. SYN15630 differed ($P \leq 0.05$) and surpassed the cv. M5947. Regarding seedling emergency in the field, depending on the cultivars evaluated, cv. SYN15630 provided higher values, compared to the others. Furthermore, cv. NS5959 exceeded the SEF obtained for cv. M5947. Authors such as Vanzolini and Carvalho (2002) related the low quality of seeds to the initial development of seedlings and reduced emergency in the field; according to these authors, the low vigor of the seeds reflects in marked failures in the plant populations.

In the first order significant interaction (I × C) for accelerated aging (AA) of seeds, a quadratic regression behavior was found for the cvs. NS5959 and SYN15630, with maximum AA points of 88.73% and 68.67%, provided by the slurry volumes of 437.50 and 210.00 mL/100 kg of seeds, respectively (Figure 4). In contrast, cv. M5947 showed a cubic behavior to regression, in which the AA values decreased until

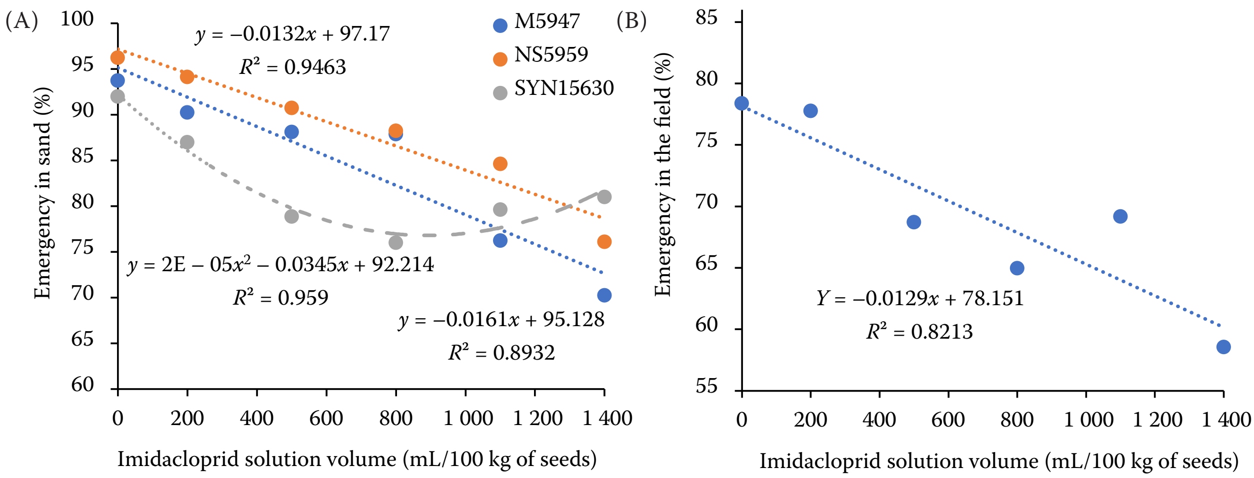

Figure 3. Seedling emergency in the sand, in the function of the seed treatment with different imidacloprid slurry volumes and the soybean cultivars (A); and seedlings emergency in the field, in the function of the seed treatment with different imidacloprid slurry volumes (B)

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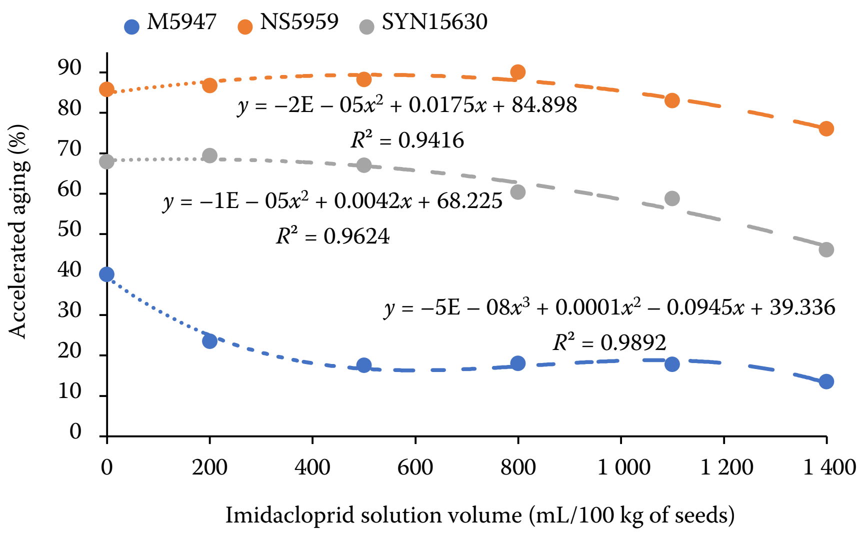

Figure 4. Percentage of normal seedlings in the accelerated aging test of soybean seeds, in the function of the seed treatment with different imidacloprid slurry volumes and the cultivars

the volume of a solution of 500 mL/100 kg of seeds, stabilising, then, until the volume of 1 100 mL/100 kg seeds, from which there was a new decrease in this response variable (Figure 4).

The effective evolution of the soybean cultivars inside of the imidacloprid slurry volumes for the accelerated aging (AA) of seeds, it is observed that cv. NS5959 presented a higher average for the control (0 mL/100 kg of seeds) and in the solution volume of 200 mL/100 kg of seeds. On the contrary, for the other slurry volumes, cv. SYN15630 differed and overcame the others ($P \leq 0.05$). Cv. M5947 showed lower AA averages, compared to cvs. NS5959 and SYN15630 regardless of the volume of insecticide evaluated. However, for cv. SYN15630, the increase in slurry volume promoted an increase in the percentage of normal seedlings. In consideration of the results, for the other cultivars, there was a trend indicating that high slurry volumes promote the decline of seed vigor.

These results agree with Dan et al. (2010) when they verified that the aging of treated seeds promotes the reduction of normal seedlings, especially after storage. However, high vigor seeds still perform better in unfavourable conditions since the organisation capacity of plasma membranes remains maintained, ensuring seed integrity (Marcos-Filho 2015a).

The maximum physiological potential is reached close to seed maturity since, after this stage, deterioration begins. In these circumstances, factors such as harvest time, environmental conditions, drying, and storage procedures are responsible for the deterioration process (Marcos-Filho 1999, 2015b). Also, it is important to note that aging caused by deterioration promotes delay in the germination process, reduced embryo growth, and susceptibility to environmental stresses (Maia et al. 2007).

Regarding the performance of seeds in the field, defined according to the physiological quality and vigor of the seeds, Brzezinski et al. (2017) pointed out that seeds of high vigor have superior performance under stress conditions because, after the imbibition process, they can maintain the integrity of the membranes since the deterioration occurs quickly in this condition.

Finally, it is concluded that the treatment of soybean seeds with increasing slurry volumes of imidacloprid reduces their germination potential. On the other hand, seed storage for 30 days negatively influences the seed germination of cv. NS5959 and positively influences the seeds of cv. SYN15630. The storage of soybean seeds treated with different imidacloprid slurry volumes increased the levels of electrical conductivity. Besides, increasing the volume of imidacloprid solution in seed treatment reduces its physiological potential, with direct effects on the seedling emergency in the sand and the field.

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