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Physiological potential of soybean seeds over storage after industrial treatment[1]

Lucas Caiubi Pereira[2]*, Mayara Mariana Garcia[2], Alessandro Lucca Braccini[2], Gláucia Cristina Ferri[2], Andreia Kazumi Suzukawa[2], Danilo Cesar Volpato Marteli[2], Thaisa Cavalieri Matera[2], Renata Cristiane Pereira[2], Larissa Vinis Correia[2]

ABSTRACT - The aim of this study was to evaluate the effect of industrial seed treatments on the physiological potential of soybean seeds over storage. Four mixtures of agrochemical products in association with two fertilizers were tested. The agrochemical product mixtures were carbendazim/thiram + imidacloprid/thiodicarb; pyraclostrobin, thiophanate-methyl, and fipronil; thiophanate-methyl/fluazinam + bifenthrin/imidacloprid; and metalaxyl-m/fludioxonil + thiamethoxam. The two fertilizers were 7% N, 16% P_2O_5, 0.6% Co, and 2.5% Mo; and 1% Co, 10% Mo, and 7% P_2O_5. The experiment was carried out in a completely randomized design in a split-plot arrangement in time, with four replications. The treatments were allocated in the plots, while the storage periods (0, 30, 60, 90, and 120 days) constituted the split-plots. The following tests were carried out in each period: first count of germination, germination, accelerated aging, emergence speed index in sand substrate, and final seedling emergence in sand substrate. Seed germination and vigor declined over the storage period, especially after industrial treatment. Pesticide mixtures of a carbendazim/thiram fungicide base and an imidacloprid/thiodicarb insecticide base most impaired seed physiological potential throughout storage, regardless of fertilizer use in the industrial treatment.

Index terms: *Glycine max*, micronutrients, germination, vigor.

Potencial fisiológico de sementes de soja durante o armazenamento após tratamento industrial

RESUMO - O objetivo no trabalho foi avaliar o efeito do tratamento industrial sobre o potencial fisiológico de sementes de soja durante o armazenamento. Quatro misturas de defensivos agrícolas (carbendazim/thiram + imidaclopridо/tiodicarbe; piraclostrobina, tiofanato metílico e fipronil; detiofanato-metílico/fluazinam + bifentrina /imidacloprido e metalaxil-m/ fludioxonil + thiamethoxam) associadas a dois fertilizantes (7% N, 16% P_2O_5, 0,6% Co, 2,5% Mo e 1% Co, 10% Mo e 7% P_2O_5) foram testadas. Adotou-se o delineamento experimental inteiramente casualizado em parcelas subdivididas no tempo, com quatro repetições. Nas parcelas foram alocados os tratamentos e nas subparcelas os períodos de armazenamento (0, 30, 60, 90 e 120 dias). Em cada período foram realizados os testes de primeira contagem, germinação, envelhecimento acelerado, índice de velocidade de emergência e emergência de plântulas em substrato de areia. A germinação e o vigor das sementes de soja são reduzidos ao longo do armazenamento, sobretudo após o tratamento industrial. Caldas à base de carbendazim/thiram + imidacloprido/tiodicarbe prejudicam o potencial fisiológico das sementes ao longo do armazenamento, independentemente do emprego de fertilizantes no tratamento industrial.

Termos para indexação: *Glycine max*, micronutrientes, germinação, vigor.

Introduction

The use of high quality seed plays an important role in plant stand and, consequently, in agricultural crop yield. However, sowing of seeds rarely occurs in areas free of threats to plant health, leading the producer to make use of chemical fungicide or insecticide seed treatments. In this scenario, soybean (*Glycine max*) stands out as the main agricultural

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[2]Universidade Estadual de Maringá, Av. Colombo, 5790, 87020-900 - Maringá, PR, Brasil.
*Corresponding author <lucascaiubi@yahoo.com.br>

commodity crop in the use of seed treatments (Nunes, 2016). It is estimated that more than 95% of the area sown to soybean in Brazil makes use of chemically treated seeds (Henning et al., 2010), and industrially treated seeds represented 66% of total seeds sold in 2015 (França-Neto et al., 2015).

There are numerous beneficial results from covering seeds with fungicides and insecticides for plant health protection in crops, both in reducing pathogen transmission and in defense against fungus and insect attacks during storage or in the initial stages of the crop (Bays et al., 2007; Pereira et al., 2009; Henning et al., 2010). A priori, industrial treatments of soybean available in the market do not adopt additional standardized covering of seeds with cobalt and molybdenum, nutrients that are indispensable for success in biological fixation of atmospheric nitrogen in symbiosis between soybean and bacteria of the genus *Bradyrhizobium* spp. (Sfredo and Oliveira, 2010). Nevertheless, as low application rates of these elements are required, demand for the use of these elements together with chemical treatment is growing, above all when the seed lots are treated in accordance with producer demand.

Information in the literature regarding the physiological and agronomic performance of soybean seed treated immediately before sowing is not scarce (Pereira et al., 2007; Binsfeld et al., 2014). However, unlike the treatment performed on the farm itself, industrial seed treatment occurs months before sowing (Strieder et al., 2014), a practice that may lead to deterioration of the seed lot.

After undergoing industrial treatment, soybean seeds are stored, the main purpose of which is to provide ambient conditions favorable to maintenance of the physiological and plant health quality of seed lots up to the time of sale (Carvalho and Nakagawa, 2012). After physiological maturity, seed deterioration begins, culminating in an inevitable, continuous, and irreversible process (Delouche and Baskin, 1973), and it is not possible to recover the level of vital activity lost over time.

Seed deterioration results in a series of deleterious changes that occur over time (aging), which may be accelerated or attenuated according to the environment, especially in relation to temperature and relative humidity, as well as in relation to the presence of insects and microorganisms that consume seed reserves (Marcos-Filho, 2015). For the soybean crop, seeds are normally stored in bags of seed placed in conventional warehouses, i.e., without special atmospheric controls, thereby exposing seeds to oscillations in relative humidity and in temperature. These oscillations not only accelerate the aging process, but may also favor infection from fungi and pests, which jointly bring about loss of vigor and germination (Fessel et al., 2003).

In spite of the beneficial effects of fungicides and insecticides in crop protection, harmful effects they may have on the physiological potential of soybean seeds have not infrequently been reported (Dan et al., 2010; Dan et al., 2011; Avelar et al., 2011), associated or not with fertilizers (Bays et al., 2007; Farooq et al., 2012; Binsfeld et al., 2014). However, little is known regarding the combined effect of these products on seed physiological performance over the storage period.

In this respect, the hypothesis established in this study is that reduction in vigor and germination potential of soybean seeds is accelerated after they are covered with fungicides, insecticides, polymers, and fertilizers. The aim of the present study, therefore, was to evaluate the effect of combinations of commercial products used in industrial treatment on the physiological potential of soybean seeds during storage.

Materials and Methods

The cultivar used in the experiment was NA 5909 RR. The trial was conducted in a completely randomized experimental design in split-plots in time, with four replications. Treatments consisted of the combination between five storage periods (0, 30, 60, 90, and 120 days) with four combinations of agrochemical pesticide products normally used in industrial treatment of soybean seeds and two fertilizer formulations. The industrial seed treatments used, including the respective combinations of pesticides, fertilizers, and volumes of the seed-coating mixtures, are shown in Table 1. Also known as "technologies", such combinations consisted of the mixture of fungicide, insecticides, and seed-coating polymers (dry powder and liquid) as follows: Technology I – carbendazim 150 g. L^{-1} + thiram 350 g. L^{-1} (Derosal Plus®, rate of 200 mL. 100 kg^{-1}), imidacloprid 150 g. L^{-1} + thiodicarb 450 g. L^{-1} (Cropstar®, rate of 500 mL.100 kg^{-1}), polymer (Peridiam 306, rate of 200 mL. 100 kg^{-1}) and a powder seed coating (Sepiret PF, rate of 170 g.100 kg^{-1}); Technology II – pyraclostrobin 25 g. L^{-1} + thiophanate-methyl 225 g. L^{-1} + fipronil 250 g. L^{-1} (Standak Top®, rate of 200 mL. 100 kg^{-1}), a polymer (Florite Green®, rate of 200 mL. 100 kg^{-1}), and a powder seed coating (Sepiret PF, rate of 200 g. 100 kg^{-1}); Technology III - thiophanate-methyl 350 g. L^{-1} + fluazinam 52.5 g. L^{-1} (Certeza®, rate of 200 mL. 100 kg^{1}), bifenthrin 135 g. L^{-1} + imidacloprid 165 g. L^{-1} (Rocks®, rate of 350 mL. 100 kg^{-1}), a polymer (Peridiam 306, rate of 200 mL .100 kg^{-1}) and a powder seed coating (Sepiret PF, rate of 200 g. 100 kg^{1}); and Technology IV – metalaxyl-m 10 g. L^{-1} + fludioxonil 25 g. L^{-1} (Maxim XL rate of 62.5 mL. 100 kg^{-1}), thiamethoxam 350 g. L^{-1} (Cruiser 350 FS®, rate of 156.25 mL. 100 kg^{-1}), the polymer (Florite Green®, rate of 125 mL. 100 kg^{-1}) and a powder seed coating (Sepiret PF, rate of 200 g. 100 kg^{-1}). Fertilizer I: 7% N, 16% P_2O_5, 0.6% Co, 2.5% Mo (rate of 200 mL. 100 kg^{-1} of seeds); and Fertilizer II: 1% Co, 10% Mo, and 7% P_2O_5 (rate of 200 mL . 100 kg^{-1} of seeds).

Table 1. Industrial treatment of soybean seeds with the respective volumes of seed-coating mixtures.

Treatment	Description	Volume of seed-coating mixture (mL. 100 kg^{-1})
T1	Control	-
T2	Technology I	900
T3	Technology I + Fertilizer I	1100
T4	Technology I + Fertilizer II	1100
T5	Technology II	400
T6	Technology II + Fertilizer I	600
T7	Technology II + Fertilizer II	600
T8	Technology III	750
T9	Technology III + Fertilizer I	950
T10	Technology III + Fertilizer II	950
T11	Technology IV	343.75
T12	Technology IV + Fertilizer I	543.75
T13	Technology IV + Fertilizer II	543.75

Technology I: [carbendazim + thiram] + [imidacloprid + thiodicarb]; Technology II: [pyraclostrobin + thiophanate-methyl] + fipronil; Technology III: [thiophanate-methyl + fluazinam] + [bifenthrin + imidacloprid], and Technology IV: [metalaxyl-m + fludioxonil] + thiamethoxam, Fertilizer I: 7% N, 16% P_2O_5, 0.6% Co, and 2.5% Mo; and Fertilizer II: 1% Co, 10% Mo, and 7% P_2O_5.

The seeds were treated in a continuous lot seed-coating device (Cimbria Centricoater CC10) and were then placed in Kraft paper bags and kept in laboratory ambient conditions (mean ambient temperature of 22 ºC and mean relative humidity of 66%), simulating conventional storage. The physiological potential of the seeds was evaluated in each one of the five storage periods (0, 30, 60, 90, and 120 days) by means of the tests described below.

The germination test was conducted with 400 seeds distributed in eight subsamples of 50 seeds for each treatment so as to meet the criteria established in the Rules for Seed Testing (Brasil, 2009). Using this same procedure, the first count of germination was carried out on the fifth day after the beginning of the test. In both tests, only the seedlings with a main root and above ground part greater than 3 cm with the presence of at least 2 seminal roots were considered normal (Nakagawa, 1999).

The accelerated aging test was conducted with eight subsamples of 50 seeds per treatment. Before being placed for germination in the germination test, the seeds were arranged on stainless steel screens inserted within plastic boxes containing 40 mL of distilled water (Krzyzanowski et al., 1991). After that, the boxes were placed in a water-jacketed incubator (model 3015, VWR, USA), regulated at 41 ± 1 ºC for 48 hours (Marcos-Filho, 1999). Evaluation was made on the fifth day after sowing, counting the seedlings considered normal. Results were expressed in percentage of normal seedlings.

The emergence speed index in sand substrate was conducted with eight subsamples of 50 seeds for each treatment. In this test, the sand used was first washed and placed in plastic trays under greenhouse conditions and moisture was maintained with moderate irrigations. Daily notations were made of the number of normal emerged seedlings up to 15 days after sowing, according to Nakagawa (1999). The results were expressed as proposed by Maguire (1962).

Seedling emergence testing in sand substrate was carried out through sowing in trays containing washed sand, with eight subsamples of 50 seeds for each treatment. Normal emerged seedlings were counted at 15 days after sowing, according to Nakagawa (1999). Results were expressed in percentage.

The data of the response variables were subjected to analysis of variance in line with its basic presuppositions (normally distributed errors with a mean of zero and variance in common). For that purpose, the Shapiro-Wilk test was used for the normality test and the Levene test for the homogeneity of residual variances (Banzatto and Kronka, 2008). Except for the emergence speed index, all the other variables exhibited heterogeneity of variances and were, therefore, previously transformed in arcsine. However, in the tables, the original mean values were used.

Within the same storage period, the means were compared by subjecting them to the Scott-Knott test at 5% probability. Regression analysis was used to compare the storage periods to check the fit of the polynomial models (linear and quadratic) and non-linear models (logarithmic and exponential) for dependent variables. The models adopted were those that proved to be significant with non-significant deviations of regression at the level of 5% probability, according to Banzatto and Kronka (2008).

Results and Discussion

Up to 60 days of storage (Table 2), none of the treatments used compromised the sales potential of the seeds, since the mean values of normal seedlings in the germination test were above 80%, the value established by the Brazilian Ministry of Agriculture (Ministério da Agricultura, Pecuária e Abastecimento) as the minimum assurance for sale of soybean seeds in Brazil (Brasil, 2005). This result corroborates Zambon (2013) and Strieder et al. (2014), who recommend performing the industrial treatment a maximum of 60 days before the beginning of sowing so as to minimize possible effects toxic to plants from the mixtures placed on the seeds. However, in addition to the control (T1), seven other treatments (T3 and T4, T5, T7 and T8, T10, and T11) remained suitable for sale up to 90 days, with treatments T1, T4, and T11 standing out in a positive way.

Table 2. Normal seedlings in the germination test (%) and in the seedling emergence test in sand substrate in response to industrial seed treatments in five storage periods.

	Germination					Emergence in sand substrate				
	0	30	60	90	120	0	30	60	90	120
T1	92.50 a	87.25 a	85.25 a	81.25 a	77.52 b	100.00 a	98.00 a	93.50 a	90.50 a	85.25 b
T2	90.00 b	85.50 b	83.00 b	77.00 b	71.54 c	97.50 a	93.50 a	90.00 b	87.00 b	83.00 b
T3	90.50 b	88.75 a	84.50 b	80.75 b	77.21 b	95.50 a	91.00 a	89.00 b	84.50 b	84.50 b
T4	93.00 a	90.25 a	86.75 a	83.75 a	80.89 a	99.00 a	97.50 a	93.00 a	90.00 a	86.75 a
T5	90.50 b	88.50 a	87.00 a	82.25 a	77.82 b	96.00 a	91.00 a	90.00 b	85.00 b	87.00 a
T6	90.75 b	86.50 b	84.00 b	78.50 b	73.56 b	98.50 a	97.50 a	94.50 a	90.50 a	84.00 b
T7	93.00 a	91.25 a	88.00 a	83.25 a	78.77 b	94.00 a	91.50 a	88.50 b	84.50 b	88.00 a
T8	93.75 a	91.50 a	89.75 a	83.50 a	77.80 b	98.00 a	95.00 a	90.50 b	86.00 b	84.75 b
T9	92.75 a	88.50 a	86.25 a	77.75 b	70.13 c	93.00 b	90.00 b	88.00 b	82.00 b	86.25 a
T10	96.00 a	91.25 a	86.50 a	82.25 a	78.23 b	100.00 a	100.00 a	97.00 a	93.50 a	86.50 a
T11	93.75 a	88.50 a	86.25 a	83.75 a	81.37 a	96.50 a	93.00 a	93.50 a	89.25 a	83.75 b
T12	89.00 b	85.50 b	81.50 b	77.00 b	72.76 c	88.50 b	86.00 b	82.50 b	81.50 b	79.00 b
T13	96.00 a	87.50 a	82.50 b	77.50 b	75.21 b	100.00 a	99.25 a	96.00 a	91.00 a	82.50 b

Mean values followed by the same lowercase letter in the column do not differ significantly by the Scott-Knott test at the level of 5% probability at a fixed storage period.

In relation to the addition of fertilizers to the industrial treatments, in the period in which all the treatments proved to be suitable for sale (up to 60 days), it can be seen in Table 2 that in the four technologies tested, Fertilizer II provided germination results equal to the results of the control (T13 was an exception at 60 days), as well as greater than or equal to the treatments in which only the fungicides, insecticides, and polymers were used. Identical behavior was observed for treatment T7, in which the addition of Formulation II also resulted in a percentage of normal seedlings greater than or similar to that of the standard treatment (T5) in the germination test. For the other technologies, the effects of the fertilizers ranged from neutral to harmful.

Analysis of germination data over the storage period allowed equations of decreasing linear regression to be fitted (Table 3), signaling that there was reduction in germination over the 120-day storage period. It was found (Table 3) that the treatments that share the products of Technology I (T2, T3, and T4), as well as of Technology III (T8, T9, and T10), were those that led to the biggest reductions in seed germination potential for each day of storage. This condition worsened when fertilizers were added, as the angular coefficients of the equations show (Table 3).

Pereira et al. (2009) observed that antifungal commercial products based on carbendazim, thiophanate-methyl, fludioxonil + mefenoxam, thiabendazole + thiram, and carboxin + thiram did not affect soybean seed germination and emergence when analyzed immediately after treatment. The same was indicated by Avelar et al. (2011), who did not observe adverse effects from the fungicides based on metalaxyl-m + fludioxonil, the same used in Technology IV, on soybean seeds over 180 days of storage. However, after addition of the insecticide thiamethoxam to the seed-coating mixtures, the latter authors indicated reductions in germination and vigor, signaling that the harmful effects may be related, alone or in combination, to the phytotoxic nature of this insecticide or to the increase in the volume of the seed-coating mixture.

In part, what was indicated above, suggests that the greater reductions observed in Technologies I and III (Table 3) may be associated with the high volumes of the seed-coating mixtures, which ranged from 750 to 950 mL. 100 kg^{-1} for the first and from 900 to 1100 mL. 100 kg^{-1} for the second. According to Embrapa (2011), the volume of the coating mixture for soybean seeds should not exceed 600 mL. 100 kg^{-1}, the limit of aqueous solution tolerated so that damage to the membranes does not occur. Such recommendation is based on studies with aqueous solutions; however, current seed-coating mixtures are composed of different products already formulated in a liquid medium, whose osmotic potential is thus different from the aqueous solutions evaluated in that study. Recently, Segalin et al. (2013) affirmed that volumes of up to 1400 mL. 100 kg^{-1} of seeds have already been tested in the soybean crop and did not cause damage to seed quality. However, it should be emphasized that in that study, the seeds were not subjected to storage periods, as the seeds of this study were.

Neither harmful effects nor benefits of the seed treatments tested were found in emergence in sand substrate up to 30 days of storage (Table 2), except for treatments T9 and T12. As the respective pairs of these treatments (T7 and T8 for T9, and T11

Table 3. Linear regression equations for the industrial treatments of soybean seeds according to storage periods.

Treat.	Linear Regression Equations		
	Germination	Emergence	First Count
T1	Y= 91.92 - 0.019 x. r^2 0.99	Y= 100.00 - 0.019 x. r^2 0.95	Y= 84.94 - 0.123 x. r^2 0.94
T2	Y= 90.72 - 0.157 x. r^2 0.98	Y= 97.78 - 0.130 x. r^2 0.98	Y= 86.51 - 0.352 x. r^2 0.92
T3	Y= 91.27 - 0.165 x. r^2 0.99	Y= 94.52 - 0.093 x. r^2 0.89	Y= 82.19 – 0.266 x. r^2 0.92
T4	Y= 92.98 - 0.164 x. r^2 0.99	Y= 99.10 - 0.101 x. r^2 0.97	Y= 96.60 - 0.334 x. r^2 0.87
T5	Y= 91.97 - 0.116 x. r^2 0.96	Y= 93.38 - 0.116 x. r^2 0.89	Y= 88.38 – 0.152 x. r^2 0.96
T6	Y= 90.82 - 0.133 x. r^2 0.96	Y= 90.82 - 0.019 x. r^2 0.91	Y= 86.68 - 0.145 x. r^2 0.98
T7	Y= 94.92 - 0.141 x. r^2 0.96	No fit	Y= 85.23 - 0.170 x. r^2 0.98
T8	Y= 96.22 - 0.157 x. r^2 0.99	Y= 98.08 - 0.101 x. r^2 0.98	Y= 89.67 - 0.206 x. r^2 0.98
T9	Y= 98.92 - 0.177 x. r^2 0.96	Y= 92.84 - 0.089 x. r^2 0.81	No fit
T10	Y= 95.72 - 0.187 x. r^2 0.99	No fit	Y= 83.72- 0.133 x. r^2 0.98
T11	Y= 92.39 - 0.092 x. r^2 0.97	Y= 95.41 - 0.086 x. r^2 0.85	Y= 89.01 - 0.151 x. r^2 0.95
T12	Y= 89.27 - 0.134 x. r^2 0.99	Y= 88.04 - 0.076 x. r^2 0.98	Y= 81.01 - 0.135 x. r^2 0.91
T13	Y= 93.13 - 0.148 x. r^2 0.93	No fit	Y= 84.61 - 0.187 x. r^2 0.93

and T3 for T12) did not differ statistically among themselves, or from the control (Table 2), this response probably arose from the addition of Fertilizer I to the respective seed-coating mixtures.

The combined results of the germination and emergence tests at time zero show that the seed lot tested had high initial germination potential (Table 2). In this regard, Marcos-Filho et al. (2009) indicated that high quality seed lots had lower fluctuation in the percentage of emerged seedlings. Taylor and Salanenka (2012), furthermore, affirm that, unlike tests with paper as a substrate, in emergence, sand attenuates the concentration of active ingredients near the seeds, reducing possible phytotoxic effects from the seed-coating mixtures. However, with storage between 60 and 90 days (Table 2), i.e., as seed deterioration advanced, the emergence test in sand substrate allowed lots to be differentiated in different levels of vigor. The control treatment (T1) stood out in a positive way, as well as the treatments T4, T6, T10, and T13.

Up to 60 days of storage, in first count of germination (Table 4), all the treatments based on Technologies II, III, and IV had values equivalent to the untreated control (Table 2). In contrast, only the combination of fungicide, insecticides, and polymers in Technology I had treatments with values of normal seedlings lower than the untreated control (T1),

.Table 4. Normal seedlings in the first count of the germination test (%) and the emergence speed index for the industrial seed treatments in five storage periods.

Treat.	First count					Emergence speed index				
	0	30	60	90	120	0	30	60	90	120
T1	86.75 a	80.50 a	77.25 a	71.25 a	70.25 a	13.47 a	12.83 a	12.36 a	11.44 a	10.60 a
T2	81.50 b	76.75 b	70.25 b	62.00 b	36.76 c	13.17 a	11.15 b	8.03 b	4.65 c	4.15 c
T3	78.00 b	75.25 b	69.00 b	59.75 b	55.38 c	12.15 b	10.39 b	8.00 b	7.39 c	4.75 c
T4	89.75 a	76.50 a	81.75 a	73.50 a	61.05 b	12.11 b	10.92 b	4.59 c	3.79 c	2.24 c
T5	87.75 a	83.25 a	80.50 a	76.75 a	67.85 b	13.52 a	13.08 a	12.76 a	11.25 a	6.44 b
T6	86.75 a	82.25 a	78.75 a	72.00 a	69.90 a	13.37 a	13.11 a	12.72 a	12.34 a	7.18 b
T7	85.00 a	80.25 a	76.50 a	68.25 b	64.40 b	13.36 a	12.69 a	10.41 b	9.82 b	5.21 c
T8	90.25 a	83.50 a	77.75 a	69.25 b	64.01 b	13.54 a	12.87 a	10.59 b	9.77 b	4.99 c
T9	85.00 a	81.50 a	75.25 a	57.25 b	51.56 c	13.41 a	13.26 a	12.79 a	11.10 a	9.43 a
T10	83.00 a	79.25 a	77.50 a	72.00 a	67.36 b	13.37 a	12.74 a	10.30 b	9.63 b	6.75 b
T11	91.00 a	84.00 a	79.00 a	73.50 a	69.37 a	13.10 a	12.49 a	11.84 a	11.17 a	6.29 b
T12	83.00 a	77.00 a	71.25 b	67.50 b	62.30 b	11.59 b	10.97 b	9.99 b	9.22 b	6.53 b
T13	88.00 a	80.00 a	71.25 b	62.50 b	58.11 b	13.16 a	12.22 a	9.74 b	9.05 b	6.79 b

Mean values followed by the same lowercase letter in the column do not differ significantly by the Scott-Knott test at the level of 5% probability, at a fixed storage period.

with a clear exception, when the combination was used with Fertilizer II (Table 2).

In relation to emergence speed index, treatments T3, T4, and T12 were the only ones that led to decreases in vigor immediately after the treatment, and treatment T2 joined them at 30 days. In contrast, for accelerated aging, the highest percentage of normal seedlings was observed for treatment T6 (Technology II + Fertilizer I) up to 90 days of storage, the period in which the control (T1) stood out in a positive way (Table 5).

In regard to regression analysis (Table 3), all the treatments led to a decrease in the mean of normal seedlings superior to the untreated control (T1), regardless of the test used for evaluation of physiological quality. However, if we compare the angular coefficients of the four mixtures tested commercially (T2, T5, T8 and T11) with the angular coefficients of their respective pairs with fertilizer (Table 1), we find that for each day of storage, the addition of micronutrient-based formulations led to greater decreases both in the germination test (Table 3) and in the accelerated aging test (Table 6). Nevertheless, for the emergence test in sand substrate, this reduction was less with the application of fertilizers (Table 6), while in the first count, a pattern of behavior was not observed since the results ranged from beneficial to harmful according to the technology and the fertilizer (Table 6).

In regression analysis (Tables 5 and 6), when the deviation of regression was significant, it was not possible to obtain a statistical model able to establish a functional relation with the values found (Banzatto and Kronka, 2008). It may be supposed in these cases that only additional physiological analyses in shorter intervals would make it possible to investigate if the absence of fit is an authentic lack of structuring of the data, the fruit of recoating the seeds, or if it arises from the period used (30 days), which was insufficient for picking up this tendency.

For all the physiological quality tests (germination, emergence in sand substrate, first count, and emergence speed index), among the combinations tested, the treatments based on Technology I (T2, T3, and T4) exhibited the greatest decreases in seed physiological potential, probably due to the combination of high volumes of the seed-coating mixtures with the phytotoxic nature of the active ingredients used (carbendazim/thiram + imidacloprid/thiodicarb). In this respect, Segalin et al. (2013) and Marcos-Filho (2015) indicate that high volumes of seed-coating mixture promote damage to membranes because of the high speed of initial imbibition, whereas Dan et al. (2010) and Dan et al. (2011) found that soybean seeds covered with imidacloprid and

Table 5. Normal seedlings in the accelerated aging test (%) for industrial seed treatments and storage periods.

Treat.	0	30	60	90	120
T1	76.00 b	67.75 c	61.00 b	52.75 b	49.75 a
T2	78.75 b	67.50 c	58.00 c	47.50 b	27.88 c
T3	71.50 b	57.00 d	54.75 c	39.25 c	25.90 c
T4	83.50 b	74.75 b	65.25 b	44.25 b	23.74 c
T5	82.00 b	70.75 b	65.75 b	51.50 b	34.86 b
T6	90.00 a	80.75 a	73.25 a	62.25 a	29.51 c
T7	79.50 b	72.00 b	65.25 b	55.25 b	32.65 b
T8	72.00 b	66.00 c	56.75 c	47.75 b	35.13 b
T9	71.25b	61.00 d	50.50 c	39.75c	23.68 c
T10	65.25c	55.50 d	41.00 d	27.00 d	15.22 d
T11	76.00 b	66.75 c	60.75 b	52.50 b	29.30 c
T12	83.50 b	72.25 b	62.75 b	50.00 b	29.49 c
T13	76.75 b	64.50 c	53.75 c	42.25 c	32.95 b

Mean values followed by the same lowercase letter in the column do not differ significantly by the Scott-Knott test at the level of 5% probability, at a fixed storage period.

Table 6. Linear regression equations for the industrial treatments of soybean seeds according to storage periods.

Treatment	Linear Regression Equations	
	Emergence speed index	Accelerated aging
T1	Y= 13.59 - 0.024 x. r^2 0.99	Y= 73.70 - 0.193 x. r^2 0.95
T2	Y= 12.59 - 0.968 x. r^2 0.92	Y= 80.81 - 0.419 x. r^2 0.98
T3	Y= 12.11 – 0.597 x. r^2 0.98	Y= 71.17 – 0.455 x. r^2 0.97
T4	No fit	Y= 88.01 – 0.492 x. r^2 0.97
T5	No fit	Y= 84.12 - 0.389 x. r^2 0.98
T6	No fit	Y= 96.66 - 0.405 x. r^2 0.93
T7	No fit	Y= 82.75 - 0.461 x. r^2 0.98
T8	Y= 14.65 - 0.073 x. r^2 0.93	Y= 74.03– 0.409 x. r^2 0.99
T9	Y= 14.19 - 0.381 x. r^2 0.92	Y= 72.40 - 0.450 x. r^2 0.99
T10	Y= 13.96 - 0.057 x. r^2 0.97	Y= 65.13 - 0.394 x. r^2 0.98
T11	No fit	Y= 89.01 - 0.151 x. r^2 0.95
T12	Y= 12.24 - 0.448 x. r^2 0.93	Y= 86.07 - 0.444 x. r^2 0.91
T13	Y= 13.26 - 0.054 x. r^2 0.98	Y= 75.01 - 0.341 x. r^2 0.99

analyzed on the same day on which they were treated did not exhibit adverse effects on vigor and germination. However, with storage, physiological potential was drastically reduced.

Castro et al. (2008), however, obtained superior physiological quality results in soybean seeds treated with imidacloprid. In that study, the authors used seed-coating mixtures that contained only the imidacloprid-based insecticide product, with volumes that did not exceed 300 mL. 100 Kg^{-1} of seed. Such results, therefore, may explain the superior results of physiological quality of Technology I in relation to Technology III (also with an imidacloprid base), because in addition to containing other insecticide substances in the seed-coating mixture, in the latter, the volume of the seed-coating mixture was less. This reinforces the signs that the adverse effects of the seed-coating mixtures result from the combination of different factors and not only the active ingredients, as, for example, the storage period and conditions and the volume of the seed-coating mixture.

In spite of their plant health protective functions, commercial formulations of some pesticides may also be capable of mediating beneficial effects on plant metabolism and morphology (Munkvold, 2009), generating the phenomenon called "phytotonic effect" (Castro et al., 2008). In this respect, it is plausible that the germination performance of the treatments that achieved the minimum assurance of germination beyond 60 days of storage (Table 2) may be the result of a possible stimulant effect of the seed-coating mixtures used, especially treatments T4 and T11, which even exceeded the untreated control (T1) at 120 days of storage.

From Table 2, it can be seen that while for Technology I, both fertilizers benefitted germination (Table 2) so as to allow sale of the seed lot at 90 days, a contrary effect was observed in Technology IV, in which only T11 achieved the minimum assured germination in this period. In contrast, for Technology II and III, Fertilizer II had a neutral effect, while in the treatments in which Fertilizer I was added, germination reached values lower than 80% of normal seedlings. Moreover, superior germination performance was obtained in associating Fertilizer I and Technology II (T6) in the accelerated aging test (Table 5), compared to the results of their pairs of technology (T5 and T7).

Bays et al. (2007) reported a neutral effect after the application of cobalt, molybdenum, and boron on seeds in chemical treatment, whereas Binsfeld et al. (2014) observed superior vigor in soybean lots when treated with a nutrient complex, which, in addition to these micronutrients, contained nitrogen and phosphorus, as in the fertilizers used in this study. However, these elements cannot be credited with the positive effect on physiological potential observed in this study, nor in that of Binsfeld et al. (2014) since, during emergence, cotyledons are the structures responsible for seedling nutrition (Marcos-Filho, 2015).

However, polynutrient nutritional complexes, as in the fertilizers used in this study, may contain additives such as amino acids and organic acids, substances that at determined concentrations do not need to be listed on the product label, but are able to trigger a phytotonic effect on soybean seeds. This hypothesis is reinforced by a study conducted by Ludwig et al. (2011), who observed that application of amino acids on soybean seeds had effects on seed germination potential ranging from neutral to positive.

Among the commercial treatments tested (T2, T5, T7, and T11), in the germination test, treatment T11 was the only one that remained suitable for sale over 120 days of storage, maintaining its position in the group of superior results also in the tests of emergence in sand substrate, first count of germination, and emergence speed index up to 90 days. In contrast, in relation to accelerated aging up to 90 days, T11 was equal to the control, but classified in inferior groups. Part of these results can be explained by the lower volume of seed-coating mixture used in Technology IV, which minimizes the damage caused to membranes.

Positive effects on seed quality with the insecticide thiamethoxam (Technology IV) were also indicated by Dan et al. (2010), Dan et al. (2011) and Avelar et al. (2011). Castro and Pereira (2008) summarize that together with aldicarb, thiamethoxam is considered a bioactivator product that can lead to increases in biomass. However, biotests with this active ingredient conducted in tomato seeds sensitive to gibberellin, auxin, and cytokinin signaled that this molecule does not act in the plant in the same way as these three groups of growth promotors (Castro and Pereira, 2008), but appears to increase water uptake and stomatal resistance, improving the water balance of the plant.

Conclusions

Germination and vigor of soybean seeds decrease over the storage period, above all after industrial treatment.

Seed-coating mixtures based on carbendazim/thiram + imidacloprid/thiodicarb hurt the physiological potential of seeds over the storage period, regardless of the use of fertilizers in the industrial treatment.

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