

## Treatment with fungicides and insecticides on the physiological quality and health of wheat seeds<sup>1</sup>

Julia Abati<sup>2\*</sup>, Claudemir Zucareli<sup>2</sup>, José Salvador Simoneti Foloni<sup>2</sup>,  
Fernando Augusto Henning<sup>3</sup>, Cristian Rafael Brzezinski<sup>2</sup>, Ademir Assis Henning<sup>3</sup>

**ABSTRACT** – Seed treatment with insecticides and fungicides has become an important practice for ensuring initial plant stand in establishing crops. In this context, the aim of this study was to evaluate the influence of chemical seed treatment with insecticides and fungicides on the physiological quality and health of the seeds of wheat cultivars. Seeds of the wheat cultivars BRS Pardela and BRS Gaivota were used, subjected to the following chemical treatments: 1- control, 2- carboxin + thiram + imidacloprid + thiodicarb, 3- carbendazim + thiram + imidacloprid + thiodicarb, 4- fipronil + thiophanate-methyl + pyraclostrobin, 5- triadimenol + imidacloprid + thiodicarb, 6- fipronil, and 7- imidacloprid + thiodicarb. Physiological quality was evaluated by tests of germination, accelerated aging, the length and dry weight of shoots and roots, and seedling emergence in the field. Seed health quality was evaluated by the blotter test method. The seeds of the wheat cultivars tested respond differently to the chemical treatments in regard to effects on germination and vigor. The treatment with triadimenol + imidacloprid + thiodicarb is harmful to seedling development. For the BRS Gaivota cultivar, the seed treatment with carboxin + thiram + imidacloprid + thiodicarb; and carbendazim + thiram + imidacloprid + thiodicarb improved seedling establishment in the field compared to the control.

Index terms: *Triticum aestivum* L., seed treatment, germination, vigor.

## Tratamento com fungicidas e inseticidas na qualidade fisiológica e sanitária de sementes de trigo

**RESUMO** - O tratamento de sementes com inseticidas e fungicidas tem se tornado uma prática importante para assegurar o estande inicial de plantas na implantação das lavouras. Diante disso, o objetivo foi avaliar a influência do tratamento químico de sementes com inseticidas e fungicidas sobre a qualidade fisiológica e sanitária de sementes de cultivares de trigo. Foram utilizadas sementes das cultivares de trigo BRS Pardela e BRS Gaivota, submetidas aos seguintes tratamentos químicos: 1- testemunha, 2- carboxin + thiram + imidacloprido + tiodicarbe, 3- carbendazim + thiram + imidacloprido + tiodicarbe, 4- fipronil + tiofanato metílico + piraclostrobina, 5- triadimenol + imidacloprido + tiodicarbe, 6- fipronil e 7- imidacloprido + tiodicarbe. A qualidade fisiológica foi avaliada por meio dos testes de germinação, envelhecimento acelerado, comprimento e massa seca de parte aérea e raiz, emergência de plântulas em campo e a qualidade sanitária pelo método do *blotter test*. As sementes das cultivares de trigo testadas respondem de forma diferenciada aos tratamentos químicos quanto aos efeitos na germinação e vigor. O tratamento com triadimenol + imidacloprido + tiodicarbe é prejudicial ao desenvolvimento de plântulas. Para a cultivar BRS Gaivota o tratamento de sementes com carboxin + thiram + imidacloprido + tiodicarbe e carbendazim + thiram + imidacloprido + tiodicarbe favorece o estabelecimento das plântulas em campo, em relação a testemunha.

Termos para indexação: *Triticum aestivum* L., tratamento de sementes, germinação, vigor.

### Introduction

The wheat (*Triticum aestivum* L.) crop holds an important position in the world and Brazilian domestic grain market. In Brazil, annual production of the cereal crop oscillates between five and six million tons, with 90% of this production centered in the

southern region of the country (Embrapa, 2014). Nevertheless, around 50% of the wheat consumed within Brazil is imported (CONAB, 2014). Thus, there is great socioeconomic interest in the country in increasing its production so as to meet domestic demand and because of the benefits generated from growing it (Barbieri et al., 2013).

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<sup>2</sup>Departamento de Agronomia, UEL, Caixa Postal 6001, 86051-990 - Londrina, PR, Brasil.

<sup>3</sup>Embrapa Soja, Caixa Postal 231, 86001-970 - Londrina, PR, Brasil.

\*Corresponding author <juliaabati@yahoo.com.br>

Together with the use of quality seeds, the application of fungicides and insecticides through seed treatment is of utmost importance in obtaining high yields. This is because fungicides and insecticides act in protection against seed pathogens, whether from storage or present in the soil, and also act against the initial attack of pests specific to the soil (Antonello et al., 2009; Menten and Moraes, 2010; Pereira et al., 2010).

Chemical seed treatment is considered one of the most effective methods of control of fungi and insects. Nevertheless, Kashypa et al. (1994), in studies on wheat seeds, and Ludwig et al. (2011) on soybean seeds, found that some chemical products, when applied on the seeds, may lead to reduction in germination and hinder seedling survival. According to Antonello et al. (2009) and Deuner et al. (2014), who worked with maize seeds, the phytotoxicity effect depends on the product used and the time that the seeds were stored. According to Goulart (1988), the fungicide triadimenol, although it was effective for seedling protection (providing for total absence of disease lesions on wheat), proved to be phytotoxic, leading to the rise of abnormal seedlings in the germination process.

In contrast, Matos et al. (2013) observed that maize seeds treated with different fungicides exhibited germination and vigor superior to the untreated control in the cold test. This difference may be explained as a result of the chemical products controlling the pathogens present in the seeds since these seeds exhibited low quality of health.

Associated with chemical treatment, some insecticides, in addition to providing pest control in the crop, may act physiologically, assisting both initial growth and plant development (Dan et al., 2012). Corroborating this affirmation, Hossen et al. (2014) verified a greater percentage and speed of germination in wheat seeds treated with the insecticide thiamethoxam in relation to untreated seeds. Furthermore,

Barros et al. (2005) found similar results in common bean seeds treated with the insecticide fipronil.

The practice of seed treatment is widely adopted in various crops; however, in the wheat crop, the results are still in the initial stages, especially with the advent of new formulations and chemical products that are being introduced on the market, as well as new cultivars. It is noteworthy that together with the use of these new products and formulations, there is a trend of recommending the use of a smaller quantity of seeds per area, especially for these new cultivars. Thus, the correct recommendation of products for seed treatment becomes essential in assuring uniform germination and seedling emergence, especially considering the possibility of occurrence of stress conditions, seed colonization by fungi, and pest attack. It is also important to recommend the use of seeds of high physiological and genetic quality associated with physical purity and health to obtain crops with adequate stand and vigorous seedlings (Scheeren et al., 2010; Dan et al., 2012).

In light of the above, the aim of this study was to evaluate the influence of chemical treatment with insecticides and fungicides on the physiological quality and health of seeds of wheat cultivars.

## Material and Methods

The experiment was conducted at the Seed and Grain Technological Center and in the experimental field of the Empresa Brasileira de Pesquisa Agropecuária (Embrapa), at the National Soybean Research Center (Embrapa Soja) in Londrina, Parana, Brazil.

The wheat cultivars used were BRS Pardela and BRS Gaivotá, evaluated separately, with seven chemical seed treatments, involving fungicides and insecticides (Table 1).

Table 1. Active ingredients, commercial names, and application rates used for seed treatment of the wheat cultivars BRS Pardela and BRS Gaivotá.

Treatments	Active ingredient (a.i.)	Commercial name	Commercial product application rate <sup>1</sup>
T1	Control	-	-
T2	carboxin <sup>2</sup> + thiram <sup>2</sup> + imidacloprid <sup>3</sup> + thiodicarb <sup>3</sup>	Vitavax-Thiram <sup>®</sup> + Cropstar <sup>®</sup>	250 + 100
T3	carbendazim <sup>2</sup> + thiram <sup>2</sup> + imidacloprid <sup>3</sup> + thiodicarb <sup>3</sup>	Derosal Plus <sup>®</sup> + Cropstar <sup>®</sup>	200 + 100
T4	fipronil <sup>3</sup> + thiophanate methyl <sup>2</sup> + pyraclostrobin <sup>2</sup>	Standak Top <sup>®</sup>	100
T5	triadimenol <sup>2</sup> + imidacloprid <sup>3</sup> + thiodicarb <sup>3</sup>	Baytan <sup>®</sup> + Cropstar <sup>®</sup>	250 + 100
T6	fipronil <sup>3</sup>	Fipronil <sup>®</sup>	100
T7	imidacloprid <sup>3</sup> + thiodicarb <sup>3</sup>	Cropstar <sup>®</sup>	250

<sup>1</sup>Commercial product application rate: mL.100 kg<sup>-1</sup> of seeds.

<sup>2</sup>Active ingredients of the fungicide class.

<sup>3</sup>Active ingredients of the insecticide class.

Chemical treatment of the seeds was carried out in plastic bags in which the products were added over the seeds, followed by shaking until the seeds were completely covered, with volume of the mixture of 600 mL.100 kg<sup>-1</sup> of seeds (product + water).

After the seed treatment, the physiological quality and health of the seeds was evaluated by the following tests:

*Germination:* carried out on four subsamples of 50 seeds per replication, for a total of 600 seeds per treatment. The seeds were distributed over germitest paper with a water volume for soaking at the quantity of 2.5 times the dry weight of the substrate, in the form of rolls. The seeds were placed in a germinator at a temperature of 20 °C for eight days. After that, evaluations were carried out according to the recommendations of the Rules for Seed Testing [Regras para Análise de Sementes] (Brasil, 2009), and the results were expressed in percentage of normal and abnormal seedlings.

*Accelerated aging:* four subsamples of 50 seeds per replication were used, placed in gerbox type plastic boxes, with screen supports, containing 40 mL of distilled water at the bottom. The seeds were uniformly distributed on the screen in a single layer. After that, they were placed in a water-jacketed incubation chamber at 42 °C for 60 hours (Marcos-Filho, 1999). After this period, they were subjected to the germination test.

*Length of total seedling, shoot, and root:* four subsamples of 25 seeds per replication were used. The germination paper was moistened with distilled water at 2.5 times the dry weight of the substrate. The seeds were arranged on the upper third in

the lengthwise direction of the paper. The rolls were placed in plastic bags which were arranged vertically in the germinator for seven days at 20 °C. After that, the length of the normal seedlings was checked (total length of the seedling, of the shoot, and of the root) with a ruler in millimeters. The results were expressed in centimeters per seedling.

*Dry matter weight of the shoot and root:* this was carried out on the normal seedlings obtained in the test of seedling length. After measuring seedling length, the remaining part of the seed was removed and the shoot and root were separated. These were then placed in paper bags and placed in an air circulation laboratory oven, where they remained for 24 hours at a temperature of 80 °C (Nakagawa, 1999). At the end of this period, dry weight was checked on a precision scale accurate to 0.0001 g, and the results were expressed in mg per seedling.

*Seedling emergence in the field:* seeds were sown in April 2013, using a density of 300 seeds per m<sup>2</sup>. Plots consisted of six rows of six-meter length, spaced at 0.20 m, for a total area of 7.2 m<sup>2</sup> per plot. Only the four center rows were considered as useful area, leaving the 0.5 m at the beginning and end of the plot as a border. At 15 days after sowing, the total number of emerged seedlings was counted in a total area of 0.75 m<sup>2</sup>, which was composed of three subsamples of 0.25 m<sup>2</sup>, and the result was expressed in seedlings per m<sup>2</sup>. The data on mean daily temperature and rainfall during the period of sowing and seedling emergence were obtained from the meteorological station of Embrapa Soja, located at approximately 2000 m from the experiment, in Londrina (Figure 1).

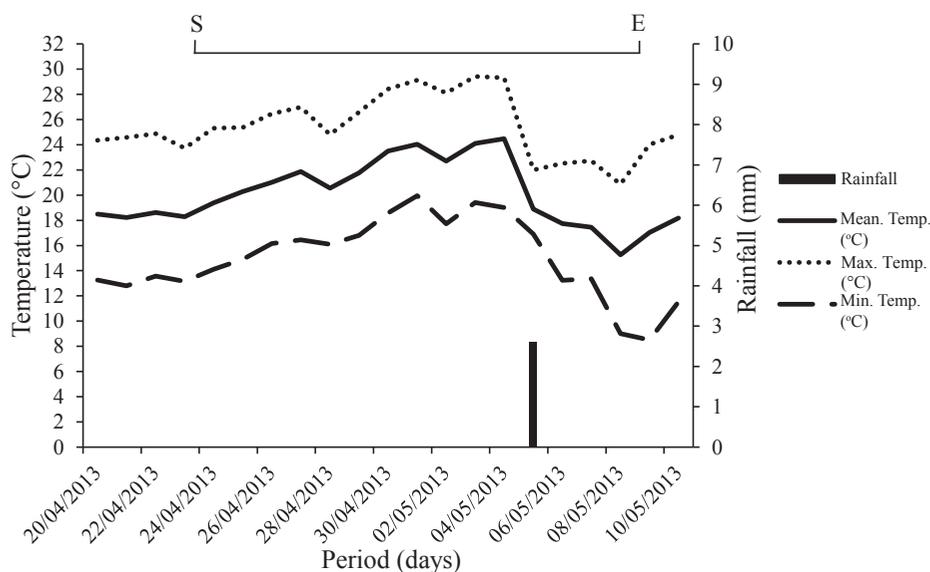


Figure 1. Maximum, minimum, and mean daily temperature (°C) and rainfall during the period of seedling emergence in the field. S: sowing; E: emergence.

*Seed health analysis:* the method used was the blotter test. The seeds were arranged in gerbox type plastic boxes, duly disinfected with sodium hypochlorite at 1.05%. The boxes contained three sheets of filter paper moistened with distilled water and autoclaved. Ten gerboxes with 20 seeds taken at random were used, with three replications, for a total of 600 seeds per treatment. After that, the seeds were placed in an incubation chamber where they remained for seven days at a temperature of 20 °C ( $\pm 2$  °C) under continuous fluorescent light (Henning, 2005). Fungi were identified with the aid of a binocular stereoscopic microscope and optical microscope. The results were expressed in percentage.

A completely randomized design was used for laboratory evaluations, with three replications and, for the field evaluation, a randomized block design was used, with four replications. The data obtained were subjected to analysis of variance in an independent way for each cultivar, and the mean values were compared by the Tukey test at 5% probability. Analyses were carried out by the computational program Sistema para Análise de Variância (System

for Analysis of Variance) - SISVAR (Ferreira, 2011).

## Results and Discussion

The presence of pathogenic fungi was not observed in seed health analysis; there was only the presence of fungi of the genus *Epicoccum* sp. and *Alternaria* sp. According to Celano et al. (2012), these fungi are considered saprophytes in the wheat crop and are not pathogenic agents.

The absence of pathogenic fungi, both in the treated seeds and in the control, may be associated with the use of commercial seed lots produced with high physiological quality and health. According to França-Neto et al. (2010), the use of healthy seeds, associated with physiological, genetic, and physical quality, is fundamental for obtaining an adequate stand of plants, and is thus a focus of seed industries.

The effects of chemical seed treatments on physiological quality in the cultivars BRS Pardela and BRS Gaivotá are shown in the table of analysis of variance (Table 2).

Table 2. Summary of analysis of variance (mean squares) for the traits of physiological quality in seeds of wheat cultivars as a result of chemical treatment of seeds.

Mean Squares						
----- BRS Pardela -----						
S.V.	DF	G	AS	AA	TSL	SL
Treatment	6	34.55**	21.96**	166.04**	57.31**	9.62**
Error	14	4.95	1.95	19.85	12.65	0.39
Mean		93.33	4.57	48.28	24.91	10.09
CV (%)		2.38	30.57	9.23	14.28	6.25
S.V.	DF	RL	SDW	RDW	DF	EF
Block <sup>1</sup>					3	607.23 <sup>ns</sup>
Treatment	6	21.46 <sup>ns</sup>	0.54 <sup>ns</sup>	0.54 <sup>ns</sup>	6	1071.23 <sup>ns</sup>
Error	14	9.29	0.37	0.27	18	418.79
Mean		14.82	6.81	5.15		269.14
CV (%)		20.57	9.02	10.11		7.60
----- BRS Gaivotá -----						
S.V.	DF	G	AS	AA	TSL	SL
Treatment	6	74.66**	31.63**	508.98**	43.27**	5.78**
Error	14	2.57	2.52	59.38	8.18	0.39
Mean		93.00	4.42	68.80	27.03	9.44
CV (%)		1.72	35.87	11.20	10.58	6.66
S.V.	DF	RL	SDW	RDW	DF	EF
Block <sup>1</sup>					3	2266.47*
Treatment	6	18.12 <sup>ns</sup>	0.12 <sup>ns</sup>	0.30 <sup>ns</sup>	6	2016.57*
Error	14	6.39	0.23	0.14	18	520.69
Mean		17.59	5.90	6.05		257.85
CV (%)		14.37	8.23	6.31		8.85

ns, not significant, \*\* and \*, significant at 1% and 5% probability, respectively, by the F test.

<sup>1</sup> Randomized blocks.

G: germination test; AS: abnormal seedlings; AA: accelerated aging; TSL: total seedling length; SL: shoot length; RL: root length; SDW: shoot dry weight; RDW: root dry weight; EF: seedling emergence in the field.

In relation to the germination test in the cultivar BRS Pardela, the seeds treated with triadimenol + imidacloprid + thiodicarb (T5) showed inferior results; however, they were not significantly different from the other treatments. As for BRS Gaivotá, seed treatment with carboxin + thiram + imidacloprid + thiodicarb (T2) brought about significant reduction in seed germination. Possibly, this reduction in germination was due to association of the fungicides carboxin + thiram and triadimenol with the insecticides imidacloprid + thiodicarb, because these insecticides separately (T7) did not show a negative effect (Table 3).

Still, all the values obtained for germination were above the standard established for commercialization of wheat seeds, which require minimum germination of 80% (MAPA, 2013).

The results of the germination test corroborate the evaluations of abnormal seedlings (Table 3). This indicates that some treatments may have led to phytotoxicity, which caused abnormality in seedlings. For soybean, França-Neto et al. (2000) reported that the phytotoxic effect of the chemical treatments may reduce seed quality. Fungicides of the triazole group, when used in seed treatment, may cause phytotoxicity to wheat seedlings, and may lead to reduction of the mesocotyl and cracks in the leaf tip (Picinini and Fernandes, 2003). Similar results were described by Goulart (1988) upon observing that the fungicide triadimenol applied on wheat seeds led to an increase in abnormal seedlings, as well as the appearance of seedlings with twisted, thick, and broadened leaves.

Table 3. Mean values of the properties of physiological quality and health of seeds of wheat cultivars as a function of the response to chemical seed treatment.

----- BRS Pardela -----						
Treatment <sup>1</sup>	G (%)	AS (%)	AA (%)	TSL (cm)	SL (cm)	
T1	91 ab	7 c	52 ab	27.18 a	10.80 a	
T2	92 ab	7 c	50 ab	26.16 a	11.10 a	
T3	97 a	1 a	54 a	25.67 a	11.09 a	
T4	96 a	2 ab	52 ab	26.07 a	10.52 a	
T5	89 b	8 c	35 c	15.10 b	6.08 b	
T6	91 ab	6 bc	41 bc	27.10 a	10.64 a	
T7	97 a	2 ab	54 a	27.12 a	10.42 a	
----- BRS Gaivotá -----						
Treatment <sup>1</sup>	G (%)	AS (%)	AA (%)	TSL (cm)	SL (cm)	EF (plants/m <sup>2</sup> )
T1	93 bc	5 ab	53 b	29.52 a	10.25 a	217 b
T2	83 d	10 c	65 ab	27.91 a	10.05 a	279 a
T3	96 abc	2 a	69 ab	28.80 a	10.15 a	285 a
T4	94 abc	4 ab	84 a	28.66 a	9.84 a	264 ab
T5	96 abc	2 a	85 a	18.71 b	6.33 b	247 ab
T6	92 c	7 bc	53 b	29.10 a	9.78 a	254 ab
T7	97 a	1 a	73 ab	26.56 a	9.72 a	259 ab

The mean values within each column followed by the same letter do not differ among themselves by the Tukey test ( $p \leq 0.05$ ).

<sup>1</sup>Treatments: 1: control (without seed treatment); 2: carboxin + thiram + imidacloprid + thiodicarb; 3: carbendazim + thiram + imidacloprid + thiodicarb; 4: fipronil + thiophanate methyl + pyraclostrobin; 5: triadimenol + imidacloprid + thiodicarb; 6: fipronil; 7: imidacloprid + thiodicarb.

G: germination test; AS: abnormal seedlings; AA: accelerated aging; TSL: total seedling length; SL: shoot length; EF: emergence in the field.

In the accelerated aging test in the cultivar BRS Pardela, treatment 5 (triadimenol + imidacloprid + thiodicarb) led to reduction in vigor in comparison to the control. However, for the seeds of BRS Gaivotá, this treatment did not result in the same behavior (Table 3). This treatment together with T4 (fipronil + imidacloprid + thiodicarb) led to smaller reductions in viability and greater seed vigor (Table 3).

For the data on total seedling length and shoot length, in both cultivars, the treatment with triadimenol + imidacloprid + thiodicarb (T5) differed from the others and from the

control, showing lower total length and shoot length of the seedlings (Table 3). Similar results were observed by Silva et al. (1993), where the fungicide triadimenol reduced the length of the coleoptile and of the mesocotyl in wheat and barley, and by Rampim et al. (2012) who, studying three wheat cultivars, observed that triadimenol led to lower values of shoot length, regardless of the cultivar tested. According to these authors, such results indicate that this product may cause a phytotoxic effect to wheat seedlings.

In regard to seedling emergence in the field, there was a

significant effect only in the cultivar BRS Gaivotá, in which T2 (carboxin + thiram + imidacloprid + thiodicarb) and T3 (carbendazim + thiram + imidacloprid + thiodicarb) led to greater plant populations per m<sup>2</sup> in relation to the control (Table 3).

Furthermore, for this variable, the control (T1) led to lower values of emerged seedlings in the field, though not differing statistically from treatments 4, 5, 6, and 7 (Table 3). This result may be explained due to the absence of chemical treatment on the seeds for, although there was high initial seed health, there was a reduction in emergence because, when they are exposed to uncontrolled environmental conditions, such as occurs in the field (Figure 1), they are subject to the action of soil pathogens. This therefore indicates the importance of seed treatment to obtain an adequate plant stand.

In light of the above, it may be seen, in relation to germination and accelerated aging, that the cultivars respond in a differentiated manner to the different chemical products. Furthermore, some of the formulations used with fungicides and insecticides may lead to phytotoxicity in wheat seedlings.

## Conclusions

The wheat cultivars tested respond in a differentiated manner to the chemical seed treatments in regard to the effects on germination and vigor.

The treatment with triadimenol + imidacloprid + thiodicarb is harmful to seedling development.

In the cultivar BRS Gaivotá, the seed treatments with carboxin + thiram + imidacloprid + thiodicarb, and carbendazim + thiram + imidacloprid + thiodicarb improve seedling establishment in the field compared to the control.

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