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Full Length Article

Intense Continuous Shading is Detrimental to the Development and Production of White Oat Grains

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Abstract

Knowledge of the tolerance of annual cultures to less intense solar radiation is essential to achieving sustainable and more productive systems. The objective of this study was to assess grain performance and yield of white oat plants under intensities and orientations of artificial shading ranges during their growth cycle. The experiments were carried out during the 2020 and 2021 growing seasons, using white oat (*Avena sativa* L.) cultivar URS Altiva. The study used a completely randomized design, in a 3x2+1 factorial arrangement, with three shading intensities: low (25%), medium (50%), high (75%), two orientations: north/south, east/west, and additional treatment (control – under full sun). There were losses mainly under high shading intensity, e.g., a reduction in the Soil Plant Analysis Development (SPAD) and Normalized Difference Vegetation Index (NDVI) at 70 days after the onset of solar radiation restriction, leading to delayed leaf senescence by 42 days (2020) and 21 days (2021); reduced plant height by up to 19.1% (2020) and 21.7% (2021); reduced dry weight at pre-harvest (2021) up to 36.1%, in comparison to the control (274.3 g) and reduction of yield components, number and grain weight. The orientation of the north/south ranges differed from that of the east/west ones by their detrimental effect on grain growth and yield components. In conclusion, low shading intensity throughout the cycle on white oat plants did not cause significant damage to plant development and yield, while high intensity affected them negatively, particularly in the north/south orientation.

Keywords: Adaptability; While oat; Grain yield; Solar radiation; Orientation

Introduction

White oat (*Avena sativa* L.) is gaining importance, mainly in the southern region of Brazil due to its nutritional and economic values. Increased cultivation of white oat is reflected by greater grain production and yield (CONAB 2023), release of new cultivars through genetic breeding programs, a wider range of cultivation systems and use of crop as pasture for animals (Danielowski *et al.* 2021). In addition, white oats are characterized as an optional winter species in the region, especially because few crops are grown in the winter after harvest of summer crops (Balbinot Junior *et al.* 2009).

Integrated production systems are alternatives for optimizing production areas (Bi *et al*. 2019). For example, integrated crop-forestry systems, in which trees are planted between bands of annual crops or perennial crops, can provide short- and long-term financial returns (Burner *et al.* 2018). Therefore, some local environmental factors may be affected in these systems, *e.g*., the incidence of direct solar radiation, which may vary, especially for plants grown in the lower stratum or among tree rows. This variation in solar radiation is due, among other factors, to the sun's position, time of day, direction of the rows, distance between the trees, height and shape of the canopy and wind-induced crown movement (Way and Pearcy 2012; Cordeiro *et al.* 2015).

Solar radiation is crucial for photosynthetic performance, which is vital for plants (Stirbet *et al*. 2020), because it is responsible for producing the carbohydrates required for plant growth and development (Taiz *et al.* 2017). Crop yield, in turn, is directly related to the production of photosynthates by leaves and/or green organs of plants and their subsequent efficiency to synthesize reserves for grain development (Goffman *et al.* 2005). Thus, plants have internal mechanisms for increasing light use efficiency when there is low intensity or mechanisms for reducing and repairing damage induced by more intense light (Nevo *et al.* 2012). One of these mechanisms is a change in leaf chlorophyll content in response to low light intensity. According to Xie *et al.* (2022), plants that are tolerant to low light conditions or have shade prevention mechanisms that present lower a/b chlorophyll ratio and a higher ratio between

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photosystems II and I; in this sense, they differ from plants under optimal light conditions.

Assessing the tolerance of white oat crops cultivated under low solar radiation or shading can provide new possibilities for cultivation in integrated grain production systems. However, black oat plants grown together with tree species under limited solar radiation in integrated systems presents reductions of leaf area index, growth rate, net assimilation and leaf weight (Sgarbossa *et al*. 2020), which causes a reduction in plant dry matter and yield, when they are intercropped with native trees (Nicodemo *et al.* 2016). In addition, a study evaluating the performance of white oat grown close to eucalyptus rows in an agroforestry system, reported an increase in plant yield that was proportional to the largest distance between the white oat plants and the eucalyptus plants (Deiss *et al*. 2016).

With this background, there is a need for studies on the adaptation and tolerance of white oat to shading, considering the constant release and use of modern and more productive cultivars. Thus, the objective of this study was to assess grain yield performance of white oat plants of the cultivar URS Altiva under simulated shading conditions (intensities and directions) throughout the continuous cycle.

Materials and Methods

Setup and conduction of the experiments

The study was conducted at the Center of Agricultural Sciences (CAV) at Santa Catarina State University (UDESC), in Lages, Santa Catarina state, under geographic coordinates 27°52’ South latitude, 50°18' West Longitude, and average altitude of 930 m. The municipality has an average annual temperature of 15.7°C and precipitation of 1500 mm, according to information from the Climate Atlas of Southern Brazil (Wrege *et al.* 2012). The soil of the site is characterized as an aluminic humic Cambisol with clayey texture (Embrapa 2013). Weather data on temperature, precipitation, and solar radiation incident throughout conduction of the experiments (Fig. 1), was compiled according to data published by the National Institute of Meteorology (INMET 2019).

The experiments were carried out on the white oat (*Avena sativa* L.) cultivar URS Altiva in the 2020 and 2021 growing seasons. This cultivar was released in 2015 and is characterized by early cycle, great height and moderate lodging resistance (Danielowski *et al.* 2021). The cultivar URS Altiva stands out for its high grain yield potential and hectoliter weight (Carvalho *et al*. 2023; Lucas *et al*. 2023).

Sowing was carried out in August 2020 and July 2021. Fertilization was carried out according to recommendations of the Soil Chemistry and Fertility Commission (CQFS 2016), based on soil analysis, aiming at 6 t ha^{-1} of grain yield potential. Soil chemical characteristics determined by analysis were pH 6.5; SMP index 6.1; phosphorus 18.7 mg dm^{-3}; potassium 173.2 mg dm^{-3}; H+Al 4.0 cmol dm^{-3}; CEC pH 7.0 of 18.4 cmol dm^{-3}; organic matter 3.1% and base saturation (V) 48.0%. Fertilization at sowing consisted of 400 kg ha^{-1} of the NPK 5-20-10 formulation; twice N topdressing fertilization were used (30 kg ha^{-1}), split between the tillering and stem elongation stages.

Weed control during crop development was performed by hand. Phytosanitary treatment was applied to control insects and diseases according to technical indications for the oat (MAPA 2009). Regarding water availability, complementation was performed through manual irrigation according to the need of the plants.

The shading structures were made of wood, with 1.5 × 1.5 m width and length, similar to those described by Varella *et al.* (2011). These structures differed in terms of distance between boards (shading intensities) and installation orientation (direction of boards – east/west or north/south). The structures were installed at a height of 0.6 and 1.2 m from the soil level, which was increased as the plants grew taller.

Harvest was performed by cutting the plants present in the usable plot when the plants of the control treatment (no shading) were between the hard dough (87) and soft dough (85) stages in the 2020 and 2021 growing seasons, respectively (Zadoks *et al*. 1974). These growth stages were chosen aiming to escape herbivory by birds.

Experimental design

The experiment used a completely randomized design in a 2 × 3 + 1 factorial arrangement, with two orientations of the shading bands, north/south (N/S) and east/west (E/W) directions; three levels of limited solar radiation, namely low (25%), medium (50%) and high (75%) and additional treatment (control) with plants grown under full sun. Four replicates were used, in a total of 28 experimental plots. The research design was based on research feasibility, related to number of factors, available space, and regular assessments. The local conditions were chosen considering the greatest possible homogeneity of soil and incident solar radiation. The plants were shaded at stage 11 *i.e.*, when its presented with the first unfolded blade-shaped leaf, according to the phenological scale of Zadoks (Zadoks *et al.* 1974) and they remained under such conditions until the end of the cycle. Each usable experimental plot was composed of a 1.0 m row, with inter-row spacing of 0.20 m, within a larger area 1.2 m wide and 2.0 m long, containing 10 sowing rows.

Assessments

The plants were assessed at different times. Assessments were carried out weekly (SPAD, NDVI and height), monthly (leaf area and dry weight) and after the plants had been cut (yield components and grain retention on sieves). After the shading structures were installed over the plants, the following assessments were made on a weekly basis: plant height (distance from the base of the plant at ground level to the end of the last leaf or panicle); indirect chlorophyll

assessments, based on the SPAD index, using a SPAD handheld meter (SPAD-502 Plus, Konica Minolta), while NDVI was determined with an NDVI handheld meter (NDVI 310, PlantPen). The assessments were made in three plants per experimental unit, previously demarcated throughout the period, between seven and 70 days after installation of the structures.

Plant dry weight and leaf area were determined monthly, 30 and 60 days after shading had been applied. Plant leaf area was determined by measuring leaf blades with photosynthetically active tissue, obtained from three plants per experimental unit, using a bench-top leaf area meter (LI-3100, Lincoln, Nebraska, USA). After that, the plants (shoot) were dried in a forced air circulation oven at 65 ± 2°C to constant weight for dry weight quantification using a 0.01 g precision balance (model BL3200H, Shimadzu, Japan).

For assessment of yield components, the number of panicles and spikelets was determined by counting the plants collected in each experimental plot. Subsequently, the plants were dried at 65 ± 2°C for 72 h in a forced air circulation oven. The harvest index (HI) was determined by weighing plant dry matter (corresponding to biological yield), threshing, and separating and weighing the grains. The following formula was used (1):

$$\text{HI}\ (\%) = \left(\frac{\text{GY}}{\text{DWt}}\right) \times 100 \qquad (1)$$

Where HI is harvest index, GY is grain yield and DWt is total dry weight (Takeda *et al*. 1980).

Grain weight or yield was determined after manual threshing and thousand grain weight (TGW) was determined by counting and checking the weight of one thousand grains in each plot. Sieve analysis was made with oblong sieves with diameters of 0, 1.75, 2.0, 2.2, 2.5, 3.0 mm × 22 mm, followed by weighing the grains retained on each sieve, with results expressed as a percentage.

Statistical analysis

The data were submitted to the normality test of the residuals and homogeneity of variances, and then submitted to analysis of variance by the F-test at 5% significance. When significant, the means of the variables shading levels and shading orientation were compared by Tukey's test ($P < 0.05$). Comparison was made between treatment means using Dunnett's test ($P < 0.05$). Results of post-harvest analyses were presented proportionally to the control, which was a standard of 100%. The statistical software R (R Core Team 2018) was used.

Results

Chlorophyll indexes

There was variation in the results for the SPAD index (Fig. 2) and NDVI (Fig. 3) of white oat plants assessed over time, regarding shading levels and orientations (Table S1). This finding is in line with the weekly assessment performed after the condition was imposed, for both years (Table S1; Table S4). The SPAD index in plants under full sun (control) was higher in the first weeks of assessment performed after the beginning of shading, while in the final weeks of crop development, there was an inverse effect, with higher SPAD in plants under high shading intensity. In 2021, the initial difference was limited to high shading intensity, particularly in the north/south orientation (N/S) and at the end of assessments in the east/west (E/W) one (Fig. 2). It is noteworthy that at 70 days after the beginning of shading, the plants were already in stages close to maturity, but in 2021 there was delayed plant maturity.

In general, white oat plants grown under low shading intensity presented higher means for SPAD index, in comparison to high intensity, with an increment ranging from 6.6 to 18.0%, depending on the evaluation period. There was a difference between the orientations at 14, 56, 63 and 70 days; the E/W direction had the highest means for SPAD, in comparison to the N/S orientation, whose means ranged from 41.2 to 52.2 and from 38.7 to 49.7, respectively (Table S2).

For NDVI, there was a difference between plants grown under different shading intensities and control (full sun), especially in 2020. There was a higher contrast between treatments and control at 70 days (2020): most of the shaded plants presented a similar behavior was identified by the SPAD index (Fig. 2) but higher NDVI than when under full sun (Fig. 3).

The chlorophyll index, indirectly assessed through NDVI, had an effect in 2020 at 28, 42 and 63 days, with significant interaction between the factors shading levels and orientations. At 28 days, under east/west orientation and high level of shading, NDVI differed from the other days (means 6.8) with a lower value of 6.5. This also occurred under low intensity in the N/S shading orientation at 42 days, with a value of 6.7 and 6.4% less than the means of the other days. On the other hand, at 63 days the NDVI of white oat plants was reduced between orientations at the high level of shading, with a reduction (5.6%) for N/S, and between the levels: it was lower under low shading intensity (6.9), in contrast with the high level (7.2) (Table S3).

Growth

Height of the plants grown under different shading levels and orientations differed from that of the control treatment at the beginning and end of the cycle. Initially, the shaded plants had greater height, but at final stages of the cycle, height was lower as compared to control (plants under full sun). This trend was confirmed in both the growing seasons (Fig. 4). The height of white oat plants, evaluated throughout the cycle, had a significant interaction between shading intensities and orientations in 2020 and isolated factors in 2021. In the first growing season, there was significance for plant height among treatments at 42 days after the start of shading. Plant height was lower under high shading intensity in the E/W orientation,

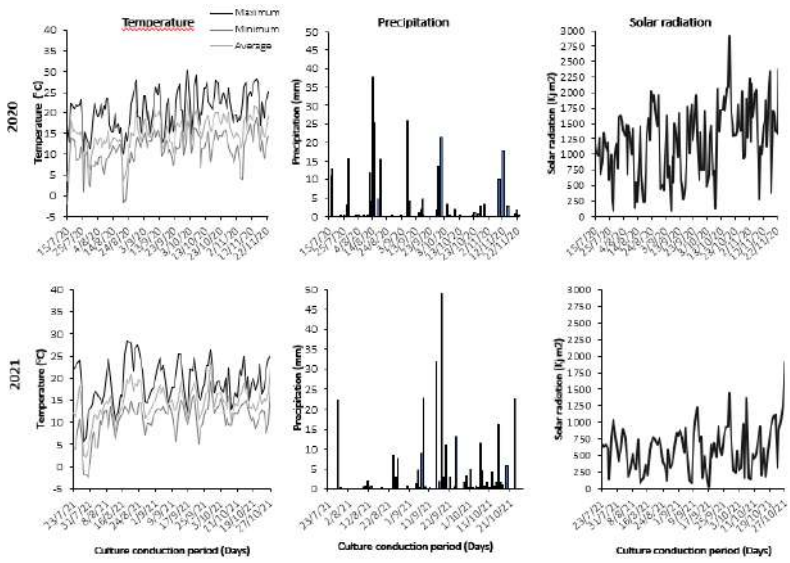

Fig. 1: Temperature maximum (T° MAX), minimum (T° MIN) and average (T° MEAN), rainfall and global solar radiation global during experiments with artificial continuous shadding on white oat. Lages-SC, 2020/20 e 2021/21 growing seasons
Source: Adapted from INMET (2019)

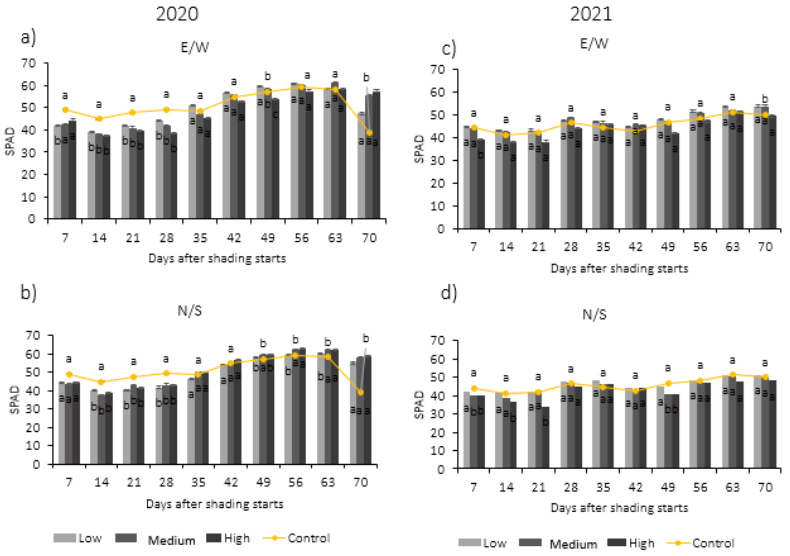

Fig. 2: Evolution of the SPAD index of white oats grown under different shading levels and orientations during the cycle, in the 2020 and 2021 growing seasons
*Bars represent standard error of mean for each treatment; *Same letters do not differ between treatments and control by Dunnett's test ($p \leq 0.05$)

with reductions ranging from 11.3 to 19.1%. In 2021, there was a detrimental effect of shading in most assessments, when the plants were under high shading intensity, with a reduction of up to 21.7% at the time. Regarding the orientations, the lowest plant height averages were found for N/S (<6.7%), compared to east/west (Table S4).

There was an effect for leaf area (LA) and dry weight (DW) of white oat plants, at 30 and 60 days after the onset of light restriction, except for LA at 60 days; there was a difference between the E/W and N/S orientations. The reduction of LA and DW under high solar radiation ranged from 3.9 to 45.6 and 46.6 to 92.9%, respectively, under low light intensity, between the years and the evaluation period (Table 1). For LA of plants at 30 days (2020), there was a negative effect of shading at medium and high intensity, with averages between 93.3 and 108.4 $cm^2.plant^{-1}$, compared to plants under full sun (138.0 $cm^2.plant^{-1}$). Similar results were found in 2021, when LA of shaded plants ranged between 87.6 and 130.8 cm^2 $plant^{-1}$, compared to the control treatment in the same year (165.3 cm^2 $plant^{-1}$). However, there was higher LA at 60 days (2021), with 296.7 and 289.9 cm^2 $plant^{-1}$ for E/W and N/S orientations, respectively (Table 1).

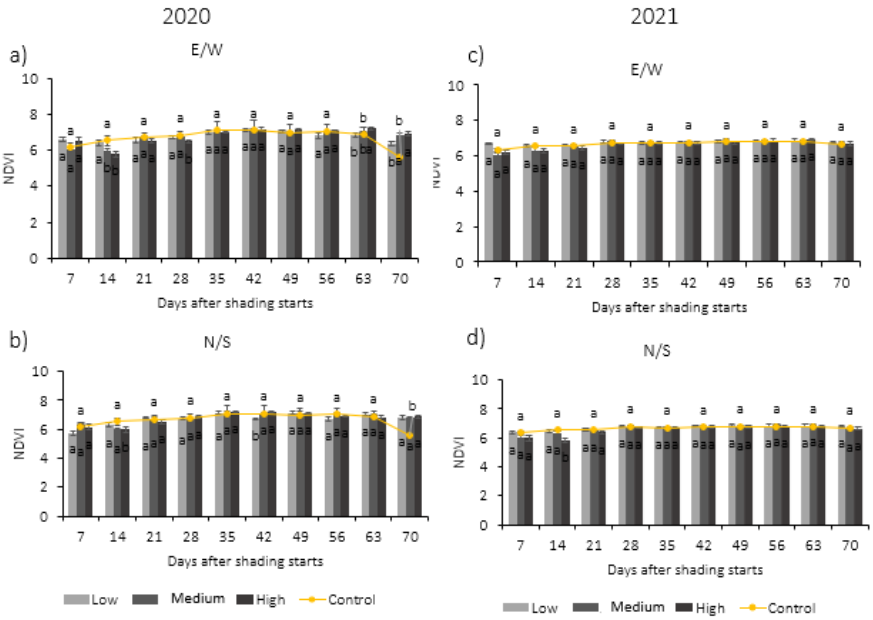

Fig. 3: Evolution of the NDVI of white oats grown under different shading levels and orientations during the cycle, in the 2020 and 2021 harvests
*Bars represent standard error of mean for each treatment; *Same letters do not differ between treatments and control by Dunnett's test (p≤0.05)

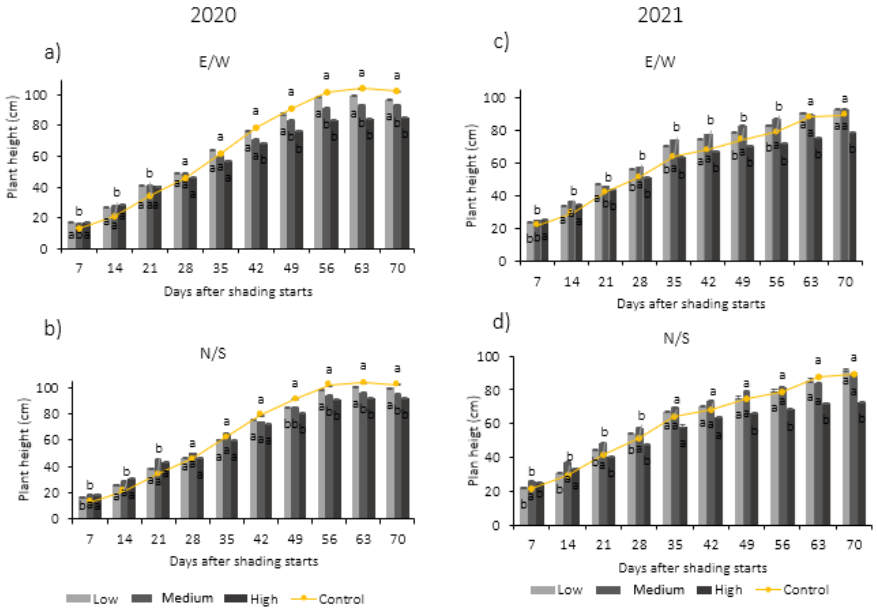

Fig. 4: Height of white oat plants grown under different shading levels and orientations during the cycle, in the 2020 and 2021 harvests
*Bars represent standard error of mean for each treatment; *Same letters do not differ between treatments and control by Dunnett's test (p≤0.05)

Plant DW differed from the control as it showed lower results, mainly at medium and high levels of shading, indicating negative effects of shading at 30 and 60 days and at pre-harvest. In 2020, DW of white oat plants under medium and high shading intensities presented the lowest averages at 30 and 60 days after the start of shading. Compared to the control group (0.8 g plant^{-1}), there were reductions by up to 46.3 and 43.7% for the shaded plants at medium and high intensity levels, respectively (Table 1). In 2021, at both assessments and at the end of the cycle, the lowest averages for plant DW were found under high shading level (75%), with reductions by up to 55.8% (30 days), 28.7% (60 days) and 36.1% at end of cycle (Table 1). For final DW (2021), there was an effect for the shading orientations: DW was increased by 9.1% under the E/W orientation, in comparison to shading under the N/S orientation (213.7 g) (Table 1).

Table 1: Leaf area and dry weight of white oat plants grown under different shading levels and orientations during the cycle, in the 2020 and 2021 harvests

Orientation	Shading levels																	
	2020							2021										
	Low		Medium		High		Average		Low		Medium		High		Average			
	Leaf area 30 days (cm^2 $plant^{-1}$)																	
East/West	126.98	ns	113.90		98.72	*	113.20	ns	155.61	ns	130.79	*	103.44	*	129.95	a		
North/South	119.31		108.43	*	93.30	*	107.01		122.59	*	132.61		87.59	*	114.26	b		
Average	123.15	A	111.17	AB	96.01	B			139.10	A	131.70	A	95.52	B				
Control				137.96									165.25					
	Leaf area 60 Days (cm^2 $plant^{-1}$)																	
East/West	176.02	ns	176.22		168.91		173.72	a	228.67	ns	296.69	*	211.25		245.54	ns		
North/South	147.64		169.75		158.76		158.71	b	211.04		289.90	*	211.99		237.64			
Average	161.83	ns	172.98		163.83				219.85	B	293.30	A	211.62	B				
Control				151.51									174.41					
	Dry weight 30 days ($g.plant^{-1}$)																	
East/West	0.66	ns	0.55	*	0.40	*	0.53	ns	0.75	ns	0.53	*	0.43	*	0.57	ns		
North/South	0.63	*	0.50	*	0.47	*	0.53		0.55		0.58		0.31	*	0.48			
Average	0.64	A	0.52	B	0.43	B			0.65	A	0.55	A	0.34	B				
Control				0.80									0.77					
	Dry weight 60 days ($g.plant^{-1}$)																	
East/West	3.17	ns	2.70	*	2.01	*	2.63	ns	2.72	ns	2.77		1.81		2.43	ns		
North/South	3.05		2.43	*	2.18	*	2.55		2.35		2.54		1.67		2.19			
Average	3.11	A	2.57	B	2.10	C			2.54	A	2.66	A	1.74	B				
Control				3.73									2.44					
	Final dry weight ($g.plant^{-1}$)																	
East/West	209.06	ns*	193.19	*	168.21	*	190.15	ns	276.05	ns	238.93		184.69	*	233.19	a		
North/South	182.28	*	177.82	*	180.99	*	180.36		232.91	*	242.34		165.98	*	213.74	b		
Average	195.67	ns	185.50		174.60				254.48	A	240.59	A	175.33	B				
Control	255.16								274.27									

*Same lowercase letters in the column and uppercase in the row do not differ by Tukey's test ($p \leq 0.05$); *means differ from the control by Dunnett's test ($p \leq 0.05$). ns: non-significant statistical difference

Yield components

The number of panicles per linear meter in 2021 under low shading intensity was higher, with 98.6% panicles per linear meter, while the remaining medium and high levels reached only 89.3 and 82.6% of the total percentage of panicles for plants under full sun. The number of panicles of plants shaded mainly under high intensity (82.6%) differed from that of the control (100%), with a reduction by 17.4%, regardless of orientation (Table 2).

For number of grains per linear meter (NG), there was significance for the interaction between the factors in 2021: shaded plants were negatively affected, except under low intensity in the E/W orientation. It was found that under low shading in the N/S orientation, the number of grains was lower, compared to E/W and the control, with 72.7% in comparison to the control with 100% (Table 2). In addition, with shading in the E/W and N/S orientations under high intensity, the NG per linear meter was affected, with percentages of 54.5 and 48.7%, while under low shading, the percentages were 101.6 and 72.7%, respectively, considering the NG under full sun as 100%. The NG per linear meter under low solar radiation level in the E/W orientation (101.6%) did not differ from that of the control (100%).

The weight of white oat grains per linear meter (WG) was negatively impacted on the shaded plants in comparison to the control for both seasons tested, especially under medium and high intensities (Table S4). In 2021, there was a difference between intensities; under low shading level, grain weight had a higher proportion than the other levels, with 78.6%, compared to the control *i.e.*, 100% (Table 2).

In both growing seasons, TGW of white oat shaded mainly under medium and high intensity levels, in both orientations, differed from that of the control, indicating a negative effect of these conditions on thousand grain weight (Table 2). TGW presented an interactive effect between shading levels and orientations for the 2020 growing season. TGW for plants under high shading intensity in both orientations, and the mean in N/S, presented a lower percentage in comparison to the other intensities and the control (100%): 69.6% in E/W (high intensity), and 81.1 (medium intensity) and 78.7% (high intensity) in S/N. In the second growing season, TGW was 84.3 and 86.9%, for E/W and N/S, respectively, in comparison to the control (100.0%).

For HI of white oat plants, there was an effect between shading levels for orientations in 2020, and for shading intensities in 2021. The HI (2020) under N/S shading (105.3%) was higher, compared to E/W (97.7%). Furthermore, in 2021, the HI was 73.4 and 76.1%, for S/N and E/W, respectively. The HI under low shading intensity (2021) differed from high intensity, with 84.1 and 67.1% of the total HI of plants grown under full sun radiations (Table 2).

For percentage of grain retention on different sieves, there was a difference for the control (full sun) when compared to the values found for the shaded plants. The

Table 2: Percentages of number of panicles and grains, grain weight, thousand grain weight (MMG) and harvest index (HI) of white oats grown under different shading levels and orientations during the cycle, in the 2020 and 2021 harvests

Orientation	Shading levels														
	2020						2021								
	Low		Medium		High		Average		Low		Medium		High		Average	
	Number of Panicles per linear meter (% in relation to the control)															
East/West	104.86	ns	101.72		96,46		101,01	ns	102,65	ns	91,31		83,54	*	92,50	ns
North/South	110.38		94.11		87,91		104,87		94,44		87,67	*	81,67	*	87,83	
Average	93.25	B	104.05	AB	111,53	A			98,55	A	89,34	B	82,61	B		
Control				100.00									100.00			
	Number of grains per linear meter (% in relation to the control)															
East/West	86.32	ns	95.61		8,72		90,22	ns	101,64	aA	75,52	aB*	54,48	aC*	77,21	a
North/South	83.02		93.04		97,48		91,18		72,71	bA*	75,90	aA*	48,72	aB*	65,78	b
Average	84.67	ns	94.33		93,10				87,17	A	75,71	B	51,60	C		
Control				100.00									100.00			
	Mass of grains per linear meter (% in relation to the control)															
East/West	75.93	ns*	83.15		61,58	*	73,55	ns	90,63	ns	63,14	*	45,02	*	66,26	ns
North/South	75.52	*	75.90	*	75,50	*	75,64		66,62	*	65,08	*	41,08	*	57,59	
Average	75.72	ns	79.53		68,54				78,62	A	64,11	B	43,05	C		
Control				100.00									100.00			
	Thousand grain weight (% in relation to the control)															
East/West	87.87	aA*	87.06	aA*	69,60	aB*	81,51	ns	88,87	ns	83,39	*	80,74	*	84,33	ns
North/South	92.52	aA	81.14	aB*	78,66	bB*	84,10		91,34		85,58	*	83,79	*	86,90	
Average	90.20	A	84.10	A	74,13	B			90,11	ns	84,48		82,27			
Control				100.00									100.00			
	Harvest index (% in relation to the witness)															
East/West	91.55	ns	109.04		92,36		97,65	b	89,73	ns	72,09	*	66,34	*	76,06	ns
North/South	103.97		106.55		105,49		105,34	a	78,54		73,59	*	67,90	*	73,35	
Average	97.76	ns	107.80		98,93				84,14	A	72,84	AB	67,12	B		
Control				100.00									100.00			

*Same lowercase letters in the column and uppercase in the row do not differ by Tukey's test ($p \leq 0.05$). ns: non-significant statistical difference

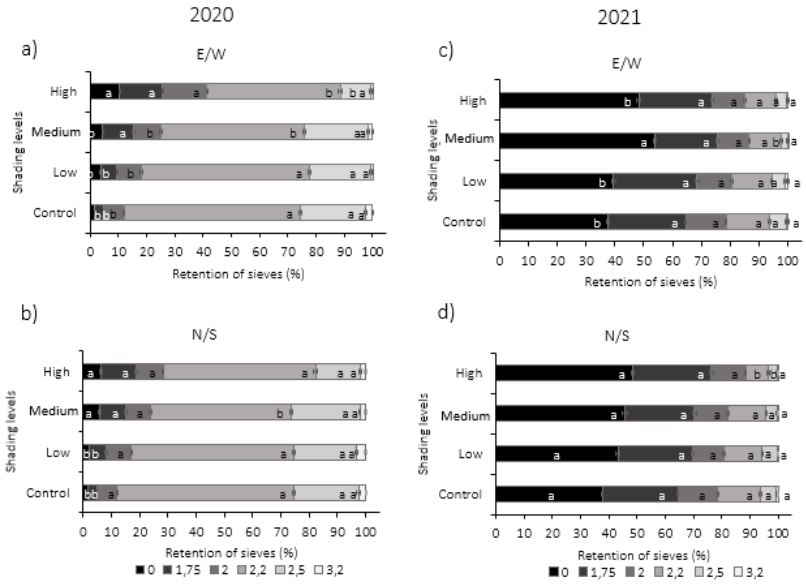

Fig. 5: Sieve retention of grains from white oat plants grown under different shading levels and orientations during the cycle, in the 2020 and 2021 harvests

*Bars represent standard error of mean for each treatment; *Same letters do not differ between treatments and control by Dunnett's test ($p \leq 0.05$)

percentage of grain retention on 1.75 mm sieves was higher under high and medium shading intensities, with a mean of 13.8 and 10.2% between orientations, compared to the control (1.6%). A similar effect was found for non-retained grains (empty grains), whose percentage under high level was 8.4%, which accounts for an increase by 6.8% in comparison to the control (Fig. 5). This indicated that the shaded plants produced small grains.

Grain size assessment through sieve retention (2020) showed an effect on shading intensities of the sieves of 2.5; 2.2; 1.75 and 0, and interaction for sieve diameter of 2.0 mm (Fig. 5). The grain retention on 2.5 and 2.2 sieves was higher

under low intensity (21.4%) and differed from the percentage for high intensity shading (13.3%). In addition, the percentage of grain retention on 1.75 and 0 sieves (non-retained) of shaded plants at medium and high levels was higher than that of the control, with increases of up to 38.0%, indicating that grains produced by shaded plants had lower accumulation of reserves (Fig. 5). This confirmed the results obtained in the first harvest. In 2021, grain retention percentage differed between the levels only on sieves of 2.5 and 2.2 mm. Highest retention means were found for the sieve of 2.5 mm under low intensity (5.2%) and the sieve of 2.2 mm under low (13.5%) and medium (12.3%) intensities (Table S5), confirming that plants under shading produce grains with lower accumulation of reserves (Fig. 5).

Discussion

The environments in integrated or forest grazing systems present dynamic local environmental conditions, changing the structure of plants, compared to full-sun cultivation. This indicates that the morphological adaptation capacity of the species in shaded environments should be included in the crop management analyses chosen in such a system (Garcez Neto *et al.* 2010). It was found that high shading intensity affected the development and yield of grains, but under low intensity, there were no significant deleterious effects, indicating the possible adaptation of the plants to such condition. This indicated that high shading intensities (75%), together with N/S orientation, in general, caused damage to chlorophyll indexes, plant height and plant dry weight, grain yield and yield components. The white oat plants grown under full sun (control) showed better performance in comparison to the shaded plants, especially when compared to intense shading. For the orientations, the results indicated that shading provided by E/W simulation bands did not impair crop development, compared to N/S, which was disadvantageous to plant development.

Weather conditions that vary throughout the years, such as precipitation and temperature, also interfere with plant responses (Matysiak 2006). The sowing time of white oat crops characterizes the temperature range, length of day and cumulative thermal sum, which influence the speed of development and growth, and the earliest cultivars are the most sensitive to such environmental variations (Sponchiado 2017). It was found that in 2021, there was lower availability of sunlight and lower average temperature. This fact may have caused slower plant development and higher yield losses, compared to 2020 and intensified the condition of reduced sunlight on plants.

Chlorophyll content is related to the photosynthetic efficiency of plants, since they are pigments that act in the photochemical phase of photosynthesis and are specialized in light capture (Taiz *et al*. 2017). The assessments of the SPAD and NDVI showed negative effects of high shading intensity on the chlorophyll content of the plants immediately after the onset of light limitation, indicating possible stress by changing the condition, but with subsequent recovery (Fig. 2–3). Corroborating the present results, Yang *et al.* (2020) found that sensitive wheat cultivars under higher shading and duration intensities presented a reduction in chlorophyll content and damage to the leaf photosynthetic apparatus. Wang *et al.* (2020) found that under a 30 and 50% reduction in sunlight after heading, corn plants showed a yield reduction of up to 70%, in biomass accumulation, leaf chlorophyll, soluble protein content and the activity of enzymes related to nitrogen metabolism, such as nitrate reductase, glutamine synthetase and glutamate synthase. However, these studies have reported an increase in the translocation of assimilates stored in the vegetative organs in the previous period.

At the end of the crop cycle, the highest chlorophyll index in shaded plants may account for delayed leaf senescence and increased plant cycle. It is known that to advance the senescence process, there is a need to reduce the leaf chlorophyll content, because there is a higher rate of degradation of these pigments (Jespersen *et al*. 2016). These findings corroborate those of Yasin *et al.* (2019), who reported that some weed species needed a longer period to start flowering when submitted to reduced daily solar radiation. Moreover, Inurreta-Aguirre *et al*. (2018) found that wheat plants produced in agroforestry systems with a mean reduction of 50% of solar radiation reached maturity later in comparison to full-sun cultivation, ranging from days to week, depending on crop growing seasons. Zhang *et al.* (2019) found that the shading of early flowering mutant plants and premature senescence of rice cultivar caused delayed leaf senescence and increased yield because there was less accumulation of reactive oxygen species in these plants. These results refer to the differences in thermal time, observed in the agroforestry system, air warming during the night and air cooling during the day.

A low solar radiation intensity on plants interferes with their photosynthetic rate (Mu *et al.* 2010; Li *et al.* 2014; Wang *et al.* 2015; Poorter *et al.* 2019; Jumrani and Bhatia 2020) and causes reductions in growth (Yasin *et al.* 2019), dry weight, and grain yield (Jumrani and Bhatia 2020). The results for height and dry weight of white oats were impaired under shaded conditions (especially under high level of intensity). The initial height of the shaded plants indicated the effect of stem etiolation. Later, as the cycle progressed, the shaded plants showed lower height and dry matter accumulation, in comparison to plants grown under full sun, a fact that may be related to limited growth resulting from the use of reserves for grain formation.

A greater height of plants under high shading intensity at the beginning of development may have been due to responses to the shadow avoidance syndrome, in which there is relocation of the photosynthates to promote greater internode elongation, together with a reduction in diameter (Stamm and Kumar 2010; Zhang *et al.* 2020). These results corroborate those reported by Li *et al.* (2010) when testing shading levels of 0, 8, 15 and 23% on

a wheat crop from booting growth to maturity, in which the stems were elongated and had lower dry weight per internode unit of the plants.

The effects for dry weight accumulation in plants may be directly related to a reduction in the source-sink ratio (Asseng *et al.* 2017). The beneficial effect of the low shading level may be related to the favorable effect of this condition to plants for the photosynthetic rate and possibly for a decrease in saturation (Kanniah *et al*. 2012). The oat is C3 photosynthetic metabolism plant (Castro *et al*. 2012), showing saturation of CO_2 assimilation under conditions with 30 to 40% of the total radiation, corresponding to 600–900 µmol m^{-2} s^{-1} (Ilic and Fallik 2017). Similar results were found in weeds under low light intensity conditions (6 to 20% solar radiation) or under low photosynthetic photon flow density (PPFD), which gives lower biomass production under such conditions (Yasin *et al.* 2019).

Reduction in the number of panicles in plants may be closely linked to the interference of absence of light over the leaf production rate in the main stem and leaves in tillers. This fact was reported by Tilley *et al.* (2019) for wheat plants, which showed a lower number of leaves under continuous shading. This is due to the production of new tillers when plant reserves are higher than necessary for their growth (Charles-Edwards 1984). Thus, the accumulation of photosynthates may have been impaired by the intense restriction of solar radiation, and limited development of new tillers in plants, as reported in oat (Dietz *et al.* 2023) and *Urochloa decumbens* and *U. brizantha* species (Martuscello *et al.* 2009).

Continuous solar radiation restriction on the plants during development affects spikelet pollination (Kobata *et al*. 2013; Deng *et al.* 2021) and consequently, grain formation. Also, there was an increase in the number of empty spikelets and lower spikelet filling (Wang *et al.* 2015). These facts can justify the results for reduced number of grains per linear meter (NG) of shaded white oat plants (Table 2). Nicodemo *et al.* (2016), when growing black oat in integrated crop-forestry systems, found an increase in plant dry matter, thousand grain weight, number of grains per panicle and panicles per square meter, when spaced further apart from rows of native trees, where the photosynthetically active radiation available to oats was higher, indicating harmful effects of high levels of shading when the plants were closer to the trees.

The weight of white oat grains (quantified per linear meter of cultivation – WG) under continuous shading was sensitive to the condition, as well as TGW in both agricultural years, whose values were decreased at the highest shading levels. Considering the importance of the source in the source-sink ratio during plant development and grain formation, the results can be related to the harmful effect of limiting the amount of source tissues by imposing the solar radiation restriction (Asseng *et al.* 2017). These results may be due to reduced photosynthetic rate and damage to the photosynthetic apparatus, together with decreased chlorophyll content and stomatal conductance (Yang *et al.* 2020). Thus, it may have affected the tillering phase, the differentiation of the floral primordium, anthesis and grain filling. These stages are characterized by determination of the number of panicles per plant, spikelets, and number of grains, and grain weight, respectively (Danielowski *et al.* 2021). Yang *et al.* (2019) when growing wheat in agroforestry systems reported that when close to the trees, the plants reduced their chlorophyll content, leaf area index, photosynthetic rate and stomatal conductance, plant biomass, number of grains and thousand grain weight, when compared to full-sun cultivation.

A reduction in the harvest index (HI) of the shaded plants indicates that this condition affected the destination of photosynthates to the grains, as well as the partition of the vegetative parts to the grains, owing to lower rates for growth and accumulation of photosynthates in the plants (Labra *et al*. 2017). Inurreta-Aguirre *et al.* (2018) found reduction in the yield of wheat plants when assessing the effect of full-sun and agroforestry cultivation systems on genotypes, with means of 203.0 and 62.0 $g.m^{-2}$ respectively, in 2016. This can be seen by the reduction of tillering, spikelets, and number of grains. In addition, they found variation in the HI values in both systems and years. Under the agroforestry system, the mean harvest index was 0.29 and 0.21, while plants under full sun presented values of 0.23 and 0.28, for 2015 and 2016, respectively.

Shading, especially at high intensity, has been shown to be harmful to the development and production of grains by white oat plants. The condition of continuous limited solar radiation, that is, during all plant development, may have directly resulted in the reduction of the source-sink ratio (Asseng *et al.* 2017). According to Sandaña and Pinochet (2011), the reduction in biomass accumulation in plants, arising from lower interception of photosynthetically active radiation, has a negative impact on grain yield. This situation was found to occur in the present work. Therefore, the effects of shading on white oat plants, on grain yield and yield components indicated that white oat has mean sensitivity (dependent on the intensity of light restriction) to the shading condition during the cycle – a fact that has been confirmed in two crop growing years.

Conclusion

The chlorophyll index in the leaves (evaluated by optical sensors) confirms that intense shading results in damage to photosynthesis with negative consequences to the yield of white oat grains of the cultivar URS Altiva. A low level of (continuous) shading during the cycle of white oat plants does not cause significant damage to their development and production (leaf area (LA), dry weight (DW), NG, WG, TGW and HI). Nonetheless, continuous shading of the plants in the N/S orientation was more harmful to the production process of white oats than shading in the E/W orientation.

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Author Contributions

All authors actively contributed to this paper's formulation, discussion of findings, and composition, collectively assuming responsibility for its content.

Conflict of Interest

All authors declare no conflict of interest.

Data Availability

Data presented in this study will be available on a fair request to the corresponding author.

Ethics Approval

Not applicable to this paper.

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Efeito do regulador de crescimento cloreto de clormequate nos atributos morfométricos e produtivos de plantas de soja

Effect of growth regulator chlormequat chloride in morphometric and production attributes of soybean

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RESUMO: O cultivo de soja em regiões de altitude estimula o crescimento de estruturas vegetativas e diminui as reprodutivas. O uso de reguladores de crescimento favorece a efetivação de estruturas reprodutivas. Objetivou-se avaliar variáveis morformétricas e componentes de rendimento de soja sob aplicação do regulador de crescimento cloreto de clormequate. O experimento foi conduzido em casa de vegetação no munícipio de Lages (SC), Brasil, na safra 2018/19. O delineamento experimental foi em blocos ao acaso com cinco repetições, seguindo o arranjo fatorial 2 x 5 x 3: a) duas cultivares de soja, b) cinco doses do cloreto de clormequate (0, 25, 50, 75 e 100 g i.a. ha^{-1}), c) três estádios de aplicação do regulador (V9, R1 e R3). Os resultados obtidos demonstraram que as aplicações no estádio V9 nas doses 75 e 100 g i.a. ha^{-1} tiveram maior efeito na redução de altura de plantas, pela redução do comprimento de entrenós, sem afetar o número de nós e de vagens por planta, e ainda incrementando o diâmetro da haste principal, desse modo aplicações nesses estádios podem controlar o excesso de crescimento vegetativo e evitar os problemas do acamamento.

Palavras-chave: Estratégias de aplicação. Fitorreguladores. Inibidores de giberelinas.

ABSTRACT: The cultivation of soybean in high places stimulates the growth of vegetative structures and decreases reproduction ones. The use of growth regulators favors the effectiveness of reproduction structures. Current research evaluates morphometric variables and yield components of soybean by the growth regulator chlormequat chloride. Experiment was conducted in a greenhouse in Lages SC Brazil during the 2018-2019 harvest. Experimental design comprised randomized blocks with five replications, factorial scheme 2 x 5 x 3: a) two soybean cultivars, b) five doses of chlormequat chloride (0, 25, 50, 75 and 100 g i.a. ha^{-1}), c) three applications of regulator (V9, R1 and R3). Results showed that applications of stage V9 at doses 75 and 100 g i.a. ha^{-1} had a greater effect on the decrease of plant height, decrease of length of internodes, without any effect on the number of nodes and pods per plant. It also increases the diameter of the main stem. Consequently, applications of these stages may control excess of vegetative growth and prevent other issues.

Keywords: Inhibitors of gibberellins. Phytoregulators. Strategies for application.

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INTRODUÇÃO

A cultura da soja (*Glycine max* (L.) Merr) é a principal *commodity* agrícola brasileira, sendo responsável por 34,98% das exportações do agronegócio brasileiro no ano de 2020 (AGROSTAT, 2021) e seu cultivo se estende de Norte a Sul do país. Na região Sul do Brasil, nos Estados do Paraná, Rio Grande do Sul e Santa Catarina é comum encontrar locais de produção de soja acima de 700 m de altitude, tais áreas se caracterizam por produtividades médias elevadas.

Condições de umidade elevada, e nebulosidade durante o período vegetativo nessas regiões e de manejo, como excesso de nutrientes no solo, ou alta densidade de plantas, podem proporcionar crescimento vegetativo considerável, que pode tornar os entrenós frágeis, podendo não resistir à força exercida sobre eles, cedendo ao peso das estruturas acima, resultando o acamamento (MUNDSTOCK; THOMAS, 2005), sobretudo quando associado a condições climáticas severas, como ventos e chuva intensa. Segundo Souza *et al*. (2013) cultivares de soja de maior altura são mais sensíveis ao acamamento, principalmente em condições de excesso de nitrogênio disponível e de maior densidade populacional.

O acamamento pode provocar autossombreamento, reduzindo a fotossíntese líquida e o período de duração de vida da folha, acarretando redução de fotoassimilados disponíveis ao enchimento de grãos, além de proporcionar um microclima de maior umidade, que é favorável à depreciação dos grãos de soja e incidência de doenças. O prejuízo no rendimento de grãos ocasionado pelo acamamento de plantas é função direta do estádio de desenvolvimento em que ocorre o problema e de sua intensidade, especialmente quando o acamamento se dá no início do enchimento de grãos (BALBINOT JÚNIOR, 2012; MARQUES, 2019; SOUZA *et al*., 2013).

As giberelinas induzem o alongamento dos entrenós através da promoção do alongamento celular (TAIZ; ZEIGER, 2017), em consequência aumentando a altura de plantas, sendo o hormônio mais associado ao aumento de altura das plantas. O uso de reguladores de crescimento pode ser uma estratégia viável para redução de altura de plantas, visando reduzir o acamamento sem provocar fitotoxicidade e manter o rendimento de grãos (SOUZA *et al*., 2013).

O cloreto de clormequate é um composto do grupo químico amônia quaternária, que atua na primeira etapa da biossíntese de giberelinas, atuando em diferentes estágios dessa etapa, bloqueando a síntese do ent-kaureno, que é precursor das giberelinas, inibindo a síntese do mesmo (HOPKINS; HÜNER, 2004; RADEMACHER, 2016; TAIZ; ZEIGER, 2017).

A aplicação do cloreto de clormequate na soja pode proporcionar plantas com menor altura e aumento do diâmetro da haste principal, características que podem reduzir a incidência de acamamento, e maior número de vagens, maior número de grãos por planta e grãos mais pesados (SOUZA *et al*., 2013). A resposta à aplicação do cloreto de clormequate pode variar

conforme o genótipo, o estádio fenológico da aplicação e condições de manejo. Moreira *et al*. (2020) observaram que aplicações no estádio R1 foram mais eficazes na fixação de estruturas reprodutivas sob restrição hídrica, que aplicações mais tardias nos estádios R3 e R5.

Em vista disso, torna-se necessário buscar resultados que auxiliem na tomada de decisão quanto à aplicação de reguladores de crescimento na cultura da soja. A hipótese do trabalho é de que a aplicação do cloreto de clormequate tem maior efeito sobre as plantas de soja no estádio fenológico V9 (nono nó da haste principal com folha expandida) que nos estádios R1 e R3 (início de florescimento e de formação das vagens) (FEHR; CAVNESS, 1977), reduzindo altura de plantas, incrementando o diâmetro de haste, características que podem reduzir o índice de acamamento e maximizando a partição de fotoassimilados, favorecendo o rendimento de grãos e seus componentes.

Objetivou-se no presente estudo avaliar o efeito do regulador de crescimento cloreto de clormequate nas características morfométricas e componentes de rendimento em função das doses e estádios de aplicação do cloreto de clormequate em duas cultivares de soja.

2 MATERIAL E MÉTODOS

O experimento foi conduzido no Centro de Ciências Agroveterinárias, na Universidade do Estado de Santa Catarina (CAV/UDESC), em casa de vegetação, no município de Lages, Santa Catarina, 27°48’58”S e 50°19’34”O, altitude de 930 m.

As unidades experimentais foram compostas por 150 vasos com 0,005 m³ de capacidade, que foram preenchidos com 5 kg de solo seco, classificado como Cambissolo Húmico Alumínico (EMBRAPA, 2017). A adubação seguiu o recomendado pelo Manual de Adubação e Calagem para os Estados do Rio Grande do Sul e Santa Catarina (CQFS – RS / SC, 2016), conforme prévia análise química do solo objetivando rendimento de grãos de 5.000 kg ha^{-1} para a cultura da soja.

A semeadura foi realizada dia 29/11/2018 com cinco sementes por vaso, tratadas previamente com inseticida a base de fipronil (Standak® BASF®); composto de fungicidas carbendazim + tiram (Derosal Plus® Bayer®); e inoculante a base de bactérias fixadoras de nitrogênio *Bradyrhizobium japonicum* (cepa Semia 5079) e *Bradyrhizobium elkanii* (cepa Semia 5019) (Masterfix® Stoller®, concentração 5 x 109 UFC ml^{-1}) na dose de 100 ml para 100 kg de sementes.

Quando as plantas apresentavam duas folhas expandidas foi realizado o desbaste mantendo apenas uma planta por vaso. A irrigação foi realizada de forma manual, mantendo a umidade do solo próxima a 80% da máxima umidade gravimétrica de cada vaso, pela diferença entre o peso do solo seco e o peso do solo com umidade saturada, os tratamentos com inseticida e fungicida seguiram as recomendações para a cultura e a temperatura média do ambiente foi

de 25 ± 10 °C. O delineamento experimental foi em blocos casualizados, com cinco repetições, no esquema fatorial 2 x 5 x 3:

a) Duas cultivares de soja, a BMX Zeus IPRO, grupo de maturação 5.5, com ciclo precoce, de hábito indeterminado, e TMG 7062 IPRO, do grupo de maturação 6.2, e ciclo médio, considerada de hábito semi-determinado;
b) Cinco doses do regulador de crescimento cloreto de clormequate (Tuval® da empresa Microquímica com 10% de ingrediente ativo) - 0, 25, 50, 75 e 100 g i.a. ha^{-1} (0; 250; 500; 750; e 1000 ml da dose comercial, respectivamente);
c) Três estádios fenológicos distintos de aplicação, V9 (nono nó com trifólio desenvolvido), R1 (início do florescimento), R3 (início de formação das vagens).

O regulador de crescimento cloreto de clormequate foi aplicado via pulverizador costal de parcelas experimentais do modelo Herbicat H® pressurizado por CO_2 à pressão constante de 30 lb pol^{-2}, com bico único do tipo leque, regulado para volume de calda equivalente a 200 L ha^{-1}. As aplicações do regulador foram realizadas sempre entre 8 e 9 horas da manhã, com temperatura entre 20 e 25 ºC dentro da casa de vegetação.

Foram avaliadas ao final do ciclo (R8) as características morfométricas de altura de plantas, comprimento médio dos entrenós (CE), diâmetro de haste principal (DHP), número de nós (NN) e número de ramos na haste principal (RP). Os componentes do rendimento avaliados foram o número de vagens por planta (NVP); número de grãos por vagem (NGV); peso de cem grãos (PCG). Ao final do ciclo também foi determinada a matéria seca da parte aérea das plantas, e o índice de colheita (IC), determinado pela produção de grãos por planta, dividida pela matéria seca total da parte aérea da planta, transformados e expressados em percentagem.

Os dados foram previamente testados quanto à normalidade pelo teste de Shapiro-Wilk, e homogeneidade pelo teste Bartlett, apresentando distribuição normal. Após isso, os dados foram submetidos à análise de variância (ANOVA) pelo teste F a 5% de significância e, quando detectadas variações significativas, as médias foram contrastadas pelo teste de Tukey a 5% de probabilidade de erro, o fator quantitativo (dose do regulador) foi submetido a análise de regressão polinomial a 5% ($P < 0,05$) no *software* de análise de dados R (R-Core Team, 2020).

3 RESULTADOS E DISCUSSÃO

Segundo a análise de variância (Tabela 1), houve efeito de interação entre estádio de aplicação e dose do cloreto de clormequate (ExD) nas variáveis altura, número de nós da haste principal (NN), diâmetro da haste principal (DHP) e peso de cem grãos (PCG). Foi observada interação tripla (CxExD) para o comprimento médio dos entrenós (CE) e número de vagens por planta (NVP), enquanto apenas para o índice de colheita houve efeito isolado da cultivar, logo a aplicação não demonstrou efeito para esta variável, bem como nas variáveis ramos por planta

(RP) e número de grãos por vagem (NGV), que não foram influenciadas por nenhum dos fatores.

Tabela 1. Resumo da análise de variância das variáveis: Altura, NN, RP, CE, DHP, NVP, NGV, PCG e IC obtidos de plantas de soja em função das cultivares testadas, estádio e dose de aplicação do regulador de crescimento cloreto de clormequate, em ambiente de casa de vegetação. Lages (SC)

FV [1/]	GL	ALTURA	NN	RP	CE	DHP	NVP	NGV	PCG	IC
Bloco	4	8,62	0,92	0,68	0,01	0,10	0,190	0,1	2,8	<0,001
Cultivar	1	14800,66*	798,10 *	0,10^{ns}	0,34*	2,46*	2939,3 *	0,01^{ns}	7,8^{ns}	**0,200***
Estádio	2	179,48*	0,32^{ns}	5,60^{ns}	2,25*	1,27*	28,9^{ns}	0,03^{ns}	2,0^{ns}	0,001^{ns}
Dose	4	101,82*	2,35^{ns}	1,85^{ns}	0,15^{ns}	0,33^{ns}	119,3^{ns}	0,07^{ns}	4,9^{ns}	0,001^{ns}
C x E	2	36,68^{ns}	0,88^{ns}	4,48^{ns}	0,12^{ns}	0,23^{ns}	62,3^{ns}	0,02^{ns}	1,4^{ns}	0,003^{ns}
C x D	4	96,13^{ns}	0,99^{ns}	2,09^{ns}	0,08^{ns}	0,33^{ns}	66,6^{ns}	0,8ns	1,8^{ns}	0,001^{ns}
E x D	8	**67,11***	**9,50***	4,04^{ns}	0,47*	**0,55***	42,2^{ns}	0,08^{ns}	**7,0***	0,001^{ns}
C x E x D	8	32,92^{ns}	1,39^{ns}	3,37^{ns}	**0,21***	0,17^{ns}	**43,5 ***	0,08^{ns}	4,5^{ns}	0,001^{ns}
Resíduo	116	19,91	1,05	2,32	0,08	0,19	20,1	0,06	3,0	0,001
Efeito de cultivar		(cm)	(n°)	(n°)	(cm)	(mm)	(n°)	(n°)	(g)	(-)
TMG 7062		72,28a	18,68a	4,96	3,31a	6,62a	36,40b	2,41	18,43	0,55b
BMX ZEUS		52,41b	14,06b	4,90	3,22b	6,37b	45,25a	2,42	17,86	0,62a
Média		62,34	16,37	4,93	3,26	6,49	40,8	2,41	18,09	0,58
CV (%)		7,16	6,27	30,88	8,99	6,73	10,99	9,86	9,62	5,06

[1/] Fontes de variação (FV); número de nós (NN); número de ramos por planta (RP); comprimento médio de entrenós (CE); diâmetro da haste principal (DHP); número de vagens (NV); número de grãos por vagem (NGV); peso de cem grãos (PCG); e índice de colheita (IC).
ns - não significativo a P > 0,05; * - significativo a P < 0,05.

Foi observada interação entre estádio e dose, além do efeito isolado da cultivar na altura final de plantas, a aplicação do regulador no estádio V9 apresentou redução linear de altura, na ordem de 7, 44 cm a cada 100 g i.a. ha^{-1}. No estádio R1 foi observada resposta quadrática, onde a altura mínima estimada foi de 58,74 cm, com a dose de 55,09 g i.a. ha^{-1} do cloreto de clormequate (altura mínima = -b/2*a), e houve a redução de 9,90% em relação à testemunha. As aplicações no estádio R3 não tiveram efeito significativo na altura de plantas (Figura 1).

A maior eficiência na redução de altura final de plantas sob aplicações no estádio V9, e a ausência de diferença significativa no estádio R3, demonstram a ausência de efeito do regulador quando aplicado em estádios mais tardios. Souza *et al*. (2013) observaram redução significativa na altura de plantas de soja sob aplicação de reguladores de crescimento no estádio R1, quando comparada à aplicação no estádio R3. Ao testar 5 reguladores de crescimento Marianno (2016) obteve maior redução de altura com o uso do cloreto de clormequate no estádio R1, enquanto as aplicações nos estádios mais tardios, R3 e R5, não apresentaram redução significativa. No estádio V9 as doses 75 e 100 g i.a. ha^{-1} foram as mais efetivas na redução na altura final de plantas, enquanto no estádio R1 houve redução significativa apenas

na dose 50 g i.a. ha^{-1}, sendo que as doses superiores não foram eficientes em reduzir a altura de plantas.

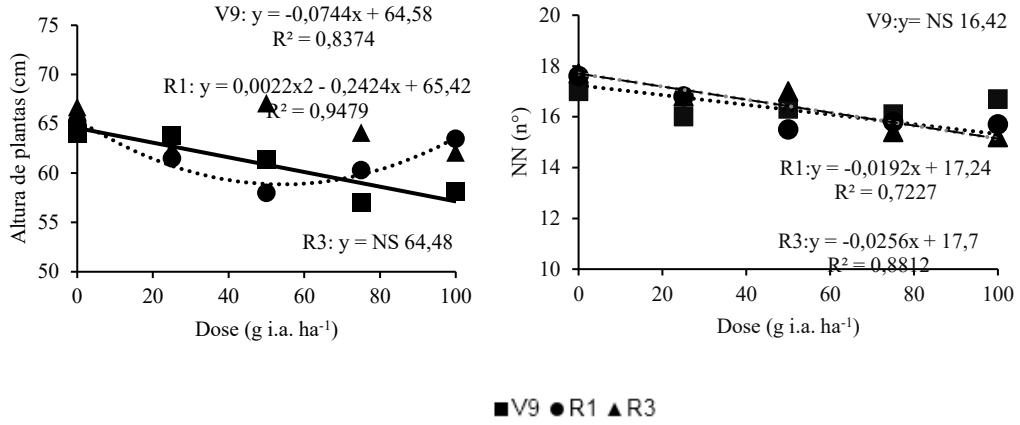

Figura 1. Altura de plantas e número de nós na haste principal (NN) em função do estádio e dose de aplicação do cloreto de clormequate na média das cultivares de soja testadas em casa de vegetação. Lages (SC).

Para o número de nós também houve interação entre estádio e dose de aplicação do cloreto de clormequate; o incremento das doses nos estádios mais tardios, R1 e R3, reduziu o número de nós. No estádio R1 houve taxa de decréscimo de 2 nós a cada 100 g i.a. ha^{-1}, enquanto no estádio R3 esta foi de 2,5 nós para cada 100 g i.a. ha^{-1} do regulador. O número de nós é proporcional à altura de plantas, desde que associado ao comprimento de entrenós (MARQUES, 2019).

Nos estádios R1 e R3 não foram observadas reduções significativas na altura, porém o aumento das doses do regulador influenciou no número de nós, e houve aumento do comprimento dos entrenós apicais, como forma de compensação. A aplicação mais precoce, no estádio V9, não teve efeito significativo sobre o número de nós. Os resultados sugerem que nas plantas submetidas à aplicação do regulador no estádio V9, o comprimento dos entrenós foi menor, uma vez que houve redução de altura, porém o número de nós não teve redução significativa. Tal característica pode ser considerada positiva, uma vez que o número de nós é considerado um componente de rendimento secundário e está correlacionado ao número de vagens por planta (MUNDSTOCK; THOMAS, 2005; OKI *et al*., 2018). Embora Moreira *et al*. (2020) não tenham encontrado efeito do regulador cloreto de clormequate (aplicado sobre plantas nos estádios V5, V9 e R1) sobre os caracteres NN, NGP, PCG e IC, ressalta-se que é importante manter constante o NN nas plantas visando-se manter o potencial produtivo das cultivares em decorrência de alteração de ambiente ou de algum tratamento específico (YANG *et al*., 2021).

A fim de melhor observar o efeito dos reguladores no comprimento de entrenós, foi analisada para cada cultivar a interação entre dose e estádio de aplicação (Figura 2). Na cultivar BMX Zeus as aplicações no estádio V9 reduziram o comprimento de entrenós na ordem de 0,5 cm a cada 100 g de i.a. ha^{-1}, esse decréscimo pode ser explicado pela redução de altura sem reduzir o número de nós.

A aplicação do regulador no estádio R3 apresentou resposta cúbica entre as doses aplicadas, na dose 25 g i.a. ha^{-1} houve redução de 10,91% em relação à dose 0, porém na aplicação das doses 50 e 75 g i.a. ha^{-1} observou-se aumento de 7,37 e 6,19%, respectivamente, em relação à testemunha, enquanto na dose 100 g i.a. ha^{-1} não houve diferença significativa, tal comportamento pode evidenciar um efeito compensatório de doses mais elevadas em estádios mais tardios de aplicação do regulador, relacionado ao menor número de nós neste estádio.

Na cultivar TMG 7062 sob aplicação do regulador nos estádios V9 e R1 foi observado encurtamento dos entrenós, com o incremento das doses na taxa de 0,38 cm a cada 100 g i.a. ha^{-1} em ambos os estádios. A aplicação no estádio R3 resultou em comportamento quadrático, com ponto de mínima de 3,38 cm, na dose 20 g i.a. ha^{-1} (menor comprimento = -b/2*a), enquanto as doses 75 e 100 g i.a. ha^{-1} demonstraram aumento significativo de 8,38 e 10,69% no tamanho de entrenós quando comparadas à testemunha.

Observou-se comportamento similar entre as cultivares nos estádios V9 e R1, o comprimento de entrenós menor ocorreu em consequência da menor altura, e não houve redução no número de nós em relação ao tratamento no estádio R3, sendo observado aumento do comprimento de entrenós em ambas as cultivares, no entanto de forma mais expressiva na cultivar TMG 7062, tal resposta pode ocorrer em virtude do atraso na senescência foliar dessa cultivar em relação à BMX Zeus, que tem ciclo mais precoce devido ao seu grupo de maturação inferior (5.5).

O incremento das doses no estádio R3 apresentou maior redução no número de nós, porém como resposta compensatória os entrenós apicais apresentaram maior elongação. Souza *et al*. (2013) observaram correlação linear positiva entre o comprimento de entrenós e altura de planta, tal relação pode explicar o comportamento desta variável. O alongamento dos entrenós pode tornar o caule mais frágil pela redução de lignina e celulose na sua estrutura, tal característica não é desejada, e pode aumentar o risco de acamamento (LIU *et al*., 2017).

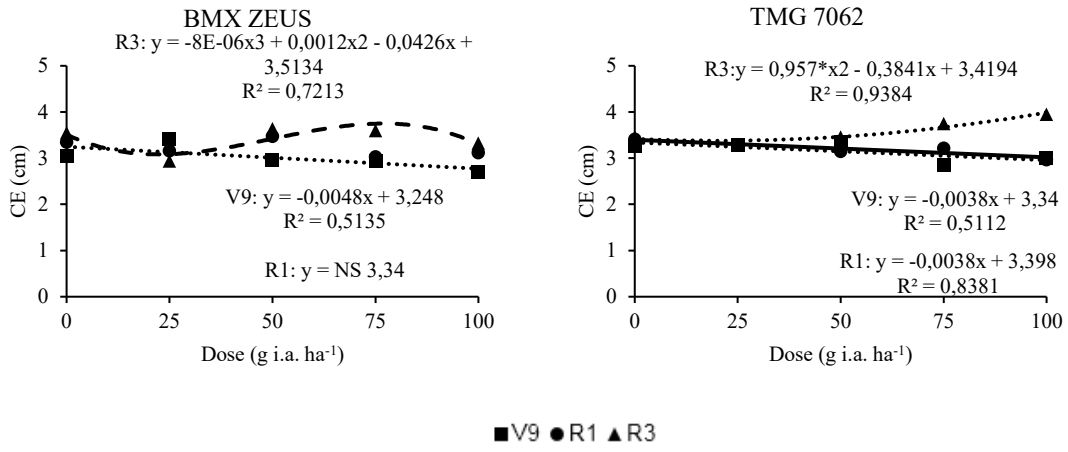

Figura 2. Comprimento de entrenós (CE) das cultivares de soja BMX Zeus e TMG 7062 em função do estádio e dose de aplicação do cloreto de clormequate em ambiente de casa de vegetação. Lages (SC).

Foi observado comportamento linear crescente entre o aumento da dose do regulador com aumento do diâmetro da haste principal (DHP) no estádio V9, com taxa de acréscimo de 0,57 mm para cada 100 g i.a. ha^{-1}. A aplicação do regulador no estádio R1 teve resposta quadrática, apenas na dose 25 g i.a. ha^{-1} houve tendência de incremento no DHP, sendo o ponto de máxima o diâmetro 6,67 mm (diâmetro máximo = -b/2*a) com a dose 40,5 g i.a. ha^{-1}, as doses 75 e 100 g i.a. ha^{-1} apresentaram tendência de redução de 3,55 e 6,50%, respectivamente, em relação à dose 25 g i.a. ha^{-1}. No estádio R3 a aplicação não demonstrou efeito significativo (Figura 3).

O resultado corrobora os de Souza *et al*. (2013), que observaram maior aumento do diâmetro da haste principal na primeira época de aplicação (R1), quando comparado à segunda época de aplicação de reguladores de crescimento, também foi observada correlação positiva com o número dos entrenós. Kaur *et al*. (2015) obtiveram incremento de 23,8% no diâmetro da haste com o uso do regulador cloreto de clormequate em relação à testemunha, porém não observaram diferença significativa entre os estádios de aplicação. Em trabalho com restrição hídrica na cultura da soja, Marianno (2016) obteve incremento no diâmetro da haste com o cloreto de clormequate aplicado no estádio R3, sinalizando possível efeito mitigatório desse regulador.

O diâmetro da haste pode favorecer a resistência ao acamamento, e os reguladores de crescimento podem atuar desviando os carboidratos que seriam destinados ao aumento de altura para o aumento do diâmetro e aumento da matéria seca na haste (WANG *et al*., 2016). Ramesh e Ramprasad (2013) observaram incremento de 28,4% no acúmulo de matéria seca da haste de plantas de soja mediante a aplicação da dose 137,5 g i.a. ha^{-1} do regulador cloreto de clormequate, em relação à testemunha.

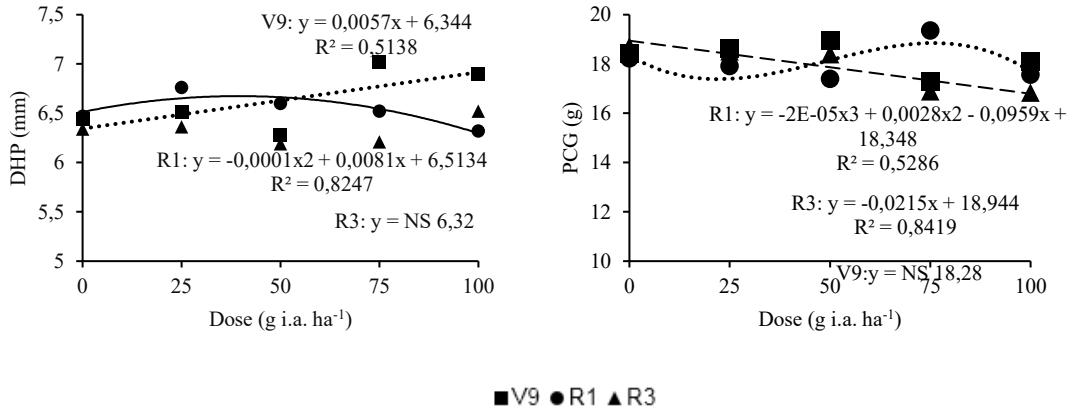

Figura 3. Diâmetro da haste principal (DHP) e peso de cem grãos (PCG) de soja em função do estádio e dose de aplicação do cloreto de clormequate em ambiente de casa de vegetação. Lages (SC).

Para o peso de cem grãos (PCG) as doses aplicadas no estádio V9 não tiveram efeito significativo, assim como Linzemeyer Júnior *et al*. (2008), que também não observaram efeito

no PCG com a aplicação do regulador de crescimento trinexapac-ethyl em estádio vegetativo. No estádio R1 houve comportamento cúbico, a aplicação da dose 75 g i.a. ha^{-1} incrementou 6,25% no PCG em relação à testemunha, enquanto as doses 25, 50 e 100 g i.a. ha^{-1} apresentaram respectivamente redução de 1,70, 4,55 e 3,62% para a variável. Tarun *et al*. (2016) observaram que a aplicação do regulador cloreto de clormequate incrementou em 17% o PCG de soja em relação à testemunha.

Para as aplicações no estádio R3 houve decréscimo no peso de cem grãos na ordem de 2,15 g a cada 100 g i.a. ha^{-1} de aumento da dose do regulador de crescimento. A redução no estádio mais tardio de aplicação (R3) corrobora com Souza *et al*. (2013), que obtiveram maior redução na massa de mil grãos (MMG) na aplicação mais tardia de reguladores de crescimento. A aplicação mais tardia do regulador de crescimento pode provocar a retenção de fotoassimilados em outros tecidos da planta, ou deslocamento para órgãos vegetativos, em detrimento dos grãos, tornando-os menos pesados (SOUZA *et al*., 2013). As aplicações mais tardias (R1 e R3) apresentaram maior altura final e maior comprimento de entrenós, em relação às efetuadas no estádio V9, independente da cultivar, sendo assim não houve priorização do enchimento de grãos, logo isso explica a redução no PCG.

No número de vagens por planta (NVP) foi observada interação entre dose e estádio para cada cultivar separadamente. Na cultivar BMX Zeus as aplicações nos estádios V9 e R3 não apresentaram comportamento significativo de dose, apenas efeito simples de estádio, sendo o NVP de 43,7 e 46,7, para V9 e R3, respectivamente. As aplicações no estádio R1 apresentaram comportamento linear positivo entre a dose aplicada e o número de vagens, houve acréscimo na taxa de 15 vagens a cada 100 g i.a. ha^{-1} de cloreto de clormequate (Figura 4).

Esses dados corroboram os de Kaur *et al*. (2015), que encontraram incremento significativo no número de vagens por planta sob aplicação do cloreto de clormequate no início da floração (R1) quando comparado à aplicação no início da formação das vagens (R3). Tarun *et al*. (2016) também obtiveram com o uso do cloreto de clormequate incremento no número de vagens por planta na ordem de 13,55%, comparado à testemunha, em aplicação no estádio vegetativo. A ausência de resposta na aplicação V9 pode estar relacionada à não ocorrência de efeito significativo no número de nós por planta encontrado nesta condição. A cultivar BMX Zeus apresenta senescência foliar antecipada quando comparada à cultivar TMG 7062, fator que limita a formação de novas vagens após a aplicação em R3.

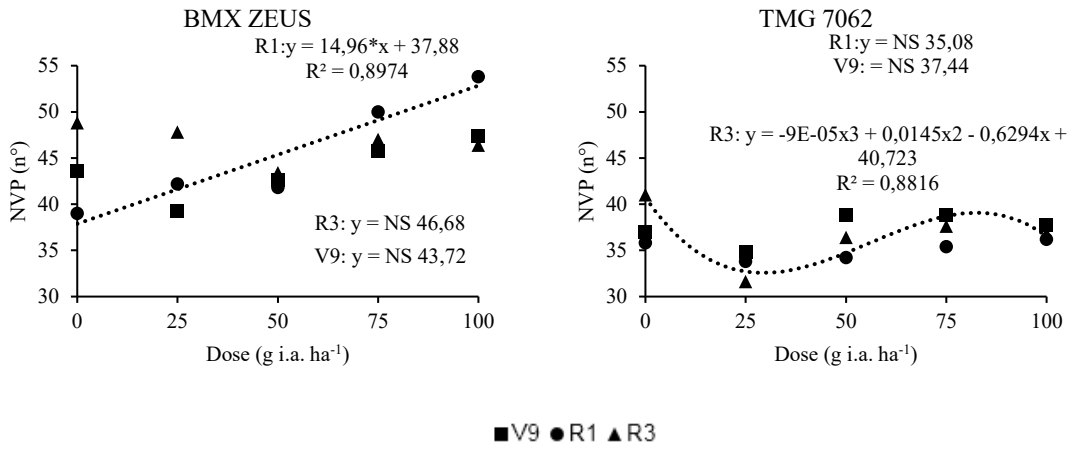

Figura 4. Número de vagens por planta (NVP) em função do estádio e dose de aplicação do cloreto de clormequate em ambiente de casa de vegetação. Lages (SC).

Para a cultivar TMG 7062, as aplicações no estádio R3 apresentaram resposta cúbica, a dose 25 g i.a. ha^{-1} teve redução no número de vagens de 13,06% em relação à testemunha, enquanto as doses 50, 75 e 100 g i.a. ha^{-1} não diferiram da testemunha ($P > 0,05$).

A resposta da cultivar TMG 7062 pode estar relacionada ao seu maior número de nós e atraso na senescência foliar que permite maior aporte de fotoassimilados para produção de vagens. Esses resultados divergem de Linzmeyer Júnior *et al*. (2008) e Souza *et al*. (2013), que não observaram efeito significativo nessa variável sob aplicação de reguladores de crescimento.

Não foi observado efeito significativo dos tratamentos no número de ramos por planta, número de grãos por vagem e no índice de colheita, sendo essas características que podem variar mais em função de outros fatores de manejo e genótipo cultivado.

No presente trabalho foram estabelecidas correlações entre as variáveis, a análise de correlação de Pearson (Tabela 2) demonstrou correlação positiva entre a altura com o número de nós e comprimento dos entrenós, como observado a redução de altura nas aplicações no estádio R1 influenciou na redução no número de nós. Segundo a análise de correlação o número de vagens por planta foi o componente que mais influenciou no índice de colheita, segundo Corassa *et al*. (2019) o número de vagens por planta é componente que tem maior influência no rendimento de grãos, e deve ser o de maior atenção em relação ao manejo. Foi observada correlação negativa entre o número de vagens por planta e número de grãos por vagem.

Tabela 2. Correlação de Pearson entre as variáveis altura de plantas, número de nós (NN), comprimento médio de entrenós (CE), número de ramos por planta (NRP), diâmetro da haste principal (DHP), número de vagens por planta (NVP), número de grãos por vagem (NGV), peso de cem grãos (PCG) e índice de colheita (IC) mediante aplicação do cloreto de clormequate. Lages (SC)

	Altura	**NN**	**CE**	**NRP**	**DHP**	**NVP**	**NGV**	**PCG**	**IC**
Altura	1,00	**0,85****	**0,38***	-0,07	0,13	-0,11	0,16	0,08	-0,03
NN	-	1,00	0,02	-0,13	0,32*	-0,14	0,06	0,13	-0,05
CE	-	-	1,00	0,08	-0,21	0,01	0,12	-0,02	0,01
NRP	-	-	-	1,00	0,00	0,03	0,05	-0,09	-0,03
DHP	-	-	-	-	1,00	0,02	-0,08	-0,03	0,01
NVP	-	-	-	-	-	1,00	**-0,42***	-0,21	**0,57***
NGV	-	-	-	-	-	-	1,00	-0,32	0,02
PCG	-	-	-	-	-	-	-	1,00	0,06
IC	-	-	-	-	-	-	-	-	1,00

** e * = diferem estatisticamente pelo teste t a 0,01 e 0,05, respectivamente; ns: não significativo.

4 CONSIDERAÇÕES FINAIS

As aplicações do cloreto de clormequate no estádio V9 são mais eficazes na redução de altura de plantas, especialmente nas doses 75 e 100 g i.a. ha^{-1}, sem afetar o número de nós, de vagens por planta e o peso de cem grãos, além de incrementar no diâmetro da haste principal, que é uma importante característica de resistência ao acamamento.

Aplicações mais tardias como no estádio R3 se mostram pouco efetivas. Os resultados demonstram que a resposta ao cloreto de clormequate é influenciada pelo genótipo, sendo distinta entre eles. A cultivar BMX Zeus apresenta maior índice de colheita e de seus componentes e mostra maior equilíbrio na partição entre biomassa reprodutiva e vegetativa. Apesar da eficácia da aplicação no estádio V9, futuros trabalhos podem elucidar dúvidas sobre a aplicação sequencial do cloreto de clormequate, como é recomendado no seu uso comercial na cultura do algodão (*Gossypium hirsutum* L.). Além de reduzir o risco de acamamento o uso de reguladores nos estádios mais responsivos pode otimizar a partição de fotoassimilados, aumentando a eficiência produtiva da cultura.

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Efficacy of growth regulators in a lodging-sensitive wheat cultivar: grain yield, crop economic profitability and flour industrial quality

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Abstract

Growth regulators in annual crops such as wheat are being used for reducing plant lodging. Thus, this study aimed to assess the effects of applying different growth regulators on grain yield, industrial quality of flour, and the economic viability of a lodging-sensitive wheat cultivar, namely TBIO-Pioneiro. The RDB experimental design consisted of five treatments and nine replicates. The treatments used the following regulators: trinexapac-ethyl (T1), ethephon (T2), prohexadione calcium (T3), chlormequat chloride (T4) and a control (untreated) (T5). The rates were determined according to the manufacturer's recommendations for other crops. Plant height was reduced with application of regulators, for example, trinexapac (93.1 cm), and control (100.7 cm). This same growth regulator tested increased TGW (36.2 g) over the control (33.6 g) making plant lodging lowest (from 34%-control to about 10%-treated). Grain yields with application of growth regulators were higher than control (3.1 t ha^{-1}), ranging from 148.9 to 158.9% (means regulator-treated of 4.8 t ha^{-1}). Regarding grain alveograph indices, there was an increase in gluten strength (W) with the use of ethephon (309 10^{-4}J), prohexadione (309.3 10^{-4}J), and chlormequat (309 10^{-4}J), compared with control (240.3 10^{-4}J). The economic return on investment was higher than the control, of up to 35.7%. It is concluded that application of prohexadione, ethephon and chlormequat changes the gluten strength of wheat grains and that the growth regulators tested are efficient in controlling lodging, reducing plant height and increasing grain yields, thus providing productive stability and higher financial returns in wheat cropping.

Keywords: Chlormequat chloride. Ethephon. Prohexadione-calcium. Trinexapac-ethyl. *Triticum aestivum* L. Economic performance.
Abbreviations: CAV_Centro de Ciências Agroveterinárias; CEPA_Centro de Socioeconomia e Planejamento Agrícola – Epagri/Cepa; DAA_Days after application; EPAGRI_Empresa de Pesquisa Agropecuária e Extensão Rural de Santa Catarina; FMI_Foreign matter and impurities; GA_Gibberellin; GI_Gross income; GV_Grain volume; GY_grain yields; HW_Hectoliter weight; IA_Lodging index; L_Extensibility; M%_Grain moisture content; NDVI_Normalized difference vegetation index; P/L_Tenacity/extensibility ratio; P_Tenacity; RDB_Randomized block design; SPAD_Soil plant analysis development; TGW_Thousand grain weight; UDESC_Universidade do Estado de Santa Catarina/Santa Catarina State University; V%_base saturation; W_Gluten Strength

Introduction

Current increasing yields achieved by wheat (*Triticum aestivum* L.) producers are associated with the higher productive potential of modern cultivars and use of a greater number of crop inputs, such as nitrogen fertilization (Zagonel and Fernandes, 2007). This is due to increasing intensification of winter cereal production systems for the purpose of achieving higher yields (Subedi et al., 2021).

The use of high nitrogen doses and greater sowing density rates enhance plant growth of this cereal, which leads to lodging (Rodrigues et al., 2013; Fioreze and Rodrigues, 2014). The stem of lodged plants bends toward the ground as a result of lower resistance, occurrence of winds, and the weight of water accumulated in mature spikes (Espindula et al., 2010; Peake et al., 2020). Lodging is associated with wheat grain yields, quality of grains during the grain filling stage, reduced photosynthesis efficiency and translocation of assimilates to the spikes and, after maturation, it favoring deterioration, preharvest grain germination and, consequently, harvest losses (Zagonel and Fernandes, 2007).

One of the alternatives to reduce wheat plant lodging without the need to reduce nitrogen fertilization or sowing densities is the application of growth regulators. Regulators are synthetic substances not produced by plants and which affect their growth (Taiz et al., 2017). Plant growth regulators reduce plant height by reducing cell elongation or the cell division rate, and they are antagonistic to gibberellins and auxins (Rademacher, 2000).

Growth regulators can be classified into two groups: ethylene-releasing compounds (ethephon) and gibberellin (GA) biosynthesis inhibitors. Among GA biosynthesis inhibitors, there are compounds from the quaternary ammonium group (chlormequat chloride, mepiquat chloride and AMO-1618) and the phosphono group (chlorphenium chloride), which block GA synthesis before *ent*-kaurene, i.e., phase 1 of GA biosynthesis; the group of nitrogen-containing

heterocyclics, such as ancymidol (pyrimidines), cyclases (norbornane diazetines) and triazole (paclobutrazol and uniconazole), which inhibit the *ent*-kaurene oxidation into *ent*-kaurenoic acid, in phase 2 of GA biosynthesis; and derivatives of acylcyclohexadiones, such as trinexapac-ethyl and prohexadione calcium, which inhibit 2-oxoglutarate-dependent dioxygenases in phase 3 of gibberellin biosynthesis (Rademacher, 2000).

Concerning the use of growth regulators, previous reports have argued that they reduce plant height (Zagonel and Fernandes, 2007; Penckowski et al., 2010; Marco-Júnior et al., 2013; Chavarria et al., 2015; Hawerroth et al., 2015, Subedi et al, 2021), reduce the length of the second and third internodes (Schwerz et al., 2015) and culm length (Espindula et al., 2010), and reduce plant lodging (Penckowski et al., 2010; Zhang et al., 2014; Hawerroth et al., 2015; Corbin et al., 2016; Marolli et al., 2017; Ahmad et al., 2020). In addition, they can increase grain yield (Penckowski et al., 2010; Zhang et al., 2014; Chavarria et al., 2015).

Studies on growth regulators to obtain responses about their effects on cereal yields are essential in view of the good effects found for plant morphology and reduced lodging (Mykhalska et al., 2020). There is also little information about the relationship between productive traits, industrial flour potential, and the technical-economic viability of using these products. Thus, the aim of this study was to assess the effect of applying different growth regulators on a lodging-sensitive wheat cultivar on grain yield, industrial quality of grains and flour, and the economic viability of using these products.

Results

Chlorophyll content, plant height and lodging

The analysis of variance showed that there was no significant effect for chlorophyll B, NDVI and plant height at 14 DAA of different growth regulators applied to the wheat crop (Table 2).

It can be seen that chlorophyll A and total chlorophyll achieved lower values, 34.2% and 40.6% respectively, after application of ethephon, compared with control (without growth regulator). The application of prohexadione-calcium, with 35.5% of chlorophyll A and 42% of total chlorophyll, did not differ from the other treatments. Likewise, trinexapac-ethyl did not differ from the other growth regulators with respect to total chlorophyll, with 42.5% over the mean (Table 2).

Wheat plant heights at 14 days after application of growth regulators did not exhibit differences, with 52.0 cm on average. However, at 21 DAA, the untreated wheat plants and the ones sprayed with chlormequat chloride were taller, with 86.8 and 86.2 cm in height, respectively. This result did not differ from that of application with ethephon: 85.3 cm. The shortest plant height was achieved with application of trinexapac-ethyl, which accounts for a 4% reduction when compared with control (Table 2). Such result remained until harvest, i.e., plant height with application of trinexapac-ethyl at harvest was lower (93.1 cm), with a reduction of 7.5% in comparison to the control plants, which were 100.7 cm tall (Table 2).

With respect to the percentage of plant lodging, the lowest values were found with applications of trinexapac-ethyl and prohexadione-calcium: 8.6 and 12.5%, respectively. However, the highest lodging rate was found (34.2%) without application of growth regulators (control), a result that did not differ from the one after application with chlormequat chloride, with 25.2% (Table 2).

Moisture content in wheat grains, after application of growth regulators, was higher when applying chlormequat chloride, i.e., there was an increase of 1.5% in comparison to the seeds of the control treatment, i.e., 14.0% (Table 2).

Grain yield components

For the hectoliter weight (HW) of wheat grains, the highest values were found for application of trinexapac-ethyl and prohexadione-calcium, 77.9 and 77.5 kg 100 l^{-1} of grains, respectively, which did not differ from the grains of wheat plants that received ethephon application, which showed HW of 76.7 kg 100 l^{-1}. The increased HW values achieved with trinexapac-ethyl and prohexadione-calcium regulators compared with control were 3.7 to 3.1% higher, respectively (Table 2).

TGW after application of trinexapac-ethyl on wheat plants was 36.2 g, i.e., 7.0% higher than control, but it did not differ from the TGW of wheat grains treated with other plant growth regulators (Table 2). The application of growth regulators increased wheat grain yield from 3.1 t ha^{-1} (control) to 4.8 t ha^{-1} (mean of all regulators), which accounts for an approximate increase of 50% in GY (Table 2).

Farinography and economic yield

Concerning the alveograph variables of wheat flour obtained from the grains of plants treated with growth regulators, there was no significant effect for tenacity (P), extensibility (L) and for the tenacity/extensibility ratio (P/L) (Table 3). There was a significant effect only for gluten strength (W, 10^{-4}J), i.e., the application of prohexadione-calcium (309.3 10^{-4}J), ethephon (309.0 10^{-4}J) and chlormequat chloride (309.0 10^{-4}J) provided higher gluten strength compared to control (240.3 10^{-4}J) and the treatment with trinexapac-ethyl (240.3 10^{-4}J) (Table 3).

The process of analysis of the economic viability of use of each growth regulator is described in Table 2. The treatment with chlormequat chloride indicated higher net income, with 601.19 US$.$ha^{-1}$, and return of investment of 35.9%, followed by prohexadione-calcium with 35.7%, trinexapac-ethyl with 34.2% and ethephon with 32.2%, when compared with producer standard treatment. It can be seen that regardless of the growth regulator used, net income and return on investment are higher than the ones achieved with the standard treatment (Table 4).

Discussion

The occurrence of lodging in cereal crops such as wheat interferes with crop yield and, consequently, with grain quality (Mykhalska et al., 2020). Therefore, application of growth regulators is an alternative way to prevent plant lodging.

According to the results found for chlorophyll content in wheat plants after application of ethephon, which is a product characterized by ethylene release (Rademacher, 2000), it can be argued that it may influence the leaf senescence process. Ethylene is a plant hormone considered to be a positive regulator of leaf senescence (Jibran et al., 2013), and it is also important in the senescence of bracts and floral glumes and in the maturation of wheat grains (Beltrano et al., 1994).

Chlorophyll is associated with plant photosynthesis efficiency owing to the pigments that act in the photochemical phase

of photosynthesis, which are specialized in capturing light (Taiz et al., 2017). Leaf chlorophyll contents decrease as the senescence process advances, when degradation of these pigments is accelerated (Jespersen et al., 2016). According to Schippers et al. (2015), senescence is an important process in agricultural crops because it affects grain yield and quality. According to these authors, delayed senescence allows greater assimilation of carbohydrates and, consequently, higher crop yields.

However, the use of other growth regulators did not have a harmful effect on the chlorophyll contents of plants. This can occur since these products favor the interception of sunlight by the leaves, because of the plants' upright structure, as reported by Fioreze and Rodrigues (2012). These authors studied plant density changes associated with application of trinexapac-ethyl in wheat crops (in the culm elongation phase) and found that the plants exhibited a more intense green color in leaves, with SPAD index ranging between 48.4 and 50.4. Chavarria et al. (2015) found increases of chlorophyl A, B and total chlorophyl contents, of 4.4, 5.4 and 4.5%, respectively, in wheat cv. Mirante plants sprayed with trinexapac-ethyl.

For plant height, the growth regulators applied in the wheat crop were especially effective in inhibiting gibberellins synthesis. Thus, with reduced levels of gibberellins, plants do not reach a stature considered as normal, i.e., they develop shorter and thicker stalks, which ensure more resistance to adverse conditions (Khodanitska et al., 2021). In addition, a shorter plant height allows reduction of losses caused by plant lodging and favors the interception of sun radiation (Zagonel and Fernandes, 2007; Peake et al., 2020).

Thus, these results corroborate the ones found by Espíndula et al. (2010), who studied the effect of application of chlormequat chloride and trinexapac-ethyl on wheat crops. The authors found that increased doses of both plant growth regulators provided a reduction of plant stalk length, with mean values ranging from 62.0 to 76.8 cm, compared with control, as well as a shorter peduncle length. Likewise, Chavarria et al. (2015) found that the application of trinexapac-ethyl in wheat cultivars Quartzo and Mirante reduced plant height by 16.9 and 15.8%, respectively.

Lower plant height is related to a lower percentage of plant lodging (Zagonel and Fernandes, 2007), which explains the findings. Ahmad et al. (2020) reported that the application of ethephon in wheat crops resulted in reduced plant height, less lodging, larger internode diameter and higher lignin content in such crops. In addition, there was a negative correlation between lignin content and lodging, that is, high lignin levels contribute to increased plant resistance to lodging.

The percentage of grain moisture that resulted from the application of prohexadione-calcium can be associated with the regulation of cytokinin contents in the plants, delayed leaf senescence and, consequently, delayed moisture loss in grains. The advance and rate of plant senescence may vary, but plant hormones are key factors that can delay or accelerate this process (Taiz et al., 2017). According to Peleg et al. (2011), leaf senescence is retarded when cytokinin contents increase. As reported by Grossmann et al. (1994) and Bekheta et al. (2009), prohexadione-calcium application increases the concentration of cytokinin and abscisic acid in plant tissues. This is due to reduction of ethylene hormone production (Grossmann et al., 1993).

Hectoliter weight (HW) of wheat grains is used in several countries as a traditional measure to determine grain quality (Costa et al., 2008). According to Normative Instruction no. 7 of August 15, 2001 (Brasil, 2001) wheat grains are classified into three types, based on the minimum HW value, moisture, extraneous matters and impurities. HW values ranging from 70 to 74.9 kg 100 l^{-1} classify wheat grains as type 3, ranging 75 to 77.9 kg 100 l^{-1} as type 2, and over 78 kg 100 l^{-1} as type 1. Thus, all grain volumes obtained with and without application of growth regulators meet the minimum quality value for HW. Stefen et al. (2014) found HW values for wheat grains of cv. Mirante between 72.0 and 77.9 kg 100 l^{-1} when they examined the effects of two different dosages and timing of application of foliar nitrogen and different growth regulators. However, the regulators did not influence differences in HW in the grains produced.

The results found for HW, TGW and yield indicate that the application of growth regulators in wheat has a favorable effect on these attributes. This may be due to lower plant height and, consequently, lower lodging rate, as well as better interception of solar radiation (Zagonel and Fernandes; 2007) owing to the angle of the flag leaf, i.e., arranged more uprightly, and changes in the plant foliar architecture (Peneckowski and Fernandes, 2010).

The results of this study agree with the results found by Penckowski et al. (2010), who reported increased grain yields of wheat cv. Avante, with mean values ranging from 5725 to 6234 kg ha^{-1} when they evaluated the time of application of trinexapac-ethyl and nitrogen doses. Stefen et al. (2014), however, assessed the effect of two different dosages and timing of application of foliar nitrogen and different regulators and found that growth regulators did not affect the TGW and grain yield of cv. Mirante. Similarly, Buzetti et al. (2006) did not find interference in rice yield with application of chlormequat chloride, and Hawerroth et al. (2015) in white oats with application of trinexapac-ethyl. It should be noted that the results of research with growth regulators and their effects on grain yield and quality has a strong component, which is the crop year-genotype interaction. In crop years more favorable to the occurrence of lodging, that is, when in fact there is greater plant lodging, grain yield and quality in the control treatment are more negatively impacted, and where growth regulators are applied, the difference in grain yield increases positively. This was found to occur in this study (Table 2, data on lodging and grain yield).

The use of growth regulators can influence plant growth and the industrial quality of wheat, and it may change the alveograph properties of wheat (Penckowski et al., 2010). In the present study, there was alteration only in gluten strength (W) (Table 3), unlike the finding reported by Penckowski et al. (2010) after application of trinexapac-ethyl, which promoted increased extensibility (L), reduced tenacity/extensibility ratio and increased gluten strength for cv. BRS 177, while cv. Avante did not exhibit alveograph changes. Similarly, Stefen et al. (2014) found that the application of trinexapac-ethyl in cv. Mirante did not provide changes in the flour's W index. Such results suggest that the response of W depends on the cultivar planted (Penckowski et al., 2010; Stefen et al., 2015). Thus, cv. TBIO-Pioneiro did not alter the flour's W index after application of trinexapac-ethyl, but the application of prohexadione-calcium, ethephon and chlormequat chloride increased W by nearly 28% (Table 3).

Application of growth regulators reduces the occurrence of lodging in wheat crops and enables adjustments in nitrogen

Table 1. Chemical analysis of the soil in the experimental area using growth regulators in wheat cultivation.

pH-water	pH-SMP	P	K	H + Al	CEC pH 7.0	V%	O.M.
		(mg dm-^{3})	(mg dm-^{3})	(cmol dm-^{3})	(cmol dm-^{3})	%	%
5.6	5.4	19.4	99	8.3	22	62	3.4

pH SMP = SMP-buffer index (Shoemaker-McLean-Pratt); CEC = cation exchange capacity; V% = base saturation; O.M. = organic matter.

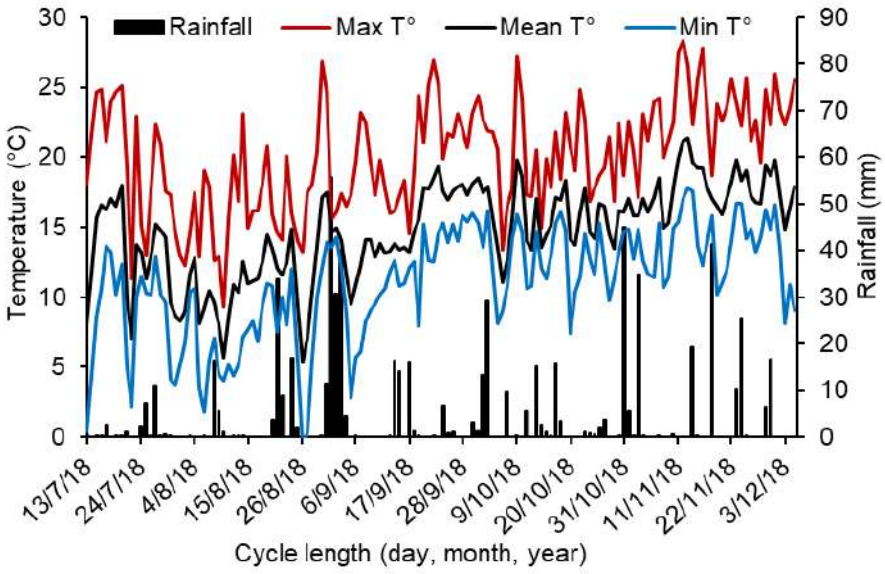

Fig 1. Rainfall, minimum (min T°), mean (mean T°) and maximum (max T°) temperatures recorded during the wheat growing period (2018 growing season). Source: INMET, 2019.

Table 2. Chlorophyll A and B, total chlorophyll, NDVI, plant height at 14, 21 DAA and at harvest, lodging, foreign matters and impurities (FMI), moisture (%), hectoliter weight (HW), thousand-grain weight (TGW) and grain yields (GY) of wheat produced with application of different growth regulators.

Variables	Control		Trinexapac		Prohexadione		Ethephon		Chlormequat		Mean	CV (%)*	P>f
Chlorophyll A	36.32	a	35.85	a	35.55	ab	34.18	b	36.58	a	35.70	3.17	<0.01
Chlorophyll B	7.30	ns	6.63	-	6.45	-	6.40		6.83		6.72	10.31	0.583
Total Chlorophyll	43.63	a	42.48	a b	42.00	ab	40.58	b	43.40	a	42.42	4.00	0.003
NDVI	7.27	ns	7.17		7.08		7.02		7.17		7.14	3.23	0.198
Height (14DAA) (cm)	54.44	ns	50.78		51.78		50.44		52.78		52.04	8.75	0.350
Height (21DAA) (cm)	86.78	a	83.33	c	84.00	bc	85.33	ab	86.22	a	85.13	1.59	<0.01
Height (harvest) (cm)	100.67	a	93.11	c	94.11	bc	95.56	bc	96.44	b	95.98	2.33	<0.01
Lodging (%)¥	34.17	a	8.61	c	12.50	c	16.11	bc	25.22	ab	19.32	6.44	<0.01
FMI (%)	10.59	a	3.74	b	2.05	b	1.67	b	2.62	b	4.14	6.17	<0.01
Moisture (%)	14.03	b	14.60	b	14.22	b	13.91	b	15.51	a	14.45	2.43	<0.01
PH (kg 100 l^{-1})	75.13	b c	77.89	a	77.46	a	76.73	ab	74.68	c	76.38	1.84	<0.01
TGW (g)	33.64	b	36.19	a	35.46	ab	34.93	ab	35.62	ab	35.17	4.33	0.013
GY (t ha^{-1})	3.09	b	4.83	a	4.88	a	4.60	a	4.91	a	4.46	14.77	<0.01

*Means followed by different letters in the rows differ from each other by Tukey's test ($p<0.05$); ¥: arcsine transformed data $[(x/100)^{0.5}]$.

Table 3. Wheat flour alveograph indices resulting from application of plant growth regulators.

Variables	Control	Trinexapac	Prohexadione	Ethephon	Chlormequat	Mean	CV (%)	P>f
Tenacity (P)	165.7^{ns}	165.7	155.3	155.3	155.3	159.5	19.9	0.982
Extensibility (L)	53.0 ^{ns}	53.0	57.0	57.0	57.0	55.4	6.4	0.391
Gluten strength (W, 10^{-4}J)	240.3b	240.3b	309.3a	309.0a	309.0a	281.7	4.3	<0.01
P/L ratio	3.13^{ns}	3.13	2.73	2.73	2.73	2.89	20.6	0.791

*Means followed by different letters in the rows differ from each other by Tukey's test (p<0.05). CV: coefficient of variation.

Table 4. Economic viability (US$) of applying growth regulators in wheat crops.

Treatment	Expenses	Gross income	Net income	Return on
	----------(US$ ha^{-1}) ----------			Investment (%)
Producers' standard treatment	733.18	842.08	108.90	-
Trinexapac-ethyl	747.74	1317.46	569.72	34.23
Prohexadione-calcium	1062.77	1332.52	269.75	35.72
Ethephon	883.00	1257.11	374.12	32.25
Chlormequat chloride	738.23	1339.42	601.19	35.93

fertilization and sowing density for higher production potential, and reduces quantitative and qualitative losses of wheat grains caused by lodging. The reason is that lodging can reduce photosynthesis efficiency (Ingver et al., 2010; Packa et al., 2015) and translocation of photoassimilates, and increases the deterioration and germination of seeds and grains in the preharvest phase (Zagonel and Fernandes, 2007). As a result of the benefits achieved with application of growth regulators, there is a positive economic return, irrespective of the product chosen, as can be seen in the present study, which shows economic returns of 32.3 to 35.9 % for wheat cultivar TBIO-Pioneiro, when compared with control (Table 4).

Materials and Methods

Field trial and growth conditions

The experiment was conducted in the experimental site of the *Centro de Ciências Agroveterinárias- CAV* [Center of Agricultural and Veterinary Sciences] in the Santa Catarina State University - UDESC, in Lages -SC, at coordinates 27° 52' South latitude, 50° 18' West longitude and mean altitude of 930 m during 2018. The soil is characterized as an Aluminum Humic Cambisol with clayey texture (Embrapa, 2013). Temperature and rainfall data during the experiment, as shown in Figure 1, were recorded by the National Institute of Meteorology (Inmet). The lodging-sensitive wheat cultivar was TBIO-Pioneiro, one of the most cultivated wheat species in Brazil, used as flour in baking processes, but considered as sensitive to moderately sensitive to plant lodging (Kuhnem et al., 2020). A growth regulator is used with a view to achieving grain yield greater than 3 t ha^{-1}. Only TBIO-Pioneiro was used, because in lodging-tolerant cultivars, it is not technically-economically sensible to apply growth regulator to mitigate plant lodging.

Conduction of study and experimental design

Prior to sowing, which was conducted on July 13, 2018, the seeds were treated with a commercial product based on pyraclostrobin + thyofanate methyl + fipronil (Standak Top®, 5-45-50 g ai. 100 kg^{-1} of seeds, respectively). Base fertilization consisted of an application of 400 kg ha^{-1} of NPK 5-20-10 fertilizer. For foliar fertilization, 60 kg ha^{-1} of nitrogen was used, divided between the plant tillering and elongation phases, according to recommendations of the *Comissão de Química e Fertilidade do Solo* (CQFS, 2016) [Soil Chemical and Fertility Commission] and technical information for wheat cultivation (Kuhnem et al., 2020), targeting a grain yield of 5 t ha^{-1}.

A phytosanitary treatment was carried out according to technical recommendations for wheat crops (Kuhnem et al., 2020). The treatments with application of growth regulators were performed between stages GS31/32, according to Zadoks, Chang and Konzak scale (1974), corresponding to the period in which plants have the first visible node and the second perceptible node in the main plant stem.

The experiment used a randomized block design with five treatments and nine replications. The treatments consisted in applying the following growth regulators: trinexapac-ethyl (Moddus®, 125 g a.i. ha^{-1}), ethephon (Ethrel®, 480 g a.i. ha^{-1}), prohexadione-calcium (Viviful®, 130 g a.i. ha^{-1}), chlormequat chloride (Tuval®, 25 g a.i. ha^{-1}) and control (without application). The experimental plots consisted of five 10-m long sowing rows spaced 0.20 m.

Traits measured

The analyses performed after application of the growth regulators were NDVI (Normalized Difference Vegetation Index), chlorophyll A and B and total chlorophyll at 14 and 21 days after application (DAA); plant height (measured from the plant base, close to the ground, to the spike extremity at 14 and 21 DAA, and at the physiological harvest maturity); lodging index through visual evaluation, using the equation IA (%) = I x A x 2, where "I" is the vertical angle formed by the plant stalk in relation to the ground, with scores ranging from 0 to 5 (0 for absent inclination and 5 when all plants were lodged); "A" is the plot area with lodged plants, with scores ranging from 0 to 10 (0 for no lodging and 10 when all plot area contained lodged plants; and "2" is the correction coefficient to percentage, according to the methodology adapted from Moes and Stobbe (1991).

After harvesting, the percentage of moisture was determined by oven drying at 105 °C for 24 h, and using the equation M% = (iW – fW)/iW*100, where iW is the initial weight of the sample and fW, weight after drying; hectoliter weight, using a 0.25 L hectoliter scale and the equation HW = (GHW x 100)/ HW, where GHW refers to the weight of grains measured by the hectoliter scale, and GV is grain volume expressed in kg 100 l^{-1}; grain yield (weight of harvested grains >1.75 mm); percentage of foreign matters

and impurities (FMI); thousand-grain weight (TGW) with percentage of grain moisture corrected to 13%.

Flour quality was analyzed by alveograph indices using the method 54-30 of AACC (1995; Stefen et al., 2015), for determination of gluten Strength (W, 10^{-4}J), Tenacity (P), Extensibility (L) and Tenacity/Extensibility Ratio (P/L).

The economic viability of using different growth regulators in wheat crops was also assessed for each hectare of production. Gross income (US$) was determined on the basis of the grain yield obtained for each treatment; net income, by the difference between gross income (US$) and expenses (US$); and return on investment through the following formula: (gross income with investment less gross income without investment) / (100 * gross income with investment). Data were collected from Epagri/ Cepa for fixed costs (based on April 2021) of a medium-technology farm, and price of a 60-kg bag of wheat marketed in the period, corresponding to 16.32 US$ (83,49 R$) (Cepa, 2021). The costs of growth regulators used per hectare were calculated on the basis of 2021 selling prices: trinexapac-ethyl (Moddus®) = 14.57 US$ ha^{-1}, prohexadione-calcium (Viviful®) = 329.95 US$ ha^{-1}, ethephon (Ethrel®) = 149.82 US$ ha^{-1} and chlormequat chloride (Tuval®) = 5.06 US$ ha^{-1}.

Statistical analysis

The results were subjected to analysis of variance (Anova) by the F-test, and when significant differences were found, Tukey's test was used ($p<0.05$).

Conclusion

Application of growth regulators in lodging-sentitive wheat cv TBIO-Pioneiro is effective for controlling lodging (trinexapac-ethyl, prohexadione-calcium and ethephon), reducing plant height (trinexapac-ethyl, prohexadione-calcium, ethephon and chlormequat chloride) and increasing grain yield (trinexapac, prohexadione, ethephon and chlormequat). Application of prohexadione-calcium, ethephon and chlormequat chloride as growth regulators positively change the gluten strength of wheat flour. Application of growth regulators in wheat plants provides higher financial return in grain production.

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Physiological potential of wheat seeds produced under light restriction

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ABSTRACT: The low radiation intensity can negatively affect the production of photoassimilates in plants for allocation and synthesis of reserves into seeds. This study aimed to evaluate the physiological quality of wheat seeds produced by plants imposed on conditions of light reduction in different phenological stages, remaining until the end of the cycle. The experiment was carried out in a greenhouse in 2018, with a completely randomized design, with 11 treatments and 20 repetitions. The treatments consisted of 11 seed lots: 10 of plants grown in artificial shade (70%) started in the Zadoks code's phenological stages: 23; 30; 35; 45; 55; 60; 65; 70; 75; 80 and control lot (without shading). The seeds were analyzed for germination and vigor. Seed germination was not affected. There was a reduction in vigor through the tests of accelerated and cold aging when the shading started between stages 55 and 65, but with a minimum vigor of 93 and 94%, respectively. It is concluded that the restriction of light during the development of wheat plants does not affect the germination of the seeds produced but impairs the vigor mainly when imposed in the reproductive phases.

Key words: low light intensity; photosynthesis; shading; *Triticum aestivum* L.; vigor

Potencial fisiológico de sementes de trigo produzidas sob condições de restrição luminosa

RESUMO: A baixa intensidade de radiação pode afetar negativamente a produção de fotoassimilados nas plantas para a alocação e síntese de reservas nas sementes. Objetivou-se neste estudo avaliar a qualidade fisiológica de sementes de trigo produzidas por plantas impostas a condições de redução da luz em diferentes estágios fenológicos, permanecendo até o final do ciclo. O experimento foi realizado em casa de vegetação em 2018, com delineamento experimental inteiramente casualizado, com 11 tratamentos e 20 repetições. Os tratamentos consistiram em 11 lotes de sementes: 10 de plantas cultivadas em sombra artificial (70%) iniciados nos estágios fenológicos (códigos de Zadoks): 23; 30; 35; 45; 55; 60; 65; 70; 75; 80 e lote testemunha (sem sombreamento). As sementes foram analisadas quanto à germinação e vigor. A germinação das sementes não foi afetada. Houve redução do vigor através dos testes de envelhecimento acelerado e frio foi quando o sombreamento iniciou entre os estágios 55 e 65, mas com um vigor mínimo de 93 e 94%, respectivamente. Conclui-se que a restrição da luz durante o desenvolvimento das plantas de trigo não afeta a germinação das sementes produzidas, mas prejudica o vigor principalmente quando imposto nas fases reprodutivas.

Palavras-chave: baixa intensidade luminosa; fotossíntese; sombreamento; *Triticum aestivum* L.; vigor

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Introduction

Wheat (*Triticum aestivum* L.) is the second most produced cereal worldwide, with world production of 731.62 million tons in the 2018 season (FAO, 2020). In Brazil, the annual production was 5.4 million tons in the same year, according to data from the National Supply Company (Conab, 2020).

The satisfactory growth and productivity of agricultural crops is dependent on local environmental conditions, among which the importance of luminosity stands out. It is indispensable in the photosynthetic process for the production of photoassimilates by plants, required for vegetative growth and development of reproductive organs of agronomic interest, such as seeds (Marcos Filho, 2015).

Low light incidence conditions are related to factors intrinsic and extrinsic to the crop, as well as conditions of greater plant density in sowing, latitude, seasons, integrated crop-forest systems and less hours of sunshine or greater cloudiness (daytime cloudiness index), that normally occur in the high fields in Rio Grande do Sul and Santa Catarina even in the months of June, July and August (Custódio et al., 2009). In this way, the development of crops, such as wheat, can be compromised.

In wheat, some studies point out the reduction of the photosynthetic rate (Mu et al., 2010) as the main negative effect of shading, resulting in less biomass production under conditions of 50% reduction in luminosity (Asseng et al., 2017). However, other authors have observed increase in grain yield in low-intensity shading, due to the delay in wheat leaf senescence (Xu et al., 2016; Arenas-Corraliza et al., 2019) and increase in leaf area and growth index the plants (Li et al., 2010; Arenas-Corraliza et al., 2019).

However, there are no studies that assess the physiological quality of cereal seeds produced in shade conditions. In canola, a smaller number of grains per plant being compensated in greater weight per grain under shading conditions (Labra et al., 2017). Regarding seed quality, Bellaloui et al. (2012) found an increase in germination in most tested soybean cultivars, with a 50% limitation in the reduction of light. However, under conditions of restricted lighting, in the aforementioned surveys, a reduction in grain yield was observed. Therefore, the definition of the sowing schedule can be an escape from the occurrence of light restriction, especially in critical stages (between anthesis and seed filling), both for grain yield and the seed quality to be produced.

Studies related to the characterization of the seed quality produced under conditions of low level of light incident on crops during their development are scarce. Thus, based on the hypothesis that longer periods of shading during the development of wheat plants cause a reduction in the physiological seed quality, the objective was to evaluate the physiological quality of wheat seeds produced by plants growing in 70% light reduction (continuous) at different phenological stages.

Materials and Methods

The study was carried out in a greenhouse during 2018, at the Center for Agroveterinary Sciences, of the Santa Catarina State University (CAV/UDESC) in the municipality of Lages, SC, Brazil, located under geographical coordinates 27° 52' South Latitude, 50° 18' West Longitude and average altitude of 930 m.

Sowing was carried out on April 16, 2018 corresponding to Julian Day (JD) number 106. A photoperiod suitable for the species was used, with constant complementary artificial lighting during the crop cycle, with 10 hours of darkness and 14 hours of light. 10 seeds were sown per pot, followed by thinning, maintaining only 5 plants in each pot. For seed treatment, a commercial product based on pyraclostrobin + thiophanate- methyl + fipronil (Standak Top, 5 g a.i. ha^{-1} + 45 g a.i. ha^{-1} + 50 g a.i. ha^{-1}) was used. Daily irrigation was performed manually, according to the requirement of the plants. The temperature and relative humidity (RH) records, made by the Datalogger (TENMARS, TM305U) of the greenhouse, during the crop growing season, are shown in Figure 1.

Based on the interpretation of the results of the soil analysis, liming and fertilization was carried out, according to recommendations of the Soil Chemistry and Fertility Commission (CQFS RS/SC, 2016), corresponding to 6.8 tons per hectare of limestone, and the fertilization with 20.74 kg ha^{-1} of K_2O and 31.32 kg ha^{-1} of P_2O_5 and 60 kg ha^{-1} of nitrogen (urea), being divided into base and cover applications. The cover fertilizations were carried out at stage 20 (beginning of tillering - JD117), according to the scale of Zadoks et al. (1974), and at stage 30 (start of elongation of the stem-JD 131), using 0.5 g of urea and 0.5 g of KH_2PO_4 (Anhydrous Potassium Monobasic Phosphate) per pot. For phytosanitary treatments,

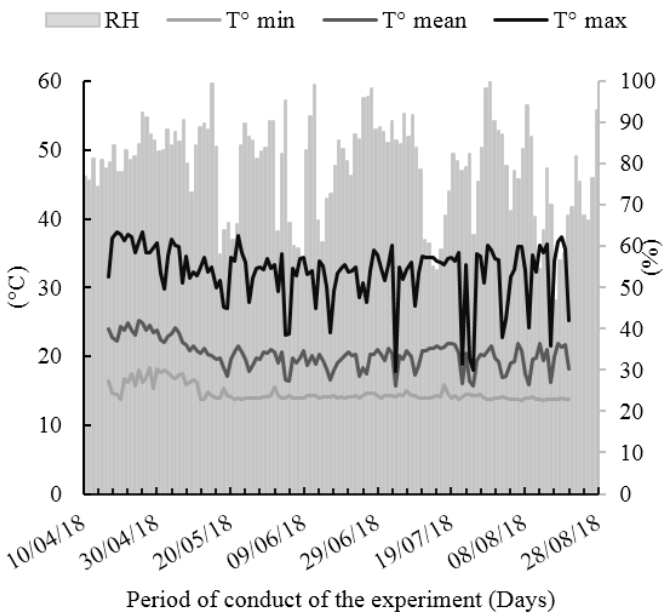

Figure 1. Averages of relative humidity (RH) and temperatures (minimum - T° min, mean - T° mean and maximum -T° max) recorded in the greenhouse during the experimental wheat cropping, during the 2018 season.

applications of trifloxystrobin + tebuconazole (Native 60 g a.i. ha^{-1} + 120 g a.i. ha^{-1}) and thiamethoxam + lambda-cyhalothrin (Engeo Pleno, 5.64 g a.i ha^{-1} + 4.24 g a.i. ha^{-1}) on May 25 (JD 145); metconazole (Caramba, 72 g a.i. ha^{-1}) on June 28 (JD 179); and trifloxystrobin + cyproconazole (Sphere Max, 92.75 g a.i. ha^{-1} + 40 g a.i. ha^{-1}) on July 30 (JD 211), for powdery mildew control (*Blumeria graminis* f. sp. *tritici*).

Artificial shading was imposed on plants with the use of black colored shade net with 70% fiber density, allowing only 30% of the available solar radiation to be exceeded (conferred via PAR- Photosynthetically Active Radiation and PPFD-photosynthetic photon flux density). The shade net was installed on a 1.50 m high wooden structure, necessary to house the pots containing the plants on all net sides.

Harvesting was carried out when the seeds were in the harvest maturity, on August 20, 2018, corresponding to JD 232. After the manual threshing of the ears, the seeds were dried in a forced air convection oven at 35 °C until their mass stabilized, standardizing humidity close to 13%. After that, the seeds of the plants in the pots of each treatment were homogenized, forming the respective seed lots as the average sample, and subsequently separated into 4 repetitions to compose the working samples.

The experiment was conducted in a completely randomized design with 11 treatments and 20 replicates. The treatments consisted of 11 seed lots. Ten seed lots originated from plants that artificial shading condition of 70% of the light was imposed, at different phenological stages, according to the scale of Zadoks et al. (1974), being: 23 (tillering - 3rd week after emergency - WAE); 30 (start of elongation of the culm - 4th WAE); 35 (fifth visible node - 5th WAE); 45 (full rubber - 6th WAE); 55 (half of the emerged inflorescences - 7th WAE); 60 (beginning of anthesis- 8th WAE); 65 (half of complete anthesis - 9th WAE); 70 (beginning of the 10th SAE grain development); 75 (milky grain - 11th WAE); 80 (beginning of the development of grain in mass - 12th WAE). A control treatment was also established with plants that shading was not imposed (Figure 2). Each experimental plot was composed of a unique pot containing five wheat plants of the cultivar Esporão, category C1, cycle of 128 days, average height of 90 cm, moderately resistant to lodging and moderately susceptible to powdery mildew.

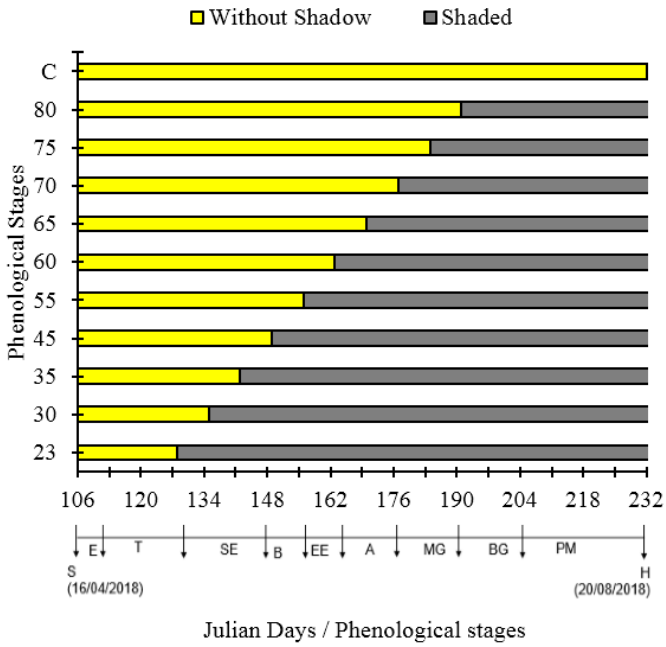

Figure 2. Representation of the phenological stages of beginning of the condition of light restriction imposed on wheat plants to obtain seed lots. Sowing (S); Emergency (E); Tillering (T); Stem elongation (SE); Booting (B); Emergency of the Ear (EE); Anthesis (A); Milky Grain (MG); Bulk grain (BG); Physiological Maturity (PM); Harvest (H). C: Control Treatment.

Evaluated characteristics

The following analyzes were proceeded:

- Thousand Seeds Weight (TSW): weighing eight repetitions of 100 seeds. Each replicate was weighed and multiplied by 10 (Brasil, 2009);
- Germination Test (G): eight replicates of 50 seeds for each treatment, distributed between sheets of specific paper (Germitest®) previously moistened with deionized water using an amount equivalent to 2.5 times its weight; left in the germinator for eight days at a temperature of 20 °C, evaluating the percentage of normal seedlings (G) abnormal (Ab) and dead seeds (DS) at the end of the period (Brasil, 2009). For abnormal seedlings and dead seeds, data were transformed into $((x + 1,5)/100)^{0,5}$;
- First Germination Count (FGC): percentage of normal seedlings at four days after the start of the germination test (Krzyzanowski et al., 1999);
- Germination Speed Index (GSI): daily count (24 h intervals) of normal seedlings germinated over the duration of this germination (Maguire, 1962);
- Accelerated Aging (AA): 400 seeds per treatment, kept at 42 °C, under 100% humidity for 60 h (Krzyzanowski et al., 1999), and submitted to the germination, with evaluation at eight days;
- Cold Test (CT): eight replicates of 50 seeds, maintained at a temperature of 10 °C during seven days (Krzyzanowski et al., 1999), following for germination, and evaluated a eight days;
- Electrical Conductivity (EC): four replicates of 50 seeds per treatment, added to 75 mL of deionized water, for 24 h at a temperature of 25 °C, and measurement of the EC with a bench conductivity meter (Quimis, Model 0795A2). The results in expressed as $\mu S\ g^{-1}\ cm^{-1}$ (Krzyzanowski et al., 1999);
- Tetrazolium: four replicates of 50 seeds placed between three sheets of Germitest® paper, moistened with 2.5 times their weight, and kept for 18 h at a temperature of 20 °C, to perform the pre-wetting. Afterwards, the seeds were cut, through a longitudinal bisection along the embryo and in ¾ of the endosperm; soaked in a solution of 0.5% chloride of 2,3,5 triphenyl tetrazolium during 3 hours, and then classified as viable and non-

viable, according to the color of the embryo (Brasil, 2009). The results were expressed as a percentage of viable seeds (Brasil, 2009);

- Shoot Length (SLS); Seedling main Root Length (SRL); and Total Seedling Length (TSL): on the fifth day of the germination test (Brasil, 2009), the aerial part (shoot), main root of 20 wheat seedlings, was measured, with eight replicates per treatment (Krzyzanowski et al., 1999);
- Seedling dry weight (SDW): drying in a forced air oven at a temperature of 65 °C until its mass stabilizes (Krzyzanowski et al., 1999).

Statistical analysis

Data were firstly subjected to normality and homogeneity tests, and, subsequently, to the analysis of variance by the F test ($p < 0.05$), with the percentage transformed into a arcsin of $(x/100)^{0.5}$. If there was a significant difference, the averages of the lots were compared by the Skott-Knott test ($p<0.05$), using the statistical software Sisvar (Ferreira, 2011).

Results

No statistically significant difference was observed ($p> 0.05$) for germination (G), percentage of abnormal seedlings (AB), dead seeds (DS) and viability by the tetrazolium test for seed lots from wheat plants grown under shading started at different phenological stages (Figure 1 and Table 1).

The thousand seeds weight (TSW) of the seed lots from shaded plants showed lower means than the control (38.7 g), except for the seed lot obtained from shading from stage 35 (Zadoks et al., 1974) (Table 1). The seed lots corresponding to the stages of onset of shading in 23, 30 and 35 showed an increase of 34.5, 54.9 and 74.3%, respectively, when compared to those produced by shaded plants from stage 60 (anthesis) with 21.3 g. A more intense negative effect was observed in the TSW of seeds produced by shaded plants between phenological stages 45 and 65 (booting and half of complete anthesis), whose reduction varied from 53.4 to 81.5%, compared to the control (38.7 g).

For the assessment of vigor by the first germination count test (FGC), significant higher values were observed for seed lots from wheat plants imposed on shading in phenological stages 23, 30 and 35 (tillering at fifth visible node), and 80 (grain by mass), with percentages of germination between 94 and 96%. These lots showed greater vigor than the control (without shading) by the test, whose percentage of normal seedlings in the FGC was by 81%. As for the batches obtained from shading starting at stages 45 to 75 (booting with milky grain), there was no difference with the control.

Germination speed index (GSI) for seed lots obtained from plants with shading starting at stages 23 (tillering), 30 and 35 (stem elongation) and 80 (grain mass) showed higher results than the control (11.8), with an average increase of 4.4% in the index. The seeds of the lots corresponding to the shading period started between stages 45 (rubber) and 75 (milky grain) did not differ from the control.

Electrical conductivity (EC) showed significant higher values than the control for all the lots of wheat plant seeds submitted to shading in different phenological stages, characterizing their lower vigor (Table 1). The seed lot obtained from the shaded plants from stage 55 (half of the emerged inflorescence) showed a higher EC value (60.3 µS g^{-1} cm^{-1}), representing an increase of 84.5% in relation to the seeds produced by wheat plants without shading (control) whose EC value was 32.7 µS g^{-1} cm^{-1}.

Seed vigor verified by the AA test showed the lowest percentages, varying from 93 to 98%, for the plant lots growing in the shading condition from stage 45 (booting), 60 and 65 (anthesis), respectively (Table 2). In the cold test (CT) a reduction in seed vigor was observed only for the lot of seeds obtained from wheat plants produced under the condition of

Table 1. Thousand seed weight (TSW), first germination count (FGC), germination (G), abnormal seedlings (AB), dead seeds (DS), viability by tetrazolium (TZ), germination speed index (GSI) and electrical conductivity (EC) of wheat seeds produced under different periods of shading.

Phenological stages	TSW (g)	FGC	G	AB (%)	DS	TZ	GSI	EC (µS g^{-1} cm^{-1})
23	28.71 c	96 a	99 ns	1 ns	0 ns	99 ns	12.34 a	50.3 b
30	33.07 b	96 a	100	0	0	100	12.33 a	42.4 c
35	37.21 a	94 a	100	0	0	99	12.31 a	50.7 b
45	25.26 d	88 b	99	1	0	99	12.03 b	54.4 b
55	23.88 d	88 b	98	1	1	96	12.05 b	60.3 a
60	21.35 d	85 b	98	1	1	99	11.92 b	44.2 c
65	23.24 d	86 b	99	1	0	99	11.96 b	53.5 b
70	27.52 c	90 b	99	1	0	99	12.09 b	44.4 c
75	28.42 c	90 b	99	1	0	99	12.03 b	45.5 c
80	32.49 b	96 a	99	1	0	99	12.33 a	43.8 c
C	38.75 a	81 b	99	1	0	99	11.81 b	32.7 d
LSD	5.24	8.5	2.3	1.9	1.2	2.8	0.4	10.1
CV (%)	12.4	7.9	5.5	29.1	20.2	5.2	2.3	10.4
Mean	29.1	90	99	1	1	99	12.1	89.8

* Same letters in the column do not differ from each other by the Skott-Knott test at $p<0.05$. ns: not significant ($p>0.05$). C: Control Treatment; LSD: least significant difference; CV (%): Coefficient of variation.

Table 2. Accelerated aging (EA), cold test (CT), main root length (RL), aerial part (SL – shoot), total (TSL) and seedling dry weight (SDW) of wheat seeds produced under different periods of shading.

Phenological stages	AA	CT	RL	SL	TSL	SDW
	(%)			(cm)		(mg pl^{-1})
23	99 a	100 a	19.06 b	15.4 b	34.47 b	20.88 c
30	100 a	100 a	21.03 a	14.83 b	35.86 a	25.99 b
35	99 a	100 a	20.80 a	14.81 b	35.62 a	24.46 b
45	99 a	99 a	20.32 a	14.68 b	35.00 b	20.51 c
55	98 b	100 a	20.33 a	14.47 b	34.81 b	19.56 c
60	93 c	96 b	18.72 b	14.46 b	33.18 b	17.39 c
65	95 b	100 a	19.44 b	16.65 a	36.09 a	18.83 c
70	98 a	99 a	20.23 a	15.35 b	35.58 a	20.56 c
75	99 a	100 a	20.64 a	15.03 b	35.68 a	22.60 c
80	99 a	99 a	21.11 a	15.21 b	36.32 a	26.50 b
C	100 a	100 a	20.33 a	14.55 b	34.88 b	32.14 a
LSD	3.1	2.1	1.4	1.7	2.3	4.7
CV (%)	5.9	4.4	5	8.2	4.7	14.8
Mean	98	99	20.2	15	35.2	0.4

* Same letters in the column do not differ from each other by the Skott-Knott test at $p<0.05$. ns: not significant ($p>0.05$). C: Control Treatment; LSD: least significant difference; CV (%): Coefficient of variation.

light restriction started at stage 60 (beginning of anthesis), with vigor of 96% in the final evaluation (Table 2).

Length of the main root (RL) of the wheat seedlings decreased by 6.2, 7.9 and 4.4% in the seed lots whose plants were subject to the shading condition at stages 23, 60 and 65, respectively, when compared to those obtained from non-shaded plants, with 20.3 cm (Table 2). In relation to the length of the aerial part (shoot) of seedlings (SL), an increase of 14.5% was observed for the seed lots of wheat plants shaded in stage 65 (half of the complete anthesis), compared to the control, whose SLS was 14.5 cm. Higher values were observed for the seminal roots, whose structure is the first to be emitted during the germination process, in relation to the aerial part (shoot).

Total length of wheat seedlings (TSL) of seed lots obtained from plants growing under shading in stages 30 (stem lengthening), 65 (half of complete anthesis), 70 (beginning of grain development), 75 (milky grain) and 80 (grain by mass) showed an increase by 2 to 4.1% compared to the control. The seedlings of wheat seeds from plants grown without shading condition had a TSL of 34.9 cm.

The SDW (seedling dry weight) was significantly reduced by 84.8% for seed lots from wheat plants submitted to shading condition, regardless of the phenological stage at which it was started, compared to the control (32.14 mg). The greatest negative effects for wheat SDW were observed when light restriction was started between stages 45 to 75, corresponding to booting and milky grain.

Discussion

The effect observed in TSW when limiting light to plants in the early stages of wheat crop development may be due to the effect of plant compensation, since the reduction in the number of seeds (data not shown) may have resulted in seeds with higher weight. The fact that the accumulation of biomass in the plant decreases, due to the lower intercepted photosynthetically active radiation, subsequently impacts the grain yield due to the lower remobilization of photoassimilates accumulated in the vegetative parts of the plant (Asseng et al., 2017).

Similar results were obtained for canola crop under 70% limitation of light during anthesis, whose variation in the increase in TSW was from 47 to 61%, due to the higher rate and period of seed filling (Labra et al., 2017). The light restriction on plants during the anthesis period corroborates the results of Asseng et al. (2017) whose reduction in the number of wheat grains per area was 17 and 28%, for shadings of 50 and 90%, respectively, during the stages of booting up to anthesis. Therefore, the effect of light restriction in the periods close to flowering probably occurred due to the reduction of the photosynthetic rate of shaded plants (Guo et al., 2014), especially of the flag leaf, whose photoassimilates have a high relationship with the filling of wheat seeds (Guo et al., 2014).

The results observed for the FGC and GSI characterize the reduction of seed vigor, since lots with less vigor have lower germination speed and percentage of normal seedlings in the first count (Krzyzanowski et al., 1999) (Table 1). In a study with the herbicide metsulfuron-methyl on wheat plants in the tillering stages and 75% of the visible ears, Albrecht et al. (2010) observed that the vigor of produced seeds, evaluated by the FGC, varied between 84 and 94% of germination in the two growing seasons, lower percentage when applied in the reproductive stages. In relation to the GSI, Abati et al. (2014) found similar results, with an average of 12.7 with linear reduction when submitting seeds to higher levels of osmotic potential.

The EC assessment is efficient in classifying seed lots according to vigor (Sponchiado et al., 2014). The highest values of EC were observed for the seed lots with low vigor. Because they have less integrity of the integument membranes, less capacity for structuring after soaking in

water and consequently greater release of exudates to the outside of the cells (Marcos Filho, 2015) (Table 2).

The EC values of wheat seeds from shaded plants were higher than that observed by Barbieri et al. (2013) that observed a variation of the EC of the seeds of four cultivars, with values between 15 and 30 µS g^{-1} cm^{-1}, when evaluating the effect of population reduction of cultivars. In comparison to those observed in this research, seed lot control with 32.7 µS g^{-1} cm^{-1} and plant seed lot shaded from the phenological stage 55 with 60.3 µS g^{-1} cm^{-1} (Table 2).

The AA evaluations allowed to separate high and low vigor seed lots, in which deterioration of low vigor seeds is faster when subjected to the unfavorable condition of high temperature and relative humidity. However, the less vigorous lots reached a minimum of 93% in the AA. More intense reductions in seed vigor by the AA test were observed for wheat cultivars BRS Umbu and BRS Tarumã when submitted to defoliation, in which the minimum percentage in AA was 46% and 49%, respectively (Rodolfo et al., 2017), due to greater photosynthetic limitation.

In the CT, seeds with low vigor show greater loss of leachate during the reorganization of the membranes, due to their slower repair, when submitting in conditions of low temperature and high humidity (Marcos Filho, 2015). However, Brunes et al. (2015) did not observe a significant effect when studying the quality of the seeds with the application of Boron in wheat plants, although the results obtained were maintained above 90% of vigor by the test.

Observations in the batches that originated seedlings with reduced initial performance are related to the lower capacity to transform the reserve tissues and incorporate it into the embryonic axis (Bewley et al., 2013). According to Marcos Filho (2015), the evident demonstration of the decline in physiological potential is the reduction in the speed of germination of viable seeds, followed by the shorter length of seedlings and the occurrence of abnormality in germination.

The sensitivity of the reproductive stages of the wheat was observed due some treatments, which reflected in the final quality of the seeds produced. Likewise, different treatments for assessing vigor by seedling development were observed by Olivoto et al. (2017), with splice different nitrogen sources applied on wheat plants topdressing. These authors observed that the application of nitrogen in the most advanced stages results in greater accumulation of proteins in the seeds and, consequently, greater vigor of its seeds. As observed with the cultivar Quartz, in which the average for SLS was 8.9 cm with application of ammonium nitrate in the tillering, tillering and booting and, tillering, booting and flowering; for SRL, when applied to the taper, 10.8 cm; and SDW with urea splitting in tillering and rubber and, tillering and flowering, with 12.6 and 12.1 mg per seedling.

Considering the seed formation, vigor tests are carried out in batches to verify the intensity of the factors that reduce their physiological quality, in advance of the reduction of the germinative potential (Krzyzanowski et al., 1999). It is because seed lots may differ in vigor, even not differing in germination potential (Carvalho & Nakagawa, 2012). Bellaloui et al. (2012) observed a reduction in the percentage of germination, depending on the genotypes, due to the lower temperature of the environment, when evaluating soybean seeds produced under a 50% limitation of light. In addition, there was a reduction in protein content, due supposedly to lower photosynthetic rate, assimilation of carbon and nitrogen.

Based on the relationship among the Zadoks phenological codes and the evaluated seed traits (Table 3), a positive relationship of the Zadoks code was observed only with AB. These results pointing to the light restriction imposed on plants during seed formation, lower quality seeds are produced on basis in abnormal seedlings (AB) originated during germination process. Among the all seed quality traits evaluated, a positive association of TSW x G; TSW x AA; TSW x RL; TSW x SDW; FGC x GSI; G x (TZ, GSI, AA, RL and TSL); AA x (CT, RL and SDW); CT x TSL; RL x TSL and RL and SDW. The negative relationship were observed of TSW x DS, TSW x EC; G x AB; G x DS; AB x GSI; DS x TZ; DS x AA; DS x CT; DS x TSL; TZ x EC and EC x SDW.

Table 3. Correlation of Pearson among Zadoks phenological stages (Zad), TSW, FGC, G, AB, DS, TZ, GSI, EC, AA, CT, RL, SL, TSL and SDW from wheat seeds produced under different periods of shading.

	TSW	FGC	G	AB	DS	TZ	GSI	EC	AA	CT	RL	SL	TSL	SDW
n→	40	40	40	40	40	40	40	40	40	40	80	80	80	80
Zad	-0.31	-0.40	-0.42	0.57	0.10	-0.15	-0.49	-0.20	-0.32	-0.26	0.13	0.21	0.26	-0.10
TSW	-	0.24	0.69	-0.52	-0.56	0.35	0.31	-0.59	0.73	0.48	0.61	-0.22	0.38	0.92
FGC		-	0.53	-0.49	-0.34	0.24	0.98	0.17	0.41	0.26	0.37	0.12	0.41	0.03
G			-	-0.78	-0.78	0.64	0.56	-0.24	0.58	0.52	0.56	0.18	0.62	0.50
AB				-	0.22	-0.34	-0.55	0.06	-0.32	-0.26	-0.46	0.17	-0.29	-0.29
DS					-	-0.66	-0.33	0.32	-0.59	-0.56	-0.41	-0.45	-0.68	-0.49
TZ						-	0.22	-0.60	0.10	-0.11	0.05	0.24	0.22	0.30
GSI							-	0.17	0.42	0.28	0.37	0.09	0.39	0.08
EC								-	-0.21	0.14	-0.15	0.19	0.00	-0.71
AA									-	0.71	0.74	-0.25	0.48	0.70
CT										-	0.50	0.29	0.65	0.42
RL											-	-0.22	0.72	0.61
SL												-	0.51	-0.26
TSL													-	0.36

* The values in red color are negatively correlated and in blue color are positively correlated, by T test ($t<0.05$).

Taken together, the results indicate sensitivity in the reproductive stage of wheat plants to define seed vigor due light restriction. Similar to results obtained by Rodolfo et al. (2017), who found no effects on the germination of the seeds produced, but there was a reduction in vigor, probably due to the lower content of carbohydrates (starch) in the seeds, by limiting the source-sink relationship of wheat plants, manipulating the source factor through defoliation between the tillering and booting stages. Thus, in the stages that precede the anthesis, the formation of floral structures occurs (Marcos Filho, 2015), later double fertilization, that is, the union of the nuclei spermatic (n) with the oosphere (n) and the polar nuclei (2n), to then start cell division, growth of the zygote (2n) and the reserve tissue, the endosperm (3n) (Berger, 2003).

The endosperm is the tissue that supports the development and germination of the embryo originating from the fertilization of one of the male gametes that arrive through the pollen tube and the central cell of the embryonic sac (Berger, 2003). It develops in the central region of the embryonic sac through intense cell divisions (Bewley et al., 2013). The efficient accumulation of carbohydrates in the seed is necessary for the proper development of the embryo, providing chemical energy until the establishment of the autotrophic seedling (Goffman et al., 2005). In this way, the photosynthetic activity of the flag leaf must be sufficient to supply photosynthesis for the formation and filling of seeds, even under reduced light, and then, there is no damage in the formation of this seed (Xu et al., 2016), because more than 60% of the photosynthesis product (glucose) destined for a seed is produced by the flag leaf (Fang et al., 2006).

Conclusions

The continuous shading in wheat plants does not interfere with the germination of the seeds produced, but there is a negative effect on the seed vigor and the thousand seeds weight, depending on the phenological stage that light restriction starts.

In general, under shading the physiological quality of the seeds produced is high, except, when the shading starts between the rubber and the anthesis (stages 45 to 65), wheat plants are sensitive to light restriction. This can be useful in collaboration to growers for definition of sowing schedule, as an escape for critical stages.

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Compliance with Ethical Standards

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Role of metabolization and conjugation of herbicides in tolerance of white oats to 4-Hydroxyphenylpyruvate Dioxygenase (HPPD) enzyme inhibitors

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Abstract

The success of chemical control is dependent on crop tolerance to herbicides and their efficiency in controlling weeds. Thus, knowledge of the tolerance level of cultivated plants and the tolerance mechanism involved is crucial for safe herbicide use. The objective of this study was to evaluate the effect of metabolization (malathion) and conjugation (NBD-Cl) inhibitors on the tolerance of white oats to the herbicides mesotrione and tembotrione. The experiment was carried out under field conditions in the 2021 crop, in an RBD, with nine treatments, namely: T0- control (no application); T1- malathion; T2- NBD-Cl; T3- mesotrione; T4- mesotrione+malathion; T5- mesotrione+NBD-Cl; T6- tembotrione; T7- tembotrione+malathion and T8- tembotrione+NBD-Cl. The plant characteristics were evaluated for plant intoxication, SPAD index, levels of chlorophyll *a, b* and total carotenoids, yield components, yield and industrial grain quality. Symptoms of intoxication in oat plants started at 7 DAA and increased up to 14 DAA in treatments containing mesotrione or tembotrione. Plant recovery started at 21 DAA, and it was intensified at 28 DAA. Intoxication levels were not affected by the addition of the metabolization inhibitors malathion (43.1%) and NBD-Cl (36.2%). There was no change in other variables tested under the control treatment. Based on these findings, it can be concluded that there was no change in the level of tolerance of white oats to the herbicides mesotrione or tembotrione after application of malathion and NBD-Cl. This outcome suggests that metabolization or conjugation is not the main mechanisms that make white oat tolerant to HPPD enzyme inhibitors.

Keywords: *Avena sativa*; differential metabolism; GSTI; herbicide detoxification; malathion; NBD-Cl; P450.
Abbreviations: AGROFIT_Informações sobre agrotóxicos fitossanitários registrados no Ministério da Agricultura, Pecuária e Abastecimento; ALS_acetolactate synthase enzyme; AVENACOR_industrial grain yield; C*a*_chlorophyll *a;* C*b*_chlorophyll *b*; CFB_Clima temperado com verão ameno e falta de estação seca; CONAB_Companhia Nacional de Abastecimento; CQFS_Comissão de Química e Fertilidade do Solo; CT_total carotenoids; DAA_days after application; DHI_dehulling index; G>2MM_grains>2mm; GSTI_glutathione S-transferase; HI_harvest index; HPPD_hydroxyphenyl-pyruvate dioxygenase enzyme; HW_hectoliter weight; IBGE_Instituto Brasileiro de geografia e estatísticas; MDA_malonaldehyde; NBD-Cl_4-chloro-7-nitrobenzofurazan; NGS_number of grains per spikelet; NPM_number of panicles per m²; NSP_number of spikelets per panicle; RDB_randomized block design; SPAD_Soil Plant Analysis Development; TGW_thousand-grain weight; UPFA_Universidade de Passo Fundo; U%_grain harvest moisture.

Introduction

White oat (*Avena sativa*) is widely used in crop rotation, mulching, pasture, silage, hay and grain production systems for animal (Pacheco et al., 2021) and human feed (Kim et al., 2021). In Brazil, white oat is grown during the winter, mostly in the states of Rio Grande do Sul, Paraná and Mato Grosso do Sul, with an expected production of 1.15 million tons for the 2022 harvest (Conab, 2022).

Historically, there was a significant increase in oat-growing areas in Brazil, from 40.3 thousand hectares year^{-1} in the 1970s to 503.4 thousand hectares year^{-1} in the 2022 harvest, i.e., the harvested area increased by 12.5 times (IBGE, 2022). Yield in the same period increased from 1,000 kg ha^{-1} to 2,281 kg ha^{-1} (Conab, 2022). Despite the increased production of oat grains, there are factors that pose limitations to crop production, for example, weed interference, which reduces the yield and quality of crops of agricultural interest (Kadam et al., 2021).

Integrated weed management is used in oat cultivation to reduce the negative effects of interference, but it needs to be complemented with chemical control, an important method in modern agriculture (Fu et al., 2019). However, there is a lack of selective herbicides available for post-emergence use in oat crops; therefore, control is limited to two mechanisms of action: auxin-mimicking and acetolactate synthase (ALS)-inhibiting herbicides (Agrofit, 2022), which have specific control for Magnoliopsida (Dicots) weeds.

However, recent studies have recommended herbicides that inhibit the biosynthesis of carotenoids and plastoquinones, particularly the inhibitors of 4-hydroxyphenyl-pyruvatedioxygenase (HPPD), because they are selective for winter crops and efficient in controlling weeds, and they offer low toxicity to the environment (Ahrens et al., 2013; Lindell et al. 2015; Ndikuryayo et al., 2017; Schmitz et al., 2015). Also, they have been shown to be highly selective for oat crops (Buba et al., 2021; Soltani et al., 2011; Pedroso et al., 2021).

HPPD-inhibiting herbicides include the herbicides mesotrione and tembotrione, which are widely used in maize as they have a broad spectrum of weed control, including Poeceae (Williams et al., 2010). These herbicides inhibit the HPPD enzyme, which converts 4-hydroxyphenyl pyruvate into homogentisate - a precursor in the biosynthesis of plastoquinone and tocopherol in plants (Lee et al., 1998). Plastoquinone is required for the biosynthesis of carotenoids, which protect chlorophyll by absorbing excited electrons released during photosynthesis. Depletion of carotenoids leads to photo-oxidation of chlorophyll, which results in in bleaching, followed by necrosis and plant death (Dankov et al., 2009).

The selectivity of HPPD-inhibiting herbicides is mostly due to differential metabolism in tolerant plants, such as corn (Matte et al., 2018; Lu et al., 2020), and resistant biotypes of wild radish (*Raphanus raphanistrum*) (Lu et al., 2020) and grain sorghum (*Bicolor sorghum*) (Pandian et al., 2020). This could be the mechanism responsible for the selectivity of these herbicides to white oat.

Differential metabolism in herbicide-tolerant plants, also called detoxification or herbicide metabolism is the main mechanism responsible for the selectivity of herbicides used at post-emergence in crops of agricultural interest (Nandula, 2017). Differential metabolism is characterized by the ability of plants to rapidly metabolize the phytotoxic chemical (herbicide) into a less toxic or non-toxic product (Edwards et al., 2000; Hatzios et al., 2004).

According to Jablonkai (2015), herbicide detoxification can be divided into three phases in plants, namely: Phase I: also called metabolization, in which primary metabolism converts biologically active molecules into less active compounds through hydroxylation performed by Cytochrome P450 monooxygenases; Phase II: the products resulting from phase I through oxidation, reduction and hydrolysis, are converted into less toxic molecules; the best known process is conjugation to glutathione, carried out by the enzyme glutathione S-transferase - GSTI or glucosyl transferase - GT (Cobb et al., 2011); Phase III: Phase II conjugates are transformed into non-toxic secondary conjugates or linked to insoluble residues. Phase IV: Phase III products can be compartmentalized in the cell (compartmentalization).

Differential metabolism in herbicide-tolerant or resistant plants can be investigated by using malathion and/or 4-chloro-7-nitrobenzofurazan (NBD-Cl). These products are used for inhibiting the cytochrome P450 enzymes and the activity of the glutathione-S-transferase enzyme, respectively; they inhibit phases I and II of the detoxification process, reversing maize tolerance to HPPD inhibitors (Ma et al., 2012; Oliveira et al., 2018), wild radish (Lu et al., 2020) and grain sorghum (Pandian et al., 2020).

Thus, the objective of this study was to evaluate the effect of metabolization (malathion) and conjugation (NBD-Cl) inhibitors on the tolerance of white oat to HPPD-inhibiting herbicides.

Results

Oat intoxication

There was a significant treatment effect for the variable intoxication of white oat plants at 7, 14, 21 and 28 DAA (Table 1). The first symptoms of plant intoxication were found at 7 DAA in treatments containing the herbicides mesotrione or tembotrione. The control, malathion and NBD-Cl treatments showed no injuries to the oat plants (Table 1). Treatments containing mesotrione (mesotrione, mesotrione+ malathion and mesotrione+NBD-Cl) showed the highest levels of intoxication with 33.1, 38.8 and 36.2% respectively, compared to treatments with tembotrione (tembotrione, tembotrione+malathion and tembotrione+NBD-Cl), which showed only 5.8, 6.0 and 6.4% of intoxication (Table 1).

At 14 DAA, intoxication levels were higher, except for the control, malathion and NBD-Cl treatments, which did not show symptoms of intoxication (Table 1). The highest levels of intoxication were found in treatments containing the herbicide mesotrione (mesotrione at 63.1%, mesotrione+malathion at 68.1% and mesotrione+NBD-Cl at 65%) compared to those containing the herbicide tembotrione (tembotrione at 6.8%, tembotrione+malathion at 7.3% and tembotrione+NBD-Cl at 6.6%) (Table 1).

At 21 DAA, there was a reduction in the levels of intoxication of the treatments compared to the previous period, indicating the recovery of injuries by the oat plants. The highest levels of intoxication were found in plants treated with mesotrione+malathion (43.1%) and mesotrione+NBD-Cl (36.2%), followed by mesotrione at 28.1%, and later with treatments containing the herbicide tembotrione (tembotrione+ malathion at 2.6%, tembotrione at 2.4% and tembotri-

Table 1. Intoxication of white oat plants (%) at 7, 14, 21 and 28 days after application (DAA) of herbicides applied alone or previously treated with a metabolization or conjugation inhibitor. 2021/2021 season, Lages – SC.

Treatment	7 DAA		14 DAA		21 DAA		28 DAA	
	Intoxication (%)							
Control	0.0	c	0.0	c	0.0	d	0.0	b
Malathion	0.0	c	0.0	c	0.0	d	0.0	b
NBD-Cl	0.0	c	0.0	c	0.0	d	0.0	b
Mesotrione	33.1	a	63.1	a	28.1	b	6.5	a
Mesotrione + Malathion	38.8	a	68.7	a	43.1	a	10.4	a
Mesotrione+NBD-Cl	36.2	a	65.0	a	36.2	a	8.2	a
Tembotrione	5.8	b	6.8	b	2.4	c	0.4	b
Tembotrione+Malathion	6.0	b	7.3	b	2.6	c	0.5	b
Tembotrione+NBD-Cl	6.4	b	6.6	b	1.5	c	0.3	b
p-value	<0.001*		<0.001*		<0.001*		<0.001*	
C.V. (%)	11.7		7.8		12.3		27.5	

*Significant at p<0.05 by the F-test; ^{ns} non-significant by the F-test; identical letters in the column do not differ from each other at p<0.05 by Tukey's test; CV: coefficient of variation.

one+NBD-Cl at 1.5%), which did not differ from each other (Table 1).

At 28 DAA, there was a reduction in the levels of plant intoxication in comparison to the other periods of assessment, indicating more intense plant recovery. The highest levels of intoxication were found in the treatments mesotrione+malathion at 10.4%, mesotrione+NBD-Cl at 8.2% and mesotrione at 6.5%, compared to the other treatments that did not differ from each other (Table 1) and resulted in nearly zero intoxication.

Photossyntetic pigments

There was a significant effect (p<0.05) of treatment for the SPAD index of white oat plants only at 7 and 14 DAA (Table 2). At 7 DAA, the highest SPAD index was measured in the following treatments: control (39.41), malathion (39.42) and NBD-Cl (39.65), while the lowest value was measured in the Mesotrione treatment (31.31) (Table 2). However, the other treatments showed no significant difference from the untreated control (Table 2).

At 14 DAA, treatments containing mesotrione (mesotrione with 12.78, mesotrione+malathion with 10.46 and mesotrione+NBD-Cl with 14.31) had the lowest SPAD index compared to the other treatments, which did not differ from each other (Table 2). At 21 and 28 DAA, there was no difference (p>0.05) between treatments (Table 2); they showed a mean SPAD index of 38.9 and 45.5, respectively.

For concentrations of chlorophyll *a* (C*a*), chlorophyll *b* (C*b*), total carotenoids (CT) and malonaldehyde (MDA) in white oat plants at 21 DAA, there was no significant difference (p<0.05) between treatments (Supplementary 1). The average treatment values corresponded to 106.4, 30.6 and 26.7 µg g FM^{-1} for C*a*, C*b* and CT respectively, and 22.9 nmol ml^{-1} of MDA.

Growth, yield and yield components

There was no significant effect (p>0.05) of treatment for the following variables: plant height, harvest index (HI), number of panicles per m² (NPM), number of spikelets per panicle (NSP) and number of grains per spikelet (NGS) (Supplementary 2). The average value of the treatments corresponded to 107.8 cm for plant height, 0.47 for HI, 522 NPM, 30.3 NSP and 1.1 NGS. There was also no significant effect (p>0.05) for grain harvest moisture (U%), thousand-grain weight (TGW) and grain yield, and the mean values were 11.7% of U%, 37 g for TGW and 5907 kg ha^{-1} for grain yield (Supplementary 3).

Grain quality

There was no significant effect (p>0.05) for the variables related to quality and industrial yield of white oat grains owing to the treatments tested (Supplementary 4). The average values of the treatments corresponded to: grains higher than 2 mm by sieve (G>2) = 90.5%; hectoliter weight (HW) = 50.5 kg hL^{-1}; dehulling index (DHI) = 75.4%; industrial grain yield (Avenacor) = 4034 kg ha^{-1} of caryopsis.

In summary, the findings of this study indicate a significant effect (p<0.05) for plant intoxication (Table 1) and SPAD index (Table 2) as a result of the application of the herbicides mesotrione and tembotrione, either alone or with previously treated with malathion or NBD-Cl. These results clearly show the appearance of symptoms in the plants and their subsequent recovery, allowing the white oat plants to complete their cycle without affecting grain yield and yield components, and without affecting grain quality. The herbicides mesotrione and tembotrione were selective to white oat crop.

Table 2. SPAD index of white oats at 7, 14, 21 and 28 days after application (DAA) of herbicides applied alone or previously treated with a metabolization or conjugation inhibitor. 2021/2021 season, Lages – SC.

Treatment	7 DAA		14 DAA		21 DAA	28 DAA
	SPAD Chlorophyll Index					
Control	39.41	a	37.29	a	37.25 ns	44.07 ns
Malathion	39.42	a	38.21	a	40.79	46.34
NBD-Cl	39.65	a	38.55	a	39.72	44.21
Mesotrione	31.31	b	12.78	b	36.85	46.29
Mesotrione + Malathion	36.16	ab	10.46	b	38.63	46.20
Mesotrione+NBD-Cl	33.80	ab	14.31	b	38.97	47.95
Tembotrione	37.38	ab	37.22	a	39.40	45.44
Tembotrione+Malathion	34.41	ab	36.92	a	41.02	44.63
Tembotrione+NBD-Cl	35.50	ab	35.12	a	37.32	44.53
p-value	0.003*		<0.001*		0.506^{ns}	0.279^{ns}
C.V. (%)	7.8		10.4		8.1	4.9

*Significant at p<0.05 by the F-test; ^{ns} non-significant by the F-test (p>0.05); Different letters in the column differ from each other at p<0.05 by Tukey's test; CV: coefficient of variation.

Discussions

Symptoms of intoxication started at 7 DAA, in treatments containing the herbicides mesotrione or tembotrione, alone or with previous treatment using malathion or NBD-Cl (Table 1). Visual symptoms were characterized by bleaching and/or purplish stains on tissues produced after application of the treatments. The symptoms are similar to those described by De David et al. (2017) for *Illex paraguariensis*, Pandian et al. (2020) for sorghum, Teixeira et al. (2017) for sweet sorghum and Wang et al. (2020) for wheat, sprayed with carotenoid biosynthesis inhibitor herbicides.

Tissue bleaching is due to the photo-oxidation of chlorophyll which results from the absence of carotenoids that protect chlorophyll from the excited electrons released during photosynthesis (Dankov et al., 2009; Wang et al., 2020). Carotenoid depletion is the result of applying the HPPD-inhibiting herbicides mesotrione and tembotrione. HPPD is responsible for converting 4-hydroxyphenyl pyruvate into homogentisate, i.e., the precursor of plastoquinone, which is required for carotenoid biosynthesis (Lee et al., 1998; Godar et al., 2015).

To assess the intoxication of oat plants at 7 DAA, the treatments were separated into three groups (Table 1) in terms of increasing levels of intoxication, namely: without herbicides (0%), treatments containing tembotrione (5.8 to 6.4%) and treatments containing mesotrione (33.1 to 38.8%). It was found that intoxication levels were not affected by the metabolization inhibitors tested, only by the herbicides. A similar behavior was found at 14 DAA, except for the increase in intoxication levels in treatments containing tembotrione (6.6 to 7.8%) and mesotrione (63.1 to 68.7%) in comparison to the previous assessment period (Table 1).

The externalization of symptoms after 7 DAA, together with increased levels of symptoms up to 14 DAA, is due to the mechanism of action of the herbicides mesotrione and tembotrione; they inhibit the synthesis of new carotenoids after application, but they do not affect the carotenoids formed prior to application (Dan Hess, 2000; Grossmann et al., 2007; Hawkes, 2007; De Paula et al., 2020). Thus, plants continue to grow, but new tissues are produced without the presence of carotenoids, resulting in increased levels of intoxication. A similar behavior was reported by De David et al. (2017) for *Illex paraguariensis* and, by Teixeira et al. (2017) for sweet sorghum under increasing rates of tembotrione.

At 21 DAA, there were reduced intoxication levels for treatments containing tembotrione (1.5 to 2.6%) and mesotrione (28.1 to 43.1%) in comparison to the previous evaluation period (Table 1). This finding suggests the onset of plant recovery, which is intensified at 28 DAA (Table 1). Plant recovery occurs as a result of the production of new tissues with the absence of visual symptoms of intoxication, leading to a reduction in intoxication levels. A similar behavior was found in wheat cultivars “Quartzo” and “Pioneiro” sprayed with 0.8 L ha^{-1} of clomazone; however, recovery started at 14 DAA, and it was intensified at 21 DAA (Galon et al., 2015).

Plant recovery resulting from the production of new tissues (green) without symptoms is a strong indication that the herbicides being used are not inhibiting the site of action; the route is reestablished and starts producing carotenoids again. The non-inhibition of the herbicidal site of action on the plant may be due to reduced translocation, and/or increased differential metabolism and/or

compartmentalization of the herbicide by the plant in vacuoles or cell walls (Délye et al., 2013). The mechanisms of detoxification by the action of cytochrome P450 are responsible for the selectivity of HPPD-inhibiting herbicides in crops of agricultural interest (Ahrens et al., 2013), e.g., corn (Lu et al., 2020). They also thought to be responsible for the resistance of wild radish (*Raphanus raphanistrum*) (Lu et al., 2020), grain sorghum (*Sorgum bicolor*) (Padian et al., 2020) and Palmer amaranth (*Amaranthus palmeri*) (Küpper et al., 2018) biotypes to HPPD-inhibiting herbicides; However, there is no information available on white oat crops.

To test differential metabolism in plants, experiments were conducted with the C^{14}-labeled herbicide molecule, which was applied to tolerant and susceptible plants. It was subsequently assessed and quantified for absorption, translocation and metabolism (Nandula et al., 2017; Shergill et al., 2018; Lu et al., 2020). As an alternative, differential metabolism in plants can be investigated by means of metabolization inhibitors, a method applied in this work. The application of malathion and/or 4-chloro-7-nitrobenzofurazan (NBD-Cl) inhibits the cytochrome P450 enzymes and the activity of the glutathione-S-transferase enzyme, respectively, inhibiting phases I and II of the metabolism process, thus reversing tolerance by plants (Shergill et al., 2018).

The application of malathion (43.1%) or NBD-Cl (36.2%) reduced the recovery speed of plants sprayed with mesotrione, compared to this herbicide alone (28.1%) at 21 DAA (Table 1), but the intoxications for the other treatments and evaluation periods did not change (Table 1). This result indicates a possible effect of detoxification on the tolerance of white oat to mesotrione. However, the same outcome did not occur for tembotrione. Unlike the findings in this work, the application of malathion or NBD-Cl reverses the tolerance of the crop to HPPD inhibitors in maize (Ma et al., 2012; Oliveira et al., 2018), wild radish (Lu et al., 2020), grain sorghum (Pandian et al., 2020) and Palmer amaranth (Shergill et al., 2018; Küpper et al., 2018).

The onset of intoxication symptoms at 7 DAA and their intensification at 14 DAA in treatments containing the herbicides mesotrione and tembotrione, followed by the recovery process at 21 and 28 DAA, can also be evidenced by the SPAD Index (Table 2). It is measured by the difference in light transmitted through a leaf using two detectors at wavelengths from 650 to 940 nm, generating a leaf greenness index (Blackmer et al., 1995). In other words, the SPAD index is positively correlated with leaf greenness and chlorophyll, which is degraded by the absence of carotenoids in the tissues sprayed with carotenoid biosynthesis inhibitor herbicides. This variable was also not affected by the application of malathion or NBD-Cl metabolizing inhibitors (Table 2).

The concentrations of chlorophyll *a* (C*a*), chlorophyll *b* (C*b*) and total carotenoids (TC) in white oat plants at 21 DAA, were not altered by the application of the herbicides mesotrione and tembotrione, alone or with previous treatment using malathion or NBD-Cl (Supplementary 1). This outcome indicates recovered tissue and absence of symptoms at the apex of the plant. The same occurred for malonaldehyde (MDA) concentration at 21 DAA (Supplementary 1). MDA is commonly used as a measure of oxidative stress in physiological systems (Ghani, 2017) as it is generated from lipid peroxidation in plant cells under different types of stress (Carvalho et al., 2011). The findings show the absence of lipid peroxidation in the evaluated tissue of white oat plants as a result of the treatments when compared to control (Supplementary 1). This outcome allows the plant to complete its cycle without interfering with yield. This behavior is opposite to what is expected in plants treated with carotenoid biosynthesis inhibitor herbicides, which reduce tocopherol and plastoquinone levels, leading to carotenoid depletion and consequently leaf bleaching (reduction in chlorophyll and carotenoid levels) and plant death arising from oxidative stress (lipid peroxidation) (Beaudegnies et al., 2009; Dankov et al., 2009; Wang et al., 2020).

The assessment of herbicide selectivity must be based on plant intoxication symptoms and oxidative stress parameters (Agostinetto et al., 2016). In addition, crop yield and product quality need to be quantified. There are products that affect crop yield without causing visual symptoms, while others cause injuries that are recoverable, thus not affecting yield (Negrisoli et al., 2004). Also, they may stimulate plant development when applied at specific rates (Duke et al., 2006). The parameters relative to yield components (Supplementary 2), yield (Supplementary 3), and grain quality (Supplementary 4) did not differ from those of the control when applying the herbicides mesotrione and tembotrione, either alone or in previous treatments with malathion or NBD-Cl. These findings indicate crop tolerance to the application of the herbicides mesotrione and tembotrione alone and absence of tolerance reversal after application of the metabolization inhibitors malathion and NBD-Cl.

Differential metabolism is considered to be the main mechanism responsible for tolerance and resistance of species to HPPD inhibitors, but this fact was not found to occur in this work. However, Kramer et al. (2014) reported that the HPPD isoenzyme, encoded by the gene *avhppd-03* in *Avena sativa*, has a reduced binding affinity for mesotrione; thus, it is more easily tolerated by the species. In addition, Matringe et al. (2005), Siehl et al. (2014), Dreesen et al. (2018) and Hanwkes et al. (2019) reported the use of the HPPD-resistant gene of *Avena sativa* in association with a modified gene of *Pseudomonas fluorescens* for the development of transgenic soybean (*Glycine max*)

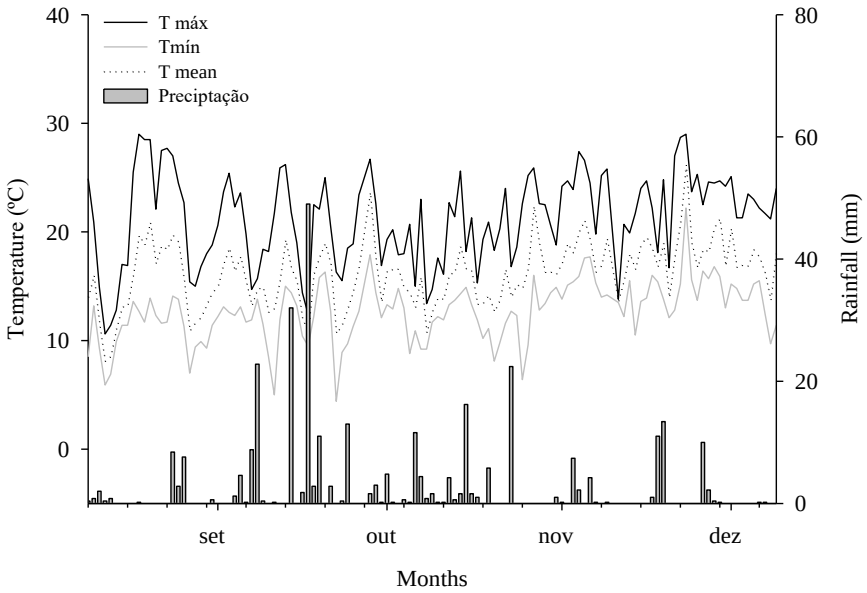

Fig 1. Rainfall, maximum, minimum and mean air temperatures from sowing to harvesting the experimental white oat growing during the 2021/2021 season, in Lages - SC. T max; Maximum temperature; T min: minimum temperature; T mean: mean temperature.
Source: Inmet, 2021;

resistant to HPPD inhibitors or, for the same purpose, the use of only the HPPD "*Avena sativa 339*" with deletion of a single amino acid in the N-terminal domain (Hawkes et al., 2016). Thus, the low enzymatic affinity of the HPPD isoenzyme of *A. sativa* may be the main mechanism responsible for the tolerance of the crop to the herbicides mesotrione and tembotrione. This may explain the occurrence of some symptoms of intoxication and subsequent recovery.

Based on these findings, further research should be conducted on the use of (C^{14})-labeled herbicide molecules to evaluate their differential absorption and translocation and to prove the occurrence of differential metabolism in oat plants. There should be studies on the enzymatic affinity of HPPD in oats, and on the sequencing of the HPPD gene in oats and susceptible plants, to elucidate the complete mechanism of crop tolerance to this group of herbicides.

Materials and Methods

Characterization of the field

The experiment was carried out at an experimental site at the State University of Santa Catarina CAV/UDESC, in the city of Lages – SC, under the following geographical coordinates: 27° 52' South latitude; 50° 18' West Longitude; and average altitude of 930 m, during the crop year of 2021. The climate of the region, according to the Köppen classification, is Cfb (temperate climate, with mild summers). Figure 1 shows the meteorological data (maximum, average, and minimum temperatures, and rainfall) in Lages during the time when the experiments were conducted.

The experiment was implemented in an aluminic humic Cambisol (Embrapa, 2017) with the following characteristics: pH in water: 6.3; Ca: 9.1 $cmol_c\ dm^{-3}$; Mg: 4.0 $cmol_c\ dm^{-3}$; Al: 0.0 $cmol_c\ dm^{-3}$; K: 143.3 mg dm^{-3}; P: 35.2 mg dm^{-3}; CEC: 16.6 $cmol_c\ dm^{-3}$; OM: 2.8% and clay: 50%.

Experimental design and treatments

The crop was sown under a no-tillage system (soybean stubble) using a randomized block design (DBC), with four replications. The treatments are listed below: T0-control (no application); T1-malathion (Dipil®, 1000 g active ingredient (a.i.) ha^{-1})); T2- 4-chloro-7-nitrobenzofurazan – NBD-Cl (Alfa Aesar®, 270 g a.i. ha^{-1}); T3- mesotrione (Callisto®, 168 g a.i. ha^{-1}); T4-mesotrione+malathion (168 + 1000 g a.i. ha^{-1}); T5-mesotrione+NBD-Cl (168 + 270 a.i. ha^{-1}); T6-tembotrione (Soberan®, 88.2 a.i. ha^{-1}); T7-tembotrione+malathion (88.2 + 1000 a.i. ha^{-1}) and T8-tembotrione+NBD-Cl (88.2 + 270 g a.i. ha^{-1}).

Plant material

This experiment was carried out with the white oat cultivar UPFA Fuerza, launched in 2015. It has a semi-erect growth habit, with high stature and medium cycle, and it is resistant to leaf rust (Danielowski et al., 2021).

Conduction of study

Before sowing, the seeds were treated with fipronil+pyraclostrobin+thiophanate-methyl (Standak Top®, 200 ml 100 kg^{-1} of seeds) and sown mechanically

on August 9, 2021; 350 viable seeds per m² of the white oat cultivar UPFA Fuerza were used. Fertilization was performed with 400 kg ha^{-1} of the N-P_2O_5-K_2O 5-20-10 formula, and later, at the beginning of tillering and stem elongation, topdressing nitrogen fertilization was performed with 45 kg ha^{-1} of N, following the recommendations of the Soil Chemistry and Fertility Commission (CQFS, 2016) for a productive potential of 5 t ha^{-1} of seeds. Each plot consisted of 5 rows that were 5 m long, with 0.2 m inter-row spacing, spaced 0.5 m apart from each other.

The phytosanitary management of the crop was carried out by applying fungicides and insecticides whenever necessary (through monitoring) to ensure the intended yield potential, following the technical indications for white oat (Danielowski et al., 2021).

Herbicide application (mesotrione or tembotrione) was carried out when 50% of the plants had 3-4 fully expanded leaves (09/11/2021), and 0.5% v/v vegetable oil (Áureo®) was added to the spray mixture. The 4-chloro-7-nitrobenzofurazan (NBD-Cl; diluted in acetone) an metabolization inhibitor via glutathione-S-transferase (GST) enzymes was applied 48 hours before herbicide application, while the application of malathion, an inhibitor of cytochrome P450 monooxygenase (P450) enzymes, preceded the application of herbicides by two hours, based on the work by Shergill et al. (2018). The treatments were applied with the aid of a CO_2-pressurized backpack sprayer (Herbicat, Brazil), with a pressure of 30 KPa, fitted with six 80.02 flat jet tips VS (TeeJet®, USA), calibrated for a spray volume of 200 L ha^{-1}.

Traits measured

At 7, 14, 21 and 28 DAA (days after application) of the treatments, plant intoxication was evaluated using the percentage scale from 0 to 100% (SBCPD, 1995), where 0% represents the absence of symptoms and 100%, the death of all plants. Concomitantly, the SPAD index was measured on the last fully expanded leaf of the plant, using a SPAD-502 Plus chlorophyll meter.

At 21 DAA, the last fully expanded leaf was collected from five plants per plot, then frozen in liquid nitrogen and stored in an ultrafreezer at -80 ºC for quantification of concentrations of chlorophyll *a* (C*a*), chlorophyll *b* (C*b*), total carotenoids (TC) and malonaldehyde (MDA). C*a*, C*b* and TC were quantified by using the methodology proposed by Lichtenthaler et al. (2001), while MDA was quantified by the method proposed by Heath et al. (1968).

At stage 89 (Zadocks et al., 1974), oat plants with 0.3 m² (0.5 m x 0.6 m) in each plot were cut close to the ground and packed in paper bags. Using these sub-samples, number of panicles, spikelets and G>2 per spikelet were counted by direct counting. Then, the samples were packed and dried in an oven at 65±3 ºC to constant weight. Subsequently, total plant biomass and grain weight were weighed separately on a semi-analytical scale, and the harvest index (HI) was determined through the ratio: total grain weight (g)/total plant biomass (g).

Grain yield was estimated by harvesting the total area of each plot (5 m²); water content was corrected to 13% and converted into hectare. Thousand-grain weight (TGW) was measured on the basis of the count of 1,000 grains and subsequent weighing on a semi-analytical balance with a precision of 0.0001 g.

Grain thickness greater than two mm (G>2) was determined by weighing and subsequently sieving a sample of grains in a rectangular sieve (40 x 30 cm) with 2 mm x 20 mm thick oblong holes. The sample retained on the sieve was weighed again to estimate the percentage (%) of G>2.

Hectoliter weight (HW) was measured by weighing a grain sample previously sieved G>2 and placed in a tube of known volume (0.25 L), using the Dalle Molle hectoliter weight scale (type 40). Such weight (g) was converted (kg into 100 L), based on specific table for oat crops (Brasil, 2009).

Dehulling index (DHI) was determined by manually dehulling a sample of 5 g of G>2 (hull+caryopsis) previously dried in an oven at 60 ºC After dehulling, the grains were weighed again to determine caryopsis weight.

Industrial yield (AVENACOR, kg ha^{-1}) was estimated by calculating grain yield (GY) x G>2 x DHI.

Statistical analysis

Data were tested as assumptions of analysis of variance (normality and homogeneity), and when satisfied, they underwent analysis of variance (ANOVA) by the F-test ($p<0.05$). Data on levels of intoxication that did not satisfy the assumptions were transformed by arcsin of $(x/100)^{0.5}$, prior to ANOVA. The data, when significant, were further analyzed using Tukey's test ($p<0.05$) and the R software.

Conclusion

The application of metabolization (malathion) and conjugation (NBD-Cl) inhibitors delayed the recovery process of plants sprayed with mesotrione. This result suggests that metabolization may increase the tolerance of white oat to mesotrione. The tolerance of white oat to the herbicides mesotrione and tembotrione was not reversed by the application of metabolization (malathion) and conjugation (NBD-Cl) inhibitors, indicating that differential metabolism is not the main mechanism of tolerance of white oats to HPPD-inhibiting herbicides.

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Physiological quality of oat seeds produced by plants treated with mesotrione or tembotrione

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Abstract

Selective herbicides may cause plant stresses as a result of changes in plant physiology or seed formation and development processes, thus affecting the quality of seeds. Thus, the aim of this study was to assess the effect of application of increasing doses of mesotrione or tembotrione on the physiological quality of white oat seeds from different cultivars. It is a field experiment carried out in two crop years (2019 and 2020), in a split split-plot randomized block design. The main plots consisted of the cultivars: FAEM 007, IPR Afrodite, UPFA Fuerza and URS Monarca. The subplots were mesotrione and tembotrione; and sub-subplots were doses of each herbicide: 0x;1/4x; 1/2x, 1x and 2 x (x= recommended commercial doses for maize, equivalent to 168.0 g a.i. ha^{-1} for mesotrione and 88.2 g a.i. ha^{-1} for tembotrione). Seed germination was 1% lower after application of the 2x dose of the herbicides, compared with control, i.e., there was a slight reduction in the germination of seeds produced in 2019. There was a significant effect of cultivar on germination and vigor. The cultivar UPFA Fuerza showed better performance than the other cultivars, with 96% germination index (GI) and 94% accelerated aging (AA) in 2019, while the best performance in 2020 was achieved by URS Monarca, with 98% GI and 97% AA. It was concluded that the oat seeds had high physiological quality in all treatments tested. The germination and vigor of oat seeds depended on the genotype being used, especially UPFA Fuerza (2019) and URS Monarca (2020), but they were not affected by the herbicides and doses tested.

Keywords: *Avena sativa* L; carotenoids; germination; herbicide; vigor.
Abbreviations: 2,4D_2,4-dichlorophenoxyacetic acid; AA_accelerated aging; a.i._active ingredient; Al_aluminum; ANOVA_analysis of variance; GI_germination index; ABRASEM_Associação Brasileira de Sementes e Mudas; AGROFIT_Informações sobre agrotóxicos fitossanitários registrados no Ministério da Agricultura, Pecuária e Abastecimento; ALS_acetolactate synthase enzyme; C1_seed category 1; C2_seed category 2; Ca_calcium; CEC_cation exchange capacity; CFB_Clima temperado com verão ameno e falta de estação seca; CONAB_Companhia Nacional de Abastecimento; CQFS_Comissão de Química e Fertilidade do Solo; CT_cold test; DEM_days from emergence to maturation; DSW_dry shoot weight; EC_electrical conductivity; EMBRAPA_Empresa Brasileira de Pesquisa Agropecuária; FAEM_Faculdade de Agronomia e Medicina Veterinária Eliseu Maciel; GSI_germination speed index; IPR_Instituto Agronômico do Paraná; ISTA_International Seed Testing Association; IWM_integrated weed management; K_potassium; MAPA_Ministério da Agricultura, Pecuária e Abastecimento; MCPA_2-methyl-4-chlorophenoxyacetic acid; Mg_magnesium; NI_Normative Instruction; OM_organic matter; P_phosphorus; RAS_Regras de Análises de Sementes; UFGRS-URS_Universidade Federal do Rio Grande do Sul; SL_seedling length; SDW_seedling dry weight; VI_vigor index.

Introduction

White oats (*Avena sativa* L.) are one of the major winter crops grown in southern Brazil, and they are widely used by the food industry for human nutrition (Pacheco et al., 2021). Oat production in Brazil increased in the 2020 crop year, yielding 850 thousand tons, while a total of 1080 thousand tons were expected in 2021 (Conab, 2021). In turn, the production of certified seeds in the country in 2020 reached approximately 27.2 thousand tons, with a rate of only 38% of certified seed use (Abrasem, 2021). However, to cultivate the Brazilian area, which corresponds to 400 thousand hectares (Conab, 2021), 40 thousand tons of seeds are required just to sow this area for production of oats.

Weed plants are among the factors that negatively affect the production of certified seeds and ultimately cause the exclusion of field areas from activities such as seed production or trading of white oat seeds (Normative Instruction no. 45 of Sept. 17, 2013). Therefore, weeds in oat crops are controlled by integrated weed management (IWM), combined with herbicide application (Fu et al., 2020; Kadam et al., 2021). Nevertheless, there is a small number of selective herbicides available for use in oat crops, owing to the absence of active ingredients for control of Liliopsida (Monocots); for Magnoliopsida (Dicots), only two mechanisms of action have been registered: one of ALS inhibitors (metsulfuron-methyl) and one of auxin-mimic herbicides (2,4 D and MCPA) (Agrofit, 2021). However, the successive use of few mechanisms of action is conducive to the selection of resistant biotypes (Gaines et al., 2020); for this reason, there is a need to identify new mechanisms of action selective to white oat crops. Carotenoid biosynthesis

inhibitor herbicides offer a possible alternative (Bond et al., 2014; Schmitz et al., 2015); tembotrione and mesotrione, for example, were reported as being potential selective herbicides for winter cereals (Ahrens et al., 2013; Lindell et al. 2015; Schmitz et al., 2015; Ndikuryayo et al. 2017; Schmitz et al., 2018).

Selectivity depends on the physicochemical characteristics of the product being used, the soil and climate conditions at the time of application, genotype susceptibility, dose in use and plant development stage (Harrison et al., 1995; Oliveira Jr et al., 2011). It is noteworthy that a selective herbicide may cause direct and indirect effects on the growth and development of a crop (Agostinetto et al., 2016; Marchezan et al., 2017; Marchi et al., 2021).

Herbicides can interfere in the formation and development of seeds, affecting their germination and vigor (Albretcht et al., 2012). In this regard, Albrecht et al. (2010) found a negative effect on the vigor of wheat seeds after application of increasing doses of metsulfuron-methyl; Marchi et al. (2021) also reported negative outcomes in soybean after application of flumioxazin. But there is lack of information available on the effect of application of mesotrione or tembotrione on the physiological quality of white oat seeds produced. Thus, the aim of this study was to assess the effect of mesotrione or tembotrione applied at postemergence of white oat cultivars on the physiological quality of the oat seeds produced.

Results

Seed moisture and germination

There was a significant effect (P<0.05) for the interaction between cultivar, herbicide and dose for the variable seed moisture at harvest in the 2019 crop year, while for 2020, there was an effect only for cultivar. In 2019, the increasing doses of mesotrione resulted in a quadratic behavior for cultivar FAEM 007, and the maximum seed moisture at harvest estimated for the 1.35x dose of this herbicide was 18.5% (Figure 1a). The cultivars IPR Afrodite, UPFA Fuerza and URS Monarca, after application of increasing doses of mesotrione, exhibited an increasingly linear behavior, i.e., seed moisture at harvest was increased by 1.4%, 2.4% and 0.2%, respectively, for each gram of active ingredient (g a.i. ha^{-1}) applied (Figure 1a). The increased doses of tembotrione resulted in an average seed moisture of 13.9% for cultivar UFFA Fuerza, but for the other species, they exhibited a quadratic trend (Figure 1b). Cultivar FAEM 007 reached the minimum seed moisture at harvest estimated for the 1.6x dose (16%), while cultivars IPR Afrodite and URS Monarca exhibited the maximum moisture estimated for the 1.3x (17.3%) and 0.8x (15.3%) doses (Figure 1b). In 2019, for seed moisture at harvest, considering the simple effect of cultivar, FAEM 007 and IPR Afrodite exhibited a higher value, namely 17.1%, when compared with UPFA Fuerza and URS Monarca, which showed about 15% (Table 1). However, in 2020, cultivar IPR Afrodite exhibited a higher moisture percentage, i.e., 15.8%, when compared with the other cultivars, which had approximately 13% of moisture (Table 1).

There was a significant effect (P<0.05) of cultivar on the variables relating to the germination in the 2019 and 2020 crop years, and an effect of dose in the 2019 crop year. In 2019, UPFA Fuerza exhibited the highest seed germination (96%), while the other cultivars achieved 94% (Table 2). However, in 2020, the highest seed germination was found for URS Monarca (98%), and the lowest germination rate, for UPFA Fuerza (95%), while FAEM 007 and IPR Afrodite did not differ significantly from the other two cultivars, with 96% (Table 1). Seed germination was also influenced by the herbicide doses applied in the 2019 crop year, and the maximum estimated seed germination was 95% for the 0.5x dose for both herbicides tested (Figure 2a). There was a 1% reduction in comparison to control for application of the 2x dose (Figure 2a), but this percentage did not adversely affect seed quality. In 2020, seed germination was not affected by the different doses of herbicides being used, indicating a germination rate of 96% (Figure 2b).

Seed vigor

There was a significant effect (P<0.05) of cultivar, as shown by the accelerated aging (AA) in both crop years. In 2019, AA was divided into two vigor classes: the highest for UPFA Fuerza (94%), and the lowest for IPR Afrodite (91%) (Table 1). Cultivars FAEM 007 and URS Monarca, with 92%, did not exhibit a significant difference from the previous classes (Table 1). Similarly, in 2020, AA provided two vigor classes: the highest for URS Monarca and IPR Afrodite, with 97%, and the lowest for FAEM 007, with 94% (Table 1). In contrast, the cultivar UPFA Fuerza, with 95%, did not show a significant difference from the classes previously cited (Table 1).

Regarding electrical conductivity (EC), there was a significant effect (P<0.05) of cultivar in both crop years. In 2019, four levels of vigor resulted from the EC test: the highest for IPR Afrodite (33.1 $\mu S\ cm^{-1}\ g^{-1}$), followed by URS Monarca (43.8 $\mu S\ cm^{-1}\ g^{-1}$), UPFA Fuerza (62.6 $\mu S\ cm^{-1}\ g^{-1}$) and FAEM 007 (85.8 $\mu S\ cm^{-1}\ g^{-1}$) (Table 1). However, in 2020, three levels of vigor were found: the highest one for URS Monarca (43.7 $\mu S\ cm^{-1}\ g^{-1}$), followed by FAEM 007 (54.8 $\mu S\ cm^{-1}\ g^{-1}$) and IPR Afrodite and UPFA Fuerza, which did not differ from one another, with 71 $\mu S\ cm^{-1}\ g^{-1}$ (Table 1).

For the vigor tests, namely cold test (CT), germination speed index (GSI), seedling length (SL), there was a significant effect (P<0.05) of cultivar only in the 2019 crop year (Table 2). The cold test was efficient in segregating oat cultivars into two groups of vigor in 2019: the highest one was found for FAEM 007 (95%) and UPFA Fuerza (95%), and the lowest one. for IPR Afrodite (93%), while URS Monarca (94%) showed no significant difference from the other groups (Table 2).

With the result of the germination speed index in 2019, two groups of vigor could be formed: the first group (higher vigor) was made up of IPR Afrodite (11.3), UPFA Fuerza (10.9) and URS Monarca (10.5) while the second group (lower vigor) was composed of FAEM 007 (9.5) (Table 2).

As for the seedling performance tests in 2019, two groups of vigor were formed for seedling length (SL): the group with greater vigor was formed by FAEM 007 (23.8 cm), IPR Afrodite (24.1 cm) and UPFA Fuerza (24.8 cm) and the group with lower vigor included UPFA Fuerza (21.5 cm) (Table 2). For SDW, however, there was a significant effect (P<0.05) in 2019 and 2020 crop years (Table 2). The SDW in 2019 provided two levels of vigor: the highest one was formed by cultivars FAEM 007 (22.5 mg), UPFA Fuerza (21.6 mg), and URS Monarca (23.5 mg) while the group with the lowest vigor was formed by IPR Afrodite (19.3 mg). Similarly, in 2020, two levels of vigor were found: the highest one was formed by FAEM 007 (21.3 mg) and URS Monarca (19.6 mg), and the group with the lowest vigor was formed

by IPR Afrodite (14.6 mg), but UPFA Fuerza (18.1 mg) did not differ from the others (Table 2).

There was a significant effect ($P<0.05$) of cultivar on vigor index (VI) of seeds produced in both crop years. In 2019, UPFA Fuerza exhibited the highest VI (1231), while the VI of the other cultivars tested did not differ from one another (Table 3). In 2020, however, URS Monarca achieved the highest VI (2430), while FAEM 007 (2288) and UPFA Fuerza (2322) exhibited the lowest VI (Table 3).

Discussion

Seed moisture and germination

In both crop years, seed moisture levels at harvest were within the recommended limits (13 to 18%) for oat crops, ranging from 13.5% to 18.0% in 2019 (Figure 1) and between 12.5 and 15% in 2020 (Table 1). Harvesting oat seeds with the ideal moisture content is crucial to prevent mechanical damage caused by excess of moisture (crushing) or excessive drying (impacts and abrasion). Mechanical injuries caused by seed moisture may affect the physiological potential, physical purity and health of seed lots (Marcos Filho, 2015).

The application of herbicides caused injuries to the plants' leaves. The injuries increased as the herbicide doses increased, and they were greater in the treatments with mesotrione. Nonetheless, the plants started to recover at 14 days after application of the treatments, exhibiting new tissues without signs and symptoms of injury. The plants' life cycle continued as usual and enabled the use of seeds in the present study (data not shown).

In this regard, Negrisoli et al. (2004) underlined that while there are herbicides that affect yields and the final quality of crop products without causing visible injury symptoms, others cause injuries that are recoverable and do not affect yields or the quality of grains and seeds. Therefore, herbicide selectivity should be assessed not only on the basis of visible injuries and yields, but also by considering the quality of the harvested product, including the physiological quality of the seeds produced.

During observations of the climate conditions in the growing seasons when the seeds were produced and harvested, there was a higher cumulative rainfall rate in 2019, compared with 2020 (Figure 3), particularly during the plant reproductive stage. Such climatic factors, combined with strong winds occurring in 2019, resulted in oat plant lodging – an outcome which did not occur in 2020. Several factors can influence the physiological quality of seeds, e.g., genotype, climate conditions, nutrition, diseases and pests, crop growing practices, mechanical damage, deterioration and seed size (Pádua et al., 2010; Carvalho et al., 2012; Zucareli et al., 2014). Lodging affects plants by limiting their access to sunlight, thus impairing photosynthesis and increasing the likelihood of diseases (Ingver et al., 2010; Packa et al., 2015); also, it leads to uneven seed moisture contents at harvest, as found in 2019. Seed moisture levels were influenced by the cultivar, herbicide and doses tested (Figure 1). Orthodox seeds that remained in the field after physiological maturity may show marked variations in moisture levels as result of climate fluctuations at the end of the cycle (Marcos Filho, 2015), especially when plants are lodged. In contrast, in 2020, only the effect of cultivar was found to occur on seed moisture at the time of harvest (Table 1).

The effect of cultivar on harvest seed moisture is directly related to cultivar life cycle, as can be seen for IPR Afrodite, with a total cycle of 126.1 days from emergence to maturation (DEM); UFPA Fuerza, with 125.2 days; FAEM 007, with 124 days; and URS Monarca, with 116.5 days (Danielowski et al., 2021). DEM have an impact on the speed of development and maturation of seeds at the end of the cycle.

The physiological attributes that determine seed quality include germination and vigor (Peske et al., 2012). Concerning the germination of white oat seeds, the Normative Instruction no. 45 of Sept 17, 2013 establishes a minimum germination rate of 80% for trading oat seed lots (Brasil, 2013). Thus, regardless of cultivar, herbicide and dose, the seeds had a high germination rate ($G>94\%$) and met the quality standards defined by NI 45/2013 in both crop years, i.e., a germination rate equal to or over 80% for seeds of categories C1 and C2 (Table 1).

Spraying the 2x dose of the herbicides mesotrione or tembotrione reduced seed germination by 1% compared with the control treatment (95%) (Figure 2), in 2019; this response was not found in 2020, when only the cultivar effect ($P>0.05$) (Table 1) occurred. The slight reduction in germination reported in 2019 did not compromise the germination of seeds, whose rate was 94% (Table 1).

These results corroborate the ones reported by Albrecht et al. (2010), who found that increasing doses of metsulfuron-methyl (0 to 12 g ha^{-1}) sprayed on wheat crops at the vegetative and reproductive phases did not affect seed germination. Similarly, soybean seeds from plants treated with flumioxazin did not exhibit changes in germination rates (Marchi et al., 2021). Soybean seeds from plants treated with glyphosate alone and combined with clethodin, lactofem, chlorimuron, fluazifop and cloransulam also showed no significant difference in seed germination (Silva et al., 2018). In contrast, in a study conducted by Albrecht et al. (2012), increasing doses of glyphosate (0, 1440 and 2880 g ha^{-1} acid equivalent) applied on soybean crops at the developmental stages V6 and R2 linearly reduced seed germination.

Considering the oats assessed, cultivars UPFA Fuerza (96%) and URS Monarca (98%) exhibited a higher germination rate in 2019 and 2020, respectively (Table 1). However, it should be noted that the other cultivars achieved germination rates between 94% and 96% in both crop years (Table 1). Such white oat cultivars have an aptitude to produce grains - a fact that indirectly favors the production of quality seeds. After all, modern white oat cultivars developed by plant breeding provided an increase of grain yield ant quality, resulting in higher industrial yields. One of the characteristics appreciated by the industry of oat flakes is the high percentage of grains>2mm-sized, combined with the dehull index.

Seed size and weight may affect germination and vigor (Araújo Neto et al., 2014). According to Carvalho et al. (2012), larger seeds may have had good nutrition during their development, and probably have well-formed embryos with greater amount of reserve substances, resulting in better germination and vigor. Padilha et al. (2020) and Ehrhardt-Brocardo et al. (2022) argued that seeds with greater vigor have higher mobilization and use of seed reserves for the seedlings, which favors the development of vigorous seedlings.

Seed vigor

Seed vigor is also associated with physiological attributes. Vigor is measured by several tests which are aimed at

Table 1. Seed moisture (SM) percentage at harvest, germination index (G), Accelerated aging (AA) and Electrical Conductivity (EC) of white oats for the cultivars assessed at the average spray rate of mesotrione and tembotrione, in the 2019 and 2020 crop years, in Lages, SC.

	2019 crop year						
Cultivar	SM	G		AA		EC	
	(%)	(%)		(%)		(μS $cm^{-1}g^{-1}$)	
FAEM 007	17.1 a	94	b	92	ab	85.8	d
IPR Afrodite	17.1 a	94	b	91	b	33.1	a
UPFA Fuerza	14.8 b	96	a	94	a	62.6	c
URS Monarca	15.0 b	94	b	92	ab	43.8	b
	2020 crop year						
FAEM 007	12.7 b	96	ab	94	b	54.8	b
IPR Afrodite	15.8 a	96	ab	97	a	71.0	c
UPFA Fuerza	13.7 b	95	b	95	ab	71.0	c
URS Monarca	12.5 b	98	a	97	a	43.7	a

The same letters in the column do not differ from one another by Tukey's test at 5% probability.

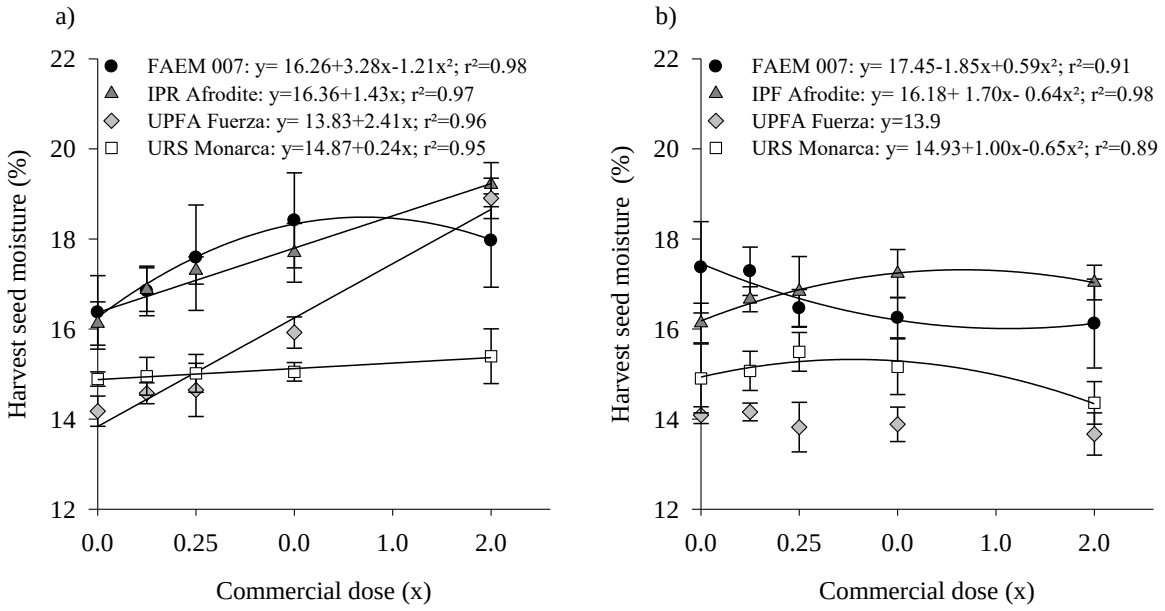

Fig 1. Seed moisture of oat cultivars at harvest as a result of doses of mesotrione (a) and tembotrione (b) sprayed in the 2019 crop year in Lages, SC. The vertical bars represent the standard error of the mean of each treatment.

Table 2. Cold test (CT), germination speed index (GSI), seedling length (SL) and shoot dry weight (SDW) of white oat cultivars, at the average spray rate of mesotrione and tembotrione, in the 2019 and 2020 crop years, in Lages, SC.

	2019 crop year							
Cultivar	CT		GSI		SL		SDW	
	(%)				(cm)		(mg)	
FAEM 007	95	a	9.5	b	23.8	a	22.5	a
IPR Afrodite	93	b	11.3	a	24.1	a	19.3	b
UPFA Fuerza	95	a	10.9	a	24.8	a	21.6	a
URS Monarca	94	ab	10.5	a	21.5	b	23.5	a
	2020 crop year							
FAEM 007	97	ns	10.9	ns	23.9	ns	21.3	a
IPR Afrodite	96		11.3		24.6		14.2	b
UPFA Fuerza	96		10.7		24.4		18.1	ab
URS Monarca	96		10.2		24.8		19.6	a

The same letters in the column do not differ from one another by Tukey's test at 5% of probability.

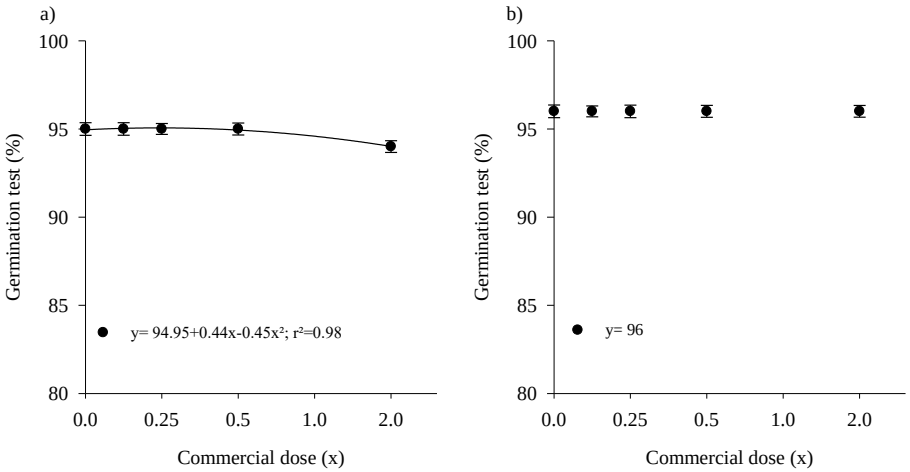

Fig 2. Germination (%) as a result of the herbicides doses sprayed, considering the average value of the oat cultivars, in the 2019 crop year (a) and the 2020 crop year (b), in Lages, SC. The vertical bars represent the standard error of the mean value of each treatment.

Table 3. Vigor index (VI) of white oats of different cultivars at the average spray rate of mesotrione and tembotrione, in the 2019 and 2020 crop years, in Lages, SC.

Cultivar	2019 crop year	2020 crop year
	VI	VI
FAEM 007	1074 b	2288 b
IPR Afrodite	1090 b	2370 ab
UPFA Fuerza	1231 a	2322 b
URS Monarca	1005 b	2430 a

The same letters in the column do not differ from one another by Tukey's test at 5% of probability.

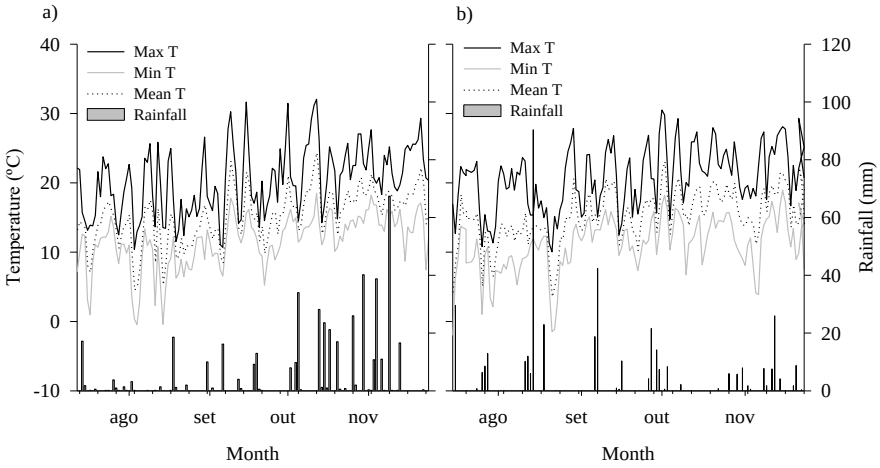

Fig 3. Rainfall, maximum, minimum and mean air temperature (daily) in the period from oat sowing to harvest for the experiment in the 2019 (a) and 2020 (b) crop years. Source: *Inmet (2019 and 2020).*

assessing the performance of seed lots under environmental conditions that are not ideally suitable for the germination of the species (Ista, 2014). Moreover, such tests may reveal significant differences in the physiological potential of seed lots with similar germination (Marcos Filho, 2015).

There were no changes to seed vigor on the basis of the doses and herbicides tested; it was only influenced by oat cultivar (Tables 1, 2 and 3). Contrary to the findings of the present study, a previous study reported that the application of different doses of glyphosate on soybean crop linearly reduced seed vigor (Albrecht et al., 2012). Similar results were found in the study by Marchin et al. (2021), in which flumioxazin sprayed on the mother plant resulted in soybean seeds with the lowest vigor, as shown by the tetrazolium and the electrical conductivity test. However, Silva et al. (2018) did not find alterations in the vigor of soybean seeds from plants treated with glyphosate and combinations (clethodin, lactofen, chlorimuron, fluazifop and cloransulam), based on the first germination count of seeds produced in two consecutive years. Albrecht et al. (2010) found reduced vigor (first count) in wheat seeds from plants treated with increasing doses of metsulfuron-methyl in the

plant reproductive phase in 2008 - a fact that did not occur again in 2009. According to the authors, this seed behavior is related to the prevailing environmental conditions in the crop area and growing seasons.

The vigor tests used in the present study, e.g., accelerated aging (AA), electrical conductivity (EC) and vigor index (VI) were efficient in segregating white oat cultivars in 2019 and 2020 (Table 1). The cold test (CT), germination speed index (GSI), seedling length (SL) and dry shoot weight (DSW) tests segregated cultivars into different groups of vigor only in 2019 (Table 2). The AA test measures the tolerance of seeds to high air humidity and temperature (41 to 45 ºC), and such exposure enhances seed deterioration in several species (Marcos Filho, 2015). Thus, seeds that exhibit higher vigor achieve superior germination after this procedure (Vieira et

Seeds that are less vigorous (or more deteriorated) according to the EC test, have lower speed of restoration of cell membrane integrity during imbibition; consequently, they release a greater number of solutes to the exterior, which results in a higher electric conductivity value (Vieira et al., 1999). Sponchiado et al. (2014) indicated that EC is efficient for use in white oats, and it can be used for assessing the physiological potential of oat seeds. The cultivar UPFA Fuerza exhibited greater vigor compared with other cultivars for AA, VI, CT, GSI, SL and DSW in 2019 (Table 1 and 2), while URS Monarca exhibited higher vigor according to the VI, AA, EC and DSW in 2020 (Table 1). For oats, no test to date is considered as entirely standardized to assess vigor, for example, for soybean (Marcos Filho, 2015). However, there are various tests that enable consistent differentiation between high- and low-vigor lots of oat seeds (Sponchiado et al., 2014). The present results show the high physiological quality achieved by the seeds in the experiments (Table 1 and 2), which results from: (i) lack of interference from the herbicides; (ii) use of seeds from genetic category obtained directly from breeders; (iii) adoption of crop growing practices that are suitable for in-field cultivation of oats (Lângaro et al., 2014); (iv) standardization of moisture levels immediately at harvest and (v) use of an adequate process breaking for seed dormancy prior to the seed evaluations.

So far, few studies have assessed the physiological quality of seeds produced from herbicide-treated plants. However, selective herbicides, when applied on the mother plant, are considered to be plant stressors (Agostinetto et al., 2016; Marchezan et al., 2017; Marchi et al., 2021). Although they may not affect a given crop, they can affect the processes of formation and development of seeds (Albrecht et al., 2012; Marchi et al., 2021). Taken together, the results point to a reduction in seed quality around 1% on germination, when compared to the control, owing to the herbicides doses applied. The treatments may minimally affect the physiological quality of the seeds, but they do not affect the performance of seed lots. This suggests that both active ingredients can be indicated as safe for use in white oat crops, considering the target weed plants and their development stage; in addition, the application method and doses previously considered as efficient in weed control in maize crops can also be used in white oat crops.

Materials and methods

Characterization of the experiment site

The seeds were produced under field conditions in the municipality of Lages-SC, at coordinates 27° 52' South latitude, 50° 18' West longitude, and mean altitude of 930 m, during the 2019 and 2020 crop years. The climate in the region, according to the Köppen classification, is Cfb (temperate climate with mild summers). Figure 3 shows the climate data (maximum, medium and minimum temperatures, and rainfall) in Lages during the periods of conduction of the study.

The soil in the experimental area is classified as an Aluminum Humic Cambisol (EMBRAPA, 2017), with the following characteristics: pH in water = 5.0; Ca = 4.7 $cmol_c\ dm^{-3}$; Mg = 1.4 $cmol_c\ dm^{-3}$; Al = 4.7 $cmol_c\ dm^{-3}$; K = 153 mg dm^{-3}; P = 26.8 mg dm^{-3}; CEC = 21.7 $cmol_c\ dm^{-3}$; O.M.= 6.6% and Clay = 27%.

Conduction of study and experimental design

Prior to sowing, the seeds (C1 category) were treated with fipronil + pyraclostrobin + thiophanate-methyl (Standak Top®), and sowed mechanically in a non-tillage system (the previous crop was soybean) on July 12, 2019 and July 15, 2020, using 350 suitable seeds per m². Each plot consisted of five 5.0-m long rows with inter-row spacing of 0.2 m and spaced 0.5 m apart from each other plot. A total of 400 kg ha^{-1} of NPK (N-P_2O_5-K_2O) formulation 5-20-10 was used for fertilization. After emergence, side-dressing nitrogen fertilization (45 kg ha^{-1} of N) was performed twice, split at the plant stages of tillering (first) and stalk elongation (second), as recommended by the Soil Chemical and Fertility Commission ("*Comissão de Química e Fertilidade do Solo*" - CQFS, 2016) for a production potential of 5 t ha^{-1} of grains.

The experiment was conducted in a randomized block design (RBD) arranged in split-split-plots with four replications per treatment. The main plot consisted of four oat cultivars: FAEM 007, IPR Afrodite, UPFA Fuerza and URS Monarca. The split-plots consisted of the herbicides mesotrione (Callisto®) and tembotrione (Soberan®), and the split-split-plot contained the herbicide doses applied, as follows: 0x (control, without herbicide application); 1/4x, 1/2x, 1x and 2x, where x corresponds to the commercial dose recommended for maize crops. Mesotrione doses corresponded to 0; 42; 84; 168 and 336 g a.i. ha^{-1} and tembotrione doses were 0.0; 22.1; 44.1; 88.2 and 176.4 g a.i. ha^{-1}.

Application of the herbicides was conducted when 50% of the plants exhibited 3-4 fully expanded leaves (on Aug 14, 2019 and Aug 17, 2020, respectively), using a CO_2-pressurized backpack sprayer (Herbicat, Brazil). The other phytosanitary managements (fungicide and insecticide applications) were carried out following the technical recommendations for cultivation of white oats (Lângaro et al., 2014).

Traits measured

At harvest (Nov 25, 2019 and Nov 23, 2020), seed moisture content (on a wet basis) was determined by using a sample collected from the split-split-plot area (5 m²), following the oven method proposed by the Brazilian Rules for Seed Testing (*Regras de Análises de Sementes-RAS*) (Brasil, 2009).

The seeds were then placed in an air circulation oven at 35 ºC to standardize their moisture content to 13%, and then they were kept for 30 days in a cold chamber (10±2 ºC and RH 50%) to prevent accelerated aging (Coelho et al., 2007) until the conduction of the tests. The oat seeds underwent a treatment for seed dormancy breaking, using the preheating method (Brasil, 2009). The seeds were subjected to the following tests:

Germination test
Carried out with four replicates of 100 seeds arranged in Germitest® paper rolls moistened with distilled water at the rate of 2.5 times the dry paper weight. The paper rolls were placed vertically in a germinator set at 20±1 °C. Normal seedlings were counted at the 10th day after sowing, following the Brazilian Rules for Seed Testing ("*Regras de Análises de Sementes-RAS "*) (Brasil, 2009).

Cold test
Carried out with four replicates of 100 seeds per treatment, arranged in Germitest® paper rolls moistened with a quantity of water equivalent to 2.5 times the dry weight of the substrate. The rolls were placed into plastic bags to prevent moisture loss and maintained in a BOD chamber at 10 °C for seven days (Krzyzanowski et al., 1999). After this period, the seeds were transferred to a germinator and maintained at 20±1 °C for seven days. After that, vigor was determined by the cold test for quantification of normal seedling percentage.

Accelerated aging
The test used transparent germination boxes with a lid (11x11x3.5 cm), containing an aluminum wire mesh suspended inside, on whose surface the seeds were arranged to form a single layer. A total of 40 mL of distilled water was poured onto the bottom of the container, which was then covered and maintained in a BOD chamber at 41 ºC for 48 h (Tunes et al., 2008). After the aging period, seed moisture content was determined by the oven method at 105±3 ºC for 24 h (Brasil, 2009). Afterwards, the seeds were arranged in Germitest® paper moistened with a quantity of water equivalent to 2.5 times the dry weight of the substrate and kept in a germinator at 20±1 °C. At the fifth and tenth days after assembling the test, normal seedling percentage was determined to measure seed vigor by the accelerated aging test.

Electrical conductivity
Measured using the mass system with four replicates of 50 seeds per treatment. Seed mass was determined by placing the seeds in plastic cups with 75 mL of distilled water and maintained them at a temperature of 25 ºC. After 24 h of soaking, the electrical conductivity of this solution was determined by using a conductivity meter, and the results were expressed in $\mu S\ cm^{-1}\ g^{-1}$ (Vieira et al., 1999).

Emergence Speed Index (in sand)
the index was determined in a greenhouse environment with four replicates of 50 seeds per treatment, sown at a depth of 3 cm. The sand was previously washed and placed in plastic trays. Moisture was maintained with irrigations as needed. The number of emerged seedlings was assessed on a daily basis after sowing until the number of emerged plants remained stable. The index was calculated according to the formula proposed by Maguire (1962): GSI = G1/N1 + G2/N2 + ... + Gn/Nn, where: GSI = Germination Speed Index; G1, G2, Gn = number of seedlings emerged in the first, second and last count and N1, N2, Nn = number of days since the first, second, etc. until the last count.

Seedling length
Four replicates of 20 seeds were sowed in the upper third portion of Germitest® paper moistened with distilled water at the rate of 2.5 times the weight of dry paper. The paper rolls containing the seeds were kept in a germinator for seven days at 20±1 °C. With the aid of a millimeter rule, ten randomly chosen normal seedlings were measured, and the results were expressed in centimeters (Nakagawa, 1999).

Shoot dry weight
Ten normal seedlings were used for measuring seedling length. They were placed in a paper bag and dried in an oven at 60±3 ºC to constant weight; afterwards, they were weighed on a semi-analytical balance (precision of 0.001 g), and the results were expressed in mg $plant^{-1}$ (Nakagawa, 1999).

Vigor index
Calculated according to the method of Abdul-Baki et al. (1973), through the equation: VI = germination (%) x seedling length (cm).

Statistical analysis

Data were tested according to the assumptions of the analysis of variance (normality and homogeneity); when these assumptions were met, the data underwent the ANOVA F-test ($P<0.05$). When significant, they were split by Tukey's test ($P<0.05$) for qualitative treatments, and regression analysis was performed for the quantitative treatments ($P<0.05$).

Conclusions

The application of increasing doses of mesotrione or tembotrione herbicides did not affect the germination and vigor of white oat seeds, but the safe dose for post-emergence application is 168 g a.i. ha^{-1} for mesotrione or 88.2 g a.i. ha^{-1} for tembotrione in production fields of white oat seeds. Of the four study cultivars, better seed physiological performance was achieved with UPFA Fuerza and URS Monarca in the 2019 and 2020 crop years, respectively.
The sampled oat seeds exhibited high germination and vigor rates, namely over 94%, regardless of herbicide or dose applied on the plants, cultivars or crop years.

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Physiological quality of soybean seeds produced under shading

Qualidade fisiológica de sementes de soja produzidas sob sombreamento

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ABSTRACT

Low light intensity reduces the production of photoassimilates by plants and consequently the accumulation of reserves in seeds. The objective was to evaluate the physiological quality of soybean seeds produced under shading initiated at different crop phenological stages. In a completely randomized design, the experiment was carried out in a greenhouse, at CAV/UDESC, during the 2018/2019 harvest, with nine treatments and 10 replications. The treatments consisted of nine seed lots, eight of which were obtained from plants submitted to continuous 70% shading, initiated at different phenological stages: stage V6 (Day Julian 07); R1 (JD 14); R2 (JD 21); R3 (JD 28); R4 (JD 35); R5 (JD 42); start of R6 (JD 49); end of R6 (JD 56), and a lot of seeds produced without shading (control). Each repetition consisted of a pot containing a soybean plant. The germination potential and vigor of the seeds produced were evaluated. Soybean seed germination was higher in the lot of seeds from shaded plants from the R4 stage (JD 35), with 98%, but the other lots presented percentages above 87%. The vigor in the accelerated aging test was lower for the lot obtained from shaded plants from V6, with 43%. The dry mass of seedlings showed a 7.0 to 13.3% reduction in relation to the control when the shading was imposed between R3 and R6. It was concluded that the continuous 70% shading in soybeans promoted higher seed germination when imposed from the R4 stage; however, the vigor was dependent on the stage that the shading started and the test used.

KEYWORDS: photosynthesis; germination; *glicyne max*; solar radiation; vigor.

RESUMO

A baixa intensidade luminosa reduz a produção de fotoassimilados pelas plantas e consequentemente o acúmulo de reservas nas sementes. Objetivou-se avaliar a qualidade fisiológica de sementes de soja produzidas sob sombreamento iniciado em diferentes estádios fenológicos da cultura. O experimento foi conduzido em casa de vegetação, no CAV/UDESC, durante a safra 2018/2019, em delineamento inteiramente casualizado, com nove tratamentos e 10 repetições. Os tratamentos foram constituídos por nove lotes de sementes, sendo oito obtidos de plantas submetidas a sombreamento contínuo de 70%, iniciado em diferentes estádios fenológicos: estádio V6 (Dia Juliano 07); R1 (DJ 14); R2 (DJ 21); R3 (DJ 28); R4 (DJ 35); R5 (DJ 42); início de R6 (DJ 49); final de R6 (DJ 56), e um lote de sementes produzidas sem sombreamento (testemunha). Cada repetição foi composta por um vaso contendo uma planta de soja. Avaliou-se o potencial de germinação e vigor das sementes produzidas. A germinação de sementes de soja foi maior no lote de sementes oriundas de plantas sombreadas a partir do estádio R4 (35 DJ), com 98%, mas, os demais lotes apresentaram percentuais acima de 87%. O vigor no teste de envelhecimento acelerado foi inferior para o lote obtido de plantas sombreadas a partir de V6, com 43%. Já a massa seca de plântulas apresentou redução de 7,0 a 13,3% em relação à testemunha, quando o sombreamento foi imposto entre R3 e R6. Concluiu-se que o sombreamento contínuo de 70% em soja promoveu maior germinação de sementes quando imposto a partir do estádio R4, porém, o vigor foi afetado de maneira dependente do estádio que o sombreamento iniciou e do teste utilizado.

PALAVRAS-CHAVE: fotossíntese; germinação; *glicyne max*; radiação solar; vigor.

INTRODUCTION

The soybean crop (*Glycine max* L.) has great global economic importance. Fabaceae production in Brazil in the 2021/2022 crop was 12 4.0 million tons, with an average productivity of 3.0 tons per hectare

(CONAB 2022). Plant growth and its productivity may suffer interference from the environmental factor, such as luminosity intensity. Solar radiation is indispensable in the photosynthetic process, through which plants synthesize carbohydrates used for their vital processes (TAIZ et al. 2017). In addition, the rate of dry matter accumulation and growth of seeds subsequently harvested are proportionally dependent on plants' source and drain ratio, i.e., the production and storage capacity of the photoassimilate during the growth cycle (ASSENG et al. 2017).

Plants can be exposed to low availability of solar radiation, for example, in crop-forest integration systems (ILF) (CORDEIRO et al. 2015); and days with high cloudiness index, as visualized by CUSTÓDIO et al. (2009) in the region of altitude fields of Rio Grande do Sul. Yet, in grain production systems with high sowing densities, a fact that causes a higher rate of self-shading among plants, mainly affecting the leaves of the rascal (SOUZA et al. 2010); and as a function of latitude, in which there is a lower incidence of solar radiation as a function of solar inclination during the winter season.

When evaluating the shading effect of 50% during the development cycle of in soybean crop, BELLALOUI et al. (2012) observed a reduction in protein and lipid content and variation of germination of the seeds produced as a function of the genotype and crop evaluated. However, CHEN et al. (2020) evaluating the shading effect on soybean crop under intercropping with corn crop, found that seeds from the tested cultivars presented radicle protrusion more quickly, longer root length and seedling dry mass. Interestingly, few studies evaluate the physiological responses of soybean seeds from the condition of luminosity attenuation starting at different stages of the development of the mother plant. Therefore, based on the hypothesis that the reduction of the amount of light incident for long periods in soybean crop reduces the quality of the seeds produced, especially when covering the reproductive stages of development, the objective was to evaluate the physiological quality of soybean seeds from plants under shading condition (70%) imposed at different phenological stages.

MATERIAL AND METHODS

Place of study and experimental conditions of obtaining seeds

The experiment was carried out in a greenhouse during the 2018/2019 harvest, at the Center for Agri veterinary Sciences - CAV, State University of Santa Catarina - UDESC, in the municipality of Lages - SC. Soybean cultivar 96Y90 (category C1 seeds), characterized as early cycle, between 130 and 140 days, maturation group 5.9, habit of growth indeterminate and tolerant to bed. Data on temperature and relative humidity during the culture conduction period were recorded by *datalogger* device (TENMARS, TM305U) (Figure 1a).

The sowing was carried out on November 28, 2018, corresponding to Julian Day (DJ) number 332. Five seeds were seed per pot, and then thinning and conducting only one plant per pot with a capacity of five liters. For the treatment of seeds, the commercial product based on pyraclostrobin + methyl thyophalate + fipronil (Standak Top, 5.0 g i. a. 100.0 $^{kg-1}$ seeds + 45.0 g i. a. 100.0 $^{kg-1}$ seeds + 50.0 g i. a. 100.0 $^{kg-1}$ seeds) and peat biological inoculant containing *Bradyrhizobium elkanii* and *Bradyrhizobium japonicum*. The irrigation was performed manually and daily according to the needs of the plants, based on the depletion of the gravimetric moisture of the soil contained in the pots.

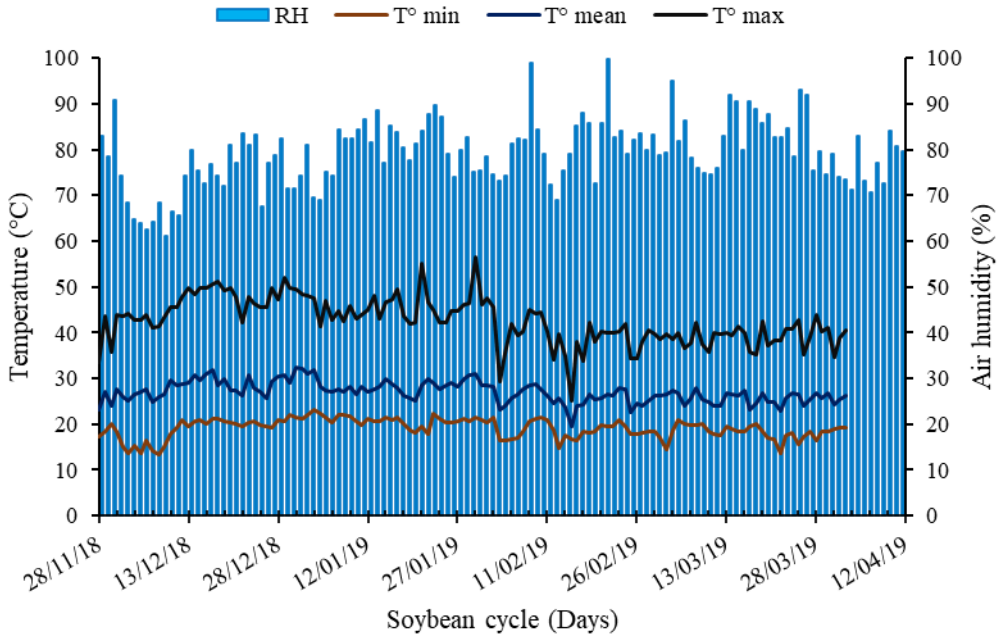
From the interpretation of the results of the soil analysis, fertilization was performed, following recommendations of the Commission of Soil Chemistry and Fertility (CQFS- RS/SC, 2016) for productivity expectation of 4000 kg $^{ha-1}$, corresponding to the application of 95.0 kg $^{ha-1}$ of K_2O, 30.0 kg $^{ha-1}$ of P_2O_5, and supplementation with 60.0 kg $^{ha-1\ nitrogen}$, divided into three applications, on November 28, December 19, 2018 and January 9, 2019 (JD 323 and 353 - 2018, and 09 - 2019). Dilutions of KH_2PO_4 (anhydrous monobasic potassium phosphate) KCl (potassium chloride) and urea were used for fertilization. For phytosanitary treatment, tebuconazole (Folicur, 150.0 g a.a. $^{ha-1}$) was used on January 10 (JD 10 - 2019) spiromesifen (Oberon, 96.0 g a.i. $^{ha-1}$) on 10 and 27 January (JD 10 and 27), and profenofos + lufenuron (Curyom, 150.0 g a.i. $^{ha-1}$ + 15.0 g a.i. $^{ha-1}$) on 16 January (JD 16).

The plants' artificial shading was performed by using black color screen, polypropylene monofilament, with fiber density of 70%, exceeding only 30% of the available solar radiation (checked via ceptometer) Light Sensor Logger LI-1500, LICOR®, Lincoln, NE, USA). The canvas was installed on a wooden structure 1.50 m high, to house the pots containing the plants.

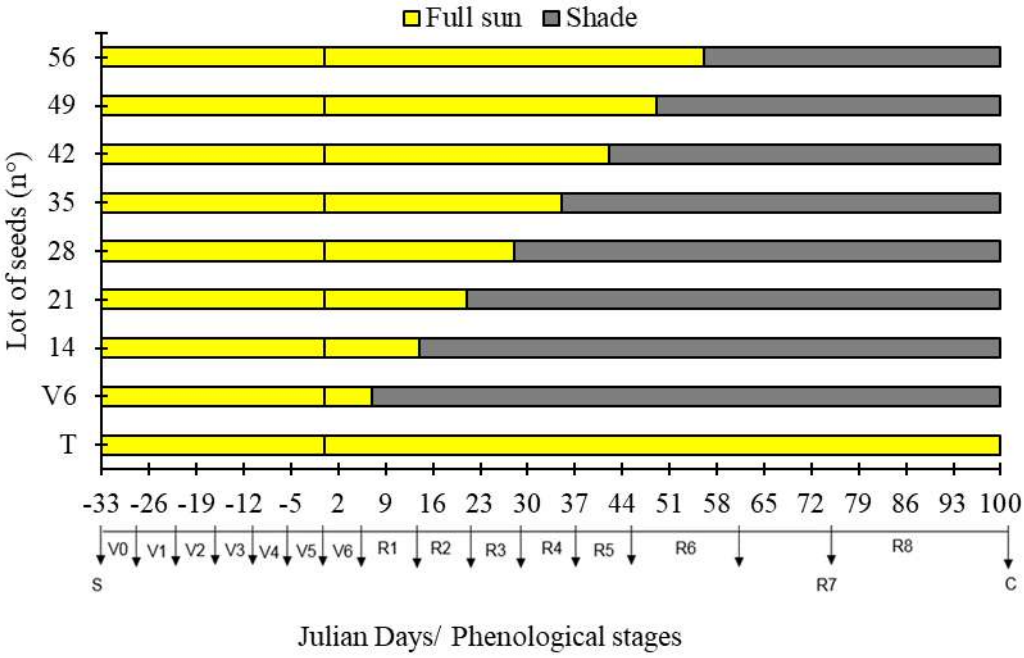
The harvest was carried out when the seeds were in harvest maturation, i.e., stage R8, in which 95% of the pods of the plant's main stem were yellow, on April 10, 2019 or JD 100. After manual threshing of the pods, the seeds were dried in an oven at 35 °C until moisture stabilization (13%). Then, the seeds of the plants of each treatment were homogenized, forming the average sample of each lot, and then separated

into four replications to make up the working samples. The size of the seeds used was standardized for seeds larger than 5.0 mm in diameter and circular sieve.

a)



b)



T: Witness. S: Sowing; C: Harvest.

Figure 1. a) Mean relative humidity (RH) and minimum (T° min), average (T° average) and maximum (T° max) recorded by a Datalogger device, in the greenhouse (unshaded environment) during driving of the experiment. b) Schematic representation of soybean seed lots formed according to phenological stages of shading starts on soybean plants. 2018/2019 growing season.

Experimental design

The experimental design used was completely randomized with nine treatments, consisting of plots of soybean seeds obtained from eight phenological stages of onset of artificial shading imposition of plants, a condition in which they remained until harvest, and the control treatment (plants conducted without shading), with 10 replications. The phenological stages at the beginning of each shading period and Julian Days (DJ) were: V6 stage (six knots on the stem from the unifoliolate node, JD 7); R1 (a flower in any knot, JD 14); R2

(flower in the first knot below the top knot with completely unrolled leaves, JD 21); R3 (pod 0.5 cm long in one of the four upper nodes with unrolled leaf, JD 28); R4 (pod 2 cm long in one of the four upper nodes with unrolled leaf, JD 35); R5 (beginning of development of seeds in one of the top four nodes with unrolled leaf, JD 42); R6 (pod with big green seed on one of the top four knots with unrolled leaf, JD 49); R6 (JD 56) (Figure 1b), whose phenological stages correspond to those described by FEHR & CAVINESS (1971). Each replication was composed of a pot containing a soybean plant of cultivar RR 96Y90.

Evaluation of seed quality

The analyses performed were:

- Weight of One Thousand Seeds (TSW): weighing of eight replicates of 100 seeds. Each repetition was weighed and multiplied by 10 (BRASIL 2009);
- Germination test (G%): 200 seeds for each treatment, divided into eight replicates of 25 seeds, distributed among pre-moistened germination paper sheets with water equivalent to 2.5 times its dough, rolled, taken to the germinator, at a temperature of 25 °C, and evaluated on the eighth day (final count). Results expressed in germination percentage (normal seedlings), abnormal seedlings and dead seeds (BRASIL 2009);
- First Germination Count (FGC): percentage of normal soybean seedlings five days after the beginning of the germination test (KRZYZANOWSKI et al. 1999);
- Accelerated aging (AA): soybean seeds were packed on aluminum canvas, so as not to overlap and use all space, contained in a gerbox under 40 ml of water. These were kept at a temperature of 41 °C for 48 hours and subsequently affected in paper rolls, according to indications for germination test, and evaluated on the eighth day for vigor percentage (KRZYZANOWSKI et al. 1999);
- Electrical Conductivity (EC): four replicates of 50 seeds per treatment, previously weighed, immersed in 75 mL of deionized water, kept at a temperature of 25 °C for 24 hours, and measured the EC of the water with a bench-top conductivity meter (Quimis, Model 0795A2). To obtain the EC of each sample, the formula was used: CE = CEs - CEa/P, being ECs: EC read by the apparatus of samples with seeds, EEC: from the pure water sample, and P: weight. Results expressed in $\mu S\ g^{-1}\ cm^{-1}$ (KRZYZANOWSKI et al. 1999);
- Length of Shoot (LS), Length of Main Root (LMR) and Length of Total Seedling (LTS): evaluation by measuring, with the aid of graduated ruler, of the aerial part, main root and total length of 20 soybean seedlings on the eighth day of the germination test, with four replications per treatment (KRZYZANOWSKI et al. 1999);
- Seedling Dry Weigth (SDW): on the eighth day of the germination test, 20 seedlings were dried per repetition, in a forced air kiln with a temperature of 65 °C until the stabilization of the mass (KRZYZANOWSKI et al. 1999). Results expressed in mg per seedling.

Statistical analysis

The data were submitted to normality and homogeneity tests, and after the analysis of variance by the F test ($p \leq 0.05$). Data that did not meet the assumptions were transformed, being the percentage through sine arc of $(x/100)^{0.5}$, and for abnormal seedlings and dead seeds through sine arc of $[(x+1.5)/100]^{0.5}$. When there was a significant difference, the lot means were grouped by the Skott-Knott test ($p \leq 0.05$) by the Sisvar software (FERREIRA 2011).

RESULTS

The results for TSW, germination and vigor by the tests of first count of germination, accelerated aging, electrical conductivity and performance of soybean seedlings showed variations depending on the stage in which the mother plants were imposed under shading during their development.

The weight of one thousand seeds (TSW) presented difference between lots. The batch of seeds from shaded plants from stage V6 presented higher TSW (163.2 g), with an increase of 4.3% in relation to the control. The lots from plants shaded in R5 and beginning of R6 presented lower TSW, with 133.5 and 131.9 g, respectively, indicating that the luminosity limitation in this period reduced on average 15.0% the seed mass compared to the control treatment (without shading), and 18.7% in relation to the lot of shaded plants from V6. In relation to lots from shaded plants in stages R2 and R3, these presented an average reduction in TSW of 7.71%, and for R4 and R6 the decrease was 10.8%, compared to the control (156.2 g) (Table 1).

For soybean seed germination, the highest percentage (98%) was obtained for the seed lot from shading imposed under the plants at stage R4, which represented an increase of 5% in relation to the control seed lot (without shading) (93%). However, negative effects of the condition of light limitation imposed from V6, R3, R5 and final of R6 on seed germination were observed, with averages of 87, 91, 89 and 87%,

respectively (Table 1). At the same time, the germination test did not check the presence of dead seeds, only abnormal seedlings. Thus, the lowest percentage of abnormal seedlings observed (2%) corresponded to the seed lot with the highest germination percentage (R4) (Table 1).

In relation to the first germination count (FGC) test, higher vigor (96%) was obtained for lots from shaded plants from the phenological stage R4 (pod with 2 cm), representing an increase of 5% in relation to the control lot (91%). The seed lots obtained from shading imposed in V6, R2, R3, R5 and R6 showed lower vigor by the test, with values between 78 and 89% (Table 1).

Table 1. Weight of a thousand seeds (WTS), germination (G), abnormal seedlings (AS), first germination count (FGC) and vigor by accelerated aging (AA) of soybean seeds produced under different phenological stages of shading imposition.

Phenological stage	TSW (g)	G (%)	AS (%)	FGC (%)	AA (%)
V6	163.2 a	87 c	13 a	84 c	43 d
R1	148.3 c	94 b	6 c	92 b	92 a
R2	143.3 c	93 b	7 c	84 c	95 a
R3	145.0 c	91 c	9 b	78 c	96 a
R4	138.8 d	98 a	2 d	96 a	76 b
R5	133.5 e	89 c	11 a	89 c	80 b
R6	131.9 e	92 b	8 b	80 c	94 a
R6	139.9 d	87 c	13 a	80 c	83 b
T	156.2 b	93 b	7 c	91 b	60 c
SMD	6.1	5.2	5.2	8.9	10.6
CV (%)	3.1	4.2	21.5	8.8	10.2
Average	144.4	91	9	86	80

*Equal letters in the column do not differ by the Skott-Knott test at p≤ 0.05; T: Witness; SMD: Significant minimum difference; CV (%): Coefficient of variation.

For the vigor of soybean seeds verified through the accelerated aging test (AA) it was observed that the batch of soybean seeds derived from plants shaded from the phenological stage V6, present or lower vigor (43%) compared with the control (60%). However, it was noted that the shading condition imposed from the R1, R2, R3 and beginning of R6 of plant development inferred the production of seeds with greater vigor by the test, with averages of 92, 95, 96 and 94%, respectively (Table 1).

Regarding the vigor of soybean seeds through the electrical conductivity (EC) test, it was observed that the seed lots from shaded plants from stages R1, R2, R4 and R5 presented higher values for the variable, with averages above 59.0 µS cm^{-1} g^{-1}, representing an increase of 6.3 to 17.5% in the EC, compared to the seed lot produced without shading (55.5 µS cm^{-1} g^{-1}). Thus, it is noteworthy that the lots in question, with the highest means of EC, are classified as of lower vigor (Table 2).

The results for seedling shoot length (SL) differed between seed lots. For seedlings derived from shading imposed from stages R1, R3 and R4, they presented higher SL, with an increase of 16.1, 25.1 and 16.6%, respectively, compared with the mean of the control (7.76 cm) (Table 2). Regarding the main root length (LMR), the lowest mean observed was for shading the mother plants from the R2 stage (8.0 cm), representing a decrease of 42.5% in relation to the control (11.4 cm). Moreover, both seed lots from r6 stage (JD 49 and 56) from the beginning of shading showed a reduction of 12.3 and 10.2%, respectively, compared to the control (Table 2).

When evaluating the total seedling length (LTS), results similar to the LMR were observed. For seedling LTS, there was a reduction of 17.7, 8.6 and 7.1%, for seed lots whose shading started in R2 and R6 (JD 49; 56) respectively, when compared to the control (19.19 cm). For seedling dry weight (SDW) it was detected lower averages for seedlings from lots and m that the shading was imposed in stages R3, R4, R5 and R6, whose decreases ranged between 7.0 and 13.3%, in relation to seedlings whose mother plants were not shaded during their development, with SDW of 116.2 mg (Table 2).

Table 2. Electrical conductivity (EC), shoot length (SL), main root length (LMR), total seedling length (LTS) and seedling dry weight (SDW) of soybean produced under different phenological stages of shading imposition.

Phenological stages	EC ($\mu S\ cm^{-1}\ g^{-1}$)	SL (cm)	LMR (cm)	LTS (cm)	SDW (mg $seedling^{-1}$)
V6	57.3 b	8.1 b	12.3 a	20.4 a	118.4 a
R1	65.2 a	9.0 a	11.5 a	20.5 a	111.5 a
R2	59.0 a	7.8 b	8.0 c	15.8 b	112.2 a
R3	53.6 b	9.7 a	11.2 a	20.9 a	108.1 b
R4	60.8 a	9.1 a	11.4 a	20.4 a	100.8 b
R5	60.4 a	8.3 b	12.5 a	20.9 a	104.5 b
R6	50.2 b	7.5 b	10.0 b	17.6 b	104.1 b
R6	53.7 b	7.6 b	10.3 b	17.8 b	112.7 a
T	55.5 b	7.8 b	11.4 a	19.2 a	116.2 a
SMD	7.2	1.4	1.9	2.6	8.8
CV (%)	6.3	8.2	8.5	6.8	4.0
Average	57.3	8.3	10.9	19.3	109.8

*Equal letters in the column do not differ by the Skott-Knott test at p≤ 0.05; T: Witness; DMS: Significant minimum difference; CV (%): Coefficient of variation.

DISCUSSION

The results for TSW of the seeds derived from plants shaded from the vegetative stage (V6) indicates a possible compensating effect, i.e., a smaller number of seeds (data not presented), but with higher mass (Table 1). Thus, it is possible to affirm the possible interference of plant etiolation induced by shading on the number of seeds produced (YANG et al. 2018). According to FRANKLIN & WHITELAM (2005) the reduction in the amount of assimilates sent to the development of reproductive parts may have been used for the growth of stems, reducing the number of seeds. This fact refers to the compensation event, in which the largest seed mass is related to the smallest number of st, in the conditions in which they were produced (LABRA et al. 2017). According to BORRÁS et al. (2004), the assimilated drained seeds the during filling phase are very dependent on the current photosynthesis. This fact may explain the negative results caused by the limitation of light in the R5 and R6 stages of soybean plant development in the TSW of the seeds (Table 1).

The results corroborate the ones observed by BIANCULLI et al. (2016) whose TSW obtained were between 110 and 130 mg by seeds of shaded plants (80% radiation restriction) during the seed filling phase. Also according to them, these values are lower than those observed for seeds produced without shading, in which the weight of each seed was on average 170 mg. This fact was observed for the other seed lots from shaded plants in the present study (Table 1).

The lots of soybean seeds from the plants submitted to shading and control lot presented germination percentages above the minimum standard required by the legislation for the commercialization of seeds, of 80%, (BRASIL 2013) (Table 1). The results for germination of seed lots of shaded plants from the end of the cell division phase and beginning of the deposition of reserves of seeds in formation (R4) indicated lower sensitivity of these to the imposed condition, a fact not visualized for the other lots. This may possibly be due to lower temperatures in the shaded environment (data not presented), during the seed filling period, and presenting beneficial condition germination (KHAN et al. 2011). Similarly, BELLALOUI et al. (2012) observed an increase in soybean seed germination produced in most genotypes, with percentages between 85 and 92%, when using 50% lumen restriction throughout the crop cycle, referring to the beneficial effect of the 10% reduction in temperature.

In addition, the increase in twinning of seeds produced under shading may be related to changes in gibberellin (GA) and abscisic acid (ABA) levels in seeds, because, according to CHEN et al. (2020), seeds obtained from shaded plants (intercropping system) showed higher expression of GA biosynthesis genes and lower for those related to ABA, during their formation, when compared to seeds produced under the sun (monoculture).

The vigor test through the FGC, together with germination, are performed under conditions considered optimal to the initial development of culture, a fact that can justify results similar behavior between the tests for the tested lots (Table 1). However, according to KRZYZANOWSKI et al. (1999) seed lots with lower percentage of normal seedlings in FGC correspond to lower vigor. PICCININ et al. (2012) observed for the FGC of soybean seeds of different genotypes and sieves, values between 68 and 91% of normal seedlings, without obtaining significant difference between treatments. CHEN et al. (2020), when studying the shading effect of soybean plants through the intercropping with corn on the seeds produced, observed greater speed in radicle emission, when they were produced under shaded condition.

The lower vigor visualized by the accelerated aging test of soybean seed lots from shaded plants from the vegetative stage (longer shaded period) (Table 1) indicates that these presented greater deterioration and lower viability under the condition of high humidity and temperature (KRZYZANOWSKI et al. 1999). According to MATERA et al. (2019) the test can be used as a prediction of the performance of soybean seeds in the field as emergence and vigor of seedlings. When testing the physiological quality of soybean seeds classified in 5.5 and 6.5 mm sieves, PICCININ et al. (2012) observed variation for vigor evaluated by the accelerated aging test only for the genotype factor, whose means varied between 50 and 86 classifying in lots of high and low vigor, as in the present study (Table 1).

Seed vigor can also be evaluated through biochemical electrical conductivity test. The high values for EC the test characterize seed lots with less vigor, that is, lower capacity of reorganization of tegument membranes after intake, and high elimination of exudates and ions from inside cells (MARCOS FILHO 2015). CATÃO & CAIXETA (2019) evaluating the electrical conductivity of soybean seeds from seven lots obtained values between 61.44 and 104.33 µS cm-1 $^{g-1}$, and higher values when testing higher temperatures and different soaking periods, numerically higher results than those observed in the present study (Table 2).

Regarding seedling performance, the low vigor observed for lots may be related to lower tissue transformation capacity in seed reserves, and incorporation into the embryonic axis (BEWLEY et al. 2013). The filling of legume seeds is dependent on the amount of photoassimilate produced during this phase (BORRÁS et al. 2004), a fact that can be observed for the SDW of seed lots from shading imposed on plants during stages R3 to R6. HENNING et al. (2010) observed differences between high and low vigor soybean cultivars related to seedling performance, in which for total length, seeds of higher and lower vigor presented an average of 25.3 and 18.7 cm, and for SDW, they were approximately 46.6 and 39.3 mg, respectively. MATHIAS et al. (2019) verified that soybean seeds of cultivar NA 5909 RG presented higher averages for seedling length, with a value of 19.6 cm, and the cultivar NS 5959 IPRO the lowest averages, of only 15.2 cm, when harvested in different periods after physiological maturity.

Considering the results obtained for germination and vigor of soybean seed lots from shaded plants, in general, it was observed that these presented variation in responses by the condition of imposed lumen restriction. According to CARVALHO & NAKAGAWA (2012) seed lots with the same germination potential may present differences in vigor. Thus, a harmful trend of the condition of lumen restriction in plants during the final stages of seed development was observed, under their vigor. However, the production of photoassimilate carried out by the crop during the seed filling period is mainly destined to these (BORRÁS et al. 2004).

With the reduction of the photosynthetic rate by lumen restriction there is a limitation in the availability of sucrose and lipid formation, which results in the increase of protein content in seeds (DELARMELINO-FERRARESI et al. 2014). This fact may explain the greater vigor obtained from the seed lots of plants imposed on the condition at the beginning of the period in question, because higher protein levels are related to the higher vigor of the seeds (MATHIAS et al. 2019). In addition, the temperature may have positively influenced the germination potential and vigor of the lot of shaded plants from R4 when compared with the control, because under shaded conditions the ambient temperature is lower, according to BELLALOUI et al. (2012) and KHAN et al. (2011), checks in the present study (data not presented).

CONCLUSION

Shading of 70% tax on soybean plants from the stage of development of the beginning of the formation of pod (R4) provided higher germination potential of the seeds produced.

The vigor of soybean seeds, according to phenological stage and test employed, was affected due to the lumen limitation imposed on plants from the seed filling stages.

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Desempenho inicial de cultivares de feijão submetidas a diferentes profundidades de semeadura

Early performance of common bean cultivars submitted to different sowing depths

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RESUMO

A profundidade de semeadura pode afetar negativamente a emergência, desenvolvimento inicial de plântulas de feijão e consequentemente o estande de plantas. Porém, o desempenho sob diferentes profundidades pode ser variável entre as cultivares. Objetivou-se neste estudo verificar o efeito de diferentes profundidades de semeadura sobre a emergência e o desenvolvimento inicial de plântulas de cultivares de feijão. O experimento foi conduzido em delineamento em blocos completamente casualizado, em esquema fatorial 4 x 4, com quatro repetições. Os fatores consistiram em quatro profundidades de semeadura (4, 8, 10 e 12 centímetros) e quatro cultivares de feijão (TAA Dama, BRS Estilo, IPR Tangará e IPR Tuiuiu). Foram avaliadas as variáveis emergência, índice de velocidade de emergência, comprimento de raiz principal, hipocótilo e epicótilo, e massa seca de plântulas. Observou-se que profundidades de semeadura entre 6 e 10 cm proporcionaram maior emergência de plântulas com 98%, e IVE de 11,3, além de favorecer o desempenho inicial de plântulas de feijão. Não houve diferença entre as cultivares para comprimento de hipocótilo e massa seca de plântulas. Concluiu-se que profundidades de semeadura entre 6 e 10 cm, de solo destorroado e descompactado, proporcionam maior emergência e consequente desempenho inicial de plântulas; no entanto, não havendo efeitos distinto das cultivares de feijão.

PALAVRAS-CHAVE: germinação; emergência; *Phaseolus vulgaris;* genótipo.

ABSTRACT

The sowing depth can negatively affect the emergence and initial development of bean seedlings and, consequently, the plant stand. However, performance under different depths may vary among cultivars. The objective of this study was to verify the effect of different sowing depths on the emergence and initial development of seedlings of bean cultivars. The experiment was conducted in a completely randomized block design, in a 4 x 4 factorial layout, with four replications. The factors consisted of four sowing depths (4 cm, 8 cm, 10 cm, and 12 cm) and four bean cultivars (TAA Dama, BRS Estilo, IPR Tangará, and IPR Tuiuiu). The variables emergence, emergence speed index, main root, hypocotyl, and epicotyl lengths, and seedling dry mass were evaluated. It was observed that sowing depths from 6 cm to 10 cm provided greater seedling emergence with 98% and IVE of 11.3, in addition to favoring the initial performance of bean seedlings. There was no difference between cultivars for hypocotyl length and seedling dry mass. It was concluded that sowing depths from 6 cm to 10 cm of unpaved and unpacked soil provide greater emergence and consequent initial seedling performance; however, there are no distinct effects for bean cultivars.

KEYWORDS: germination; emergency; *Phaseolus vulgaris*; genotype.

INTRODUÇÃO

O feijão pertencente a espécie *Phaseolus vulgaris* L., é o “tipo de feijão” em que o grão é o mais produzido e consumido no mundo (FAO 2019). A leguminosa possui grande destaque no setor agrícola, cujos grãos constituem uma fonte nutricional de proteínas, boa fonte de lisina, fibras alimentares, carboidratos complexos, minerais como cálcio e ferro, além de vitaminas do complexo B (SILVA et al. 2009), apresentado assim notória importância socioeconômica, além de ser uma cultura alternativa promissora (TAVARES et al. 2013).

Segundo estimativas da Companhia Nacional de Abastecimento (CONAB 2019), a produção

brasileira total de feijão na safra 2019/20 deve apresentar redução de 54,3 mil toneladas, correspondente a 1,8% a menos que a safra anterior. Além disto, as previsões apontam para produtividade 0,6% menor comparada com a safra 2018/19, de 1031 kg ha^{-1}. A estimativa de produção da leguminosa no estado de Santa Catarina também demonstra redução, correspondente a 7,8%, sendo a estimativa de produção de 97,4 mil toneladas na safra 2019/20.

Devido ao extenso melhoramento de plantas, o *P. vulgaris* compreende inúmeros cultivares com uma vasta gama de características morfológicas e agronômicas, incluindo diferenças na cor e tamanho da semente, bem como hábito de crescimento (PURSEGLOVE 1968), contudo a média da produtividade brasileira não aumentou mesmo com potencial produtivo elevado de cultivares atualmente comercializadas (BIEZUS et al. 2017).

A profundidade de semeadura e a distribuição de sementes na linha são os fatores que mais afetam o desempenho produtivo. Desta maneira a profundidade adequada de semeadura assegura adequada germinação e a obtenção do estande inicial apropriado ao bom desenvolvimento (RODRIGUES et al. 2016), visto que o *P. vulgaris* tem um sistema radicular baseado na raiz axial com raízes laterais tipicamente localizadas nos 15 cm superiores do solo (PURSEGLOVE 1968). Além disto, a profundidade da deposição das sementes é característica para cada espécie e quando adequada, garante germinação e plântulas uniformes na emergência (TROGELLO et al. 2013).

Profundidades excessivas de semeadura podem impedir a emergência de plântulas devido ao excesso de solo e provocar o esgotamento das reservas da semente; mas por outro lado, semeaduras muito superficiais podem expor a semente a variações ambientais excessivas, como estresse por excesso ou déficit água ou calor, e isso pode dar origem a plântulas pequenas e fracas (SILVEIRA et al. 2018). Portanto, a deposição da semente deve ocorrer em uma profundidade que permita um adequado contato com a umidade do solo, sem irregularidades que afetem a disposição das sementes e que possam provocar perdas futuras de produtividade.

Desta forma, em estudo com a cultura do feijão, BIEZUS et al. (2017) observaram para o grupo de feijão carioca médias superiores nas análises do desempenho de plântulas na maior parte das profundidades testadas (1 a 8 cm), além de indicar a profundidade de semeadura de 2 cm para os grupos Cavalo, Preto, Jalo e Carioca. Já PEDÓ et al. (2014) verificaram para as cultivares IPR Gralha e IPR Tiziu que o aumento da profundidade de semeadura reduz o desempenho das plântulas e a expressão do vigor destas. Contudo, MODOLO et al. (2011) verificaram que a profundidade de 5 cm de semeadura proporcionou melhor índice de velocidade de emergência de plântulas de feijão cultivar UTF 6.

Assim, mais uma vez se ressalta a importância dos estudos que envolvam o vigor de plântulas de diferentes cultivares e o desenvolvimento inicial das plântulas quando submetidas a distintas profundidades de semeadura (PEDÓ et al. 2014). Neste contexto, objetivou-se no presente estudo avaliar os efeitos de diferentes profundidades de semeadura sobre a emergência e o desenvolvimento de plântulas de cultivares de feijão.

MATERIAL E MÉTODOS

O experimento foi realizado em condições de campo, na Universidade do Estado de Santa Catarina - Centro de Ciências Agroveterinárias (UDESC - CAV), no período de 13 de novembro a 06 de dezembro de 2018, no município de Lages, localizado no planalto Sul do estado de Santa Catarina; cujas coordenadas geográficas são 27°52’30” de latitude sul e 50°18’20” de longitude oeste, com altitude média de 930 m. Os dados meteorológicos foram obtidos do Instituto Nacional de Meteorologia, da estação de Lages, SC. As informações de temperatura máxima, média, mínima e precipitação pluvial durante a realização do experimento são ilustrados na Figura 1.

O solo da área experimental é classificado como Cambissolo húmico alumínico (EMBRAPA 2013). As propriedades químicas do solo na camada 0-20 cm, segundo análise de solo realizada em outubro de 2018, foram apresentadas na Tabela 1. A semeadura das cultivares de feijão e adubação foi realizada manualmente, utilizando o fertilizante formulado 02-20-20, com 40 $g.m^{-2}$, de acordo com a Comissão de Química e Fertilidade do Solo – RS/SC (CQFS 2016).

O delineamento experimental adotado foi em blocos casualizados, em esquema fatorial 4 x 4 com quatro repetições. O primeiro fator foi constituído por quatro cultivares de feijão (TAA Dama, BRS Estilo, IPR Tangará e IPR Tuiuiu) e o segundo por quatro profundidades de semeadura (4 cm, 8 cm, 10 cm e 12 cm), totalizando 64 parcelas experimentais. As sementes utilizadas foram adquiridas e eram classificadas como categoria S1 (TAA Dama e IPR Tangará) e S2 (BRS Estilo e IPR Tuiuiu), provenientes da safra agrícola de produção 2017/18.

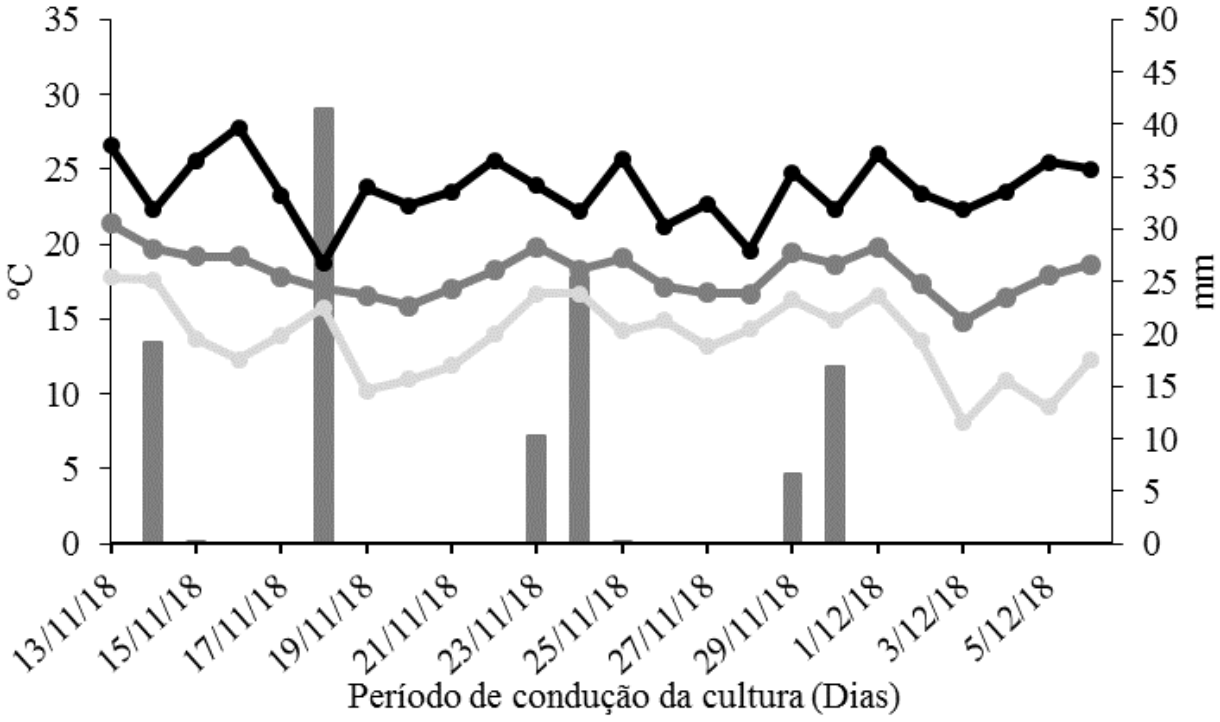

Figura 1. Precipitação pluviométrica e temperaturas (máxima, média e mínima) diárias durante o experimento. Lages, SC, 2018. Fonte: INMET 2019.

Figure 1. Daily rainfall and temperatures (maximum, average, and minimum) during the experiment. Lages, SC, Brazil, 2018. Source: INMET 2019.

Tabela 1. Análise físico-química do solo da área experimental da UDESC-CAV. Lages, SC, 2018.

Table 1. Physicochemical analysis of the soil of the experimental area of UDESC-CAV. Lages, SC, Brazil, 2018.

Amostras	Argila	V	M.O	pH (H_2O)	P Mehlich-1	K	Ca	Mg	H^++Al^{+3}	CTC (pH 7)
	----------%----------				---mg.dm^{-3}---		----------cmol$_c$.dm^{-3}----------			
0-20 cm	47	79	2,7	5,8	20,2	1,8	7,7	5,7	3,5	17,3

Cada unidade experimental foi constituída por uma linha de semeadura contendo 50 sementes. O espaçamento entrelinhas utilizado foi de 17 cm. O preparo do solo foi realizado de forma manual, com a retirada do solo até a profundidade desejada, conforme descrito por PINHEIRO et al. (2018). Posteriormente, realizou-se a adubação à lanço, semeadura manual e, então, a cobertura das sementes com solo peneirado em malha com crivo de 2 x 2 cm, com quantidade de solo correspondente a cada tratamento. O controle de plantas daninhas foi efetuado manualmente e o suprimento de água foi realizado via chuva e complementado via irrigação diária conforme a necessidade.

Quando as plântulas começaram a emergir, iniciaram-se as contagens diárias destas, para avaliação do percentual de emergência e cálculo de IVE (índice de velocidade de emergência) (MAGUIRE 1962); para avaliação do comprimento de plântulas utilizou-se a metodologia proposta por NAKAGAWA (1999). Tais avaliações foram realizadas até o momento em que houve o estabelecimento da população sem alteração no número de plântulas emergidas por três dias consecutivos de avaliação. Após esta data, procedeu-se a coleta de plântulas, coletando-se 20 plântulas por parcela, medindo-se o comprimento total das plântulas normais com auxílio de uma régua, comprimento da parte aérea (epicótilo), hipocótilo e raiz, cujos resultados foram expressos em centímetros. Após estas avaliações, tanto as partes aéreas quanto as raízes foram submetidas a secagem até massa constante, em estufa regulada a 60 °C por 72 horas.

Os resultados obtidos foram submetidos à análise de variância e avaliados pelo teste F a 5% de significância. Os dados qualitativos (cultivares) significativos foram submetidos ao teste de médias de Tukey a 5% de probabilidade de erro e os dados quantitativos (profundidades) a ajustes de regressão quando houve diferença significativa. Utilizou-se o programa estatístico R (R-CORE TEAM 2017).

RESULTADOS

A análise de variância indicou existir efeito simples de cultivar e de profundidade para as variáveis testadas (Tabela 2). A emergência das plântulas de feijão variou conforme a cultivar, sendo IPR Tangará, o genótipo com menor percentual, de 78%. Já para as cultivares TAA Dama, BRS Estilo e IPR Tuiuiu, o percentual de emergência foi superior, atingindo média de 90% (Figura 2a). A profundidade de semeadura apresentou ajuste quadrático, com acréscimos no percentual de emergência nas profundidades de 8 e 10 cm (93%), e redução em 12 cm (69%). A profundidade estimada com maior percentual de emergência (98%) para as cultivares de feijão foi em 6,7 cm (Figura 2b).

Tabela 2. Resumo da ANOVA para percentual de emergência (EM), índice de velocidade de emergência (IVE), massa seca total da planta (MST), epicótilo (EPI), hipocótilo (HIP), raiz principal (CR) e comprimento total de plântula (CTP) de cultivares de feijão cultivadas em diferentes profundidades de semeadura.

Table 2. Summary of ANOVA for emergence percentage (EM), emergence speed index (IVE), total plant dry mass (MST), and epicotyl (EPI), hypocotyl (HIP), main root (CR), and total seedling (CTP) lengths of bean cultivars cultivated at different sowing depths.

Fontes de Variação	GL	QM				
		EM	IVE			
Bloco	1	0,00	0,85			
Cultivar (C)	3	0,07$^{**/}$	6,10**			
Profundidade (P)	3	0,20**	34,00**			
C×P	9	0,01	0,77			
Erro	15	0,00	0,39			
CV (%)		5,54	6,57			
Média		87,00	9,15			
Fontes de Variação	GL	MST$^{¥}$	EPI$^{¥}$	HIP$^{¥}$	CR$^{¥}$	CTP
Bloco	3	0,17	0,01	0,11**	0,07	7,23
Cultivar (C)	3	0,25	0,18**	0,02	0,43**	19,95*
Profundidade (P)	3	1,31**	1,37**	0,24**	2,98**	244,77*
C×P	9	0,19	0,06	0,01	0,02	4,23
Erro	45	0,09	0,04	0,02	0,08	5,74
CV (%)		10,22	6,43	9,08	8,23	11,01
Média		8,72	8,76	2,14	10,97	21,77

* e ** significativos pelo teste F a 5 e 1%, respectivamente. GL: Graus de Liberdade; QM: Quadrado Médio; CV (%): Coeficiente de Variação. $^{¥}$Valores transformados por $(x+0,5)^{0,5}$.

Um comportamento semelhante ao da emergência foi observado para o índice de velocidade de emergência (IVE). Houve variação no IVE conforme a cultivar testada bem como em função da profundidade de semeadura. A cultivar BRS Estilo se destacou com maior IVE (9,9), seguida de IPR Tuiuiu. Já a cultivar IPR Tangará apresentou menor valor, sendo estes 24,4 % menor em comparação com a cultivar BRS Estilo (Figura 2c). Em relação à profundidade, o IVE apresentou comportamento quadrático, com o maior índice (IVE de 11,4) em 7,3 cm de profundidade de semeadura (Figura 2d).

O comprimento da raiz principal (CR) das plântulas de feijão apresentou diferença entre as cultivares. Para a cultivar TAA Dama observou-se redução de 22,2% no CR, em relação as cultivares BRS Estilo, IPR Tangará e IPR Tuiuiu (Figura 3a). Para o fator profundidade de semeadura, os resultados apresentaram comportamento quadrático, com maior valor para o comprimento de raiz principal (12,7 cm) obtido sobre cultivos a 10,5 cm de profundidade de semeadura (Figura 3b).

Para o comprimento do epicótilo das plântulas, IPR Tuiuiu apresentou maiores valores, não diferindo apenas da cultivar TAA Dama e IPR Tangará. Já para o genótipo BRS Estilo observou-se redução de 15,3% no comprimento quando comparado a cultivar IPR Tuiuiu (Figura 3c). Quanto ao efeito da profundidade de semeadura, observou-se ajuste quadrático, com acréscimos no comprimento do epicótilo das plântulas de feijão, sendo o máximo valor de 8 cm obtido na semeadura em 10,6 cm de profundidade (Figura 3d).

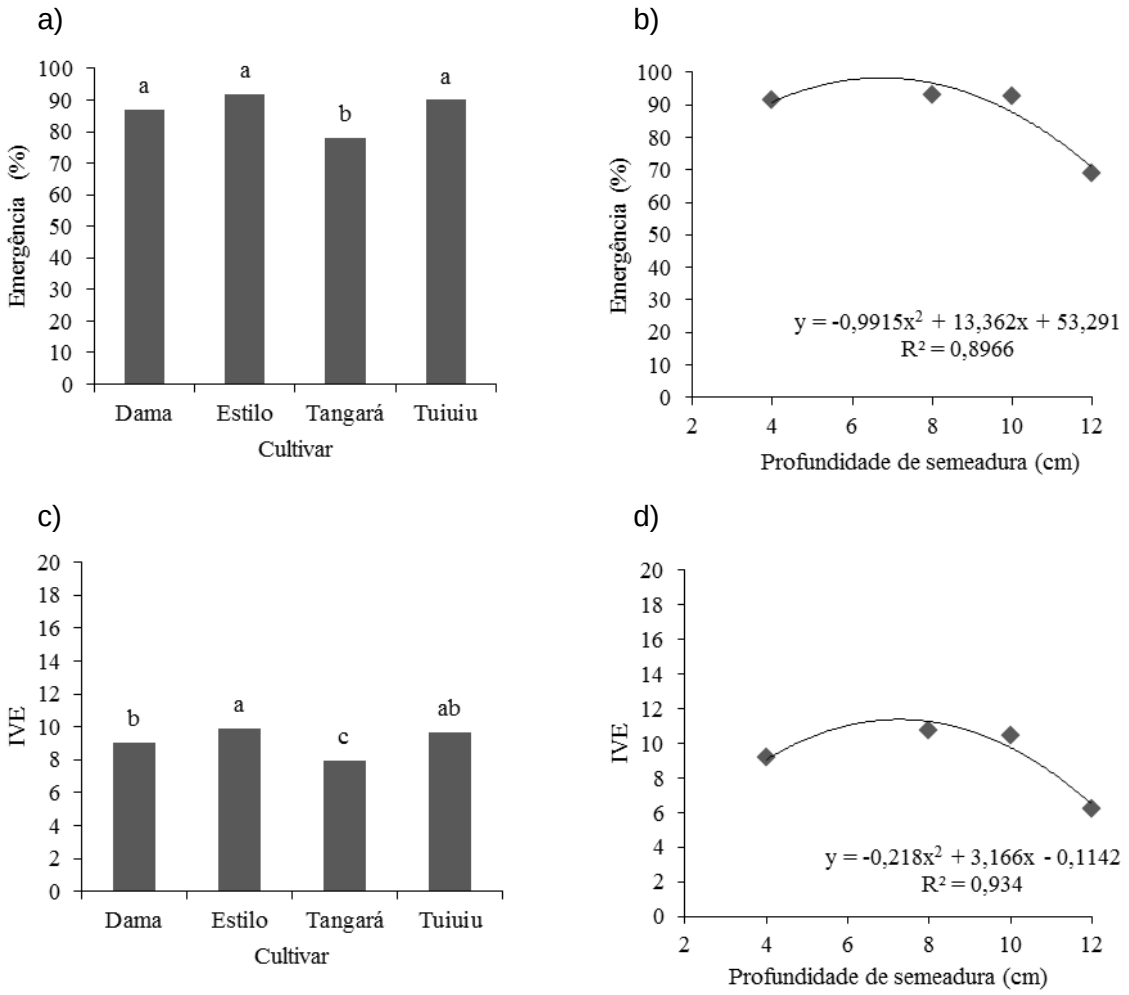

Figura 2. Percentual de emergência (a; b), e índice de velocidade de emergência (c; d) de plântulas de feijão em função de cultivares e de profundidades de semeadura.

Figure 2. Percentage of emergence (a; b) and emergence rate index (c; d) of bean seedlings as a function of cultivars and sowing depths.

O comprimento do hipocótilo foi afetado apenas pela profundidade de semeadura. Na média, as cultivares apresentaram valor de hipocótilo de 2,1 cm (Figura 3e). Observou-se comportamento linear com decréscimo de 0,10 cm no comprimento do hipocótilo das plântulas de feijão para cada um centímetro de aumento da profundidade de semeadura (Figura 3f).

A altura das plântulas de feijão apresentou comportamento variável entre as cultivares. A cultivar IPR Tuiuiu apresentou acréscimo na altura de plântulas, com média de 23,3 cm. Para a cultivar TAA Dama observou-se redução de 12,6 % na altura de plântulas, em relação a IPR Tuiuiu (Figura 4a). Em relação a profundidade, houve efeito quadrático, com os valores de maior altura observados na semeadura em 8 e 10 cm, com 23,7 e 25,9 cm. Plântulas de menor altura foram obtidas quando as semeaduras ocorreram em 12 e em 4 cm, com reduções de 26,2 e 53,2 % respectivamente, quando comparadas com a altura das plântulas com semeadura em 8 cm (Figura 4b).

A massa seca total (MS) das plântulas de feijão não apresentou diferença entre as cultivares, sendo que a média foi de 8,7 g (Figura 4c). Em relação as profundidades, a MS apresentou efeito simples e com comportamento quadrático, sendo o maior acúmulo de MS pelas plântulas obtido na profundidade de semeadura de 8,7 cm resultando em 10,1 g (Figura 4d).

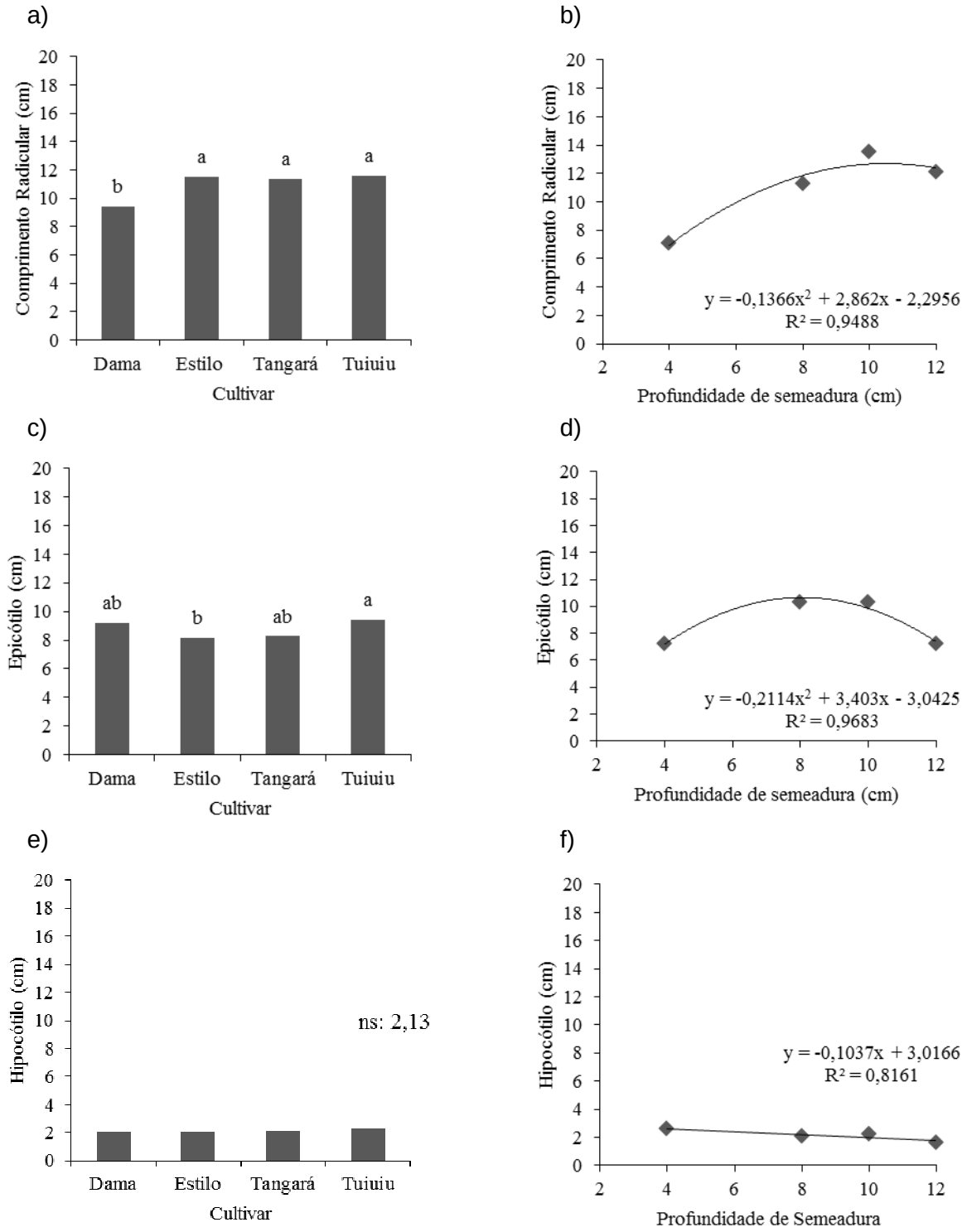

Figura 3. Comprimento de raiz (a; b), de epicótilo (c; d) e hipocótilo (e; f) de plântulas de feijão em função de cultivares e profundidades de semeadura.

Figure 3. Root (a; b), epicotyl (c; d), and hypocotyl (e; f) lengths of common bean seedlings as a function of cultivars and sowing depths.

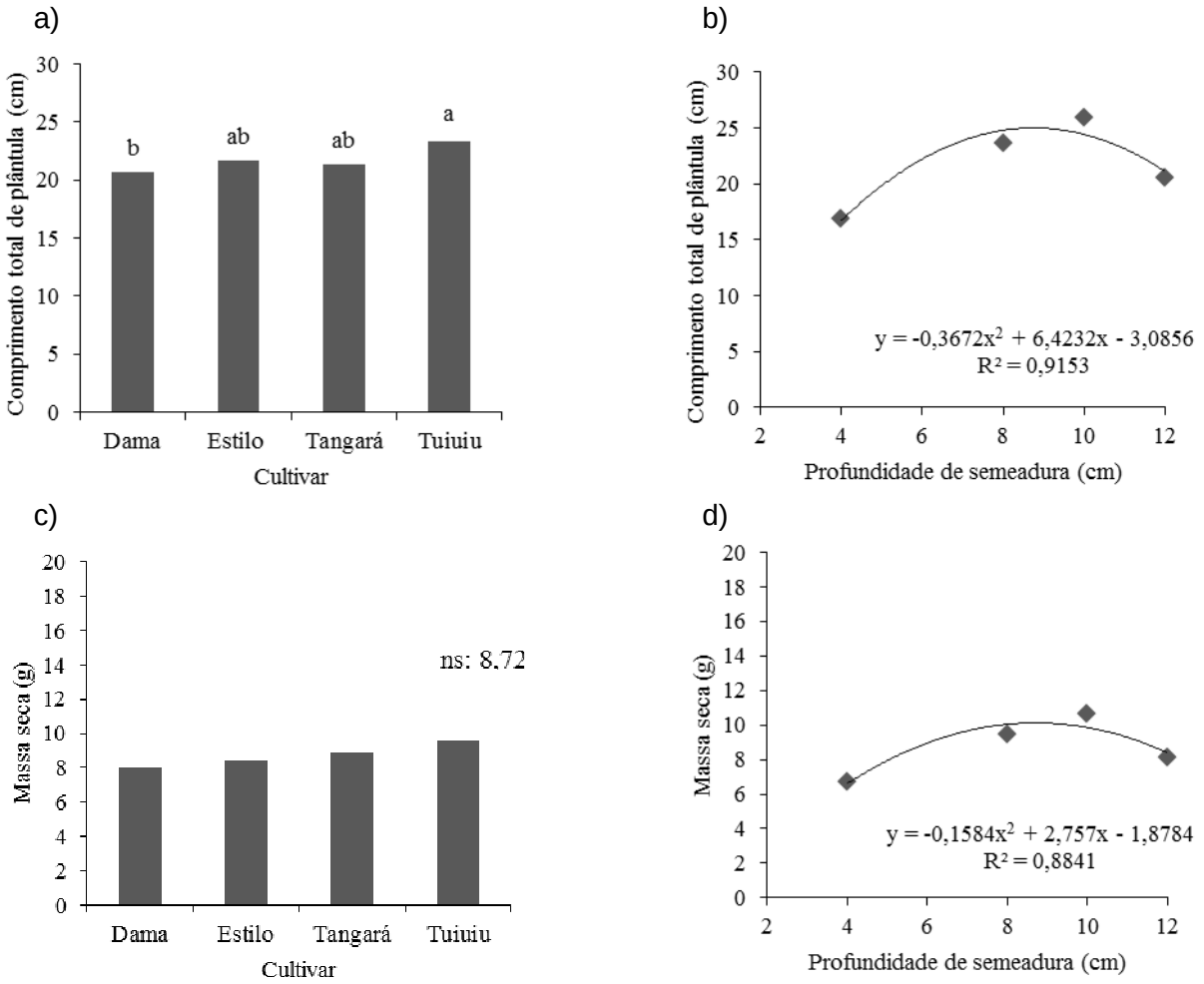

Figura 4. Comprimento total de plântulas (a; b) e massa seca (c) de plântulas de feijão em função de cultivares e profundidades de semeadura.

Figure 4. Seedling height (a; b) and dry weight (c) of common bean seedlings as a function of cultivars and sowing depths.

DISCUSSÕES

A profundidade de semeadura é específica para cada cultivar, devendo proporcionar germinação e emergência adequada às plântulas (SOUSA et al. 2007, PEDÓ et al. 2014). Neste aspecto, o desempenho apresentado entre as cultivares de feijoeiro testadas diferiu quanto aos parâmetros avaliados, como já observado por BIEZUS et al. (2017).

Conforme as observações para a emergência e IVE das plântulas de feijão, pode-se inferir sobre a interferência de fatores ambientais como temperatura e disponibilidade hídrica, além da espessura de camada de solo sobre estas. Ou seja, sob profundidades rasas as sementes ficam expostas a maior variação de temperatura e umidade, e sob semeaduras mais profundas apresentam-se limitadas pela maior camada de solo, prejudicando a emergência de plântulas ainda frágeis (TILLMANN et al. 1994, SILVA et al. 2008). Assim, a deposição das sementes deve ocorrer em uma profundidade que permita bom contato com solo úmido, mas com adequado fornecimento de oxigênio, promove boa emergência de plântulas (SILVA et al. 2008). Ademais, em profundidades maiores, segundo PEDÓ et al. (2014) a menor emergência possui relação com ao maior obstáculo físico de camadas de solo espessas, prejudicando o estabelecimento, a habilidade competitiva e o potencial produtivo da espécie.

Além disto, o vigor das sementes está relacionado com as reservas destas, e com a capacidade de expressão deste para romper as barreiras físicas, como camadas maiores de solo (PESKE et al. 2012). Ou seja, genótipos com maior capacidade de translocar e armazenar nutrientes nas sementes, apresenta maior potencial de germinação e vigor de plântulas quando sob condições adversas (MARCOS FILHO 2015). Ademais, conforme TEIXEIRA et al. (2010) observaram em sementes de feijão-caupi, a qualidade fisiológica é variável conforme cultivares. Isto pode estar relacionado aos resultados de emergência e IVE observados

para a cultivar de feijão IPR Tangará, principalmente.

Resultados semelhantes aos observados foram encontrados por ALVES et al. (2014) para emergência e IVE de plântulas de *Phaseolus lunatus* L., cujos valores foram maiores em profundidades menores, entre 2 e 3 cm, variando conforme a posição do hilo das sementes no solo. Já MODOLO et al. (2011) observaram maior IVE em 5 cm quando comparado às profundidades de 3 e 7 cm.

Já BIEZUS et al. (2017) verificaram que grupos comerciais Cavalo e Carioca apresentaram diferença no percentual de emergência em relação ao Preto e Jalo, com valores de 80% e 60%, respectivamente, na profundidade de semeadura de 4 cm, e em relação ao IVE, observaram aumento para os grupos Carioca, Preto e Jalo em profundidades de até 2 cm, e posteriormente decréscimos com o aumento da profundidade, e para o grupo Cavalo, com redução acentuada do IVE a profundidade de 1 cm. Já conforme os estudos de ALI & IDRIS (2015), não houve efeito significativo de semeaduras em profundidades de 5 e 10 cm sobre a emergência de plântulas de *Vicia faba* L..

O resultado para o máximo comprimento da raiz principal das plântulas na profundidade de semeadura de 10,5 cm pode estar relacionado ao suporte da planta em absorver água e nutrientes além de sintetizar carboidratos (HUGHES et al. 1992). Plântulas de *Erythrina verna* apresentaram redução no comprimento radicular de 9,6 cm para 7,2 cm com o aumento da profundidade de semeadura de 1 cm para 6 cm (PÊGO et al. 2015). Já para comprimento da radícula de plântulas de *Vicia faba* L. com semeaduras em profundidades de 5 e 10 cm não houve efeito significativo (ALI & IDRIS 2015).

As observações para o comprimento do epicótilo permitem inferir que pode ter ocorrido estiolamento das plântulas oriundas da semeadura nas profundidades de 8 e 10 cm, pela maior emergência, IVE e altura de plântulas. Ademais, as plântulas necessitam de hastes mais longas para superar a superfície do solo quando da sua colocação em maior profundidade (NÓBREGA & VIEIRA 1995). Semelhante ao verificado por PÊGO et al. (2015) em plântulas de *Erythrina verna*, em que houve aumento do epicótilo em semeaduras em profundidades superiores a 3 cm.

Os resultados negativos do aumento da profundidade de semeadura no comprimento do hipocótilo das plântulas de feijão indicam a diminuição da capacidade de emergir em profundidades maiores. Camadas de solo sobrepostas podem se tornar limitações ao desenvolvimento do hipocótilo, pois este deve crescer em comprimento para superar a profundidade de semeadura, e em diâmetro para adquirir força e habilidade para vencer as resistências provocadas pelo peso dos cotilédones e pelo solo (COSTA et al. 1999). Segundo KNITTLE & BURRIS (1979) as plântulas respondem aos obstáculos físicos do solo, com aumento do diâmetro e diminuição do comprimento do hipocótilo. Tais obstáculos são distintos em função da profundidade de semeadura.

A altura das plântulas de feijão cultivadas sob maiores profundidades de semeadura pode ter apresentado interferência devido a maior barreira física para emergência, além de maior demanda energética (BIEZUS et al. 2017). MODOLO et al. (2011) não encontraram diferenças ao avaliar profundidades de semeaduras de 3, 5 e 7 cm na cultura do feijão, embora observaram a tendência de plântulas com maior altura na profundidade de 5 cm. Contudo, BIEZUS et al. (2017) observaram reduções drásticas na altura de plântulas de feijão com semeaduras a partir de 2 cm de profundidade após 9 dias da semeadura.

Os resultados para a massa seca de plântulas (MSP) explicam que o acréscimo em profundidades, acima de 8,7 cm de semeadura interfere na produção de massa seca da cultura, resultando em maior gasto energético realizado pelas plântulas para romper as camadas inferiores de solo, tornando a planta debilitada, com menor acúmulo de matéria seca (GROTTA 2008). Semelhante ao encontrado por ALI & IDRIS (2015) em que a massa seca de plântulas de *Vicia faba* L. com semeaduras em profundidades de 5 e 10 cm, não apresentaram efeitos significativos. Contudo, MODOLO et al. (2011) observaram maior MSP de parte aérea de plântulas de feijão na profundidade de semeadura de 5 cm, devido provavelmente à maior altura destas, e não observou diferença para a MS de raiz, embora com tendência de aumento da MSP em maiores profundidades de semeadura. Assim como com plântulas de *Erythrina verna* em que houve redução da MSP com profundidades de até 6 cm (PÊGO et al. 2015).

As respostas observadas para o desempenho fisiológico das sementes e desenvolvimento das plântulas das diferentes cultivares de feijão testadas no presente estudo, difere dos obtidos por PINHEIRO et al. (2018) com cultivares de trigo e triticale, e por TEIXEIRA et al. (2010) com feijão-caupi, o que pode estar relacionado às condições do solo do experimento. Assim, destaca-se a relevância de estudos que avaliem o desempenho dos genótipos, definindo profundidades de semeaduras adequadas a cada espécie e cultivar.

Contudo, considerando que o experimento foi realizado com solo peneirado, os valores ideais de

profundidade que resultaram em melhor desempenho germinativo e do desenvolvimento de plântulas foram ligeiramente superiores aos encontrados nas indicações técnicas de cultivo de feijão. Fato este justificado pela condição do experimento ter sido com solo destorroado e peneirado. Cabe também salientar que o feijão, assim como a soja são espécies que apresentam germinação epígea havendo a necessidade de extensão do hipocótilo no interior do solo a empurrar os cotilédones a efetivação do processo de emergência.

CONCLUSÃO

Concluiu-se que profundidades de semeadura entre 6 e 10 cm proporcionam maior emergência e desempenho inicial de plântulas, nas condições deste estudo.

As cultivares de feijão apresentam comportamento similar em todas as profundidades de semeadura.

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Qualidade fisiológica de sementes de arroz submetidas à solução de hipoclorito de sódio

Physiological quality of rice seeds submitted to sodium hypochlorite solution

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RESUMO

Objetivou-se verificar a eficiência do hipoclorito de sódio (NaClO) na superação de dormência e na qualidade fisiológica de sementes de arroz. O experimento foi conduzido no Laboratório de Análise de Sementes, no Centro de Ciências Agroveterinárias da Universidade do Estado de Santa Catarina (CAV/Udesc). Utilizaram-se sementes de arroz cultivar SCS122 Miura, com delineamento experimental inteiramente casualizado e quatro repetições, sendo os tratamentos: T1 – sementes dormentes imersas em NaClO; T2 – sementes com dormência superada, em NaClO; T3 – sementes dormentes, em água; T4 – sementes com dormência superada, em água. O NaClO provocou aumento da condutividade elétrica (CE) para sementes com e sem dormência de 28,77 e 26,24 μS cm^{-1}.g^{-1} respectivamente, quando comparadas à imersão em água, indicando degradação das células. Sementes com e sem dormência imersas em NaClO apresentaram, respectivamente, aumento de 6,52 e 10,54 para o índice de velocidade de germinação (IVG) em relação às dormentes em água (9,2). Para a primeira contagem de germinação, a amostra com dormência em NaClO apresentou aumento de 5% em relação à imersão em água. No teste de germinação, obteve-se incremento de 5,81% para ambas as amostras com NaClO, comparadas a sementes com dormência em água (86%). Concluiu-se que o método de imersão em NaClO e em água promoveu a superação da dormência em arroz, mas afetou o vigor das sementes no teste de CE.

Palavras-chave: dormência; germinação; *Oryza sativa*.

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ABSTRACT

The aim of this study was to verify the efficiency of NaClO in overcoming dormancy and the physiological quality of rice seeds. The experiment was conducted at the Laboratory of Seed Analysis, CAV/ UDESC. Rice cultivar SCS122 Miura seeds were used, with completely randomized experimental design and four replicates. The treatments were: T1 – dormant seeds immersed in NaClO; T2 – seeds with dormancy overcome, in NaClO; T3 – dormant seeds in water; T4 – seeds with dormancy overcome, in water. NaClO increased the EC (electrical conductivity) for seeds with and without dormancy of 28.77 and 26.24 μS cm^{-1}.g^{-1}, respectively, when compared to immersion in water, indicating cells degradation. Seeds with and without dormancy immersed in NaClO presented, respectively, an increase of 6.52 and 10.54 for GSI (Germination speed index) in relation to the dormant seeds in water (9.2). For the first germination count, the sample with dormancy in NaClO showed an increase of 5% in relation to immersion in water. In the germination test, an increase of 5.81% was obtained for both samples with NaClO, compared to seeds with dormancy in water (86%). It was concluded that the immersion method in NaClO and in water promoted the overcoming of dormancy in rice, but affected the vigor of the seeds in the EC test.

Keywords: dormancy; germination; *Oryza sativa*.

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INTRODUÇÃO

O adequado e satisfatório estabelecimento das culturas a campo é dependente, entre outros fatores, da qualidade das sementes utilizadas. Elas são submetidas às análises a fim de verificar se suas características se encontram em conformidade com o padrão de qualidade definido pela legislação para, posteriormente, serem comercializadas aos agricultores (MARCOS FILHO, 2015).

O arroz (*Oryza sativa* L.) é uma cultura com relevante importância econômica para o Brasil e o mundo. Segundo dados da FAO (2018), a China é o maior produtor de arroz, seguido pela Índia e pela Indonésia, e o Brasil é classificado como o 9.º colocado. O consumo médio por pessoa chega a 60 kg/ano, exceto em países asiáticos, onde o consumo pode variar de 100 a 150 kg/ano/pessoa (SOSBAI, 2014). Segundo a Companhia Nacional de Abastecimento (CONAB, 2018), o Brasil produziu mais de 11,0 milhões de toneladas na safra 2017/2018.

As sementes de arroz apresentam dormência logo após a colheita, o que pode ser induzido pelas condições ambientais, como temperaturas baixas nos estádios iniciais da maturação e próximas a 30°C após a floração, além de inibidores e estruturas que impedem a germinação (MENEZES *et al.*, 2009). Taiz *et al.* (2017) definem a dormência como uma condição morfológica e/ou fisiológica de uma semente privada de sua germinação, mesmo estando em condições ambientais aptas para que esta ocorra.

Nessa cultura, a dormência pode ocorrer em virtude da presença de compostos fenólicos que inibem a germinação e pela limitação de oxigênio para o embrião, provocada pelo tegumento (MENEZES *et al.,* 2009). A impermeabilidade do pericarpo das sementes do cereal pode ser explicada pela sua estrutura e composição química, que impedem a passagem de água e oxigênio ao embrião, o próprio consumo de oxigênio pelo tegumento e a presença de compostos fenólicos que também o retêm na cobertura (MARCOS FILHO, 2015).

A presença de dormência dificulta as análises de germinação e vigor voltadas à comercialização das sementes (MENEZES *et al.,* 2009), além de impedir a antecipação da semeadura pelos produtores (STINGHEN, 2015). Contudo, após um período de armazenamento, entre 90 e 120 dias, a dormência das sementes de arroz é superada naturalmente (BEWLEY & BLACK, 1994).

Entre os métodos de superação da dormência em sementes de arroz descritos nas *Regras para análise de sementes* (RAS) (BRASIL, 2009), há o método do hipoclorito de sódio (NaClO). O NaClO é um componente empregado também nos laboratórios para assepsia de sementes ou outros materiais (CARNELOSSI *et al.*, 1995). Considerado um potente oxidante, que remove ou oxida os compostos inibidores da germinação das sementes, o NaClO modifica as membranas do tegumento das sementes, deixando-as mais porosas, permitindo que o oxigênio alcance o interior da semente e ocorra o desenvolvimento do embrião (HSIAO & QUICK, 1984).

Todavia sementes de arroz com dormência e sementes com dormência já superada são submetidas ao método do hipoclorito de sódio por padronização, não havendo evidências quanto ao seu efeito sobre a qualidade fisiológica de ambas, assim como na germinação e no vigor. Dessa forma, objetivou-se verificar a eficiência da utilização da solução de NaClO na superação de dormência de sementes de arroz e as respostas fisiológicas em sementes com dormência superada naturalmente pelo armazenamento.

MATERIAL E MÉTODOS

O experimento foi conduzido no Laboratório de Análise de Sementes (LAS), localizado no Centro de Ciências Agroveterinárias da Universidade do Estado de Santa Catarina (CAV/Udesc),

no município de Lages (SC). O período de condução foi de abril a junho de 2018. Utilizaram-se sementes do cultivar de arroz SCS 122 Miura, proveniente da safra 2017/2018 (recém-colhidas), caracterizadas por apresentar dormência, e da safra 2016/2017 (com um ano de armazenamento), com dormência superada pelo período de armazenamento (sem dormência).

O delineamento experimental empregado foi inteiramente casualizado (DIC), com quatro tratamentos e quatro repetições, sendo os tratamentos: T1 – sementes com dormência submetidas à concentração de 0,5% da solução de NaClO/24 horas; T2 – sementes sem dormência submetidas à concentração de 0,5% de NaClO/24 horas de imersão; T3 – sementes com dormência submetidas à imersão em água/24 horas; T4 – sementes sem dormência submetidas à imersão em água/24 horas.

Previamente às análises com o uso do hipoclorito de sódio, realizou-se o teste de germinação (BRASIL, 2009), para verificar o percentual de dormência das sementes de ambas as amostras. Assim, nessa etapa inicial, não se submeteram as sementes à pré-embebição para posterior realização do teste de germinação.

A amostra média das sementes dormentes e das sementes sem dormência foi composta por 1.400 g, conforme as recomendações de Brasil (2009). As amostras de trabalho de ambas as sementes foram obtidas pela homogeneização e subdividindo-as em quatro repetições. De cada uma destas, retiraram-se duas amostras de 5 g, uma para imersão em água e outra para imersão em NaClO. Desse modo, obtiveram-se oito subamostras para sementes dormentes e oito para sementes sem dormência.

Quatro dessas subamostras de ambas as sementes foram colocadas em imersão em 50 ml de água, e as quatro restantes em 50 ml da solução de NaClO a 0,5% conforme os tratamentos, em caixas gerbox. Posteriormente, permaneceram em estufa a 40°C por 24 horas e, após esse período, foram lavadas com água destilada para retirar possíveis resíduos do sal e, então, submetidas aos testes relacionados à germinação e condutividade elétrica (BRASIL, 2009).

Para a avaliação da qualidade fisiológica das sementes com e sem dormência submetidas à pré-embebição com água e NaClO, efetuaram-se as seguintes análises:

- Teor de água: secagem em estufa a 105°C, por período de 24 horas (BRASIL, 2009);
- Condutividade elétrica (CE): conforme descrita por Vieira & Krzyzanowski (1999), realizada com quatro repetições de 50 sementes de arroz por tratamento, imersas em 75 ml de água deionizada e mantidas sob temperatura de 25°C por 24 horas. Após esse período, utilizou-se um condutivímetro para verificar a condutividade da água de cada amostra. Os resultados foram obtidos pela fórmula: CE = (CE medida da amostra com sementes - CE medida da água pura) / peso da amostra (valores finais apresentados em $\mu S.g^{-1}.cm^{-1}$);
- Percentual de germinação: utilizaram-se oito repetições de 50 sementes para cada amostra e imersão, semeadas em rolos de papel umedecidos com água destilada, mantidas em germinador a 25°C e avaliadas aos 14 dias após o início do teste (BRASIL, 2009) (porcentagem de plântulas normais);
- Primeira contagem de germinação (PC-GER): efetivada juntamente com o teste de germinação, computando-se as plântulas normais após cinco dias da instalação do teste (BRASIL, 2009) (porcentagem de plântulas normais);
- Índice de velocidade de germinação (IVG): foi realizado com contagens diárias de plântulas normais no teste de germinação com intervalos de 24 horas a partir da constatação da primeira plântula normal e calculado conforme Maguire (1962) pela equação IVG = (G1/N1)

+ (G2/N2) + (Gn/Nn), em que G é o número de plântulas germinadas observadas em cada contagem, e N, o número de dias da semeadura a cada contagem.

Os dados obtidos foram submetidos à análise de variância e teste F, aplicando-se o teste de Tukey ao nível de 5% de probabilidade quando verificada diferença significativa entre os tratamentos.

RESULTADOS E DISCUSSÃO

As análises de germinação realizadas inicialmente à imersão em água e hipoclorito de sódio (NaClO), com as sementes de arroz cultivar SCS122 Miura com dormência, apresentaram como resultado: percentual de germinação de 32%, plântulas anormais (22%), sementes dormentes (35%) e mortas (11%); e amostra com dormência superada naturalmente pelo período de armazenamento (sem dormência): 93% de germinação, 4% de plântulas normais, 2% de sementes dormentes e 1% de mortas. Nesse teste, obtiveram-se diferença e baixos percentuais de plântulas anormais, além de sementes dormentes e mortas para a amostra de sementes com dormência, possibilitando a realização dos testes posteriores.

Ambas as amostras das sementes apresentaram teor de água de 14%, caracterizado como adequado para o armazenamento, pois preserva a viabilidade e a longevidade das sementes (CARVALHO & NAKAGAWA, 2012).

Os resultados da análise de variância referente às avaliações para condutividade elétrica (CE), primeira contagem (PC-GER), germinação (GER) e índice de velocidade de germinação (IVG) demonstram que houve diferença significativa para os tratamentos nas análises de condutividade elétrica, primeira contagem de germinação e IVG. Para as variáveis plântulas anormais (PA), sementes dormentes (SD) e sementes mortas (SM), os coeficientes de variação obtidos foram altos, por conta da maior dispersão nos dados e da presença de valores iguais a zero (tabela 1).

Tabela 1 – Resumo da Anova para análises de primeira contagem de plântulas normais (PC-GER), germinação (GER), plântulas anormais (PA), sementes dormentes (SD), sementes mortas (SM), índice de velocidade de germinação (IVG) e condutividade elétrica (CE) em amostras de sementes de arroz com dormência e sem dormência, submetidas à imersão em hipoclorito de sódio (NaClO) e água (H_2O).

Fonte de variação	QUADRADO MÉDIO						
	PC-GER	GER	PA	SD	SM	IVG	CE
Tratamentos	437,46**	64,46 ns	3,46 ns	31,13 ns	3,13 ns	3,85**	759,91**
Erro	68,16	41,48	11,73	12,48	6,05	0,69	0,15
CV* (%)	10,15	7,14	81,8	92,67	135,75	8,31	2,48
Média geral	81,31	90,18	4,19	3,81	1,81	10,02	15,69

* CV (%): Coeficiente de variação.
** Significativo.
ns: não significativo a p > 0,05 pelo teste F.

Observa-se que houve efeito significativo nos valores de CE para as sementes submetidas ao NaClO, com aumento na CE de 28,77 µS $cm^{-1}.g^{-1}$ para a amostra com dormência e 26,24 µS $cm^{-1}.g^{-1}$ para a amostra de sementes sem dormência, quando comparadas à imersão em água, cuja CE obtida foi de 0,67 e 3,20 µS $cm^{-1}.g^{-1}$, respectivamente (figura 1a).

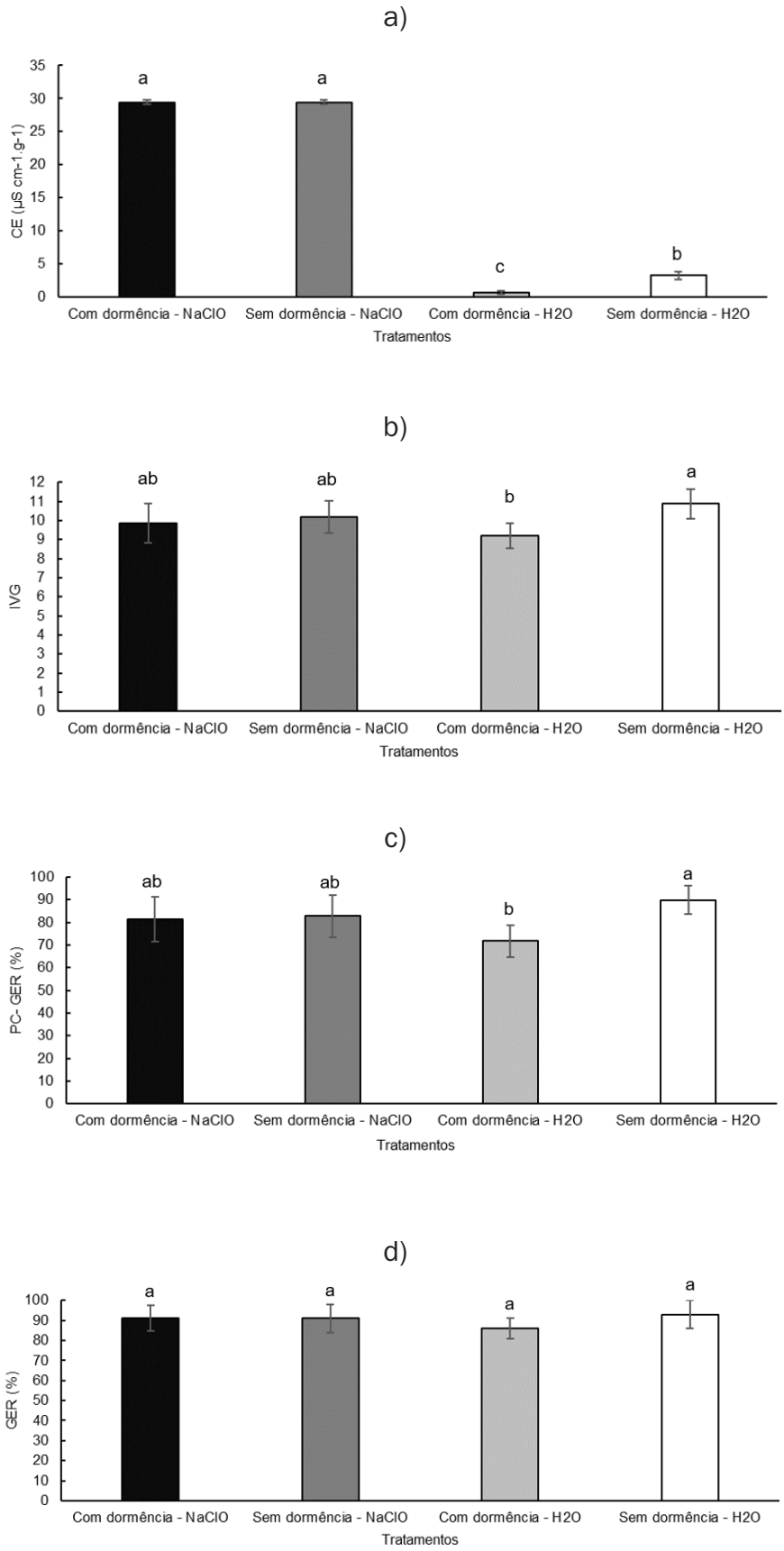

Figura 1 – Testes de condutividade elétrica (CE) (a), índice de velocidade de germinação (IVG) (b), primeira contagem de germinação (PC-GER) (c) e germinação (GER) (d), para amostras de sementes de arroz com dormência e sem dormência, submetidas à imersão em hipoclorito de sódio (NaClO) e água (H_2O).

*Médias com letras iguais minúsculas não diferem significativamente pelo teste de Tukey a 0,05 de significância.

Observaram-se, com o teste de CE, maior extravasamento de solutos das sementes, provavelmente causado pela menor integridade das células da cariopse das sementes, e redução da capacidade dessas membranas de se recompor após a embebição com o NaClO, resultando na redução da qualidade fisiológica das sementes (MARCOS FILHO, 2015). Ressalta-se que a utilização de NaClO também causou danos nas sementes sem dormência, com diferença de 2 μS $cm^{-1}.g^{-1}$ da amostra com dormência. Tais resultados corroboram os de Carnelossi *et al*. (1995), pois esses autores inferem que, embora empregado em laboratórios de análise de sementes para assepsia delas, o poder oxidante do NaClO pode atuar na permeabilidade de células.

A análise para o IVG contabilizada no teste final de germinação demonstrou diferenças significativas (figura 1b). As sementes sob imersão com NaClO com dormência e sem dormência apresentaram respectivamente aumento de 6,52% e 10,54% no índice, quando comparadas à amostra de sementes com dormência com a utilização de água, com valor de IVG de 9,2. As sementes sem dormência sob imersão com água apresentaram maior IVG, de 10,87, explicado pelo período de um ano de armazenamento a que estiveram submetidas desde a colheita. Resultados semelhantes foram encontrados por Stinghen (2015) em sementes de arroz irrigado que superaram a dormência após 90 dias da colheita.

O percentual de plântulas normais na primeira contagem de germinação (PC-GER) (figura 1c) apresentou variações em relação à dormência quando submetidas à imersão em água, na qual as sementes sem dormência (90%) tiveram incremento de 18% em relação às sementes com dormência (72%).

Em relação à germinação, a análise estatística não revelou diferença significativa entre os tratamentos após a imersão das sementes em água e em NaClO (figura 1d), como verificado no teste prévio à imersão, porém obteve-se para a amostra de sementes com dormência imersas em água menor percentual de germinação (86%). Para as amostras de sementes sem dormência imersas em água, notou-se incremento de 7% de germinação, e, para as sementes com e sem dormência em NaClO, houve em ambas incremento de 5%, quando comparadas à germinação de sementes com dormência em imersão em água. Segundo Carnelossi *et al.* (1995), a utilização de hipoclorito pode estimular a germinação das sementes ou inibi-la, pois em algumas espécies a imersão por tempo maior pode ter efeito prejudicial, enquanto em menores períodos se tornaria estimulante.

Observa-se que, independentemente do nível de dormência das sementes ao serem condicionadas no NaClO, o elevado extravasamento de solutos visualizado nelas através da condutividade elétrica não afetou o percentual de plântulas normais e o IVG, pois estes não diferiram significativamente quanto à imersão em água nas sementes com e sem dormência (figura 1).

Dessa maneira, o uso do NaClO pode ter causado oxidação nos componentes de proteção da semente, como compostos fenólicos e no tegumento, ocasionando a sua ruptura e o efeito na germinação de ambas as amostras, com e sem dormência, pois a dormência nas sementes de arroz logo após a colheita se deve à presença de compostos fenólicos (MENEZES *et al*., 2009). Esses compostos insolúveis (fenólicos) das paredes celulares têm ligações com componentes de lignina (ZHOU *et al*., 2004). O fato de a casca de arroz possuir em torno de 14,5% de lignina (SILVA *et al*., 2016) pode ter permitido a reação do NaClO através da oxidação ou pela adição de cloro nas moléculas de lignina das sementes (HISE, 1996 *apud* SOFIATTI *et al*., 2009).

O efeito do uso de NaClO pode apresentar dúvidas, conforme a concentração utilizada já observada em outras espécies. Em *Avena fatua* L., o emprego da concentração de 6% de cloro ativo causou alterações nas membranas do tegumento das sementes, proporcionando maior fornecimento de oxigênio para o embrião e, então, maior germinação (HSIAO & QUICK, 1984). A utilização de 0,5% de NaClO por 24h e a secagem a 45°C mostraram-se eficientes para a superação da dormência em azevém (*Lolium multiflorum*) armazenado até 88 dias (MARTINS *et al*., 2016). Em sementes de café, o uso de NaClO na concentração de 4% foi eficiente na degradação do pergaminho que envolvia as sementes e favoreceu a germinação (SOFIATTI *et al*., 2009). Para a germinação de sementes de mamão, a imersão sob concentração de até 6% de NaClO por 8 a 24h foi prejudicial, quanto maior a concentração (GÓMEZ, 2016). Em sementes de alface, cultivar Moreninha de Uberlândia, a utilização do hipoclorito de sódio em solução de 2% de cloro prejudicou a germinação (CARNELOSSI *et al*., 1995), e, em estudo com o cultivar Regina, a imersão em solução de hipoclorito a 1% após

o condicionamento das sementes aumentou a germinação (RODRIGUES *et al.*, 2012). No presente trabalho, fez-se uso das recomendações do RAS quanto à concentração de NaClO e à temperatura de imersão das sementes de arroz, pelo fato de ser considerado um método com potencial para a superação de dormência na cultura. Dessa forma sugere-se, para pesquisas futuras, a utilização de imersão das sementes em maiores concentrações de NaClO.

CONCLUSÕES

Conclui-se que o método de imersão de sementes em solução de hipoclorito de sódio e em água promove a superação da dormência em sementes de arroz.

O NaClO causa alterações prejudiciais na qualidade fisiológica das sementes relacionadas ao vigor, pelo teste de condutividade elétrica em sementes de arroz cultivar SCS 122 Miura.

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