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Article

Interference of Brassicaceae and Poaceae on Canola Hybrids

Interferência de Brassicaceae e Poaceae sobre Híbridos de Canola

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ABSTRACT - The objective of this study was to evaluate the initial competitive ability of canola hybrids against Brassicaceae and Poaceae species in terms of root and shoot resources. Two experiments were conducted in greenhouse, in a completely randomized design with four replications. The treatments consisted of interference conditions by root + shoot interference (root+shoot) and shoot interference (Light); and Brassicaceae and Poaceae weeds competing with hybrid of canola (Hyola 61 and Diamond) with different traits for competitiveness. The plant height, root length, dry matter of shoot and root, number of leaves and leaf area were evaluated. Data were compared using orthogonal contrasts. The turnip and volunteer canola reduced root development of the Hyola 61 in root + shoot interference, and among the Brassicaceas weeds, turnip caused greater losses. Brassicaceas weeds caused largest reductions in Diamond's hybrid growth compared to Poaceae, mainly under light interference. Brassicaceas and Poaceae weeds cause similar damage to Hyola 61 hybrid, and the injury is higher for root +shoot interference. The black oat induce shoot growth in the Diamond hybrid under shoot interference, while among the Brassicaceae weed species, there were no differences.

Keywords: root interaction, *Raphanus sativus*, *Avena strigosa*, *Lolium multiflorum*, *Brassica napus*, light.

RESUMO - O objetivo deste estudo foi avaliar a habilidade competitiva inicial de híbridos de canola quanto à interferência de plantas daninhas Brassicaceae e Poaceae por recursos do solo e da parte aérea. Foram realizados dois experimentos em casa de vegetação, em delineamento experimental inteiramente casualizado com quatro repetições. Os tratamentos constaram de condições de interferência por recursos do solo+parte aérea (raiz+parte aéra) e por parte aérea somente (Luz); e plantas daninhas das famílias Brassicaceae e Poaceae competindo com híbridos de canola (Hyola 61 e Diamond) com diferentes características para competitividade. Foram avaliados estatura das plantas, comprimento de raiz, matéria seca da parte aérea e raiz, número de folhas e área foliar. Os dados foram comparados por meio de contrastes ortogonais. Nabo e canola voluntária reduziram o desenvolvimento da raiz do Hyola 61 apenas na competição por recursos do solo+parte aérea; entre os competidores da família das Brassicaceae, o nabo causou maiores prejuízos. As plantas daninhas Brassicaceae causaram maiores reduções no crescimento do Diamond em relação às Poaceae, principalmente sob a interferência por luz. As plantas daninhas Brassicaceas e Poaceae causam danos similares ao híbrido Hyola 61, e as injúrias são maiores na competição por recursos do solo+parte aérea. A aveia preta induz crescimento da parte aérea do híbrido Diamond sob interferência da luz, enquanto que entre as espécies Brassicaceas não há distinção dos efeitos.

Palavras-chave: interação de raízes, *Raphanus sativus*, *Avena strigosa*, *Lolium multiflorum*, *Brassica napus*, luminosidade.

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INTRODUCTION

Canola (*Brassica napus* L.) is a member of the Brassicaceae family and a potential alternative to winter wheat in the southern regions of Brazil. Rio Grande do Sul is the largest producer of canola in this country, with a crop area of 43.3 thousand ha, accounting for 90% of the national production; the average yield is 1,286 kg ha^{-1} (Conab, 2017).

Weed growth negatively affects canola yield, with losses of up to 70% in the absence of weed control during the crop cycle (Hamzei et al., 2007). The main winter season weeds are turnip (*Raphanus* spp.), oat (*Avena* spp.), and ryegrass (*Lolium multiflorum*). Another problem is the occurrence of volunteer canola, which can be established in the seed bank due to high seed loss before and during harvesting (Huang et al., 2016).

Crop-weed interactions are influenced by the establishment time, plant diversity, inherent competitive ability of the species involved, allelopathy, and temporal and spatial resource availability. Therefore, the competitive ability of canola is an important factor in weed management and reduces the dependence on herbicides; resulting in a reduced environmental impact and a lower risk of herbicide resistance. Thus, canola hybrids with characteristics of high competitive ability, such as rapid early growth and increased height, early flowering, high leaf area, high levels of allelopathy, and rapid establishment of the root system (Lemerle et al., 2014), can be used to reduce weed interference and to lower herbicide dependence, thereby minimizing weed problems in the long term (Williams et al., 2008).

In the early growth stages, the supply of resources is sufficient for both crop and weed development. However, early weed emergence can result in a competitive advantage for weeds and/or cause alterations in the quality and quantity of light, triggering morphological changes in plants in response to initial interference (Green-Tracewicz et al., 2012, Roing-Villanova, Martínez-García, 2016). In such a situation, plants have developed mechanisms to tolerate or avoid shading, such as changes in leaf physiology, biochemistry, anatomy and plant morphology, and/or architecture (Roing-Villanova, Martínez-García, 2016).

In this sense, some methodologies allow the separation of interactions between plants by soil and shoot resources, thereby facilitating the understanding of the underlying mechanisms. For example, the “target plant” technique allows the partitioning of interferences between above- and belowground resources (McPhee and Aarssen, 2001). According to this technique, interferences in terms of soil resources comprise competition for water and soil nutrients as well as allelopathic compounds between plant roots. Interactions in terms of shoot resources involve competition for solar radiation and CO_2 as well as allelopathic compounds.

In this context, we tested the hypothesis that taller canola hybrids and earlier crop maturity lead to a competitive advantage against weed species of the families Brassicaceae and Poaceae when compared to shorter hybrids and medium crop maturity. Taller weeds have a higher negative impact on canola growth, and there are no differences in initial interferences in terms of above- and belowground resources. Thus, the objective of this study was to evaluate the initial competitive ability of canola hybrids against Brassicaceae and Poaceae species in terms of root and shoot resources.

MATERIAL AND METHODS

Two experiments were conducted in the greenhouse of the Department of Agronomic Sciences of the Universidade Federal de Santa Maria campus of Frederico Westphalen-RS, between June and October 2016. The experimental units were composed of plastic pots with a capacity of 5.5 L (∅ = 20 cm) filled with agricultural substrate composed of pinus bark, expanded vermiculite, peat and mineral coal (Tecnomax®).

The experimental design was completely randomized with four replicates. The treatments consisted of interference conditions with Brassicaceae and Poaceae species and two canola hybrids. For this, two experiments were carried out, using the target plant Hyola 61 hybrid (medium crop maturity and short height – low competitive ability) (Exp. I); and the target plant was the Diamond hybrid (early cycle crop, high height and rapid early growth - high competitive ability) (Exp. II). The methodology used to study the competition conditions was adapted from the “target plant technique” (McPhee and Aarssen, 2001).

The interference conditions were constituted by root + shoot interference and shoot interference only. Two additional treatment control (weed-free) where included for each interference conditions. The weeds composed of the Brassicaceae family were turnip (*Raphanus sativa*) and volunteer canola (*Brassica napus*), and Poaceae family by black oat (*Avena strigosa*) and ryegrass (*Lolium multiflorum*). The canola competitor as a volunteer crop was represented by the Hyola 433 hybrid (early cycle crop and medium height). The interference conditions were characterized by the presence of target plant (Hyola 61 or Diamond) in the center of the pot, surrounded at a distance of 4 cm by weeds. In the shoot interference condition, the canola hybrids were cultivated inside a glass 8 cm in diameter and 13 cm deep placed in the center of the pot to separate the interaction between the weed roots. In root + shoot interference there was no such separation in root. The sowing of the ryegrass occurred 14 days before sowing (DBS) of the canola hybrids, and seven DBS for the others weeds. The final weed population of were 722 and 1,200 plants m^{-2} for Brassicaceas and Poaceae, respectively.

At 21 days after the emergence of canola hybrids, the plant height (hybrids and weeds) (cm) were measure the distance from the ground to the tip of the topmost fully expanded leaf; for the root length (cm), the roots of the canola were washed in running water for further separation of other roots and substrate, and were measured from the base of the seedling to the root cap; number of leaves emitted (NF); and leaf area (AF - cm^{-2}) was measured through a leaf area determiner (Licor 3000). Subsequently, the shoot (leaves, branches and stem) and roots of the plants were packed in paper bags and placed in a drying oven at constant temperature of about 60 ^{o}C for 72 hours. After the samples were weighed to determine shoot dry matter (SDM) and root dry matter (RDM). The data of the variables evaluated in the canola hybrids were transformed in percentage in relation to the weed-free (100%).

The data were analyzed for normality by the Shapiro-Wilk test, and without normal distribution, were transformed by the square root. The data were submitted to ANOVA by the F test ($p \leq 0.05$), and if there was significance, the effects of the families and weeds were compared by orthogonal contrasts ($p \leq 0.05$) for each canola hybrid and interference condition (Table 1). The weed height variable was compared by the Duncan test ($p \leq 0.05$).

Table 1 - Orthogonal contrasts tested for interference of Brassicaceas (turnip and volunteer canola) and Poaceas weeds (ryegrass and black oats) in hybrids of canola (Hyola 61 and Diamond). Frederico Westphalen, RS, 2016

Weedy species	Contrast			
	Weed-free x Weedy species	Brassicaceae x Poaceae	Turnip x Canola	Ryegrass x Black oat
Weed-free	+	0	0	0
Turnip	-	+	+	0
Volunter canola	-	+	-	0
Ryegrass	-	-	0	+
Black oat	-	-	0	-

“+” and “-”, indicates the weedy groups of contrasts, and “0” indicates the weedy group that does not participate of contrast.

RESULTS AND DISCUSSION

Height and root length of the hybrid Hyola 61 were not altered by weed species of the families Brassicaceae and Poaceae. However, weedy species under root + shoot interference decreased Hyola 61 height and root length by 72 and 80%, respectively, in comparison to weed-free cultivation (Table 2). On the other hand, Hyola 61 on root + shoot interference had lower SDM values than Hyola 61 on shoot interference (Table 3). In terms of shoot interference, the weed species did not reduce RDM accumulation. Nevertheless, when comparing canola growth in the presence of the different Brassicaceae weed species, Hyola 61 RDM was more affected in competition with turnip than with volunteer canola when considering shoot interference.

Table 2 **-** Orthogonal contrasts for percentage of height and length of canola roots (Hyola 61 and Diamond) in interference with Brassicaceas (turnip and volunteer canola) and Poaceas weeds (ryegrass and black oats) by root+shoot and shoot interference. Frederico Westphalen, RS, 2016

Variable	Interference condition	Contrast			
		Weed-free x Weedy species	Brassicaceae x Poaceae	Turnip x Canola	Ryegrass x Black oat
		Hyola 61			
Height	Shoot	$^{(1)}$100 x 88.0^{ns}	89.6 x 85.8^{ns}	92.1 x 87.1^{ns}	72.7 x 98.9^{ns}
	Root+shoot	100 x 27.6*	31.4 x 24.2^{ns}	33.1 x 30.1^{ns}	29.2 x 19.3^{ns}
Root	Shoot	100 x 92.3^{ns}	98.4 x 84.3^{ns}	99.1 x 97.7^{ns}	88.7 x 79.9^{ns}
	Root+shoot	100 x 20.4*	19.4 x 21.3^{ns}	18.9 x 19.8^{ns}	21.3 x 21.3^{ns}
		Diamond			
Height	Shoot	100 x 115^{ns}	76.9 x 155*	85.9 x 67.9^{ns}	102 x 208*
	Root+shoot	100 x 34.6*	25.6 x 43.5*	27.0 x 24.3^{ns}	45.8 x 41.3^{ns}
Root	Shoot	100 x 71.7*	66.1 x 77.5^{ns}	60.3 x 71.8^{ns}	83.9 x 71.0^{ns}
	Root+shoot	100 x 39.7*	42.5 x 36.9^{ns}	41.2 x 43.9^{ns}	24.7 x 49.1*

$^{(1)}$ Values relative to the percentage in relation to weed-free. * or ^{ns} significant and non-significant contrasts, respectively, ($p \leq 0.05$).

Table 3 **-** Orthogonal contrasts for percentage of shoot dry matter (SDM) and root dry matter (RDM) of canola (Hyola 61 and Diamond) in interference with Brassicaceas (turnip and volunteer canola) and Poaceas weeds (ryegrass and black oats) by root+shoot and shoot interference. Frederico Westphalen, RS, 2016

Variable	Interference condition	Contrast			
		Weed-free x Weedy species	Brassicaceae x Poaceae	Turnip x Canola	Ryegrass x Black oat
		Hyola 61			
SDM	Shoot	$^{(1)}$100 x 26.0*	23.0 x 29.6^{ns}	27.5 x 17.0^{ns}	36.6 x 22.5^{ns}
	Root+shoot	100 x 3.10*	3.60 x 2.70^{ns}	3.60 x 3.60^{ns}	3.30 x 2.20^{ns}
RDM	Shoot	100 x 64.4^{ns}	63.3 x 65.8^{ns}	34.2 x 107*	70.4 x 61.2^{ns}
	Root+shoot	100 x 7.80*	6.55 x 2.97^{ns}	5.90 x 7.20^{ns}	2.50 x 3.40^{ns}
		Diamond			
SDM	Shoot	100 x 60.9*	32.8 x 88.9*	48.3 x 17.3^{ns}	48.9 x 129*
	Root+shoot	100 x 0.92*	0.70 x 1.14^{ns}	0.71 x 0.69^{ns}	1.30 x 0.97^{ns}
RDM	Shoot	100 x 79.9^{ns}	80.3 x 79.6^{ns}	41.5 x 119*	69.7 x 89.4^{ns}
	Root+shoot	100 x 2.79*	3.27 x 2.37^{ns}	4.05 x 2.70^{ns}	0.97 x 3.76^{ns}

$^{(1)}$ Values relative to the percentage in relation to weed-free. * or ^{ns} significant and non-significant contrasts, respectively, ($p \leq 0.05$).

Root + shoot interference can be expressed as the interaction between roots and shoots interaction; in addition to the competition for water and nutrients, the organic allelochemicals released from the weed roots may have contributed to the results of this study. However, using the methodology described here, it is not possible to distinguish the partition degree of interference in terms of the depletion of water, soil nutrients, and allelochemicals; the damage is basically caused by a combination of these factors.

The morphological and physiological traits of each canola hybrid influence their competitive ability against weed species. Galon et al. (2015) reported that Hyola 433 (used as volunteer hybrid in this study) was more competitive than Hyola 61 in association with turnip; however, in the interaction with ryegrass, Hyola 61 showed the highest competitive ability between all hybrids tested.

The interference effects between Brassicaceae and Poaceae species on Hyola 61, allow no conclusions about the potential of these two families to cause significant damages to canola crops. However, based on the RDM values, the damage caused by turnip is greater than volunteer canola (Tables 2 and 3).

The effects of the interaction between canola hybrids and weedy species via root + shoot interference were more pronounced than those via shoot interference only. The high density of weeds intensified the effects of competition for water and soil nutrients with canola hybrids. In addition, the allelopathic compounds released by weeds had significant effects on the cultivated crops; e.g., allelopathic compounds produced by *Raphanus* sp. have a herbicidal effect (Norsworthy, 2003; Asaduzzaman et al., 2014). The damage caused by the release of black oat allelochemicals may be related to phenolic compounds, including p-coumaric acid, syringaldehyde, and vanillin (Fragasso et al, 2012). These substances can inhibit root elongation and cell division, consequently damaging cell structures and interfering with normal plant growth and development (Li et al., 2010).

Shoot interference caused by Poaceae species induced an increase in height and SDM of the Diamond hybrid by about 100 and 170%, respectively, and black oat was more competitive than ryegrass (Tables 2 and 3). There no differences among the Brassicaceae weeds in terms of height and SDM of Diamond for shoot interference. In the root + shoot interference, height decreased by 65% compared to the weed-free control, whereas the SDM produced by the Diamond hybrid was less than 1% in the presence of weed species when compared to the weed-free control.

The growth of the Diamond hybrid can be related to altered light quality caused by larger neighbor plants (Poaceae species), which is perceived on the cotyledons that initiate the synthesis of auxin, which in turn is transported into the hypocotyl to initiate plant growth (Procko et al., 2014). The biomass of canola is proportional to the grain yield potential (Harker et al., 2011); therefore, any reduction caused by weed species will result in yield losses. However, the high-yielding canola hybrids showed higher yields in the presence of weeds when compared the low-yielding hybrids (Lemerle et al., 2017).

Root length of the Diamond hybrid was 30 and 60% less when associated with shoot and root + shoot interference, respectively, as an outcome of weedy competition (Table 2). There was no effect on root length by shoot interference for the other contrasts; however, the RDM response was similar among the different canola hybrids, where intraspecific interference (volunteer canola) generated an increase of approximately 186% in relation to turnip (Table 3). The results obtained for the Diamond hybrid are contrary to those observed by Daugovish et al. (2003), who found that weed interspecific association with canola caused greater damage than intraspecific effects. In root + shoot interference, ryegrass caused a greater reduction in Diamond root length compared to black oat, which may be associated with the rapid establishment to take up soil resources and the allelopathic compounds exuded; these effects were more significant on roots than on shoots for both canola hybrids (Amini et al., 2009).

Black oat showed the greatest potential to cause light interference due to its great height (Table 4). However, volunteer canola and ryegrass height did not differ, whereas turnip presented a smaller height. With shoot interference, neighboring plants cause shading and change the light quality by absorbing radiation at the red wavelength while reflecting the far red wavelength (Green-Tracewicz et al., 2012). The red/far red ratio of light reflected by weed species may play a significant role in the outcome of the competitive interactions between crop and weed (Cressman et al., 2011). In our study, in shoot interference by Brassicaceae species with both hybrids, the

Table 4 **-** Weedy species height (cm) under root+shoot and shoot interference with Hyola 61 and Diamond hybrids. Frederico Westphalen, RS, 2016

Weedy specie	Hyola 61		Diamond	
	Shoot	Root+shoot	Shoot	Root+shoot
Black oat	^{ns}23.3 a	25. 0a	^{ns}25.5 a	23.4 a
Ryegrass	^{ns}11.3 b	11.4 b	^{ns}8.4 b	9.4 b
Canola	^{ns}11.9 b	12.5 b	^{ns}9.5 b	10.1 b
Turnip	^{ns}6.9 c	6.5 c	^{ns}5.0 c	5.0 c

Means preceded by ^{ns} indicates no difference between interference conditions, and means followed by lower case letters compare weedy species, both by Duncan test ($p \leq 0.05$).

plant with the greater height (volunteer canola) induced an increase in root mass as a way of preventing imminent competition (Tables 3 and 4). However, in contrast to black oat, volunteer canola was not able to induce increases in hybrid height.

For Hyola 61, the weed species led to a greater reduction in SDM than RDM via shoot interference, and similar effects were observed from the Diamond hybrid (Table 3). With root + shoot interference, the negative impacts under SDM and RDM were larger than shoot interference, but the differences of the damages caused between SDM and RDM were smaller. These results are in agreement with those found by Fragasso et al. (2012), who verified a greater reduction in shoot growth compared to root growth. According to these authors, the reduction is related to the competitive ability and the production of allelopathic compounds released by weeds. For Hyola 61, no differences were found in the root + shoot interference between the two weed families, although one notable exception was observed for the shoot interference condition, where turnip caused a higher damage in RDM than volunteer canola. On the other hand, Poaceae species induced the growth of the Diamond hybrid, mainly by black oat interference, which induced the so-called "shade avoidance", which was also observed in the presence of turnip in relation to intraspecific interference.

The Diamond hybrid grew more than Hyola 61 in a weed-free environment (data not shown), and the agronomic traits such as rapid growth, great height, and short cycle contributed to better competitive ability of the Diamond. These traits provide an advantage to the canola hybrids in the presence of neighboring weeds, thus increasing crop competitiveness against weeds (Procko, et. al., 2014). The competitive ability among canola hybrids can be described by plant height, vigor, and a short cycle, as well as the synthesis of allelopathic compounds (Lemerle et al., 2014). However, allelopathy from canola hybrids had little influence on the results, since the canola density was significantly lower than the weed density.

Limited below- and aboveground resources such as water, soil nutrients, solar radiation, and allelopathic compounds are the main factors involved in weed interference. In the early growing season, weeds and crops can coexist before interference occurs; however, light quality and quantity are the first factors being modified as the plants grow, triggering morphological changes in response to neighbor plants (Green-Tracewicz et al., 2012). The responses to changes in light quality and quantity varied according to the photomorphogenic plasticity of each genotype; as a result, the genotypes modify their morphology to avoid resource competition (Ballaré and Casal, 2000).

Weedy species, in any interference condition, decreased the NL and LA values of canola hybrids (Table 5). Hybrids grown under root + shoot interference had lower LA and NL values than those grown under shoot interference, and the LA damages were higher than those observed for NL. The different weed families and species had similar effects on LA and NL of Hyola 61, except for the Brassicaceae vs. Poaceae effect on LA, which was higher in the presence of Poaceae under shoot interference. Similarly, when Diamond hybrid were grown under the influence of Poaceae species, they showed greater leaf emission and development compared to those grown in the presence of Brassicaceae species, indicating that canola hybrids can increase light interception in the presence of taller neighboring plants (Table 5). In response to changes in the light spectrum, the plants maximize light interception by increasing LA, and this effect was more evident under shoot interference, mainly between Poaceae weeds. Changes in morphophysiological traits are a response to avoid future shading and, consequently, stresses caused by competition (Merotto Jr. et al. 2009).

The distinct behavior of the hybrids against different interference conditions and weeds is a result of morphological, physiological, and metabolic characteristics that influence the ability of canola hybrids to interact with the environment in the presence of neighboring plants, whether interspecifically (volunteer canola) or interspecifically (Lemerle et al., 2017). Thus, the use of high-yielding and competitive canola hybrids represent an important strategy for weed management.

Overall, the competitive ability of canola can be altered according to its agronomics traits, types of interference, and weed families and species. We observed no distinction between the damage of the interference caused by Brassicaceae and Poaceae species on the hybrid Hyola 61. However, high injury occurred there was interference by by mutual below and above ground

Table 5 **-** Orthogonal contrasts for percentage of leaf number (LN) and leaf area (LA) of canola (Hyola 61 and Diamond) in interference with Brassicaceas (turnip and volunteer canola) and Poaceas weeds (ryegrass and black oats) by root+shoot and shoot interference. Frederico Westphalen, RS, 2016

Variable	Interference condition	Contrast			
		Weed-free x Weedy species	Brassicaceae x Poaceae	Turnip x Canola	Ryegrass x Black oat
		Hyola 61			
NF	Shoot	[(1)]100 x 67.2*	61.8 x 74.5^{ns}	58.8 x 64.7^{ns}	70.6 x 78.4^{ns}
	Root+shoot	100 x 40.2*	37.5 x 42.2^{ns}	37.5 x 37.5^{ns}	37.5 x 46.9^{ns}
LA	Shoot	100 x 26.9*	18.8 x 36.4*	23.0 x 13.0^{ns}	29.0 x 43.0^{ns}
	Root+shoot	100 x 2.80*	2.20 x 3.20^{ns}	2.40 x 1.93^{ns}	3.50 x 3.00^{ns}
		Diamond			
NF	Shoot	100 x 77.9*	68.4 x 87.5*	70.0 x 66.7^{ns}	80.0 x 95.0*
	Root+shoot	100 x 36.4*	36.4 x 36.4^{ns}	36.4 x 36.4^{ns}	36.4 x 36.4^{ns}
LA	Shoot	100 x 47.6*	30.1 x 65.0*	41.2 x 19.0*	38.8 x 91.2*
	Root+shoot	100 x 0.90*	0.58 x 1.21^{ns}	0.65 x 0.50^{ns}	1.01 x 1.42^{ns}

[(1)] Values relative to the percentage in relation to weed-free. * or ^{ns} significant and non-significant contrasts, respectively, ($p \leq 0.05$).

resources. The presence of black oat induced shoot growth in the Diamond hybrid under shoot interference, while among the Brassicaceae weed species, there were no differences. The selection of canola hybrids with higher competitive ability, such as hybrids with allelopathic characteristics, rapid growth, establishment of an extensive root system, represents a low-cost weed management strategy (Lemerle et al., 2017).

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