RUNNING TITLE:

Canola competitiveness against wild radish

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TRIAL 1B: The interaction between the seeding rate and row spacing of hybrid and open pollinated canola (*Brassica napus*) varieties on wild radish (*Raphanus raphanistrum*) growth and seed production.

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Key Words; Canola, wild radish, crop competition, seed production, seeding rate, row spacing, hybrid, open pollinated.

INTRODUCTION

Wild radish is a prevalent annual weed species infesting all cropping regions of southern Australia on neutral to acidic soils. The economic impact of wild radish is attributed to its ability to greatly reduce crop yield and quality. In addition, immature wild radish plants pose harvest and grain storage problems. Although herbicides are available to control wild radish, the protracted germination and a long seed dormancy of wild radish make it difficult to control (Reeves, Code, & Piggin, 1981). When growing in a crop, wild radish is a vigorous competitor capable of causing large reductions in crop yield. Modest wild radish densities of 7 and 200 plants m^{-2} has been found to reduce wheat (*Triticum aestivum* L.) yield by 10 and 50%, respectively (Code & Reeves, 1981; Pathan, Hashem, & Koetz), with wild radish that emerges with or shortly after the crop causing the largest reduction in yield (Cheam & Code, 1995). However, wild radish often emerges throughout the crops growing season with late-emerging plants capable of producing sufficient seed to replenish the soil seed bank (Cheam, 1986; Code & Donaldson, 1996; Reeves et al., 1981). Despite a diverse range of herbicide tolerance in F1 hybrid and open pollinated canola varieties, Australian weed surveys have found that wild radish is still present in 13% of the canola fields after all weed management practices are completed (Lemerle et al., 2001). Despite being recognized as a troublesome weed in canola, the effect canola competitiveness against wild radish and wild radish effect on canola yield is not well documented. This study was conducted to determine the effect of factorial combinations of seeding rate, row spacing and pollination type on canola yield and wild radish fecundity.

MATERIALS AND METHODS

Location

In 2018, an experiment was conducted at Cunderdin (-31.37S, 117. 14E) in the Western Australian grainbelt. Soil at the Cunderdin site was a sandy loam over a medium calcareous clay subsoil with a pH of 5.4. The long-term average annual growing season (April to October) rainfall was 333 mm (2018 data presented in Figure 1). The site had been continuously cropped under no-till production for 10 years prior to this study.

Prior to seeding

In March 2018, before weed seed bank germinating rains, the baseline seedbank of both sites were estimated by taking 4 replicate intact soil core samples (8 cm in diameter by 10 cm deep) up the center of each plot location (total of 384 soil samples per site). Soil samples from each plot (n=4) were combined to determine the wild radish seedbank. Samples were placed in shallow seedling trays that had been partially filled with weed-free potting mix to ensure drainage. The soil samples were then spread in a 2-cm-thick layer, watered with overhead micro-jet irrigation and placed outside from March to August each year. Germinated seedlings were recorded and removed at regular intervals. The census for wild radish plants ceased in August when no new seedlings emerged over a 4 week period. The number of seedlings to germinate in each tray represented the germinable wild radish seedbank and was converted to seeds per square meter for each plot. As wild radish seed banks were low and patchy, pod extracted wild radish seed was evenly spread on the soil surface of the trial site at 30 seeds/m^2 before being shallowly incorporated into the soil using rotary harrows to represent the germinable weed seed bank.

Trial establishment

In May 2018, the Cunderdin experiment was direct-seeded into cereal stubble. A factorial combination of canola cultivar, seeding rate, and row spacing was randomized in complete blocks with four replicates. Canola cultivars, Hyola 559 (Hybrid mid-maturity) and Bonito (Open-pollinated mid-maturity Triazine Tolerant (TT) variety) were seeded at 25cm and 50cm row spacings. All canola treatments were planted at one sowing depth (approx. 2cm) in an effort to minimise the confounding effects of emergence rate and early vigor differences on biomass and grain yield. Within the variety and row spacing treatments, canola was seeded at a target densities of 20, 35 and 50 viable seeds m^{-2} (on a weight basis these seeding rates equated to 3.0, 2.1 and 1.2 kg ha^{-1} and 2.8, 1.9 and 1.1 kg ha^{-1} for Hyola 559 and Bonito varieties respectively).

Both cultivars received the same fungicide/insecticide seed treatment comprising of 1 L of Cruiser Opti [210 g/L Thiamethoxam 37.5 g/L Lambda-Cyhalothrin, Syngenta Australia] and 400 mL/100 kg Maxim XL [25 g/L Fludioxonil 10 g/L Metalaxyl-M Syngenta Australia] applied per 100 kg of seed. Immediately prior to seeding, all experimental areas were treated with 2 L/ha Roundup Ultramax (Glyphosate 540 g/L, Sinochem Australia) to control all germinated weeds. Each plot was sub plotted with a no additional wild radish weed control for competition assessments and a weed-free treatment to assess the effect of factorial combinations on the crop growth and light interception of canola. The weed-free treatment was maintained using 1 L/ha Kerb (500 g/L Propyzamide, Dow AgroSciences Australia) incorporated by sowing (IBS), 1.1 kg/ha Atradex (900g/kg Atrazine, Nufarm Australia Limited) IBS followed by 1.1 kg/ha of Atradex. To control annual ryegrass (*Lolium rigidum*), 500 mL/ha of Select (240g/L Clethodim, Sumitomo Chemical Australia) applied at the 4-6 leaf stage of the canola across all treatments. Herbicides were applied using a motorized sprayer calibrated to deliver a carrier volume of 120 L water ha^{-1} at 275 kPa pressure. Each subplot size was 4m wide by 10m long.

To ensure optimal canola growth, 70 kg/ha Gusto Gold (Summit Fertilisers Australia) (analysis N – 10.2%, P- 13.1%, K- 12%, S- 7.6%, Cu- 0.07%, Zn- 0.14% and Mn- 0.01%) treated with 300ml ha^{-1} Impact (250 g/L Flutriafol, Cheminova Australia) was drilled 3cm below the seed to minimise contact with the germinating canola seed with 100kg/ha of Urea (Summit Fertilisers Australia, N – 46%) broadcast on the soil surface immediately after seeding. At the 4-6 leaf stage of the canola, 100L/ha of urea and ammonium nitrate liquid fertiliser (UAN) (Summit fertilisers Australia) (N- 32%) was evenly sprayed across the site to maintain growth. At ten weeks after Sowing (WAS), canola establishment was assessed by counting two adjacent 50cm rows over 4 replicate locations per plot. Wild radish density was assessed at 10 and 14 WAS by counting the number of plants present in four replicate a 33 x 33cm quadrants (0.11 m^{-2}) per plot. To compare the growth of the canola in the weed-free split plots, Normalized Difference Vegetation Index (NDVI) was measured at 7, 10, 13 and 16 WAS using a Crop Circle™ Handheld Optical Sensor Unit (Holland Scientific, Lincoln, NE, USA) oriented 0.8m off the ground perpendicular to the center row of the plot. NDVI quantifies vegetation by measuring the difference between near-infrared in which vegetation strongly reflects and red light which vegetation absorbs. In each plot three replicate NDVI measurements were made and reported as a plot average.

Measurements of the fractional green canopy cover (FGCC) were done using the Canopeo™ app (www.canopeoapp.com) to estimate canopy development and light interception (Patrignani & Ochsner, 2015) Canopeo™ is an image analysis tool (Mathworks, Inc., Natick, MA) that uses color values in the red–green–blue (RGB) system to measure the green canopy cover percentage. Canopeo™ images were assessed for all weed free crop treatments at 13 WAS using a 14-megapixel camera that was oriented 0.8m above the top of the crop canopy, perpendicular to the center row of the plot.

Both incoming and outgoing photosynthetically active radiation (PAR) values were measured 14WAS at the top and bottom of the canola canopy throughout the season using line quantum sensor LI-191SA (LICOR Inc., Lincoln, NE, USA). The fraction intercepted PAR (PAR) was calculated as by Monteith (1981)

$$PAR = \frac{(Io-I)}{I} \quad [1]$$

: where Io is incident PAR at the top of the canopy and I is the transmitted PAR at the bottom of the canopy.

In order to estimate the biomass and seed production of wild radish plants growing within canola competition factors, at mid pod filling, six plants were randomly subsampled from the center row of each plot and dried to constant weight in a forced draught oven at 60°C. Following drying, plants were weighed before being dissected to count the number of pods formed. From each plot, a subset of 50 mature pods had their seed extracted using a modified ‘grist mill’. The seed was then counted using at S-25 optical seed counter (Data Technologies, Kibbutz Tzora, Israel) to estimate the total seed production per wild radish plant.

At 29WAS, the whole plot (10 m length with 6 by 22-cm rows) was machine harvested to determine grain yield. Grain samples (125 g) were analysed for moisture, and oil using an Infratec™ Sofia Near Infrared Spectroscope (NIR) (FOSS analytics, VIC, Australia). To calculate the mean seed weight, approximately 7000 seeds were counted S-25 optical seed counter (Data Technologies, Kibbutz Tzora, Israel) and weighed to calculate the mean canola seed weight.

Statistical analysis

Experimental Design

A split plot design accommodating a 3 factorial (2 levels of row-spacing, 2 types of Canola variety, and 3 levels of seeding rate) experiment was implemented for herbicide applied and free blocks (Figure 1). Each block is divided into 2 main plots to which the levels of the row-spacing were randomly assigned. The main plot is further divided into 6 subplots to which 6 combinations of the canola varieties and seeding rates were randomly assigned. Therefore, each block represents a complete replicate of all 12 treatment combinations. At the Cunderdin site, the treatments were replicated 3 times for both types of blocks with and without herbicide.

Statistical Models

The data were analysed using the analysis of variance techniques for a split-plot design where the main-plot was row-spacing and the subplot accommodated the 6 levels of variety and seeding-rate combinations. The objective of the experiment was to assess the effects of row-space, variety, seeding-rate, and their interactions on a set of traits, listed below. The statistical model reflected the above-described blocking and treatment structures. The treatment means and the effects of their interactions are presented in the Tables below. The analyses for the herbicide applied and herbicide free treatments were conducted separately. The traits analysed for herbicide applied blocks are as follows: four recorded NDVI values on different dates, canopeo, two radiation interception values, canola yield, crop emergence, canola seed weight, canola moisture percentage, and two types of weed densities. Under herbicide free treatment, the analysed responses were: weed-biomass, canola yield, before treatment soil seed bank, after treatment soil seed bank, total seed production, canola seed weight, canola moisture percentage, and two types of weed densities.

The analyses were conducted using R package msanova 1.0 (VSN International Ltd, Hemel Hempstead, UK).

A.

B.

Figure 1. Aerial trial photos of the Cunderdin field site in 2018.

Results

Cunderdin rainfall data

In 2018, the Cunderdin site recorded 230mm of growing season rainfall (April – October) which is less than the 333mm long term average. This rainfall was characterized by a lack of season breaking rains in April and May. The June, July rainfall totals exceeded long term average and the crop was able to mature with good soil moisture.

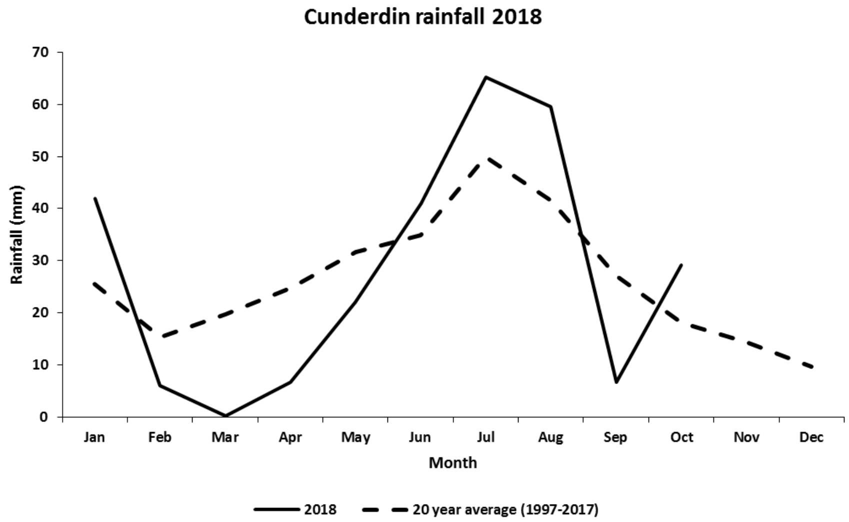

Figure 1: The rainfall totals for the 2018 season compared to the historical 20 years mean rainfall for Cunderdin in the Western Australian grainbelt.

Table 1. Mean, p-value and LSD for row-spacing, canola varieties and seed-rate on the growth of wild-radish with ***applied*** *herbicide at the location* ***Cunderdin***

Herbicide	**Variety**	**Row spacing (cm)**	**Seed Rate**	**Crop Emergence 10WAS**	**NDVI 7WAS**	**NDVI 10WAS**	**NDVI 13WAS**	**NDVI 16WS**	**Canopeo 13WAS**	**Radiation Interception (μmol m^{-2} s^{-1})**	**Radiation Interception (%)**
Herbicide applied	Open pollinated canola (Bonito)	25	0.4RR	20.2	0.193	0.373	0.717	0.727	65.7	633	0.80
			0.7RR	30.7	0.190	0.397	0.787	0.757	86.3	631	0.81
			RR	39.1	0.203	0.490	0.823	0.760	94.8	610	0.76
		50	0.4RR	37.9	0.213	0.430	0.780	0.740	81.8	667	0.83
			0.7RR	54.3	0.220	0.433	0.793	0.743	86.4	627	0.80
			RR	56.2	0.200	0.470	0.803	0.747	83.7	673	0.85
	Hybrid canola (Hyola 559)	25	0.4RR	20.5	0.170	0.337	0.720	0.720	70.4	569	0.76
			0.7RR	28.6	0.190	0.400	0.707	0.770	80.1	624	0.77
			RR	36.9	0.200	0.430	0.830	0.777	92.3	585	0.75
		50	0.4RR	24.2	0.177	0.383	0.753	0.733	74.8	656	0.84
			0.7RR	39.8	0.197	0.393	0.790	0.740	79.7	683	0.83
			RR	49.0	0.203	0.540	0.807	0.773	87.5	643	0.81
Source of variation				*P-value (LSD 5%)*							
Variety				0.012 (4.97)	NS	NS	NS	NS	NS	NS	NS
Row Spacing				0.020 (8.76)	NS	NS	NS	NS	NS	0.038 (42.7)	0.039 (0.044)
Seed Rate				< 0.001 (6.09)	NS	0.007 (0.061)	NS	< 0.001 (0.015)	< 0.001 (7.38)	NS	NS
Variety x Row spacing				0.040 (7.04)	NS	NS	NS	NS	NS	NS	NS
Variety x Seeding Rate				NS	NS	NS	NS	NS	NS	NS	NS
Row Spacing x Seeding Rate				NS	NS	NS	NS	NS	NS	NS	NS
Variety x Row spacing x Seeding				NS	NS	NS	NS	NS	NS	NS	NS

WAS – Week after sowing, NS – Not significant

*Table 2. Mean, p-value and LSD for row-spacing, canola varieties and seed-rate on the growth of wild-radish with **applied** herbicide at the location **Cunderdin***

Herbicide	Variety	Row spacing (cm)	Seed Rate	Canola Yield (t/ha)	Canola 1000 seed weight (g)	Canola Moisture (%)	Canola Oil (%)
Herbicide applied	Open pollinated canola (Bonito)	25	0.4RR	1.95	4.141	5.43	42.9
			0.7RR	2.04	4.070	5.47	43.0
			RR	1.98	4.084	5.40	42.7
		50	0.4RR	2.35	4.155	5.40	43.0
			0.7RR	2.19	4.187	5.47	42.9
			RR	2.38	4.068	5.53	42.8
	Hybrid canola (Hyola 559)	25	0.4RR	1.84	4.177	5.27	42.9
			0.7RR	1.79	4.144	5.37	43.4
			RR	2.08	4.101	5.20	42.9
		50	0.4RR	2.25	4.135	5.37	42.8
			0.7RR	2.17	4.057	5.53	42.7
			RR	2.20	4.096	5.33	43.5
Source of variation				P-value (LSD 5%)			
Variety				NS	NS	NS	NS
Row Spacing				0.001 (0.046)	NS	NS	NS
Seed Rate				NS	NS	NS	NS
Variety x Row spacing				NS	NS	NS	NS
Variety x Seeding Rate				NS	NS	NS	NS
Row Spacing x Seeding Rate				NS	NS	NS	NS
Variety x Row spacing x Seeding				NS	NS	NS	NS

WAS – Week after sowing, NS – Not significant

Table 3. Mean, p-value and LSD for row-spacing, canola varieties and seed-rate on the growth of wild-radish with ***no application of*** *herbicide at* ***Cunderdin***

Herbicide	**Variety**	**Row spacing (cm)**	**Seed Rate**	**Weed Density 10WAS (Plant/m^2)**	**Weed Density 14WAS (Plant/m^2)**	**Biomass (wild-radish) (g/plant)**	**Total WR seed production (seeds/m^2)**	**Before treatment WR soil seed bank (seeds/m^2)**	**After treatment WR soil seed bank (seeds/m^2)**	**Canola Yield (t/ha)**	**Canola 1000 seed weight (g)**	**Canola Moisture (%)**
No herbicide applied	Open pollinated canola (Bonito)	25	0.4RR	1.4	14.67	76.87	8385	40	50349	0.658	3.905	5.63
			0.7RR	1.2	12.67	51.18	1492	40	8990	1.058	3.855	5.57
			RR	1.467	9.33	51.94	2533	40	15236	1.047	3.713	5.70
		50	0.4RR	1.5	20.00	67.07	6138	40	36868	0.976	3.859	5.83
			0.7RR	0.833	23.67	54.18	6332	40	38031	0.994	3.760	5.60
			RR	1.567	15.67	55.74	3985	40	23952	1.322	3.791	5.60
	Hybrid canola (Hyola 559)	25	0.4RR	1.1	18.67	83.62	7849	40	47133	0.614	4.021	5.43
			0.7RR	0.967	10.67	42.13	3043	40	18295	0.942	3.950	5.50
			RR	1.2	11.67	53.61	3246	40	19514	1.235	3.881	5.47
		50	0.4RR	1.633	15.67	52.97	4735	40	28450	0.867	3.972	5.37
			0.7RR	1.267	10.33	63.87	3354	40	20165	1.153	3.983	5.43
			RR	0.933	11.00	54.41	4185	40	25152	1.428	4.032	5.33
Source of variation				P-value (LSD 5%)								
Variety				NS	NS	NS	NS	NS	NS	NS	0.039 (0.150)	0.002 (0.134)
Row Spacing				NS	NS	NS	NS	NS	NS	0.021 (0.127)	NS	NS
Seed Rate				NS	NS	NS	0.012 (2362)	NS	0.012 (14173)	< 0.001 0.173)	NS	NS
Variety x Row spacing				NS	0.031 (4.99)	NS	NS	NS	NS	NS	NS	NS
Variety x Seeding Rate				NS	NS	NS	NS	NS	NS	NS	NS	NS
Row Spacing x Seeding Rate				NS	NS	NS	NS	NS	NS	NS	NS	NS
Variety x Row spacing x Seeding				NS	NS	NS	NS	NS	NS	NS	NS	NS

WAS – Week after sowing, NS – Not significant

Results

Tables 1a and 1b present the estimated means, p-values and the least significant differences (LSDs) for the herbicide applied treatments, whereas Table 2 presents the results for the herbicide-free treatments.

Under herbicide application, the main effect of seeding-rate was found significant for the traits NDVI 10WAS (p-value 0.007), NDVI 16WAS (p-value < 0.001), canopeo (p-value < 0.001) and crop-emergence (p-value < 0.001). The effect of row-spacing was found significant for canola yield (p-value 0.001), crop-emergence (p-value 0.02) and both radiation interception responses (p-value < 0.04). Lastly, the effect of variety was significant only for crop-emergence (p-value 0.012). The only significant interaction effect of row-spacing and variety was for the trait crop emergence (p-value 0.04). Except for crop emergence, where all main effects were significant, the other traits had significant main effects either of seeding-rate or row-spacing.

Under the herbicide free scenario, the main effect of seeding-rate was detected for canola yield (p < 0.001), after treatment soil seedbank (p-value 0.012) and total seed production (p-value 0.012). The main effect of the row-spacing effect was significant only for canola yield (p-value 0.021). Lastly, the main effect of variety was significant for canola seed weight (p-value 0.04) and canola moisture percentage (p-value 0.002). The only significant two-way interaction of row-spacing and variety was for weed-density 14WAS (p-value 0.031).

Summary and Implications

The results of this study suggest:

1. Crop emergence was affected by pollination type (greater in open pollinated var. Bonito), increased seeding rates and increased row-spacing, with variety and row-spacing interaction.
2. Weed densities were affected by the interaction between variety and row-spacing only.
3. , Wild radish seed production was reduced by increased seeding rate.
4. In general, canola yield was increased by increasing seeding rate and row-spacing in both weedy and weed free situations.

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