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Low-Input Herbicide Management: Effects on Rapeseed Production and Profitability

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Received: 30 April 2018; Accepted: 27 June 2018; Published: 30 June 2018

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Abstract: The oilseed rape conventional system can be moved to a more sustainable one by reducing herbicide application whilst ensuring at the same time effective weed control, maintaining oilseed rape yield, and quality and increasing profitability. Over three growing season periods, two field experiments at two different Southern Italy locations were carried out. In both sites, a conventional weed-control management system (recommended label dose), four alternative low-herbicide treatments, and an untreated control were compared. We monitored weeds and crop response to herbicide treatments, and calculated the net economic return, within site and year, for each treatment. In experiment 1, a half dose of herbicide did not show any significant difference in seed yield with respect to conventional treatment in two of three growing seasons. In experiment 2, compared with the conventional system, weedy control and the lowest applied herbicide dose treatment (25% of the recommended label dose) did not underline significant differences with regard to yield level. Net returns from the half dose of metazachlor herbicide were not significantly lower than net returns from conventional treatment in experiment 1 (on a three-year average 748 vs. 812 € ha^{-1}, respectively). Our findings suggest that the herbicide dose might be cut by at least 50% in order not to jeopardize negative effects on production and economic performances.

Keywords: *Brassica napus* var. *oleifera*; yield components; sustainable crop management; weed management; profitability

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1. Introduction

Oilseed rape cultivation (*Brassica napus* L. var. *oleifera* D.C.) offers various potential production purposes, as food and feed uses [1]. Moreover, oilseed rape cultivation fits well with crop rotations based on winter cereals in Mediterranean-type environments. Oilseed rape might be also considered a valuable option from an economic point of view compared to other grain crops [2–4]. The recent establishment of oilseed processing plants have strengthened the interest in oilseed rape production in southern Europe [5–8]. However, environmental and economic sustainability of oilseed rape cropping system should be taken into account. Weed control is indeed one of the agronomic inputs that might be managed and/or reduced in order to provide a better sustainable cropping system option than the conventional one, especially in terms of production level and profitability [9]. The inappropriate management of weeds is a relevant constraint for oilseed rape productivity [10,11]. Zare et al. [12] reported that the yield reduction rate (often ranged from 30 to 70%) depends on both the weeds species type and the density of infestation. To our knowledge, especially in southern European regions, research on weed control in winter oilseed rape is almost non-existent and mostly focused in countries where oilseed rape is traditional grown ([13], and the references therein). Oilseed rape is newly introduced in rainfed cropping systems spread in Central and southern Italy, where it is generally cultivated following durum wheat [6]. The critical weed-free period for oilseed rape is from emergence to early flowering stages [14]. A wide range of weed species, including grasses and broad leave species, commonly occur in oilseed rape field [15]. One of the most common herbicides used to contrast oilseed rape weeds is metazachlor [16,17]. The successful use of metazachlor in controlling oilseed rape weeds is due to its high stability in relation to different pedo-climatic conditions (e.g., solar radiation and soil texture), up to eight weeks after treatment application [18]. The active substance might show a dual contrasting performance. On the one hand, it is assumed that active herbicide ingredient hinders a specific weed target site [18], and on the other hand, it might cause stress to the crop species [19]. In this regard, the main drawbacks related to the use of metazachlor are related to potential phytotoxicity effects during the first stages of the oilseed rape growing cycle [20,21]. Some recent microcosm studies [20,21] were carried out in order to better understand the metazachlor mode of action with regard to both physiological status and growth of a hydroponically oilseed rape cultivation in the short-term time frame. The same authors highlighted that to move the results from microcosm level to the farm level, field-scale experiments, thus including natural soils, should be set up. Field experiments have helped highlight the fact that crops undergoing pre-emergent herbicides action are able to overcome the early chemical stress conditions, hence it might be useful to consider further a mid-term time frame. Moreover, previous field-scale studies were mainly focused on the use of herbicide-resistant oilseed rape [22–26], the use of which is not allowed in European Union member states [27]. This paper analyses and assesses the consequences due to a decrease of herbicide rates towards both oilseed rape productivity and seed quality in order to reduce pesticide dependence and as a result foster environmental and economic sustainability.

2. Materials and Methods

2.1. Study Area and Sites Selection

At two field experiments (Exp. 1 and Exp. 2) carried out over three growing seasons (from 2007–2008 to 2009–2010) in two sites in southern Italy (Sardinia), weed flora dynamics and oilseed rape yield were evaluated. This area is considered representative in the Mediterranean basin in terms of climatic, edaphic, and weed flora conditions, as well as different types of cropping systems [28–30].

The selected sites (Ottava; 41° N, 9° E; 81 m a.s.l. and Ussana; 39° N, 9° E; 114 m a.s.l., respectively) differ in soil depth, soil particle size fractionation, and chemical characteristics.

In the Ottava site, soils are poorly drained [31,32] with a clay-loam texture, a phosphorous and an organic matter content equal to 38 ppm, and 1.7%, respectively. Soil depth ranges from 50 to 80 cm.

The area around Ussana site is basically characterized by Petrocalcic Palexeralf soils [32], with a loamy texture. Soil depth ranges from 90 to 120 cm, soil water content is equal to 33% at field capacity, and 17% at wilting point. According to long-term data, the climate at both Ussana and Ottava is typically Mediterranean, although the two sites differ in rainfall and thermal regime [33]. Ussana showed a rainfall trend lower (−19%) than Ottava, with a wider temperature range, because of the higher maximum and lower minimum values that occur during the entire year (Figure 1).

During the experiment period, oilseed rape was sown following durum wheat every year.

At both sites, the experiments were conducted on adjacent fields. For both experiments, a variety was selected on the basis of previous trials carried out in Southern Italy [34,35].

2.2. Experimental Design and Management

The first experiment (Exp. 1) was carried out in Ottava (experimental farm in the University of Sassari). Oilseed rape cv Kabel (Koipesol Semillas, S.A.) was sown at a density of 1,200,000 plants ha^{-1}

and depth of 2 cm on November 9, 11 and 21 in 2007, 2008 and 2009, respectively. Diammonium phosphate (200 kg ha^{-1}) was distributed with oilseed rape, and additional 100 kg ha^{-1} urea was top-dressed before oilseed rape flowering at 51 BBCH (BASF-Bayer-Ciba-Geigy-Hoechst) code stage [36]. Five herbicide treatments and a weedy control were arranged in a randomized complete block design with three replications; plot dimensions were 12 by 4.5 m.

In the first growing season, commercial formulation of metazachlor (Butisan S, 50% a.i.) was applied at three dosages and two application times: the full recommended or conventional dose (1000 g a.i. ha^{-1}, M_{PE}100), 75% of the full recommended dose (M_{PE}75), 50% of the full recommended dose (M_{PE}50), in pre-emergence, and 25% of the full recommended dose in post-emergence (POE). Two days before planting, commercial formulation of trifluralin (Triflene, 48% a.i.) was broadcasted and incorporated at 720 g a.i. ha^{-1} (TF).

Since in the first growing season, trifluralin treatment resulted in poor weed control, it was replaced in the subsequent season by another treatment of Butisan S (25% of the full recommended dose, M_{PE}25) in pre-emergence.

In all the three growing seasons, post-emergence metazachlor (POE) was applied in the field with the crop at the 4–5 leaf stage and weeds at the 2–4 leaf stage. Treatments were applied with a pneumatic backpack with TeeJet 11002E flat-fan nozzle tips (Spraying Systems Co., Wheaton, IL, USA), calibrated to deliver 140 L ha^{-1} at 276 kPa.

The second experiment (Exp. 2) was carried out at Ussana in the experimental station of the Agricultural Research Agency of Sardinia (Agris). Oilseed rape cv Kabel (Koipesol Semillas, S.A.) was sown in 18 cm row, spacing in 25 m by 6 m plots between 28 October and 22 November depending on the year (2008, 2009 and 2010). There were 34 crop rows in each experimental unit, and seeding rate was 1,200,000 seed ha^{-1}. All treatments were arranged in a randomized complete block design with four replications. In the first year, Exp. 2 had five weed control treatments. Trifluralin was applied 1 day before planting (720 g a.i. ha^{-1}; TF), and immediately after application was buried by a power harrow to an approximate depth of 10 cm. At pre-emergence, 1 day after sowing, metazachlor was applied at three doses: the conventional dose (1000 g a.i. ha^{-1}, M_{PE}100) and two further reduced doses (750 g a.i. ha^{-1} M_{PE}75; and 500 g a.i. ha^{-1}, M_{PE}50). As in Exp. 1, a post-emergence metazachlor treatment (500 g a.i. ha^{-1}, POE) was also considered. A weedy control was included in this experimental design. Herbicides were applied with a pressurized lift-mounted sprayer, Hardi NK 600 equipped with ARAG 422EF08003 Evenfan nozzles (Arag Spraying Systems Reggio Emilia, Italy) spaced 50 cm apart, calibrated to deliver 240 L ha^{-1} aqueous solution at 300 kPa. At the Ussana site, as already stated for Exp. 1, trifluralin treatment was replaced by a further pre-emergence metazachlor treatment at the rate of 250 g a.i. ha^{-1} (M_{PE}25) starting in 2009. Moreover, due to the poor performance of post-emergence treatment during the two previous growing cycles (2007–2008 and 2008–2009), in 2010, a combined treatment (POE) of clopiralid (Lontrel 72 SG, Dow AgroSciences Italia) + propaquizafop (Agil, DuPont Italia) active ingredients (200 g a.i. ha^{-1} and 100 g a.i. ha^{-1}, respectively) was applied in place of the previous post-emergence metazachlor treatment.

2.3. Measurements and Samplings

2.3.1. Effect of Herbicide on Oilseed Rape

In Exp. 1, at four and eight weeks after treatment application, the effect of herbicides on crop growth was identified by rating oilseed rape growth reduction from 0% = no injury to 100% = severe plant injury with no new growth. Data were collected from five 0.25 m^2 random quadrats per plot. In Exp. 2, at four weeks after treatment application, crop growth reduction was assessed from 0% (no injury) to 100% (severe plant injury with no new growth) on five 0.25 m^2 random quadrats per plot. Zero growth reduction reference was provided by the two border rows that surrounded each experiment, which were weeded by hand every two weeks.

2.3.2. Weed Population Dynamics and Herbicide Efficacy

In Exp. 1 at four and eight weeks after treatments application, weed biomass, and density were determined on two 0.5 m^2 quadrats per plot. Plant material was dried at 80 °C for 48 h. In Exp. 2 in each growing season, the percentage of coverage towards each weed species was visually assessed on a scale from 0 (no weed coverage) to 100 (total weed coverage). Weed coverage and composition was constantly monitored on three 0.5 m^2 areas in each plot on a monthly basis to evaluate its influence on crop yield. Reported results are the average of three measurements.

2.3.3. Seed Yield and Yield Characteristics

In Exp. 1 at crop maturity, four 0.5 m^2 sampling areas per plot were harvested to determine plant density, total aboveground biomass, seed yield, and yield components. The remaining area was individually harvested using a plot combine harvester. In Exp. 2 at oilseed rape maturity stage, in two 1 m^2 quadrats per plot, plants were counted and measured to determine crop density whereas plant height was obtained with eight measurements per plot. Crop seed yield was determined by two adjacent sub-samples of 15 m^2 by a plot combine (Wintersteiger Delta) for each plot, 1000-seeds weight was determined by hand-harvesting plants in the two adjacent middle rows of each plot. Plant samples were dried at 60 °C for 72 h and threshed by a stationary harvester. Seed yield was adjusted to 9% moisture content. The same seeds sample was used to determine seeds oil content according to [37] using Soxhlet system and petroleum ether 40–60 °C as solvent.

2.4. Economics

A net economic return assessment [38] of the seed yield was performed in order to rank the treatments considering both yield (kg ha^{-1}) and economic value. Net economic return was calculated as shown in Equation (1) by subtracting the treatment cost from the gross return (mean price × yield). The gross net return was achieved by multiplying the mean price per kilogram of product and the amount of the harvested product. Mean price per kilogram of product was obtained by taking into account mean unit oilseed rape grain prices for the 2007–2010 seasons [39]. The treatment cost was obtained by considering the suggested retail price of herbicides [40]:

$$\text{Net economic return} = (\text{mean price} \times \text{yield}) - \text{treatment cost}, \quad (1)$$

The other input costs (e.g., tillage, fertilizer, seed) were not taken into account as these costs were hypothesized to be the same across all treatments [38].

2.5. Statistical Analyses

For each experiment, weed and yield data (Dataset Repository DOI: 10.17632/54fz2czzrm.1.) were analyzed using the MIXED procedure (SAS Version 9.1, SAS Inst., Cary, NC, USA). Crop growth reduction data (expressed as percent) were arcsine transformed before analysis of variance, untransformed data were reported and discussed. Pairwise comparisons of means were conducted using Tukey's multiple comparison tests at $p \leq 0.05$ level. In Exp. 1, block and year (Y) were considered as random factors, and herbicide treatment (T) and sampling date (D) as fixed factors. In Exp. 2, the effect of herbicide treatment (T) was considered fixed and the effect of block and year (Y) was considered random. For both experiments, crop yield data was analyzed by considering herbicide treatment (T) as fixed factor and block and year as random factors. Net economic return was analyzed using GLM procedure (SAS Version 9.1, SAS Inst., Cary, NC, USA) for all the years separately. The difference among means was compared with Fisher's least significant difference test (LSD) at $p \leq 0.05$ level.

3. Results

3.1. Weather Conditions

At Exp. 1 site, total rainfall in 2008–2009 and 2009–2010 was 51% and 38% higher than the historical averages (49-year mean value), respectively, whereas in 2007–2008, total rainfall was very close to the long-term annual mean (445 vs. 479 mm year^{-1}; Figure 1a). At Exp. 2 site, significant variations in cumulated rainfall were recorded throughout the study period and in comparison to the long term series (37-year mean value). Figure 1b shows that rainfall was higher in 2008–2009 and 2009–2010 than 2007–2008. In general, the 2008–2009 and 2009–2010 cropping seasons were wetter compared to the historical trend, whereas 2007–2008 was exceptionally dry at the beginning of the oilseed rape growing cycle (November–January). Each year, during the March–June experimental period, monthly mean maximum and minimum air temperatures were <1 °C higher than the long-term means values (Figure 1a,b).

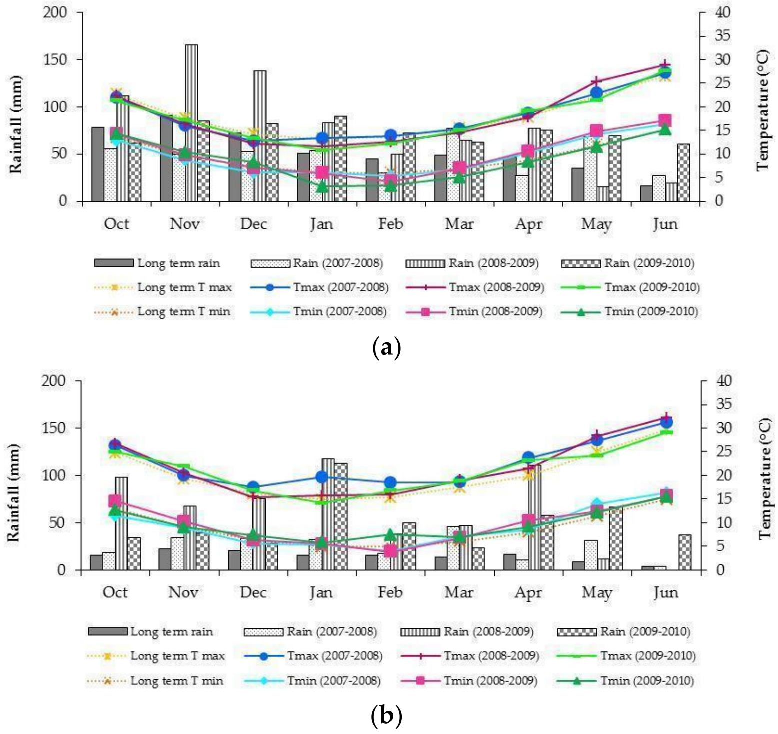

Figure 1. Rainfall (mm) and temperature (°C) for the 2007-2010 experimental period in Exp. 1 (**a**) and Exp. 2 (**b**) along with their respective long-term series (49-year, and 37-year, respectively).

3.2. Oilseed Rape Injury

We found a three-way interaction (treatment by sampling date by year) for oilseed rape injury for Exp. 1 and a two-way interaction (year by treatment) for Exp. 2; therefore, data is presented separately by treatment, sampling date, and year; and by treatment and year for Exp. 1 and Exp. 2, respectively (Table 1). In Exp. 1, in 2008, oilseed rape injury (leaf chlorosis and necrosis) was visible with the highest treatment rate, when rated at four weeks after treatment application. A similar pattern was recorded in 2009 and 2010 with significant differences among years. When evaluated at eight weeks after treatment application, metazachlor at the greatest dose caused the highest oilseed rape injury (Table 1). In Exp. 2, oilseed rape injury was evaluated at four weeks after herbicide application in all years. In 2010, metazachlor at full recommended dose ($M_{PE}100$) resulted in the greatest injury (64%), significantly different from $M_{PE}75$ and $M_{PE}50$ (Table 1). Slight or no injury was detected at the lowest herbicide doses.

Table 1. Probability values (P > F) and degrees of freedom (Df) associated with the different sources of variation in the statistical analysis of crop growth reduction (%) across Experiment 1 (and two sampling times-4 and 8 weeks after treatment application) and experiment 2 and over three growing seasons (2007–2008, 2008–2009, and 2009–2010).

Factors	Crop Growth Reduction (%)								
	Experiment 1						Experiment 2		
	4 WAT [1]			8 WAT					
	2008	2009	2010	2008	2009	2010	2008	2009	2010
M_{PE}25	-	3.3c	3.3c	-	4.1d	5.0d	-	0.0c	0.0c
M_{PE}50	6.7Bd	14.4Ab	5.6Bc	10.7c	10.0c	12.2c	13.3b	16.7bc	11.1c
M_{PE}75	61.1Ab	26.7Bb	23.3Bb	32.2b	22.0b	21.1b	31.1a	27.8b	33.3b
M_{PE}100	81.3Aa	53.3Ba	58.9Ba	59.5a	47.8a	48.9a	52.2a	61.1a	64.4a
POE	4.5Ad	2.2Bc	4.4Ac	1.1Bc	3.8Ac	4.0Ad	3.3b	0.0c	0.0c
TF	40c	-	-	35.6b	-	-	20.0a	-	-
Effects	*Df*	*P > F*						*Df*	*P > F*
Year (Y)	2	<0.0001					Year (Y)	2	<0.0001
Treatment (T)	5	<0.0001					Treatment (T)	5	0.1578
Date of sampling (D)	1	0.0142					Y × T	9	0.0297
Y × T	9	<0.0001							
Y × D	2	0.0701							
Y × T × D	15	0.0134							

Different letters indicate significant differences according to Tukey's multiple comparison tests ($p \leq 0.05$) among years (upper case, within row) and treatments (lower case, within column). [1] WAT weeks after treatment application. M_{PE}25, M_{PE}50, M_{PE}75, M_{PE}100: pre-emergence treatment with 250, 500, 750, 1000 g a.i. ha^{-1} of metazachlor, respectively. POE post-emergence treatment. TF trifluralin treatment.

3.3. Weeds Trend and Herbicide Effectiveness

3.3.1. Experiment 1

Twenty-one weed species belonging to six botanic families were identified over the three years of the experiment. Among them, only three grasses were collected. *Chenopodium album* L., *Chrysanthemum coronarium* L., *Convolvulus arvensis* L., *Fumaria capreolata* L., and *Papaver rhoeas* L. occurred in more than 95% of the plots in the field (data not shown). Weed suppression was evaluated as number of weed per m^2 and amount of aboveground biomass produced by weeds (g m^{-2} on a dry weight basis) within the herbicide regimes. Significant interactions among year, treatment, and sampling date (Table 2) were found. At four weeks after treatment application in 2008, the minimum weed density was detected in herbicide doses applied in pre-emergence, whereas the highest number of weeds per unit area occurred in the untreated control plots and in trifluralin treatment (Table 2). In 2009 and 2010, a similar trend was observed; total weed number of weeds per unit area resulted statistically higher in control and in POE treatments than the other weed control treatments. In pre-emergence treated plots, weeds density slightly increased when rated at eight weeks after treatment application, even if lower with respect to weedy, POE, and the lowest herbicide dose treated plots (Table 2). At four weeks after treatment application, the aboveground biomass produced by weeds showed statistical differences across years and treatments (Table 2). In 2008, the untreated control plots and the plots under POE and TF treatments had higher weed biomass than the pre-emergence treated plots. The same performance occurred in 2009, and 2010 when metazachlor at the label recommended dose provided good control on weeds; however, it did not show the same results when distributed at the post-emergence stage or at the lowest doses. These results were similar at eight weeks after treatment application (Table 2).

Table 2. Probability values (P > F) and degrees of freedom (Df) associated with the different sources of variation in the statistical analysis of weed density (plants m^{-2}), and weed aboveground biomass (g m^{-2}) over two sampling times (4 and 8 weeks after treatment application) and three growing seasons (2007–2008, 2008–2009, and 2009–2010) in Experiment 1.

Factors	**Weeds Density (Plants m^{-2})**						**Weeds Aboveground Biomass (g m^{-2})**					
	4 WAT [1]			**8 WAT**			**4 WAT**			**8 WAT**		
	2008	**2009**	**2010**	**2008**	**2009**	**2010**	**2008**	**2009**	**2010**	**2008**	**2009**	**2010**
$M_{PE}25$	-	13Bb	57Ac	-	66Ba	125Aa	-	31.0Ba	102.3Ab	-	637Ab	360.5Bb
$M_{PE}50$	29Bc	12Bb	74Ab	50Bd	50Ba	110Ab	0.3Bd	10.0Ba	34.8Ac	29.3Cd	417Ac	164.8Bc
$M_{PE}75$	29Bc	6Cb	52Ac	49Bd	46Bb	91Ac	0.0d	2.0b	0.0c	3.7Be	139Ad	18.5Bd
$M_{PE}100$	24Bc	4Cb	73Ab	36Be	37Bb	97Ac	0.0d	0.8b	0.0c	0.3Be	100Ad	3.8Bd
POE	48Cb	97Ba	111Aa	57Bc	69Ba	130Aa	123.0Bb	28.5Ca	187.0Aa	101.0Bc	1340Ab	225.5Bb
TF	64a	-	-	65b	-	-	47.3c	-	-	281.3b	-	-
Control	85Ba	95Ba	132Aa	73Ba	85Ba	133Aa	162.7Aa	47.0Ba	188.5Aa	481.7Ba	2617Aa	500.3Ba
Effects	*Df*	*P > F*					*P > F*					
Year (Y)	2	<0.0001					0.0002					
Treatment (T)	6	<0.0001					<0.0001					
Date of sampling (D)	1	<0.0001					<0.0001					
Y × T	9	<0.0001					<0.0001					
Y × D	2	0.002					<0.0001					
Y × T × D	15	<0.0001					<0.0001					

Different letters indicate significant differences according to Tukey's multiple comparison tests ($p \leq 0.05$) among years (upper case, within row) and treatments (lower case, within column). [1] WAT weeks after treatment application. $M_{PE}25$, $M_{PE}50$, $M_{PE}75$, $M_{PE}100$: pre-emergence treatment with 250, 500, 750, 1000 g a.i. ha^{-1} of metazachlor, respectively. POE post-emergence treatment. TF trifluralin treatment.

3.3.2. Experiment 2

In general, the coverage of weeds increased with decreased herbicide dose (Table 3), showing significant differences among years. The weed community was dominated by wild mustard (*Sinapis arvensis* L.), climbing fumitory (*Fumaria capreolata* L.), poppy (*Papaver rhoeas* L.), pot marigold (*Calendula officinalis* L.), common vetch (*Vicia sativa* L.), wild oat (*Avena fatua* L.), common canary grass (*Phalaris canariensis* L.), annual ryegrass (*Lolium rigidum* Gaudin), milk thistle [*Silybum marianum* (L.) Gaertn.], burr medic (*Medicago polymorpha* L.) and wild buckwheat (*Polygonum convolvulus* L.). Across years, the weeds species composition varied greatly with treatments and plots; thus, in the results reported herein, we focused on the main three weeds (climbing fumitory, wild mustard, poppy) since they were detected in all plots, and in all years. A one-way treatment by year interaction for climbing fumitory and wild mustard coverage was found; therefore, data is presented separately by treatment and year. There was no treatment by year interaction for poppy, therefore, that data was combined. Climbing fumitory coverage in 2008 was reduced by 90% with higher rates ($M_{PE}50$–$M_{PE}100$) of metazachlor (Table 3) while TF controlled climbing fumitory by 73%. In 2010, less consistent climbing fumitory control was noted with metazachlor. In the same year, POE treatment (clopyralid plus propaquizafop combination instead of metazachlor) again provided poor control. Wild mustard was not effectively controlled by any herbicide dose in all years. Only clopyralid plus propaquizafop applied as a post-emergent reduced wild mustard by 28%, 30% and 21% in 2008, 2009 and 2010, respectively, compared with the pre-emergence treatments. Averaged over the years, all herbicide treatments except POE showed significant effect on coverage of poppy as compared to untreated control (Table 3).

Table 3. Probability values (P > F) and degrees of freedom (Df) associated with the different sources of variation in the statistical analysis of weed coverage (%) for the main weeds species found in Experiment 2 over three growing seasons (2007–2008, 2008–2009, and 2009–2010).

Factors		Weeds Coverage (%)						
		Climbing Fumitory			Wild Mustard			Poppy
		2008	2009	2010	2008	2009	2010	
$M_{PE}25$		-	5.0Ba	35.3Ab	-	65.8Aa	30.8Bb	-
$M_{PE}50$		4.3Bb	3.8Bb	25.3Ac	67.0a	66.3a	58.5a	8.3b
$M_{PE}75$		3.8Bb	4.0Bb	38.3Aa	69.3a	64.5a	58.3a	6.3b
$M_{PE}100$		4.3Bb	0.5Bc	26.5Ac	70.5Aa	72.0Aa	55.8Ba	3.0b
POE		25.3Ba	7.5Ca	50.3Aa	43.0Ab	52.0Aa	20.8Bc	21.3a
TF		1.3b	-	-	64.8a	-	-	-
Control		31.5Aa	10.3Ba	39.0Aa	32.3Ac	44.5Ab	19.0Bc	20.6a
Effects	*Df*	*P > F*			*P > F*			*P > F*
Year (Y)	2	0.0002			0.0041			0.0517
Treatment (T)	6	0.5491			0.0053			<0.0001
T × Y	9	0.0001			0.0025			0.3273

Different letters indicate significant differences according to Tukey's HSD multiple comparison tests ($p \leq 0.05$) among years (upper case, within row) and treatments (lower case, within column). Poppy data were pooled according to a non-significant treatment by year interaction. $M_{PE}25$, $M_{PE}50$, $M_{PE}75$, $M_{PE}100$: pre-emergence treatment with 250, 500, 750, 1000 g a.i. ha^{-1} of metazachlor, respectively. POE post-emergence treatment. TF trifluralin treatment.

3.4. Yield and Yield Characteristics

3.4.1. Experiment 1

Oilseed rape yield and yield components showed significant differences among years and treatments (Tables 4 and 5). Oilseed rape treated with dosage ranging from $M_{PE}50$ to $M_{PE}100$ showed significant differences compared to weedy control in terms of yield components (Table 4). However, oilseed rape density significantly increased at lower dosage and in weedy control treatment (Table 4).

Table 4. Probability values (P > F) and degrees of freedom (Df) associated with the different sources of variation in the statistical analysis of main yield components: crop density (plants m^{-2}), number of pods per plant (n. $plant^{-1}$), number of seeds per pod (n. pod^{-1}) and 1000-seed weight (g) in Experiment 1.

Factors		Crop Density (Plants m^{-2})	Pods $Plant^{-1}$ (No.)	Seeds pod^{-1} (No.)	1000-Seed Weight (g)
Year					
2008		47c	98b	18	3.04b
2009		64b	195a	20	3.62a
2010		92a	170a	20	3.14b
Treatment					
$M_{PE}25$		88a	155b	20a	3.19b
$M_{PE}50$		64a	189a	21a	3.55a
$M_{PE}75$		61a	225a	21a	3.67a
$M_{PE}100$		51b	236a	20a	3.40a
POE		81a	98b	18b	3.10b
TF		39b	74b	16b	2.60c
Control		75a	51c	13c	2.50c
Effects	*Df*	*P > F*	*P > F*	*P > F*	*P > F*
Year (Y)	2	0.0469	<0.0001	0.8624	0.0038
Treatment (T)	6	0.0021	0.0452	<0.0001	0.0024
T × Y	9	0.2198	0.2293	0.4657	0.8964

For each parameter (treatments and years), within each column, means followed by different letters are significantly different according to Tukey's multiple comparison tests ($p \leq 0.05$). $M_{PE}25$, $M_{PE}50$, $M_{PE}75$, $M_{PE}100$: pre-emergence treatment with 250, 500, 750, 1000 g a.i. ha^{-1} of metazachlor, respectively. POE post-emergence treatment. TF trifluralin treatment.

Overall, pre-emergence treatments caused a rise of the aboveground oilseed rape biomass in all years (Table 5). In 2008, this increase was significant at metazachlor treatments compared to the control and trifluralin treatment, respectively. In 2009, the reduction in aboveground biomass was significant at the lowest metazachlor doses (both post-and pre-emergence) and control treatments (Table 5). In 2010, a trend similar to 2009 was observed. Regarding comparison among years, 2009 and 2010 resulted in significantly higher biomass production than 2008. In 2009 and 2010, the seed yield of $M_{PE}25$ and POE treatments was not significantly different from the weedy one. In 2009 and 2010 growing seasons, results showed no significant differences in seed yield among metazachlor treated plots in the range $M_{PE}50$–$M_{PE}100$.

Table 5. Probability values (P > F) and degrees of freedom (Df) associated with the different sources of variation in the statistical analysis of crop aboveground biomass (kg ha^{-1}) and seed yield (kg ha^{-1}) in Experiment 1 over three growing seasons (2007–2008, 2008–2009, and 2009–2010).

Factors		Aboveground Biomass (kg ha^{-1})			Seed Yield (kg ha^{-1})		
		2008	2009	2010	2008	2009	2010
$M_{PE}25$		-	5120b	4856a	-	1173b	1103b
$M_{PE}50$		4197Ba	5829Aa	5228Aa	1501Bb	2353Aa	1750Ba
$M_{PE}75$		4029Ba	5882Aa	5128Aa	1482Bb	2122Aa	2197Aa
$M_{PE}100$		4321Ba	5749Aa	5378Aa	1959a	2422a	2052a
POE		3707Bb	4557Ab	4231Ab	757c	1064b	939b
TF		2996c	-	-	1221b	-	-
Control		2853Bc	3874Ac	3353Bc	714c	975b	993b
Effects	*Df*	*P > F*			*P > F*		
Year (Y)	2	<0.0001			<0.0001		
Treatment (T)	6	0.0579			0.1211		
T × Y	9	0.0003			0.0161		

Different letters indicate significant differences according to Tukey's multiple comparison tests ($p \leq 0.05$) among years (upper case, within row) and treatments (lower case, within column). $M_{PE}25$, $M_{PE}50$, $M_{PE}75$, $M_{PE}100$: pre-emergence treatment with 250, 500, 750, 1000 g a.i. ha^{-1} of metazachlor, respectively. POE: post-emergence treatment. TF: trifluralin treatment.

3.4.2. Experiment 2

The effects of herbicide treatment on crop height, 1000-seed weight, and seed oil content varied depending on the year (significant T × Y interaction; Table 6), whereas no two-way interaction was found for seed yield and crop density (Table 7). Weed control, slightly increased plant height for 2008, but not in the following years, especially in 2009, when the height of the plants was lower in $M_{PE}100$ and POE. Significant differences were detected among years with the highest values recorded during 2010 (Table 6). In general, 1000-seed weight increased with pre-emergence metazachlor treatments in 2008 and 2009 (except $M_{PE}25$). Concerning seed oil content, few differences were found among treatments. In 2009, seed oil content decreased at post-emergence treatment, when compared to other treatments applied in pre-emergence and weedy control. In 2010, seed oil content was higher than previous two growing seasons, but with a significant difference only for post-emergence treated plots and weedy control plots (Table 6) with respect to previous seasons.

Table 6. Probability values (P > F) and degrees of freedom (Df) associated with the different sources of variation in the statistical analysis of crop height (cm), 1000-seed weight (g) and seed oil content (%) in Experiment 2 over three growing seasons (2007–2008, 2008–2009, and 2009–2010).

Factors		Crop Height (cm)			1000-Seed Weight (g)			Seed Oil Content (%)		
		2008	2009	2010	2008	2009	2010	2008	2009	2010
$M_{PE}25$		-	144.2Ba	159.2Aa	-	2.59b	2.89a	-	33.7b	36.4a
$M_{PE}50$		92.5Cb	143.3Ba	160.9Aa	2.63a	2.83a	2.83a	34.6a	34.7a	37.1a
$M_{PE}75$		101.7Ca	142.0Ba	156.5Aa	2.69a	2.86a	2.89a	35.0a	36.0a	38.1a
$M_{PE}100$		105.0Ca	134.8Bb	154.8Aa	2.67a	2.74a	2.86a	33.9a	36.6a	36.9a
POE		84.9Cc	138.3Bb	162.1Aa	2.46Bb	2.41Bc	2.92Aa	33.0Ba	30.6Bc	36.5Aa
TF		96.5b	-	-	2.53a	-	-	31.4b	-	-
Control		93.8Bb	147.3Aa	160.4Aa	2.53Ba	2.38Bc	3.00Aa	34.3Ba	31.3Bb	37.7Aa
Effects	*Df*	*P > F*			*P > F*			*P > F*		
Year (Y)	2	<0.0001			0.0024			7×10^{-4}		
Treatment (T)	6	0.0579			0.0905			0.024		
T × Y	9	0.0003			0.0004			0.028		

Different letters indicate significant differences according to Tukey's multiple comparison tests ($p \leq 0.05$) among years (upper case, within row) and treatments (lower case, within column). $M_{PE}25$, $M_{PE}50$, $M_{PE}75$, $M_{PE}100$: pre-emergence treatment with 250, 500, 750, 1000 g a.i. ha^{-1} of metazachlor, respectively. POE: post-emergence treatment. TF: trifluralin treatment.

Oilseed rape yield significantly differs among herbicide treatments (Table 7), showing higher results in pre- (in the range $M_{PE}50$-$M_{PE}100$) and post-emergence (POE) treated plots. Significant differences were observed among years; the highest yield was detected in 2010 and it was about three times higher than the two previous growing seasons. The two experiments were carried out during the same growing seasons, planted during the same period, and adopting the same variety. The comparison of the average oilseed rape yields suggests that each site has its own yield potential. Considering the average values of three growing seasons per site, Exp. 1 showed an average yield around 1500 kg ha^{-1}, while in Exp. 2, the yield slightly exceeded 1100 kg ha^{-1}.

Table 7. Probability values (P > F) and degrees of freedom (Df) associated with the different sources of variation in the statistical analysis of crop density (plants m^{-2}), and seed yield (kg ha^{-1}) in Experiment 2.

Factors		Crop Density (Plants m^{-2})	Seed Yield (kg ha^{-1})
Years			
2008		56	737b
2009		53	728b
2010		51	2241a
Treatments			
$M_{PE}25$		55a	1016b
$M_{PE}50$		56a	1268a
$M_{PE}75$		41c	1306a
$M_{PE}100$		49b	1294a
POE		59a	1210a
TF		54b	671c
Control		58a	1096b
Effects	*Df*	*P > F*	*P > F*
Year (Y)	2	<0.0001	0.0111
Treatment (T)	6	0.0674	<0.0001
T × Y	9	0.0817	0.5105

For each parameter (treatments and years), within each column, means followed by different letters are significantly different according to Tukey's multiple comparison tests ($p \leq 0.05$). $M_{PE}25$, $M_{PE}50$, $M_{PE}75$, $M_{PE}100$: pre-emergence treatment with 250, 500, 750, 1000 g a.i. ha^{-1} of metazachlor, respectively. POE: post-emergence treatment. TF: trifluralin treatment.

3.5. Economics

Concerning Exp. 1 (Table 8), the pre-emergence treatments, except for M_{PE}25, showed about 35 € ha^{-1} higher economic return than post-emergence, trifluralin and control, with not significant differences between conventional practice (M_{PE}100) and half dose of the same herbicide (M_{PE}50, Table 8). In Exp. 2, economic returns ranged from €287–334, €290–405, and €659–771 during 2008, 2009 and 2010, respectively. Moreover, in Exp. 2 (Table 8), control and post-emergence treatments were as well profitable as conventional practice (M_{PE}100) with no significant differences among herbicide regimes. Oilseed rape gross prices were similar (€472, €469, and €336 ha^{-1} for 2007, 2008, 2009 cropping seasons, respectively) during the analyzed cropping seasons.

Table 8. Economic aspects (€ ha^{-1} ± s.e.) of oilseed rape treated with different levels of herbicide input at Exp. 1 and Exp. 2 and over three growing seasons (2007–2008, 2008–2009, and 2009–2010, respectively).

Treatments	Net Economic Return (€ ha^{-1})					
	2008		2009		2010	
	Exp. 1	Exp. 2	Exp. 1	Exp. 2	Exp. 1	Exp. 2
M_{PE}25	–	–	524 ± 87b	288 ± 48	345 ± 58b	771 ± 129
M_{PE}50	655 ± 109a	334 ± 56	1051 ± 175a	290 ± 48	537 ± 90a	708 ± 118
M_{PE}75	620 ± 103a	327 ± 55	917 ± 153a	269 ± 45	661 ± 110a	706 ± 118
M_{PE}100	819 ± 136a	226 ± 38	1031 ± 172a	299 ± 50	586 ± 98a	680 ± 113
POE	331± 55b	287 ± 48	473 ± 79b	281 ± 47	290 ± 48b	681 ± 114
TF	566 ± 95a	311 ± 52	–	–	–	–
Control	337 ± 56b	325 ± 54	457 ± 76b	405 ± 67	334 ± 55b	659 ± 110

Means within each experiment and year followed by different letters are significantly different, according to Fisher's protected LSD test ($p \leq 0.05$). M_{PE}25, M_{PE}50, M_{PE}75, M_{PE}100: pre-emergence treatment with 250, 500, 750, 1000 g a.i. ha^{-1} of metazachlor, respectively. POE: post-emergence treatment. TF: trifluralin treatment.

4. Discussion

4.1. Oilseed Rape Injury

Our findings highlight that oilseed rape plants show substantial injuries at four weeks after application of the recommended metazachlor dose. Indeed, the highest metazachlor dose was accompanied by stunted growth and leaf chlorosis, suggesting that the recommended label dose of metazachlor caused serious effects on oilseed rape growth during the first phenological stages. Oilseed rape plants grown under metazachlor treatments of 750 g a.i. and 500 g a.i. ha^{-1} remained unaffected with no visible stunting or chlorosis. This might suggest that the photosynthesis process was not affected by low doses of metazachlor. Similar observations were reported for crops treated with other classes of herbicides [41]. Moreover, Jones et al. [42] found a decrease in phosphorus and potassium concentration in leaves of plants treated during pre-emergence phase and attributed the stunting to a reduced nutrient uptake or to a membrane leakage. Our findings are aligned with what is underscored by Vercampt et al. [20], who, on the basis of an experiment carried out in growth chamber condition, found that under exposure to the recommended metazachlor dose, the photosynthetic pigments content of oilseed rape plants underwent a variation. The decrease in photosynthetic pigments content caused a chlorosis of the leaves, indicating that the final output of photosynthetic process was addressed more to safeguard the photosynthesis system than to support the growth of the plant.

4.2. Weeds Trend and Herbicide Effectiveness

4.2.1. Experiment 1

All of the pre-emergence herbicide treatments used in Exp. 1 site across three growing seasons resulted in an efficient control of weeds, both at four and eight weeks after treatment application. In 2010, the applied treatments showed less effectiveness in controlling weeds than the previous two years, even if this increased competitiveness of weeds did not result in reduced crop yields. The relatively high weeds occurrence (both density and biomass) across all treatments in the last season (2009–2010) was probably related to the highest average temperatures (both maximum and minimum) during the first eight weeks of crop cycle (+1 °C of maximum temperature, and almost +1.5 °C of minimum temperature higher with respect to the temperature values of the same period of the previous years). Apparently, weeds were better able to benefit from mild temperatures than oilseed rape gaining a competitive advantage with regard to oilseed rape. Similarly, Lee [43] linked the dynamics of weed infestation to the higher temperatures that occurred throughout the crop-growing cycle. The author also reported that temperatures higher than a few degrees are enough to promote an early emergence and flowering time. Finally, he confirmed that higher temperatures considerably influenced the biomass production by annual species. Ramesh et al. [44] reported that both the herbicide effect on weeds and its presence in the soil was influenced by increasing temperatures ([44], and the references therein). In the first growing season, trifluralin did not have any significant effect on weed control, therefore confirming results reported by Naderi and Bijanzadeh [45], who highlighted that on the basis of a two-year study, trifluralin did not provide acceptable weed control throughout the oilseed rape growing cycle.

4.2.2. Experiment 2

Wild mustard, as also other species of *Brassicaceae* family, are poorly suppressed by metazachlor. Naderi and Ghadiri [46] showed that oilseed rape production was negatively affected by an increased number of wild mustard plants per unit area. Although in our study, a reduction in the coverage of wild mustard was noted throughout the experiment with some post-emergence treatment, we confirm that this weed represents a thorny issue due to its great competitiveness [47,48]. In order to prevent seed yield losses, it would be necessary to adopt different agricultural practices to effectively tackle this weed. Naderi and Bijanzadeh [45] put forth evidence that trifluralin at 1400 g a.i. ha^{-1} combined with two different tillage operations might successfully control wild mustard and wild oat. On the other hand, tillage operations over time might strongly impact the structure and fertility of the soil, also considering their crucial role in terms of impact in the farm energy balance [49,50]. As a consequence, appropriate crop rotations with grain cereals (e.g., durum wheat) can be effective to control wild mustard because it is easily controlled by herbicides commonly used for these crops.

4.3. Yield and Yield Characteristics

4.3.1. Experiment 1

The seed yield varied among years and treatments, indicating both biotic and abiotic influences.

The changes in seed yield were remarkable by decreasing metazachlor doses and time of application (250 g a.i. ha^{-1} both pre-emergence and post-emergence treatments). In this study, treatments carried out using high doses inhibited oilseed rape growth during the first stages of the crop, which resulted in plant density reduction in the harvest period. Notwithstanding, a rise in oilseed rape production was obtained following the same above-mentioned treatments. This might due to the fact that oilseed rape yield is highly dependent on each yield component. A reduction in plant density was likely associated with higher pod per plant due to an increased number of branches. Our results are confirmed by Diepenbrock [51], who reviewed that oilseed rape yield is able to adapt to different plant densities. Indeed, the number of branches is highly dependent on the plant habit;

thus, an increased number of plants per unit area caused a decline in the number of branches, and as a consequence a reduced number of flowers and pods [52,53]. Therefore, it is plausible that herbicide rate adjustments might be utilized to minimize weed competition in the first stages of development without compromising afterwards oilseed rape seed yield. However, the results that we have obtained are not solely attributable to the herbicide treatments.

The large differences among years within a treatment underscored by our study were consistent with recent findings relating to other environments. The oilseed rape yield is significantly influenced by the rainfall amount during the period between flowering and maturing [54,55], since there is a close and positive relation between the length of reproductive stages (flowering and maturing) and seed production. However, in typically Mediterranean climate, seasons may be either mild and wet or extremely dry. In our study, in 2008, seed filling fell during a period characterized by high temperatures and drought, affecting yield components, and as consequence seed production, as already stated by other authors [4,56] in their studies in Mediterranean-type climate.

4.3.2. Experiment 2

The lower yield obtained in 2008 compared with 2009 and 2010 was likely a consequence of different weather conditions (lower spring rains) and consequently in water soil availability for crops, so these experienced severe drought stress during the spring period soon after the beginning of flowering. In accordance with our analysis, Weymann et al. [55] showed that at the beginning of the pod and seed formation process, temperature and water-stress negatively influenced oilseed rape yield, whereas during the last phase of crop cycle, seed yield and oil content were only temperature-dependent. In 2008, rainfall absence during the period flowering-seed filling resulted in low water soil availability for crops. This environmental condition determined a reduction in production levels [55]. Indeed, at the seed-filling stage in 2010, no water-stress condition occurred because of the wetter spring season.

4.4. Economics

The fact that the analysis did not highlight any statistical difference in net returns among treatments and within each experiment, suggests that low-herbicide treatment was able to maintain yield without causing an economic loss. Our findings emphasized, as reported in other studies, that a farmer can reduce pesticide uses without undergoing significant economic losses [57–59]. Oilseed rape yield and net return were similar for both the treatments managed with the pre-emergence and the full labelled dose. These findings suggest that cropping system diversification by e.g., rotation with cereals or proposing low-herbicide managements might ensure the maintenance of good seed production as well as profitability and weed control, and hence promoting environmental and economic sustainability.

5. Conclusions

The development of sustainable agriculture from environmental and economic points of view is encouraged in Europe. Farmers are gaining interest in both attainment of economic return profits from eco-friendly technologies' adoption and crop planning that guarantees high yield. Our findings underscore that application of low-herbicide doses might guarantee good level of yield and economic return. In experiment 1, the net economic return of $M_{PE}50$ did not statistically differ from conventional treatment ($M_{PE}100$). In experiment 2, oilseed rape yields were statistically comparable in the range of the applied rates of metazachlor between $M_{PE}50$ and $M_{PE}100$. We focused on the possible production and economic effects of the application of different herbicide dose through two field trials, which can be considered representative of the Mediterranean-type environment where oilseed crop is grown in rotation with winter cereals. This study puts forth evidence that a decrease of herbicide doses might be considered a winning agronomic technique available to farmers in order to minimize environmental exploitation and maximize production level. Even if it is recognized that cutting doses of some herbicides could potentially induce the evolution of herbicide-resistant species, in the case of southern

Europe, the systematic adoption of crop rotation of oilseed rape with winter cereals is considered an agronomic strategy that is able to prevent the onset of this issue, especially when herbicides are used at reduced rates.

Author Contributions: L.L. and G.C. conceived and designed the experiments; P.A.D., G.C. and R.F. performed the experiments; P.A.D., G.C. and L.L. analysed the data; S.S. revised the paper; P.A.D. and L.L. wrote the paper.

Funding: This work was was jointly supported by the Italian Ministry of Agricultural, Food and Forestry Policies ("Bioenergie" project); and by the Sardinia Region ("Biocarburanti" project).

Acknowledgments: The authors are grateful to the technicians of the experimental farms for their technical support.

Conflicts of Interest: Authors have no competing interests to declare.

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