



Article

# Using Post-Emergence Herbicides in Combination with the Sowing Date to Suppress *Sinapis arvensis* and *Silybum marianum* in Durum Wheat

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Abstract: Wild mustard (*Sinapis arvensis* L.) and milk thistle (*Silybum marianum* (L.) Gaertn.) are two competitive broad-leaved weeds commonly found in cereals in Europe, while several weed species have developed resistance to the main herbicides that are applied on these crops. Thus, the implementation of integrated weed management (IWM) programs is of great importance. Field experiments were conducted based on a split-plot design with two factors (sowing date and herbicides). Our results showed that the density of wild mustard and milk thistle was higher in the early sowing compared to the late sowing, while the total weed density was up to 75% higher in early sowing. Moreover, the herbicides florasulam + 2.4-D and bromoxynil + 2.4-D exhibited high efficacy (>98%) against milk thistle and wild mustard, while tribenuron-methyl and florasulam + clopyralid provided greater efficacy in the late sowing compared to the early sowing. Among the four herbicides, the lowest dry biomass and grain yield of wheat were observed in tribenuron-methyl and florasulam + clopyralid, while in the weed-infested treatment, the highest values of both parameters were recorded in late sowing. Finally, the results showed that the sowing date is a cultural weed control method that should be implemented in IWM programs, since it can affect both weed density and herbicide efficacy.

Keywords: climate change; cultural practices; integrated weed management; milk thistle; wild mustard



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# 1. Introduction

Weed control in winter cereals is key to achieving high yields and, for many decades, it has been based on herbicide application. Due to the frequent application of herbicides with the same mode of action, the number of weed species with resistance to herbicides is constantly increasing. In recent years, several broad-leaved and grass weed species have developed resistance to acetolactate synthase (ALS) (e.g., sulfonylureas) and Acetyl-CoA carboxylase (ACCase) inhibitors (e.g., aryloxphenoxy-propionates (FOPs)), respectively. For instance, populations of wild oat (*Avena sterilis* L.), Italian ryegrass (*Lolium multiflorum* Lam.), and small canary grass (*Phalaris minor* Retz.) were reported to be resistant to aryloxphenoxy-propionate (FOPs) herbicides (e.g., fenoxaprop-P-ethyl, clodinafop-propargyl) [1–3]. Similarly, wild mustard (*Sinapis arvensis* L.), catchweed (*Galium aparine* L.), garland chrysanthemum (*Glebionis coronaria* L.), and corn poppy (*Papaver rhoeas* L.) are some examples of common broad-leaved weed species with resistance to various herbicides (e.g., tribenuron-methyl, mesosulfuron-methyl + iodosulfuron-methyl-sodium, and metsulfuron-methyl) belonging to the sulfonylureas class [1,4–6].

Adjustment of the sowing date is a cultivation practice that significantly contributes to weed management [7–10], as well as to the reduction in crop damage by insects [11] and diseases in cereals [12–14]. Moreover, it is also important to mention that this cultivation practice can affect grain yield [15,16] and quality parameters such as protein and gluten content [14]. Thus, due to climate change and the increased temperatures [17], an appropriate adjustment of the sowing date should be made for various crops [18,19].

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The increasing intensity of weed populations with resistance to herbicides highlights the importance of implementing integrated weed management programs in cereals production to limit this serious problem [9,20]. Interesting experimental findings addressing the impact of sowing date on the density of some weed species have been published. In a study conducted in Germany, Gerhards et al. [9] observed that the late sowing in autumn caused a noticeable reduction in black-grass (Alopecurus myosuroides Huds.) density in winter barley (Hordeum vulgare L.) and wheat compared to early sowing; it is important to point out that the sowing dates differed by about 10 days as a minimum. Similarly, Farooq and Cheema [21] examined the effects of three sowing dates on the weed density in wheat and observed that the highest values for this parameter were recorded in the early sowing. In contrast, Sharma et al. [22], in India, observed that the late sowing (December 25) of wheat caused a noticeable increase in the density and biomass of various weed species such as scarlet pimpernel (Anagalis arvensis L.) and fineleaf fumitory (Fumaria parviflora Lam.) compared to the early sowing (November 15). In another study conducted in Italy, Ingraffia et al. [10] observed that, in a wheat crop grown under a no-tillage system, the late sowing (around 20 December) increased the mean abundance of common chicory (Cichorium intybus L.) and bristly oxtongue (Helminthotheca echioides (L.) Holub; synonym Picris echioides L.) by up to 90.6% and 73.4%, respectively, compared to sowing carried out about a month earlier. In the latter study, similar results were observed for weed biomass. Thus, the above-mentioned results reveal that the impact of sowing date on weed flora depends on the weed species and the environmental conditions prevailing in an area.

Wild mustard and milk thistle (*Silybum marianum* (L.) Gaertn.) are broad-leaved weeds commonly found in cereal fields [23,24]. Both weed species exhibit high competitive ability against cereal crops such as wheat (*Triticum aestivum* L.) [23,24]. According to Zargar et al. [23], wheat yield losses due to wild mustard competition ranged from 8.9% to 56.5% depending on its density in the field. Similarly, Khan and Marwat [25] observed that milk thistle competition reduced wheat productivity by up to 37%, while its competitive ability depended on its density in the field and weather conditions, mainly rainfall, during the growing period of the crop. Based on these data, the effective control of both weed species is important to avoid high yield losses in cereal crops.

Therefore, the purpose of this study was to assess the effects of sowing (early and late) on the density of milk thistle and wild mustard, the efficacy of herbicides (tribenuron-methyl, florasulam + clopyralid, florasulam + 2.4-D, and bromoxynil + 2,4-D) against these broad-leaved weeds on the two sowing dates, and the interaction effects of both factors on durum wheat (*Triticum durum* Desf.) cultivation (e.g., plant height, relative chlorophyll content, dry biomass, and grain yield).

# 2. Materials and Methods

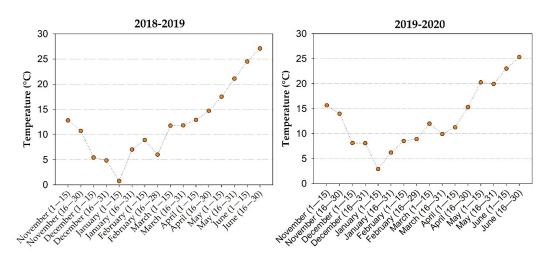
# 2.1. Experimental Site and Design

Field experiments were conducted in 2018/19 and 2019/20 at the experimental farm of the Department of Agriculture Crop Production and Rural Environment (DACPRE), University of Thessaly, in the Velestino region. In both seasons, the preceding crop was milk thistle. The latter species is a medicinal plant that is mainly cultivated for silymarin production [26], while it is also a weed with high competitive ability as mentioned above. The durum wheat variety Simeto was cultivated in both seasons. Sowing was performed with a cereal seeder machine in rows with a spacing of 0.18 m and at seeding rates of 250 and 270 kg ha<sup>-1</sup> in early and late sowing, respectively. The main cultural practices that were applied on the wheat are described in Table 1. The total recorded rainfall during the growing season (November to June) was 283.9 and 377.3 mm in 2018/19 and 2019/20, respectively, while the mean temperature for the same period was 12.4 and 13.1 °C in 2018/19 and 2019/20, respectively, as presented in Figure 1.

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Soil Properties	
sandy clay loam pH (1:1 soil to water ratio)	clay 26%, sand 38%, and silt 36% 7.4
Cultural practices	
Primary tillage	Field plowing at a depth of 25–30 cm in September
Secondary tillage	Field cultivator was used twice before sowing for seedbed preparation
Fertilization I. Basal application	The fertilizer 16–20–0 was applied at a rate of 300 kg ha <sup><math>-1</math></sup> (48, 60, and 0 kg N, P <sub>2</sub> O <sub>5</sub> , and K <sub>2</sub> O per ha, respectively) at sowing.
II. Top dressing	The inorganic fertilizer calcium ammonium nitrate (26–0–0) was applied at a rate of 300 kg ha $^{-1}$ (78, 0, and 0 kg N, P <sub>2</sub> O <sub>5</sub> , and K <sub>2</sub> O per ha, respectively) at the tillering stage (20 February in both growing seasons)

**Table 1.** Soil properties and main cultural practices applied on the wheat crop.



**Figure 1.** Mean air temperature (°C) during the growing period in 2018/19 (**left**) and 2019/20 (**right**).

The experiments were conducted based on a split-plot design with two factors and three replicates. The main plot factor was the sowing date, while the subplot factor was the herbicides. The treatments for both factors are described in Table 2, and the subplot size was 6  $\,\mathrm{m}^2$ . The early sowing was through 15 November, while the late sowing took place after the middle of November. The exact sowing dates were chosen based on the weather conditions, mainly from the rainfall recorded in early November. Moreover, the herbicides were applied at the tillering stage using a field plot sprayer with flat fan nozzles, a spray volume of 300 L ha $^{-1}$ , and a pressure of 250 kPa.

# 2.2. Sampling and Measurements

To determine the impact of the sowing date and the herbicides on the durum wheat crop, several plant parameters (height, aboveground dry biomass, relative chlorophyll content, 1000-grain weight, ear length, and seed yield) were recorded. The plant's height and ear length were measured in 5 representative plants randomly selected from the central rows of each subplot on 7 May and 5 May in 2019 and 2020, respectively. Moreover, the relative chlorophyll content (SPAD readings) was measured in flag leaves at the same sampling date using the SPAD-502 chlorophyll content meter. This method has also been used in other studies [27,28]. Moreover, to determine the dry biomass, wheat samples were

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taken by cutting plants on an area of  $0.5~\rm{m}^2$  in each subplot, and were dried in an oven at  $60~\rm{^{\circ}C}$  for  $96~\rm{h}$ .

<b>Table 2.</b> Description of treatments for sowing date and herbic
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Main Plot Factor (Sowing Date)	
Treatments	Early sowing: 13 and 7 November in 2018 and 2019, respectively Late sowing: 23 and 19 November in 2018 and 2019, respectively
Subplot factor (Herbicides)	
Treatments	Weed-infested Weed-free florasulam + 2.4-D (dose: $5 + 240$ g a.i. (active ingredient) ha <sup>-1</sup> ) bromoxynil + 2.4-D (dose: $420 + 420$ g a.i. ha <sup>-1</sup> ) tribenuron-methyl (dose: $15$ g a.i. ha <sup>-1</sup> ) florasulam+ clopyralid (dose: $3.75 + 45$ g a.i. ha <sup>-1</sup> )

The harvest was conducted mechanically (harvest width 1.4 m) on 19 June and 2 July in 2019 and 2020, respectively. The delay of the harvest in the second growing season was due to a technical failure of the experimental harvesting machine. In addition, following the harvest, the 1000-grain weight was calculated by weighing three samples of 100 grains per subplot [29].

The density and biomass of the milk thistle, wild mustard, and other broad-leaved weeds were measured on 8 May and 6 May in 2019 and 2020, respectively, on an area of 0.5 m<sup>2</sup> in each subplot. Drying of the weed samples was carried out by the same method as previously described for wheat. Finally, the herbicide efficacy (HE) was determined based on the weed biomass (WB) data using the following equation [30]:

$$\mbox{HE}(\%) = 100 \times \frac{(\mbox{WB in the weed infested plots}) - (\mbox{WB in the herbicide plots})}{\mbox{WB in the weed infested plots}}$$

# 2.3. Statistical Analysis

The data for both the herbicide efficacy and the wheat parameters are expressed as mean values of three replications. The statistical analysis was performed using SigmaPlot 12 software (Systat Software, San Jose, CA, USA). A two-way analysis of variance (ANOVA) was used to determine the effects of the sowing date, the herbicides, and their interaction on the parameters under investigation. Fisher's least significant difference (LSD) test was then used to determine the significance of the differences between the treatments for the main plot and subplot factors at p = 0.05. The data are presented separately for the sowing date and the herbicide treatments when there was no interaction between the two factors. The statistical analysis of the wild mustard, milk thistle, and total weed density was conducted according to a randomized complete block design (RCBD) with 3 replications. The LSD test was also used to determine the significance of the differences between the sowing dates.

# 3. Results

# 3.1. Weed Density

The density of the main weeds was recorded at the early and late sowing treatments. Milk thistle and wild mustard were the weed species studied in two experiments. Our results revealed that the sowing date affected the density of these weeds (Table 3). The milk thistle density was higher in the early sowing treatment compared with the late sowing in both experimental years. Similar results were recorded for wild mustard. The greatest density of this species (7 and 22 plants  $m^{-2}$  in 2018/19 and 2019/20, respectively) was observed in the early sowing. Moreover, the total weed density was up to 75% higher in the

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early sowing compared to that in late sowing. Generally, the greatest values for milk thistle, wild mustard, and total weed density were recorded in the second experimental year.

**Table 3.** Milk thistle, wild mustard, and total weed density as affected by the sowing date (early and late sowing).

		2018/19			
Sowing Date	Milk Thistle (Plants $m^{-2}$ )	Wild Mustard (Plants $m^{-2}$ )	Total Weed Density (Plants $m^{-2}$ )		
Early sowing	11.75 a <sup>1</sup>	7.00 a	57.75 a		
Late sowing	1.00 b	2.50 b	17.50 b		
LSD <sub>5%</sub>	9.190	4.478	31.342		
ANOVA					
F values	25.329 *	18.692 *	30.53 *		
		2019/20			
Sowing Date	Milk Thistle (Plants m <sup>-2</sup> )	Wild Mustard (Plants $m^{-2}$ )	Total Weed Density (Plants $m^{-2}$ )		
Early sowing	31.67 a	22.00 a	72.67 a		
Late sowing	7.00 b	7.33 b	18.00 b		
LSD <sub>5%</sub>	12.503	11.740	12.748		
ANOVA					
F values	72.053 *	28.896 *	340.453 **		

 $<sup>^{\</sup>bar{1}}$  Means followed by the same letters do not significantly differ with Fisher's least significant difference (LSD) test. ns: not significant; \* and \*\* indicate significant differences at p < 0.05 and p < 0.01, respectively.

# 3.2. Herbicides Efficacy

In both years, the herbicides bromoxynil + 2.4-D and florasulam + 2.4-D exhibited high efficacy (>99%) against milk thistle, and it is important to point out that the sowing date had no significant effect on the efficacy of these herbicides (Table 4). In contrast, the impact of the sowing date on tribenuron-methyl and florasulam + clopyralid efficacy was noticeable, because there was an increase in the efficacy of both herbicides in late sowing. Among the four herbicides evaluated in this study, tribenuron-methyl provided the lowest efficacy (49.5–72%) against milk thistle. Concerning the herbicides' efficacy against wild mustard, our results revealed that the highest efficacy was recorded for the herbicides bromoxynil + 2.4-D and florasulam + 2.4-D, while florasulam + clopyralid exhibited the lowest efficacy (82.1–86.5%). Regarding the effects of the sowing date on the herbicides' efficacy against wild mustard, our results showed that the efficacy of herbicides tribenuron-methyl and florasulam + clopyralid was greater in late sowing.

# 3.3. Relative Chlorophyll Content (SPAD Readings)

The relative chlorophyll content in 2018/19 was higher than in 2019/20. In 2018/19, the sowing date did not affect the SPAD readings ( $F_{sowing date(SD)} = 1.486$ , p = 0.235), while among the herbicide treatments, significant differences ( $F_{herbicides(H)} = 13.673$ , p < 0.001) were observed with the highest values recorded in florasulam + 2.4-D, bromoxynil + 2.4-D, and the weed-free treatments (Figure 2). The lowest SPAD values were recorded in the weed-infested treatment and were up to 9.1% lower compared to that in florasulam + 2.4-D. In 2019/20, there was an interaction between the sowing date and the herbicides ( $F_{SD \times H} = 3.417$ , p = 0.018). The SPAD values in the weed-infested treatment were about 7% higher in late sowing compared to that in early sowing. Among the four herbicides, the lowest SPAD values were observed in tribenuron-methyl and florasulam + clopyralid.

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**Table 4.** Herbicides' efficacy as affected by the sowing date (early and late sowing) and the herbicide treatments (bromoxynil + 2.4-D, florasulam + 2.4-D, florasulam + clopyralid, tribenuron-methyl, weed-free, and weed-infested).

	Herbicides Efficacy (%)					
2018/19	Sowing Date Sowing Da		g Date			
	Early Sowing	Late Sowing	LSD <sub>5%</sub>	Early Sowing	Late Sowing	LSD <sub>5%</sub>
Herbicides	Milk Thistle			Wild Mustard		
Bromoxynil + 2.4-D	99.8 aA	100 aA		100 aA	100 aA	
Florasulam + 2.4-D Florasulam + clopyralid Tribenuron-methyl Weed-free Weed-infested LSD <sub>5%</sub>	99.3 aA 85.1 bB 49.5 cB 100 aA 0 dA	100 aA 90.1 aA 57.1 aA 100 aA 0 bA	2.090	100 aA 82.1 cB 86.9 bB 100 aA 0 dA	100 aA 86.5 cA 90.3 bA 100 aA 0 dA	0.702
$\begin{array}{l} ANOVA \ (F \ values \ and \ sign \\ F_{sowing \ time(ST)} \\ F_{herbicides(H)} \\ F_{ST \ x \ H} \end{array}$	nificant differences) 4.632 * 1052.836 *** 1.652 ns			14.540 *** 8852.299 *** 5.996 ***		
2019/2020	Sowing Date			Sowin	g Date	
2017/2020	Early Sowing	Late Sowing	LSD <sub>5%</sub>	Early Sowing	Late Sowing	LSD <sub>5%</sub>
Herbicides	Milk thistle			Wild mustard		
Bromoxynil + 2.4-D Florasulam + 2.4-D Florasulam + clopyralid Tribenuron-methyl Weed-free Weed-infested LSD <sub>5%</sub>	100 aA 99.6 aA 76.7 bB 64.0 cB 100 aA 0 dA	100 aA 100 aA 83.5 bA 72.0 cA 100 aA 0 dA	2.26	98.5 aA 99.3 aA 83.4 cA 92.4 bA 100 aA 0 dA	100 aA 100 aA 85.6 bA 98.9 aA 100 aA 0 cA	-
ANOVA (F values and sign	nificant diffe	rences)				
$F_{sowing \ date(SD)}$ $F_{herbicides(H)}$ $F_{SD \ \times \ H}$	5.239 * 3.785 ns 841.083 *** 1186.084 ** 1.967 ns 1.128 ns		084 ***			

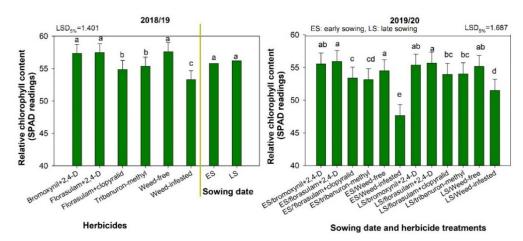
 $<sup>\</sup>overline{1}$  For each factor, means followed by the same letters (capital letter for sowing date and lower letter for herbicides) do not significantly differ with Fisher's least significant difference (LSD) test. ns: not significant; \* and \*\*\* indicate significant differences at p < 0.05 and p < 0.001, respectively.

# 3.4. Durum Wheat Growth

The statistical analysis of the plant's height data revealed that sowing time had an impact on this parameter only in 2019/20 with the highest values recorded in early sowing (Table 5). In both years, there were noticeable differences among the herbicide treatments. The lowest plant height was observed in the weed-infested treatment.

Regarding the dry biomass of the durum wheat crop, in both years, there was an interaction between the sowing date and the herbicides ( $F_{SD \times H} = 15.561$ , p < 0.001 and  $F_{SD \times H} = 4.062$ , p < 0.01, in 2018/19 and 2019/20, respectively). The dry biomass in the weed-infested treatment was about 7% higher in the late sowing compared to that in the early sowing. In contrast, the dry biomass was higher in the early sowing than in the late sowing for the florasulam + 2.4-D and bromoxynil + 2.4-D treatments (Figure 3). Among the four herbicides, the lowest dry biomass was observed in tribenuron-methyl and florasulam + clopyralid.

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**Figure 2.** Relative chlorophyll content (SPAD readings) as affected by the sowing date (early and late sowing) and the herbicide treatments (bromoxynil + 2.4-D, florasulam + 2.4-D, florasulam + clopyralid, tribenuron-methyl, weed-free, and weed-infested). Means followed by the same letters do not significantly differ with Fisher's least significant difference (LSD) test. Error bars indicate the LSD values.

**Table 5.** Durum wheat plant's height (cm) as affected by the sowing date (early and late sowing) and the herbicide treatments (bromoxynil + 2.4-D, florasulam + 2.4-D, florasulam + clopyralid, tribenuron-methyl, weed-free, and weed-infested).

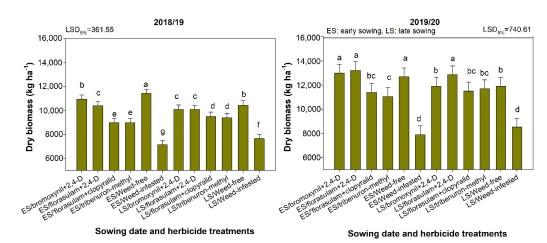
Factors/Treatments	Plant's Height (cm)			
ractors/ freatments	2018–2019	2019–2020		
Sowing Date				
Early sowing	85.1 a <sup>1</sup>	92.8 a		
Late sowing	84.9 a	90.4 b		
LSD <sub>5</sub> %	-	1.936		
Herbicides				
Bromoxynil + 2.4-D	85.1 ab	93.2 a		
Florasulam + 2.4-D	87.1 a	93.0 a		
Florasulam + clopyralid	85.6 ab	92.3 a		
Tribenuron-methyl	81.4 c	90.3 a		
Weed-free	87.1 a	92.9 a		
Weed-Infested	83.1 bc	87.9 b		
LSD <sub>5%</sub>	3.222	3.353		
ANOVA (F values and significant	differences)			
F <sub>sowng date(SD)</sub>	0.05116 ns	6.688 *		
F <sub>herbicides(H)</sub>	3.957 **	3.258 *		
$F_{SD \times H}$	0.103 ns	0.929 ns		

 $<sup>\</sup>overline{\phantom{a}}$  For each factor, means followed by the same letters do not significantly differ with Fisher's least significant difference (LSD) test. ns: not significant; \* and \*\* indicate significant differences at p < 0.05 and p < 0.01, respectively.

### 3.5. Yield and Its Components

The sowing date had no impact on ear length and 1000-grain weight (Table 6). Regarding the effects of the herbicides on these parameters, our results revealed that there were noticeable differences. The lowest values (5.63 and 5.95 cm in 2019/20 and 2018/19, respectively) of ear length were observed in the weed-infested treatment, while among the four herbicides the lowest values were recorded in florasulam + clopyralid and tribenuronmethyl. A similar trend was observed for the 1000-grain weight.

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**Figure 3.** The dry biomass (kg ha $^{-1}$ ) of the durum wheat crop as affected by the sowing date (early and late sowing) and the herbicide treatments (bromoxynil + 2.4-D, florasulam + 2.4-D, florasulam + clopyralid, tribenuron-methyl, weed-free, and weed-infested). Means followed by the same letters do not significantly differ with Fisher's least significant difference (LSD) test. Error bars indicate the LSD values.

**Table 6.** Ear length (cm) and 1000-grain weight (g) as affected by the sowing date (early and late sowing) and the herbicide treatments (bromoxynil + 2.4-D, florasulam + 2.4-D, florasulam + clopyralid, tribenuron-methyl, weed-free, and weed-infested).

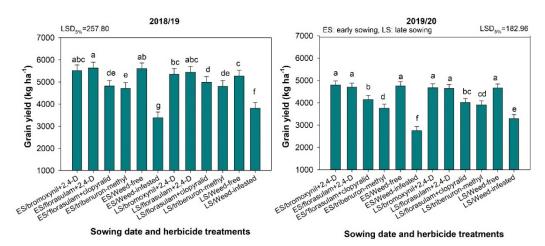
Factors/	1000-Grain Weight (g)		Ear Length (cm)	
Treatments	2018–2019	2019–2020	2018–2019	2019–2020
Sowing Date				
Early sowing	54.61 a <sup>1</sup>	53.35 a	6.34 a	6.06 a
Late sowing	54.31 a	53.32 a	6.31 a	6.05 a
LSD <sub>5%</sub>	-	-	-	-
Herbicides				
Bromoxynil + 2.4-D	56.25 a	53.67 a	6.52 a	6.21 a
Florasulam + 2.4-D	56.58 a	54.80 a	6.44 a	6.20 a
Florasulam + clopyralid	54.20 b	54.09 a	6.26 b	6.08 ab
Tribenuron- methyl	54.25 b	54.68 a	6.29 b	6.00 b
Weed-free	56.15 a	53.28 a	6.49 a	6.22 a
Weed-infested	49.34 c	49.52 b	5.95 c	5.63 c
LSD <sub>5%</sub>	1.347	1.778	0.143	0.153
ANOVA (Fvalues	and significant diffe	rences)		
F <sub>sowing dateSD)</sub>	0.630 ns	0.0036 ns	0.367 ns	0.0857 ns
F <sub>herbicides(H)</sub>	34.640 ***	10.368 ***	18.947 ***	18.694 ***
$F_{SD x H}$	0.390 ns	0.344 ns	0.926 ns	1.275 ns

<sup>&</sup>lt;sup>1</sup> For each factor, means followed by the same letters do not significantly differ with Fisher's least significant difference (LSD) test. ns: not significant; \*\*\* indicate significant differences at p < 0.001.

In 2018/19, the grain yield ranged from 3381.9 to 5636.1 kg ha $^{-1}$ , while in 2019/20, it ranged from 2746.7 to 4800.2 kg ha $^{-1}$  (Figure 4). In both years, there was an interaction between the sowing date and the herbicides ( $F_{\rm SD \times H} = 5.095$ , p < 0.01 and  $F_{\rm SD \times H} = 8.727$ , p < 0.001 in 2018/19 and 2019/20, respectively). The grain yield in the weed-infested treatment was up to 16.4% higher in the late sowing compared to the early sowing. In contrast, the grain yield was higher in the early sowing than in the late sowing for florasulam + 2.4-D, bromoxynil + 2.4-D,

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and the weed-free treatments. Among the four herbicides, the lowest dry biomass was observed in tribenuron-methyl and florasulam + clopyralid.



**Figure 4.** Grain yield (kg  $ha^{-1}$ ) as affected by the sowing date (early and late sowing) and the herbicide treatments (bromoxynil + 2.4-D, florasulam + 2.4-D, florasulam + clopyralid, tribenuron-methyl, weed-free, and weed-infested). Means followed by the same letters do not significantly differ with Fisher's least significant difference (LSD) test. Error bars indicate the LSD values.

### 4. Discussion

# 4.1. Weed Density and Sowing Date

The sowing date affected the total weed density with the highest values recorded in the early sowing. In another study, Farooq and Cheema [21] reported that at 80 days after sowing (DAS), the total weed density in the wheat crop was decreased by 35.1–45.2% on the mid-sowing date (30 November) compared with that in early sowing (15 November). The total weed density was significantly reduced by delaying seeding until December 15. Furthermore, our findings clearly demonstrated that the late sowing resulted in reductions in the densities of milk thistle and wild mustard of 64.2–66.7% and 77.9–91.4%, respectively. Similar results have also been observed for other weed species. Gerhards et al. [9] observed that the late sowing of winter cereals decreased the density of black-grass by 43% (average value) in comparison with the early sowing, while García et al. [31] reported that wheat or barley sowing in December caused a high reduction (88.0–99.3%) in great brome (*Bromus diandrus* Roth) density compared with sowing in November. In India, Singh et al. [32] observed that the highest total weed biomass was recorded in the early sowing (November 25) compared to the late sowing (December 10). Late sowing can also reduce weed seed production as reported for great brome [31].

The reduction in weed density on the late sowing date was related to the lower temperatures recorded during this period compared to that in the early sowing period. Similarly, Farooq and Cheema [21] reported that the higher weed density in the early sowing treatment was due to the higher temperatures recorded compared to that in late sowing, which favored the germination of seeds. Additionally, lower temperatures induced secondary dormancy in the seeds of black-grass, which contributed to the reduction in plant density in the late sowing date [9]. As mentioned above, the milk thistle and wild mustard density were greatly affected by the sowing date. These results were probably due to the high temperature requirements of both weed species for seed germination. According to Parmoon et al. [33] and Elahifard et al. [34], the estimated base temperatures (Tb) for the germination of milk thistle and wild mustard were 5.2 °C and 2.8–5 °C, respectively, while the optimum temperatures for both species were higher than 20 °C.

# 4.2. Herbicide Efficacy

Florasulam + clopyralid and tribenuron-methyl provided lower control of milk thistle and wild mustard than florasulam + 2,4-D and bromoxynil + 2.4-D (Figure 5). In another

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study, the herbicide bromoxynil + MCPA provided high efficacy against both species, reducing their biomass by up to 100% [35]. Similarly, florasulam + 2.4-D efficacy against wild mustard ranged from 94% to 96% [36]. Our results also indicated that the efficacy of florasulam + clopyralid and tribenuron-methyl against wild mustard was up to 86.5% and 98.9%, respectively. Moreover, the application of tribenuron-methyl (18.75 g a.i. ha<sup>-1</sup>) in a mixture with the graminicide clodinafop–propargyl led to a decrease of 63.7%–100% in wild mustard biomass [35]. In contrast, Zargar et al. [23] reported lower efficacy (75%) of tribenuron-methyl against wild mustard, while Pala [36] observed that the application of tribenuron-methyl in a mixture with pinoxaden or fenoxaprop-p-ethyl also provided low control (7476%) of this weed. It is also important to point out that the milk thistle control by the tribenuron-methyl ranged between 49.5% and 72%. Similarly, tribenuron-methyl in a mixture with clodinafop–propargyl provided poor control (64.9%) of this species [35].



**Figure 5.** (a) Experimental field in early December of 2019, (b) milk thistle density in weed-infested plots (5 May 2020), (c) necrosis of milk thistle plants in the bromoxynil + 2,4-D treatment (5 May 2020), and (d) milk thistle plant growth in tribenuron-methyl plots (5 May 2020).

The results of the present study also showed that the sowing date had no impact on florasulam + 2.4-D and bromoxynil + 2.4-D efficacy against wild mustard and milk thistle. In contrast, the late sowing date led to an increase in the efficacy of tribenuron-methyl and florasulam + clopyralid against these weed species. Similar results were observed for great brome by García et al. [37], who mentioned that the mesosulfuron-methyl + iodosulfuron-methyl-sodium provided better control of this weed species in the late sowing compared to the early sowing. It is well known that weed size is a factor affecting the efficacy of foliar applied herbicides [38,39]. The growth stage of plants at the time of application affected the efficacy of pinoxaden on black-grass in a wheat crop, with the highest control levels seen when pinoxaden was applied to black-grass at earlier growth stages [39]. Accordingly, in our study, the better efficacy of florasulam + clopyralid and tribenuron-methyl against

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broad-leaved weeds in late planting was attributable to the weeds being smaller at the time of herbicide application than they were in the early sowing.

# 4.3. Durum Wheat Growth and Yield

Both the sowing date and the use of herbicides had an impact on the growth of durum wheat in the two years. Early sowing resulted in higher values for plant height; however, weed competition in the weed-infested plots significantly reduced this measure. Similar results were also observed by Karkanis et al. [28], who mentioned that plant height in weedinfested plots was lower by up to 13% compared to that in the herbicide plots. Moreover, there was an interaction between the two factors for aboveground biomass and grain yield. In the weed-infested treatment, biomass accumulation and grain yield were higher in the late sowing compared to early sowing since the weed competition was more intense in the early sowing, negatively affecting the plant's growth and the chlorophyll content. The latter results were confirmed by the positive correlation (r = 0.795 (n = 34, p = 0.001) and r = 0.861 (n = 34, p = 0.001) in 2018/19 and 2019/20, respectively) recorded between the SPAD readings and the dry biomass. In contrast, with the use of florasulam + 2.4-D and bromoxynil + 2.4-D, the herbicides with the highest performance, the grain yield was higher in the early sowing compared to the late sowing without these differences being statistically significant. The impact of the sowing date (with or without weed competition) on wheat productivity was examined in previous studies [32,37,40,41]. In Spain, García et al. [37] observed that the early sowing of wheat in mid-October caused a reduction in the wheat yield compared to sowing in mid-November or mid-December due to the greater competitiveness of great brome on this sowing date. Similarly, in India, Singh et al. [32] reported that the wheat grain yield was higher by 18.7% in the late sowing compared to early sowing, while there were no differences between the sowing dates for 1000-grain yield, a result that was also observed in our study. Due to the high temperatures that occurred during grain filling, sowing in Iran on November 20 resulted in a lower grain yield than sowing in early November [40]. Thus, both weed competition and the environmental conditions during the growing season can have an impact on the effect of the planting date on wheat productivity.

### 5. Conclusions

The results of this study showed that the density of wild mustard and milk thistle significantly differed between the two sowing dates, with the highest values recorded in the early sowing. Florasulam + 2.4-D and bromoxynil + 2.4-D were the most effective herbicides for the control of both weed species, while tribenuron-methyl and florasulam + clopyralid were less effective. It is also important to mention that the performance of the less effective herbicides was improved in the late sowing treatment due to the decreased growth of weeds at the time of application. Regarding the effects of the two factors on biomass and grain yield in durum wheat, our results revealed that, in the early sowing, the highest values of these parameters were recorded in the weed-free, florasulam + 2.4-D, and bromoxynil + 2.4-D treatments, due to the high weed control in these treatments, while in the late sowing, the grain yield and dry biomass of durum wheat were higher in the other treatments. In conclusion, this study shows that by integrating the sowing date with herbicides, weed management was improved without reducing crop productivity.

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**Data Availability Statement:** The data supporting the findings of this study are available within this article.

**Conflicts of Interest:** The authors declare no conflict of interest.

### References

1. Travlos, I.; Tsekoura, A.; Antonopoulos, N.; Kanatas, P.; Gazoulis, I. Novel Sensor-based Method (Quick Test) for the In-season Rapid Evaluation of Herbicide Efficacy under Real Field Conditions in Durum Wheat. *Weed Sci.* **2021**, *69*, 147–160. [CrossRef]

- 2. Buddenhagen, C.E.; James, T.K.; Ngow, Z.; Hackell, D.L.; Rolston, M.P.; Chynoweth, R.J.; Gunnarsson, M.; Li, F.; Harrington, K.C.; Ghanizadeh, H. Resistance to Post-Emergent Herbicides is Becoming Common for Grass Weeds on New Zealand wheat and barley farms. *PLoS ONE* **2021**, *16*, e0258685. [CrossRef] [PubMed]
- 3. Wu, C.; Song, M.; Zhang, T.; Zhu, C.; Liu, W.; Jinn, T.; Zhao, N. Target-site Mutation and Cytochrome P450s Confer Resistance to Multiple Herbicides in Italian Ryegrass (*Lolium multiflorum* Lam.) from China. *Crop Prot.* **2022**, *161*, 106068. [CrossRef]
- 4. Sin, B.; Kadioglu, I. Trp-574-Leu mutation in Wild Mustard (*Sinapis arvensis* L.) as a result of Als Inhibiting Herbicide Applications. *PeerJ* **2021**, *9*, e11385. [CrossRef] [PubMed]
- 5. Kati, V.; Scarabel, L.; Thiery-Lanfranchi, D.; Kioleoglou, V.; Liberopoulou, S.; Délye, C. Multiple Resistance of *Papaver rhoeas* L. to 2,4-D and Acetolactate Synthase Inhibitors in four European Countries. *Weed Res.* **2019**, *59*, 367–376. [CrossRef]
- Hada, Z.; Khammassi, M.; Jenfaoui, H.; Menchari, Y.; Torra, J.; Souissi, T. Field Survey and Resistance Occurrence to ALS-Inhibiting Herbicides in *Glebionis coronaria* L. in Tunisian Wheat Crops. *Plants* 2020, 9, 1210. [CrossRef]
- 7. Bónis, P.; Balázs, F.; Balázs, J.; Kismányoky, T. Effect of Sowing Date on the Weed Infestation of Winter Wheat a in Long-term Experiment. *Acta Agron. Hung.* **2010**, *58*, 69–74. [CrossRef]
- Sharma, N.; Kumar, A.; Sharma, B.C.; Chand, L.; Sharma, V.; Kumar, M. Effects of Sowing Dates and Weed Management on Productivity of Irrigated Wheat (*Triticum aestivum*). *Indian J. Agric. Sci.* 2020, 90, 556–559.
- 9. Gerhards, R.; Messelhäuser, M.H.; Sievernich, B. Suppressing *Alopecurus myosuroides* in Winter Cereals by Delayed Sowing and Pre-Emergence Herbicides. *Plant Soil Environ.* **2022**, *68*, 290–298. [CrossRef]
- 10. Ingraffia, R.; Amato, G.; Ruisi, P.; Giambalvo, D.; Frenda, A.S. Early Sowing Can Boost Grain Production by Reducing Weed Infestation in Organic No-till Wheat. *J. Sci. Food Agric.* **2022**, *102*, 6246–6254. [CrossRef] [PubMed]
- 11. Karadjova, O.; Krusteva, H. Species Composition and Population Dynamics of the Harmful Insect Fauna (Hemiptera: Cicadomorpha, fulgoromorpha and sternorrhyncha) of Winter Triticale. *Bulg. J. Agric. Sci.* **2016**, 22, 619–626.
- 12. Cook, R.J.; Polley, R.W.; Thomas, M.R. Disease-Induced Losses in Winter Wheat in England and Wales 1985–1989. *Crop Prot.* **1991**, 10, 504–508. [CrossRef]
- 13. Gutteridge, R.J.; Hornby, D. Effects of Sowing Date and Volunteers on the Infectivity of Soil Infested with *Gaeumannomyces graminis* var. *tritici* and on Take-all Disease in Successive Crops of Winter Wheat. *Ann. Appl. Biol.* **2003**, 143, 275–282. [CrossRef]
- 14. Arata, G.J.; Martínez, M.; Elguezábal, C.; Rojas, D.; Cristos, D.; Dinolfo, M.I.; Arata, A.F. Effects of Sowing Date, Nitrogen Fertilization, and *Fusarium graminearum* in an Argentinean Bread Wheat: Integrated Analysis of Disease Parameters, Mycotoxin Contamination, Grain Quality, and Seed Deterioration. *J. Food Compos. Anal.* 2022, 107, 104364. [CrossRef]
- 15. Dwivedi, S.K.; Kumar, S.; Mishra, J.S.; Haris, A.A.; Singh, S.K.; Srivastava, A.K.; Kumar, A.; Kumar, V.; Singh, S.; Bhatt, B.P. Effect of Moisture Regimes and Sowing Dates on Wheat Physiological Process and Yield Attributes under Rain-fed Ecosystem in Eastern Indo Gangetic Plain. *Plant Physiol. Rep.* **2019**, *24*, 46–53. [CrossRef]
- 16. Ali, K.A.; Muhammad Amin, S.; Abdullah, R.A. Late Sowing Date Influence on Wheat and Triticale Crop Yields as a Draught Management Tool. *J. Saudi Soc. Agric. Sci.* **2021**, *20*, 353–358. [CrossRef]
- 17. Karkanis, A.; Ntatsi, G.; Alemardan, A.; Petropoulos, S.; Bilalis, D. Interference of Weeds in Vegetable Crop Cultivation, in the Changing Climate of Southern Europe with Emphasis on Drought and Elevated Temperatures: A Review. *J. Agric. Sci.* **2018**, 156, 1175–1185. [CrossRef]
- 18. Islam, A.T.; Islam, A.S.; Islam, G.T.; Bala, S.K.; Salehin, M.; Choudhury, A.K.; Dey, N.C.; Hossain, A. Adaptation Strategies to Increase Water Productivity of Wheat Under Changing Climate. *Agric. Water Manag.* **2022**, *264*, 107499. [CrossRef]
- 19. Lopes, M.S. Will Temperature and Rainfall Changes Prevent Yield Progress in Europe? *Food Energy Secur.* **2022**, *11*, e372. [CrossRef]
- 20. Papapanagiotou., A.P.; Damalas, C.A.; Bosmali, I.; Madesis, P.; Menexes, G.; Eleftherohorinos, I. Multiple Resistance of Silky Windgrass to Acetolactate synthase- and Acetyl-CoA Synthase-inhibiting Herbicides. *Weed Technol.* **2022**, *36*, 334–343. [CrossRef]
- 21. Farooq, O.; Cheema, Z.A. Influence of Sowing Dates and Planting Methods on Weed Dynamics in Wheat Crop. *Pak. J. Agric. Sci.* **2014**, *51*, 817–825.
- 22. Sharma, N.; Kumar, A.; Sharma, B.; Kumar, R.; Verma, A.; Kumar, M. Effects of Sowing Dates and Weed Management on Weed Growth and Nutrients Depletion by Weeds and Uptake by Wheat (*Triticum aestivum* L.) Under Shiwalik Foothills Plains of Jammu. *Bangladesh J. Bot.* **2021**, *50*, 311–318. [CrossRef]
- 23. Zargar, M.; Kavhiza, N.J.; Bayat, M.; Pakina, E. Wild Mustard (*Sinapis arvensis*) Competition and Control in Rain-Fed Spring Wheat (*Triticum aestivum* L.). *Agronomy* **2021**, *11*, 2306. [CrossRef]
- 24. Ahmad, I.; Gul, B. Competitive Ability of Wheat Crop against Different Densities of *Avena fatua* and *Silybum marianum*. *Sarhad J. Agric*. **2021**, *37*, 631–638. [CrossRef]

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25. Khan, M.A.; Marwat, K.B. Impact of Crop and Weed Densities on Competition between Wheat and *Silybum marianum* Gaertn. *Pak. J. Bot.* **2006**, *38*, 1205–1215.

- 26. Liava, V.; Karkanis, A.; Tsiropoulos, A. Yield and Silymarin Content in Milk Thistle (*Silybum marianum* (L.) Gaertn.) Fruits Affected by the Nitrogen Fertilizers. *Ind. Crops Prod.* **2021**, *171*, 113955. [CrossRef]
- 27. Islam, M.R.; Shamsul Haque, K.M.S.; Akter, N.; Abdul Karim, M. Leaf Chlorophyll Dynamics in Wheat Based on SPAD Meter Reading and its Relationship with Grain Yield. *Sci. Agric.* **2014**, *8*, 13–18.
- 28. Karkanis, A.; Veliios, E.; Grigoriou, F.; Gkrimpizis, T.; Giannouli, P. Evaluation of Efficacy and Compatibility of Herbicides with Fungicides in Durum Wheat (*Triticum durum* Desf.) under Different Environmental Conditions: Effects on Grain Yield and Gluten Content. *Not. Bot. Horti. Agrobo.* 2018, 46, 601–607. [CrossRef]
- 29. Arampatzis, D.A.; Karkanis, A.C.; Tsiropoulos, N.G. Impact of Plant Density and Mepiquat Chloride on Growth, Yield, and Silymarin Content of *Silybum marianum* Grown under Mediterranean Semi-Arid Conditions. *Agronomy* **2019**, *9*, 669. [CrossRef]
- 30. Yadav, S.K.; Lal, S.S.; Srivastava, A.K.; Bag, T.K.; Singh, B.P. Efficacy of Chemical and Non-Chemical Methods of Weed Management in Rainfed Potato (*Solanum tuberosum*). *Indian J. Agric. Sci.* **2015**, *85*, 382–386.
- 31. García, A.L.; Royo-Esnal, A.; Torra, J.; Recasens, J. Integrated Effect of Crop Sowing Date and Herbicide Stress on Fitness of *Bromus diandrus* Roth. *Span. J. Agric. Res.* **2015**, *13*, e1001. [CrossRef]
- 32. Singh, M.K.; Mishra, A.; Khanal, N.; Prasad, S.K. Effects of Sowing Dates and Mulching on Growth and Yield of Wheat and Weeds (*Phalaris minor Retz.*). *Bangladesh J. Bot.* **2019**, *48*, 75–84. [CrossRef]
- 33. Parmoon, G.; Moosavi, S.A.; Akbari, H.; Ebadi, A. Quantifying Cardinal Temperatures and Thermal Time Required for Germination of *Silybum marianum* Seed. *Crop J.* **2015**, *3*, 145–151. [CrossRef]
- 34. Elahifard, E.; Derakhshan, A.; Pakdaman Sardrood, B. Does Seed Heteromorphism Affect the Critical Temperature Thresholds for Wild Mustard (*Sinapis arvensis*) Germination? A Modeling Approach. *Botany* **2021**, *99*, 507–514. [CrossRef]
- 35. Zand, E.; Baghestani, M.A.; Soufizadeh, S.; Eskandari, A.; PourAzar, R.; Veysi, M.; Mousavi, K.; Barjasteh, A. Evaluation of Some Newly Registered Herbicides for Weed Control in Wheat (*Triticum aestivum* L.) in Iran. *Crop Prot.* **2007**, *26*, 1349–1358. [CrossRef]
- 36. Pala, F. The Effect of Post Emergence Herbicides and their Mixtures on Grass and Broadleaf Weed Control in Barley (*Hordeum vulgare* L.). *Fresenius Environ. Bull.* **2020**, *29*, 1206–1213.
- 37. García, A.L.; Royo-Esnal, A.; Torra, J.; Cantero-Martinez, C.; Recasens, J. Integrated Management of *Bromus diandrus* in Dryland Cereal Fields under No-till. *Weed Res.* **2014**, *54*, 408–417. [CrossRef]
- 38. Kudsk, P. Optimising Herbicide Performance. In *Weed Management Handbook*, 9th ed.; Naylor, R.E.L., Ed.; Blackwell Science: Oxford, UK, 2002; pp. 323–344.
- 39. Pintar, A.; Svečnjak, Z.; Šoštarčić, V.; Lakić, J.; Barić, K.; Brzoja, D.; Šćepanović, M. Growth Stage of *Alopecurus myosuroides* Huds. Determines the Efficacy of Pinoxaden. *Plants* **2021**, *10*, 732. [CrossRef]
- 40. Aslani, F.; Mehrvar, M.R. Responses of Wheat Genotypes as Affected by Different Sowing Dates. Asian J. Agric. Sci. 2012, 4, 72–74.
- 41. Zhou, B.; Sun, X.; Ge, J.; Li, C.; Ding, Z.; Ma, S.; Ma, W.; Zhao, M. Wheat Growth and Grain Yield Responses to Sowing Date-Associated Variations in Weather Conditions. *Agron. J.* **2020**, *112*, 985–997. [CrossRef]