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Critical Period of Weed Control in Maize as Influenced by Soil Tillage Practices and Glyphosate Application

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Abstract: Increasing evidence on environmental and economic benefits has raised farmers' interest in adopting alternative, less intensive soil management practices. To evaluate the influence of weed-competitive ability in response to a different tillage regime, a field study was conducted in maize under humid Central European climatic conditions in Slovenia. This study was established as a split-plot arrangement with three tillage practices (TPs) as the main plot: conventional (CN), conservation (CS), and no tillage (NT); this was combined with glyphosate application and the weed removal timing as the subplot. The weed removal timings were at the V3, V6, V9, V15, and R1 maize stages, with weed-free and weed season-long monitoring. The beginning and the end of the critical period of weed control (CPWC), based on a 5% maize yield loss rate, were determined by fitting the four-parameter log-logistic equations to the relative maize dry grain yield. The weed dry biomass from maize germination until the R1 growth stage in the NT TP was consistently lower than that in the CN and CS TP. Moreover, the NT TP resulted in a shorter CPWC (39 days after emergence (DAE)) compared to the CN (57 DAE) and CS (58 DAE). The results of CTWR (critical timing of weed removal) showed that less intensive tillage operations in the CS resulted in an earlier need for weed control (V2 and 23 DAE) compared to the CN (V3 and 39 DAE) and NT (V3 and 40 DAE). Our study suggests that the intensive tillage operations performed in the CN TP and the pre-sowing use of non-selective burndown herbicide in the NT delay the CTWR by more than 2 weeks, thus reducing the need for early post-emergence herbicide application in maize.

Keywords: weed interference; crop–weed competition; yield loss

1. Introduction

Arable weed control has been a primary management objective in crop production for centuries since, among all pests, arable weeds have the highest potential to cause serious yield losses if left uncontrolled [1]. Presently, weeds represent an even greater threat than high-yielding crop genotypes; thus, effective weed management is recognized as crucial for the ecologically sustainable intensification of agriculture [2,3].

Environmental and human health concerns related to herbicide use [4] have accelerated efforts to develop and evaluate integrated weed management systems (IWM) and tools less reliant on herbicides [5,6]. Their implementation, however, remains limited, as farmers are not willing to give up the convenience of using herbicide and adopting more complex and riskier non-chemical alternatives [7]. Certain IWM strategies do not exclude the use of herbicides but rather address the need to optimize their use and minimize their adverse environmental effects [8,9]. An advanced IWM program should, therefore, encompass chemical, cultural, and genetic tools [10] but in a more holistic approach to support the transition to long-term sustainable management of weed communities [6].

The cultivation of maize (*Zea mays* L.) plays an important role in global food production, serving as a staple crop for both human consumption and livestock feed [11]. Weeds also pose significant challenges to maize productivity, competing for vital resources, such

as nutrients, water, and sunlight, and ultimately causing substantial yield losses if not effectively managed [12]. Thus, maize has been one of the most studied crops for determining the CPWC in various environmental conditions [13–15], for different crop types [16], as well as determining the influence of individual weed species [17] and various crop and weed management practices affecting maize–weed competitive interactions [18–20].

Tillage has been employed as a key component of conventional agricultural systems and has led to the widespread degradation of soil resources, particularly soil erosion [21,22]. Thus, the adoption of alternative less intensive soil management practices is increasing due to numerous benefits for soil quality improvement, water conservation, yield stability, reductions in labour costs, and increasing biodiversity [23,24]. Soil management practices with different intensities of soil disturbance alter the vertical weed seed distribution in the soil, while seedling recruitment is influenced by environmental factors, such as soil temperature, soil water potential and exposure to light [25,26]. In addition, various tillage practices have been observed to induce notable changes in soil structure, nutrient availability, water infiltration and seedbed conditions that affect the intensity of crop–weed competition and, thus, impact the critical period of weed control [20,27].

Several studies have indicated that the adoption of conservation tillage systems results in a considerable shift in weed species composition and an increase in weed abundance, particularly perennial weed species [28,29]. However, limited attention has been given to investigating the temporal dynamics in which these changes occur, and inconsistent reports can be found. Notably, Derrouch et al. [30] reported that the prevalence of summer-germinating species exhibited a significant increase only after a minimum of 10 years of consistently implementing conservation agriculture practices. Conversely, a multi-environment study showed that non-inversion tillage led to increased weed coverage and greater weed biomass compared to inversion tillage, even in the early stages of the transitioning period [31].

The often-overlooked concept of the critical period for weed control (CPWC) could also serve as an important component of IWM programs and addresses crop–weed competition, assessment of weed infestation levels and establishment of weed control thresholds [32–35]. The CPWC represents a time interval in the crop growth cycle, during which weeds must be controlled to prevent unacceptable yield losses [10,36].

The beginning of the CPWC is indicated by the critical time for weed removal (CTWR) and represents the maximum length of time that crops can tolerate early season interference by emerging weeds without causing significant yield losses. The CPWC ends when the separately measured crop–weed competition component, the critical weed-free period (CWFP) begins, which describes the minimum weed-free period needed from the time of crop emergence until the yield is no longer affected by late-emerging weeds [33,37].

To our knowledge, there is a lack of studies that have investigated CPWC in maize, specifically focusing on the initial stage of implementing changes in weed competitive ability in the early phase of transition to less intensive tillage practices, specifically conservation and no-tillage systems. By increasing knowledge of interactions between tillage practices, weed emergence patterns and crop–weed competition dynamics, the optimal timing of weed control operations can be determined, which can contribute to the development of more sustainable IWM strategies that also consider the CPWC.

2. Materials and Methods

2.1. Study Site and Environmental Conditions

The study was conducted in 2021 in the experimental field of the Agricultural Institute of Slovenia (Infrastructure Center Jablje pri Mengšu, 46°08′33.9″ N, 14°34′21.5″ E, 309 m a. s. l.). The field is dominated by shallow to medium deep eutric brown soil on calcareous pebbles and sand. The soil is well drained with a silty-loam soil texture, comprising 25% clay, 37% silt and 38% sand. Due to their high pebble content, these soils have limited water storage capacity and are, therefore, prone to summer droughts. For several years prior to this study, the fields for the experiment were under various crop rotations with the

main crops being crimson clover, soybean and winter wheat. Soil samples were taken from depths ranging from 0 to 25 cm prior to maize sowing. Soil analysis was performed at the Agricultural Institute of Slovenia in Ljubljana for the following parameters: pH (in KCl) and available phosphorus and potassium (P_2O_5 and K_2O, respectively), employing the calcium-acetate-lactate method from Santner et al. [38]. The results showed neutral pH (7.6) and a moderate supply of phosphorus (30 mg kg^{-1} P_2O_5) and potassium (35 mg kg^{-1} K_2O). The soil organic matter content was high (4.3%). Precipitation and temperatures were measured at a weather station near the experimental field (Adcon, A753GSM, Adcon Telemetry Gmbh, Klosterneuburg, Austria), and the data were compared to long-term averages for the period 1990–2020 (Figure 1).
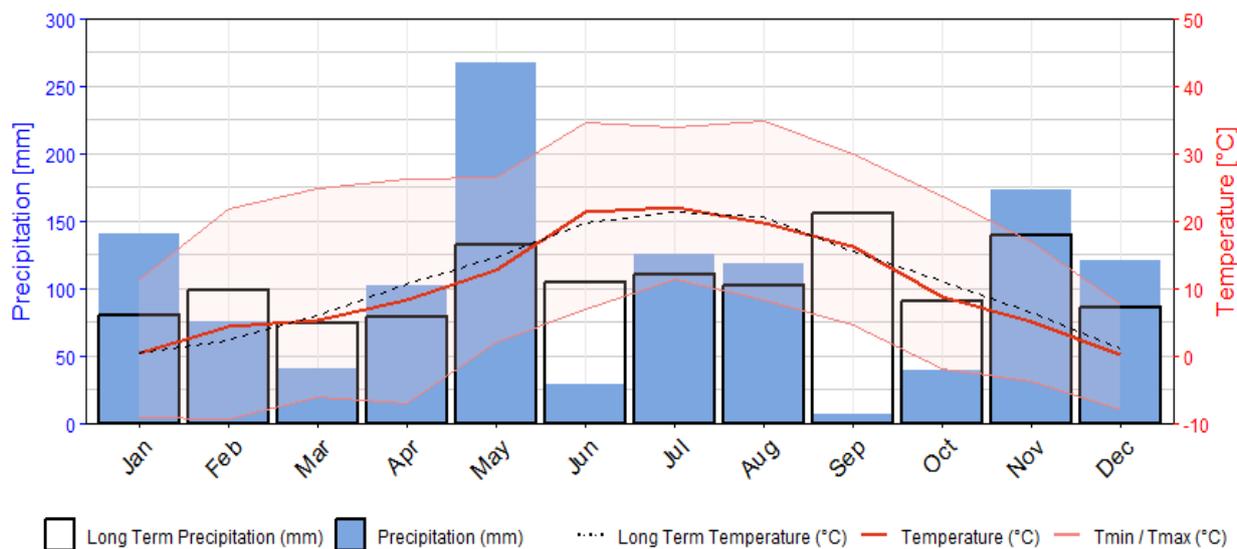

Figure 1. Mean annual temperatures and precipitation at the experimental Jablje (Slovenia) in 2021 compared to the long-term average temperatures and precipitation in the period 1990–2020.

The study site in central region of Slovenia is characterized by temperate continental climate. The long-term (1990–2020) annual mean temperature is 10.8 °C, while annual precipitation averages 1250 mm. During the study period (April to October 2021), mean temperatures and precipitation were 15.9 °C and 930 mm, respectively, while the long-term averages were 16.2 °C and 770 mm, respectively.

2.2. Experimental Design and Management Practices

The field experiment was arranged in a randomized split-plot block design, with five replications. The experimental field was in the early tillage conversion stage, with the tillage methods established in the autumn of 2018. Three different tillage practices (TPs) were used as a main plot, representing various levels of intensity of soil management: conventional (CN), conservation (CS) and no-tillage (NT). The main plots were 100 × 24 m in size and were divided into 8 m long and 3 m wide subplots, consisting of four maize rows.

Primary tillage operations were performed in March, while the seed bed was prepared immediately before maize sowing. In the CN TPs, primary tillage was performed with a plough to a 22 cm depth, and the furrows were closed with a levelling bar, while in the CS TP, the soil was prepared with a disc harrow (8 cm depth). A fine tine harrow was used to prepare the seed bed in the CN and CS TP. In the NT TP, glyphosate (1800 g a. i. ha^{-1}, Boom efekt®, Albaugh TKI, Rače, Slovenia) was used two weeks before maize sowing to control winter annual and perennial weeds. The soil in this treatment was left undisturbed, and the maize was sown directly into winter wheat stubble. Maize intermediate-maturing variety (FAO 350) DKC 4569 (Dekalb®, Bayer Crop science, AG, Monheim, Germany) was sown with a 70 cm row width and 16.5 cm in-row plant spacing, a seed rate of 86,600 seeds ha^{-1} and a depth of 4–5 cm. Prior to sowing, a basic fertilization with 250 kg of NPK 6:18:34

was performed. Maize sowing was carried out on 21 April 2021, under optimal conditions, using a direct-sowing machine. To obtain uniform germination, the entire experimental field was compacted with a Cambridge roller after sowing. Nitrogen fertilization was adjusted to crop needs, where three separate fertilizations with lower nitrogen rates were performed in a period of V3-V15. In total, 160 kg ha^{-1} of nitrogen was applied using ammonium (10.5% N) and amid (23%) mineral fertilizer.

After maize germination, 14 treatments were established, representing increasing durations of weed interference and the length of the weed-free period. The duration of weed interference period included following development stages of maize: two leaves (V2), three leaves (V3), six leaves (V6), nine leaves (V9), fifteen leaves (V15) and silk emergence (R1). The maize yields obtained at the specified corn growth stages within weed-free plots and within treatments where weeds were allowed to compete with the crop (the weedy-until stage) were compared with season-long weed-free and season-long weedy controls [10]. The determination of the maize development stage included evaluating the number of fully developed leaves per plant during vegetative growth or assessing various flowering stages during reproductive growth. This analysis was conducted by observing 10 randomly selected plants within each main plot [39].

Weeds were removed at each time point by hand hoeing and hand pulling in the rows between maize plants. After the peak emergence, from the end of June onwards, plots with the designated removal times were maintained for the remaining season through the application of Tembotrione (44 g a. i. ha^{-1}, Laudis®, Bayer AG, Monheim, Germany) and manual correction by hoeing.

2.3. Crop and Weed Measurements

The average maize growth stage was determined by examining 10 randomly selected plants within each TP. Maize development was based on the number of fully developed leaves per plant or in different generative (flowering) stages. Plant sampling and measurements (i.e., height) were carried out on 24 September 2021, when maize plants reached the full ripening stage with hard and shiny kernels and approximately 65% dry matter (BBCH 89) [40]. Ten maize plants were randomly selected within each TP and cut at ground level for the evaluation of aboveground dry matter (DM) production and height measurements. The aboveground plant parts without cobs were then dried at 60 °C for 3 days, and the DM was weighed. The experiment was conducted in conditions of natural arable weed infestation. Weed composition was regularly determined from a permanently designated area of 4.2 m^2 (3 m by 1.4 m) placed in the middle of season-long weedy plots. Weed biomass was harvested from a selected quadrat of 0.25 m^2 (0.5 m by 0.5 m) within the sampling area. Weed biomass samples were dried at 60 °C for 3 days, and the DM was weighed.

Maize harvest was performed on 1 October and 4 October 2021, using a compact plot harvester (Quantum Wintersteiger AG, Ried, Austria). The middle two maize rows were harvested within all treatments included in the experiment, and seed moisture was adjusted to 14%. Determination of CPWC was based on an acceptable yield loss (AYL) level of 5% as this limit is tolerated by producers. Maize growing degree days (GDDs) were used as the descriptive variable and calculated using the equation by Gilmore and Rogers [41]:

$$\text{GDD} = \sum \left[((\text{Tmax} + \text{Tmin})/2) - \text{Tbase} \right] \tag{1}$$

where Tmax and Tmin are the daily maximum and minimum air temperatures (°C), respectively, with a lower limit of 10 °C, and Tbase is the base temperature (10 °C). During the maize growth, (Tmax + Tmin)/2 was never lower than 10 °C; thus, no corrections were needed.

Yield data were analysed with a nonlinear regression to determine CPWC [21]; however, Cousens [42] suggested that regression analysis is more appropriate and reliable in calculating the critical period. A threshold of 5–10% AYL was used when weed control was necessary [10]. In this case, the CPWC was determined for yield losses of 2.5%, 5% and

10%. A four-parameter log-logistic model was used to analyse the relative yield data and to describe the effect of increasing duration of weed interference on maize yield:

$$Y = C + (D - C)/\{1 + \exp[B(\log X - \log E)]\} \tag{2}$$

where Y is the relative yield (percentage of season-long weed-free yield), C is the lower limit, D is the upper limit (fixed at 100), X is the GDD calculated after maize emergence, E is the GDD given a 50% response between the upper and lower limits (also known as the 'inflection point', I50) and B is the slope of the line at the inflection point.

2.4. Statistical Analysis

All data were examined for homogeneity of variance using Levene's tests prior to the statistical analysis. Due to repeated measures of plant density, plant height and crop biomass, data were then subjected to linear mixed model. Treatments represented fixed factor in the linear mixed model, while blocks were considered as random effects. The means obtained by ANOVA were compared using post hoc Tukey's HSD tests with a $p \leq 0.05$ level of significance. Statistical analysis of maize yield data was performed using the R program [43] (version 4.3.2), while nonlinear regression model was calculated utilizing the "drc" statistical add-on package [44].

3. Results and Discussion

3.1. Weed Density and Biomass Accumulation

Weed species composition and density were assessed in the season-long weedy plots in all three TPs. Broadleaf weeds were present in all TPs. A total of ten species of varied perennation were found in the experimental site, including one cryptogam (*Equisetum arvense* L.) and nine dicot species. No grass species were present. During the early spring, the weed community was dominated by winter-annual weed species. From the middle of May onwards, increasing air temperatures facilitated the germination of summer-annual weed species. Annual broadleaved weeds *Chenopodium polyspermum* L., *Lamium purpureum* L., *Veronica persica* Poir., *Amaranthus retroflexus* L. and *Galinsoga parviflora* Cav. were the dominant species throughout the duration of the experiment. Moreover, these weed species produced the majority of the weed biomass. The greatest total weed density across the season was observed in the CS TP (570 weeds m^{-2}), and the lowest was observed in the NT TP (323 weeds m^{-2}). The lowest weed infestation in NT TP is the result of the use of glyphosate. Bilalis et al. [45] also confirmed that the highest weed density in their study was in the CN and CS TP, while the lowest was in the NT TP. *L. purpureum* and *C. polyspermum* were the dominant species in the CN and CS TP. When comparing the weed density across the whole maize growing season, the peak values within all TPs were observed in the second and third assessment periods. By the end of the maize growing season, the development of emerged weeds decreased, and, furthermore, weed density decreased in September. The reduction in weed density was due to interference between the individuals [46]. Although weeds established after the CPWC did not have a considerable effect on maize yield, it should be noted that they may produce seeds that emerge in the following cropping season [47].

Dry weed biomass (DM) increased with increasing weed interference (Figure 2). Weed density and biomass were significantly higher in weedy plots. This agrees with Ahmadvand et al. [48], who stated that dry weed biomass and the number of weeds are the highest in the non-weeded control plots. The greatest weed DM was measured in CS TP in the weedy-until-R1 growth stage (405.2 g m^{-2}). In the NT TP in the weedy-until-V2 growth stage, no weeds were present due to prior glyphosate application to control perennial and winter annual weeds. Moreover, in all treatments in NT TP, the lowest DM was observed compared to CN and CS TP, which may be the result of prior glyphosate application. According to Amuri et al. [49], this can also be the result of the high residue level of previous crops, which suppresses weeds in terms of crop management.
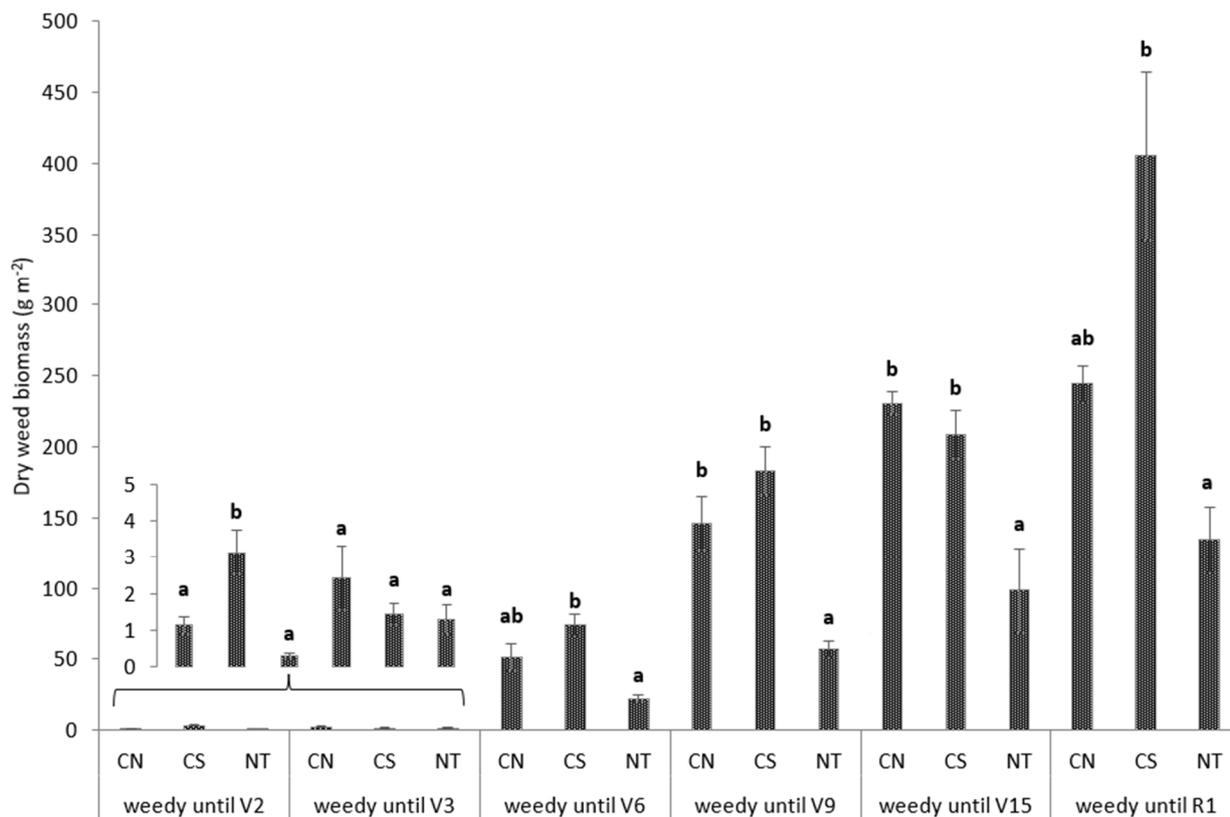
Figure 2. Weed DM (g m^{-2}) in three tillage practices (TPs) (conventional (CN), conservation (CS) and no-tillage (NT)) and treatments (maize growth stages). Data presented are means ± standard errors. Different letters (a,b) indicate significant differences ($p \leq 0.05$, Tukey's tests) between TPs in each treatment.

3.2. Maize Growth Parameters

The results of the linear mixed model showed a statistically significant effect of TP on plant density ($p \leq 0.001$), while no effect was observed for plant height ($p = 0.401$) and dry biomass ($p = 0.141$).

The greatest plant density was observed in the NT TP (9 plants m^{-2}), and the lowest was observed in the CS TP (7 plants m^{-2}), with the CN TP at an intermediate level of 8 plants m^{-2}. At the beginning of the growing season, maize plants were exposed to water stress, which led to fewer crops being established in the CN and CS TP plots with lower soil moisture. According to Djaman et al. [50], the optimum plant density in their study was approximately eight or nine plants m^{-2} for maximum maize grain yield and water use efficiency.

Although not statistically significant, maize plants in CN TP tended to be taller and more productive compared to the CS and NT plot. The plant heights observed were 289 cm, 287 cm and 281 cm for the CN, CS and NT TP, respectively. A similar trend was seen in the study of Lasisi et al. [51], where an improved water-air regime and greater mobility of minerals in deeper soil layers increased plant height in CN. In contrast, the absence of tillage in the NT increases soil compaction and reduces root density and absorption in deeper soil layers [52]. The DM of maize plants ranged from 103.5 g plant^{-1} in the CN up to 118.5 g and 154.9 g plant^{-1}, observed in the CS and NT TP, respectively. NT TP was frequently found to facilitate crop production in drier soil conditions [53], due to higher soil moisture, lower soil temperature, higher soil carbon content and soil aggregate stability [54].

3.3. Critical Timing for Weed Removal and the Critical Weed-Free Period

Maize dry grain yields were somewhat high and varied among TPs and treatments (maize growth stages) and with increasing duration of weed interference. In season-long weed-free plots, maize yields were similar and varied from 17.95 t ha^{-1} in the CN TP to 17.68 t ha^{-1} in the CS and 17.06 t ha^{-1} in the NT TP. The maize grain yield in the season-long weedy plots was significantly lower and ranged from 12.16 t ha^{-1} and 11.63 t ha^{-1} to 10.81 t ha^{-1}, as observed in the NT, CN and CS TP, respectively. Our results are similar to those of Dogan et al. [55], who reported lower maize yields with increasing duration of weed interference.

The start of the CPWC (CTWR) and ending of the CPWC (CWFP) varied among the TPs (Figure 3). The longest CPWC, based on a 5% acceptable yield loss (AYL), was determined in CN TP and corresponded to 39 to 96 DAE or the V2 to R1 leaf stages of maize. The CPWC in NT TP was 17 and 18 days shorter compared to the CN and CS plot, respectively. The suggested weed removal timing in CS TP was 17–18 days earlier compared to the CN and NT plot (Table 1). However, when AYL increased from 2.5% to 10%, the beginning of CTWR was almost the same with CN TP (+3 days), while it was significantly delayed with CS TP (+21 days) and NT TP (+11 days) (Table 1).

Table 1. Estimates of critical timing for weed removal (CTWR) and critical weed-free period (CWFP) based on growing degree days (GDD), crop growth stage (CGS) and days after emergence (DAE) influenced by the CN, CS and NT TP and treatment calculated by using a four-parameter logistic equation.

Tillage Practice	AYL (%)	GDD ($\pm$SE)		CGS		DAE	
		CTWR [a]	CWFP [a]	CTWR [a]	CWFP [a]	CTWR [a]	CWFP [a]
CN	2.5	124 (34)	928 (115)	V3	R1	39	104
	5	125 (12)	823 (83)	V2	R1	39	96
	10	148 (37)	804 (45)	V3	R1	42	95
CS	2.5	35 (40)	731 (23)	VE	V15	17	89
	5	66 (6)	633 (102)	V2	V12	23	81
	10	118 (33)	568 (48)	V3	V12	38	76
NT	2.5	95 (14)	745 (21)	V2	R1	33	90
	5	133 (10)	615 (31)	V3	V12	40	79
	10	171 (11)	489 (46)	V3	V9	44	70

[a] The CTWR and CWFP were estimated with a four-parameter log-logistic equation based on GDD at 2.5%, 5% and 10% yield loss. DAE—days after emergence, and CGS—maize growth stage.

Teasdale [56] reported that the CPWC in maize ends with canopy closure, which occurs in the V11 maize growth stage, and these findings were further confirmed by Evans et al. [18]. In our study, CWFP in all TPs ended much later, with the V12 growth stage in CS and NT TP and up to the R1 stage in the CN TP.

The variability in the studies determining the beginning and end of CPWC is influenced by the density, competitiveness and development period of the existing weed population [18,19,57]. Furthermore, the early emergence of weeds can be suppressed by the use of pre-emergence herbicides and, thus, delay the beginning of CPWC in maize [10].

In our study, increasing the period of weed interference significantly reduced yields in all TPs. Maize dry grain yield losses from weed competition were not different in various TPs and ranged from 22 to 41% less maize dry grain yield compared to the weedy-free plots. The CPWC observed for 2.5% and 5% AYL was longer compared to the 10% AYL. These results were also affirmed by Uremis et al. [58], who reported that the CPWC for 10% maize yield loss was shorter compared to the 2.5% and 5% maize yield loss.
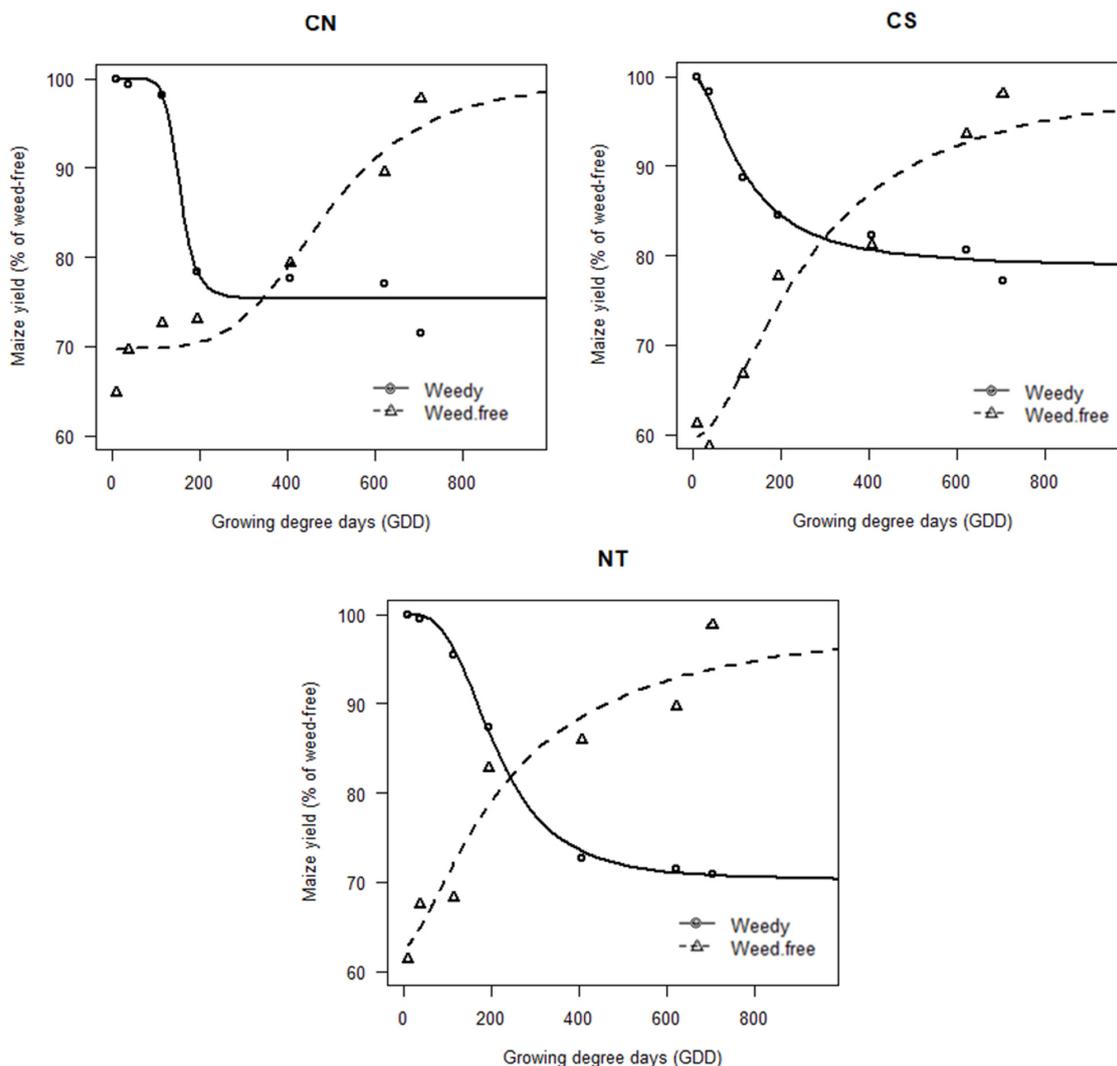
Figure 3. Maize yield (% of weed-free) response to increasing duration of weed interference as represented by growing degree days (GDD) grown in season-long weedy and season-long weed-free plots in Jablje, Slovenia. The regression lines are plotted using Equation (2), and the parameter values are presented in Table 2.

Table 2. Regression parameter estimates (±SE) for the three tillage practices characterizing the effect of duration of weed interference on the relative maize grain yield. Regression parameters presented are showing the slope (B), lower limit (C) and GDD at 50% yield reduction (I50) using the four-parameter log-logistic model (Equation (2)).

		Regression Parameters (±SE)		
Tillage Practice	**Treatment**	**B**	**C**	**I$_{50}$**
Conventional	Weedy	8.5 (3.3)	75.4 (1.4)	153 (16)
	Weed-free	−10.1 (1.1)	78.9 (5.5)	830 (17)
Conservation	Weedy	1.8 (0.6)	78.5 (1.9)	116 (21)
	Weed-free	−6.7 (14.7)	78.3 (7.4)	541 (25)
No-tillage	Weedy	3.1 (0.3)	70.2 (0.6)	209 (7)
	Weed-free	−2.8 (4.7)	79.5 (7.1)	598 (40)

4. Conclusions and Management Implications

In a time of decreasing herbicide availability, lower herbicide uses and the absence of ready-to-use non-chemical weed control measures, tillage remains one of the most important weed management strategies. To our knowledge, our study is the first to investigate the maize–weed competitive interaction, focusing on the changes in weed-competitive ability during the early phase of transition to less intensive tillage, conservation tillage and no-tillage practices.

Our study clearly showed that weed density and weed DM production in the NT plot were consistently lower than those in tilled, CN and CS plots. Moreover, using the NT TP and pre-sowing non-selective burndown, herbicide resulted in lower levels of weed competition and, thus, 17–18-day shorter CPWC compared to the CN and CS TP. Limiting the time window during which weed competition leads to substantial yield loss can potentially provide an opportunity to utilize lower herbicide doses or, where applicable, less reliant mechanical weed control methods. In contrast, the adoption of non-inversion tillage in the CS TP intensified early season weed competition and necessitated weed control almost immediately after sowing, which is more than two weeks earlier compared to the CN and NT TP. If this CTWR shift in the CS TP is not accompanied by weed control action, up to 2 t ha^{-1}, greater dry maize grain yield loss can be expected in comparison to the CN and NT TP.

It can be concluded that when reducing the tillage intensity without supplementing weed management, this will most likely result in a significant yield loss, as seen in the CS TP. Thus, when adopting alternative less-intensive tillage systems, weed management in maize should be adapted to site-specific soil conditions, crop requirements, and weed pressure. Our study also demonstrated the need for further investigation of maize–weed competition outcomes influenced by lower herbicide inputs, cultural and mechanical weed control strategies tailored to the specific TP. This can ultimately contribute to the development of an IWM system for maize less dependent on synthetic herbicides and contribute to overall resilience and sustainability of the arable cropping systems.

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