



Article Optimizing Weed Management for the New Super-Forage Moringa oleifera

Itai Shulner ^{1,2}, Evyatar Asaf ^{1,2}, Zohar Ben-Simhon ³, Miri Cohen-Zinder ⁴, Ariel Shabtay ⁴, Zvi Peleg ², and Ran Nisim Lati ^{1,*}

- ¹ Department of Phytopathology and Weed Research, Newe Ya'ar Research Center, Agricultural Research Organization, Ramat Yishay 3009503, Israel; itayshulner@gmail.com (I.S.); evya.jd@gmail.com (E.A.)
- ² The Robert H. Smith Faculty of Agriculture, Food and Environment, The Hebrew University of Jerusalem, Rehovot 7610001, Israel; zvi.peleg@mail.huji.ac.il
- ³ Newe Ya'ar Agricultural Unit Manager, Newe Ya'ar Research Center, Agricultural Research Organization, Ramat Yishay 3009503, Israel; zohar@volcani.agri.gov.il
- ⁴ Department of Ruminant Science, Beef Cattle Section, Newe Ya'ar Research Center, Agricultural Research Organization, Ramat Yishay 3009503, Israel; mirico@volcani.agri.gov.il (M.C.-Z.); shabtay@volcani.agri.gov.il (A.S.)
- * Correspondence: ranl@volcani.agri.gov.il; Tel.: +972-495-39529

Abstract: *Moringa oleifera* Lam. (moringa hereafter) is cultivated as a new summer super-forage field crop in Israel, yet no weed control protocol has been developed for it. The objective of the study was to develop an integrated weed management (IWM) practice for the moringa agro-system in arid and semi-arid regions like the Mediterranean basin. We tested various herbicides applied pre (PRE) and post (POST) crop emergence and cultivation methods for weed control, with an emphasis on crop safety. The PRE herbicides were the most effective and safe control mean. Their application resulted in minor (<5%) crop fresh weight reductions and weed cover area, compared with the control. The POST herbicides were also effective, yet their crop safety level was lower and non-consistent in some treatments. Generally, the finger weeder was less effective than the herbicide treatments and caused higher fresh weight reduction. However, this means was more effective when applied at earlier stages. Management and environmental conditions had a high impact on the moringa growth; hence, these aspects should be considered. Our results show the potential use of different herbicides and non-chemical tools and set the basis for a future IWM protocol for moringa. The wide range of options offered here can ensure economic and environmentally viable solutions for this new crop.

Keywords: finger weeder cultivation; integrated weed management; moringa; post-emergence herbicides; pre-emergence herbicides

1. Introduction

Moringa oleifera Lam. (moringa hereafter) is a tropical tree of the Moringaceae family, which consists of 13 species. It is native in North India and currently distributed along the dry tropic and subtropic areas across Southwest Asia, Northwest Africa, and Central-South America [1,2]. Moringa is known as the "Miracle Tree" as it is rich in vitamins, proteins, minerals and phytochemicals, and pharmacological compounds. The tree is used for an array of purposes including human feed, medicine, biodiesel, and water purification [3–5]. Moringa trees were introduced to the Mediterranean basin in the early 1950s as perennial trees in small-scale orchards for various medicinal purposes, utilizing the crop drought tolerance and adaption to the Mediterranean climate [6–8].

In recent years, there has been an attempt to develop moringa as a new super-forage crop for livestock feed. This trend was motivated by several studies that tested and characterized the effect of dietary supplementation of moringa-based silage on the productivity of lactating cows and lambs [9–11]. However, such a transition from a perennial crop



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Copyright: © 2021 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (https:// creativecommons.org/licenses/by/ 4.0/). grown in orchards to an intensive agro-system field crop requires the development of effective weed management protocols to ensure high and sustainable yields and profitability. Weed infestation is the main biotic factor affecting crop plants' agro-systems and can result in significant yield quantity and quality losses [12]. The weed problem is even more pronounced in forage crops, as infestation by toxic weed species [e.g., *Setaria viridis* (L.) Beauv. and *Helminthotheca echioides* (L.) Holub] can be hazardous and lead to complete yield loss [13]. While chemicals (herbicides) are the most cost-effective and efficient practices for weed control [14], to date, there is no available information on the effect of the different herbicide mode-of-action (MOA) on the moringa's development as a forage crop. Thus, it is necessary to bridge the knowledge gap and uncover the physiological and agronomical aspects involved in herbicide application to promote the transition of moringa into intensive cropping practices [15].

Despite their effectiveness, excessive use of herbicides is associated with environmental aspects and increased weed resistance [16]. Thus, to promote and ensure sustainable food production much pressure has been put on farmers to reduce the number of applied herbicides and to integrate alternative weed control methods. Furthermore, as moringa is used mainly as a food supplement, there is a market demand to reduce or eliminate the use of chemicals. Mechanical or physical methods such as cultivation, hoeing, and flaming offer a promising non-chemical alternative for weed control [17]. Being the most common practice, we selected cultivation in the current study as the non-chemical control tool. Cultivation damages the weed physically, by uprooting, covering, or cutting the plant, which finally leads to death by desiccation [17,18]. Unlike herbicides that are applied homogeneously on the entire field area, cultivation machinery operates on a limited area, mainly between the crop rows (i.e., inter-row). Thus, most of the cultivation tools do not handle weeds within the crop rows (i.e., intra-row), which can cause significant reductions in crop yield [19]. The finger weeder cultivator is an efficient tool that handles the intra-row weeds [20]. This tool has rubber fingers with different sizes and stiffness, allowing for adjustments for varying crops and soils. The finger weeder was tested in various crops and found to reduce intra-row weed densities by up to 93% [21]. However, it can be generally stated that mechanical methods are often less effective compared to herbicides; thus, they should be combined with other methods to achieve commercially acceptable control results [22]. Moreover, their efficacy is highly affected by field conditions and has a limited time window for effective treatment [23,24]. These limitations suggest that other control tools need to be combined to ensure weed control and crop yield quality. In this regard, integrated weed management (IWM) was suggested as an effective strategy to control weeds. IWM can include pre-emergence (PRE) and post-emergence (POST) herbicides with different non-chemical methods, and increase weed control to over 90% in some crops [25,26]. The main concept of IWM is to reduce the reliance on herbicides, increasing weed control efficacy and crop safety and profitability, while maintaining a more environmentally friendly method [27,28].

The development and integration of IWM required vast knowledge about the efficacy and crop safety, in terms of crop stand and yield, of each control method alone before the integration is possible. Thus, the objective of this study was to evaluate the crop-safety and efficacy of different MOA herbicides at PRE and POST application and finger weeder cultivator. This objective will set the basis for the development of IWM for moringa and sustainable production of this crop under the Mediterranean arid and semi-arid climate conditions. Altogether, our results addressed, for the first time, weed control in the superforage moringa and identified chemical and non-chemical methods that can be safely used to optimize moringa production.

2. Materials and Methods

2.1. Plant Material and Experimental Site

Moringa oleifera seeds were obtained from commercial orchards at Ein Hashofet (lat. 32°35′, long. 35°5.9′) in 2018 and from Ganei-Yohanan (lat. 35°51′, long 34°50′) in the 2019–

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2020 seasons. Experiments were conducted at the Newe Ya'ar Research Center (lat. $32^{\circ}42'$, long $35^{\circ}10'$) during the springs and summers between 2018 to 2020. The soil type at the experimental site was low-alkaline clay, with 57% clay, 23% silt, and 20% sand, pH = 7.5.

2.2. Experimental Design

Experiments were performed using a complete randomized block design with four to six replicates for the herbicide and the cultivation (finger weeder) experiments. For the herbicide experiments, all plantings were grown on 1.93 m by 5 or 7 m length raised beds during the 2018–2019 and the 2020 experiments, respectively (Figure S1 Supplemental Data). For the cultivation experiments, the plot size was 7 or 10 meters in length for the 2019 and the 2020 experiments, respectively. In all experiments, two seed lines per bed were used, 80 cm apart. A tractor-mounted NG plus 2 planter (Monosem Inc., Edwardsville, KS, USA) was used for sowing, and drip irrigation and other common forage cultural practices were used [11]. The distance between seeds on the sowing line was 10 cm with a total number of 200,000 seeds per hectare (20 seed per m^2). The critical dates of the main operations in the field experiments, including sowing, herbicide/cultivation application, crop injury evaluation, weed density evaluation, and harvesting dates are listed in Table S1 (Supplemental Data). Hand weeded checks were maintained to estimate the level of yield loss due to weed infestation and herbicide treatments. Additionally, untreated control checks were maintained to estimate weed control efficacy. High and low temperatures were recorded from a weather station located in the research center to detect any interactions between environmental conditions and crop injury (Figure S2 Supplemental Data).

2.3. Chemical Weed Control Treatments

The crop safety for moringa and the weed control efficacy included seven herbicides for PRE assay in the 2018 season (n = 4) as a preliminary experiment (Table S2, Supplemental Data). Four of the most suitable ones were validated during the 2019 season (n = 6). Treatments were applied one day after sowing and were activated with irrigation (300 m³ ha⁻¹) by a sprinkler system. No precipitation was documented throughout all the field experiments. Four herbicides were selected for the POST assay. All PRE and POST herbicides were selected for their wide usage and high weed control efficacy in summer field crops in Israel. Herbicides were sprayed at the 4–6 leaves stage in both 2019 and 2020 experiments (n = 6). In all experiments, herbicides from various MOA were applied once (Table S2, Supplemental Data). Herbicides were sprayed using a motorized back sprayer equipped with a 2 m-wide ground spray rod and a Teejet-110015 nozzle (Spraying Systems Co., Wheaton, IL, USA) with a spray volume of 200 L ha⁻¹ and maintained at a spray pressure of 310 kPa.

2.4. Non-Chemical Weed Control

Cultivation treatments were tested in 2019 and 2020, both for the intra-row and the inter-row areas. In 2019, a preliminary experiment was performed for assessing moringa safety and weed control efficacy of sequential cultivation treatments. Three treatments were applied, single (once), double (twice), and triple (three times), at 14, 25, and 45 days after emergence (n = 5). In 2020, four cultivation treatments were applied: early single, late single, early double, and late double (n = 6). The early and the late first cultivations were performed 6 and 13 days after crop emergence, at the 2nd and 4th leaf stage, respectively. The second treatment was performed 13 days after the first one. Table S3 (Supplemental Data) details the treatment's timing and number of applications. The intra-row cultivation treatments were performed using a 370 and 310 mm stiff (93 Shore A) finger weeder (K.U.L.T., Vaihingen, Germany), mounted at a distance of 35 and 31 cm between the fingers, respectively (Figure S1 Supplemental Data). To provide weed control over the entire bed width, the inter-row weeds were treated by side tillers (20 cm wide), mounted at a distance of 10 cm from the crop row sides, and goosefoot tillers (25 cm wide) mounted between the rows and on the bed edges. Driving speed during treatments was 2–3 km h⁻¹.

2.5. Plants Evaluation

Moringa yield (leaves and stem fresh weight) was evaluated 54–80 days after sowing (DAS) (Table S1, Figure S1 Supplemental Data). Samples were taken from one m² area in the middle of the bed, by cutting the plants 20 cm above the soil surface, and defoliating the leaves from the woody parts (stem and petioles). Moringa's initial crop stand was evaluated 20 days after emergence or before the first POST (herbicide or cultivation) treatment, respectively. Weed cover and biomass were determined 25 and 45 DAS, respectively. Weed cover was determined by visual estimation and biomass was determined by harvesting the above-ground parts of the plants. Weed biomass was recorded after 72 h of drying at 65 °C. Additionally, weed density was determined by counting the number of weeds before and after treatments over the entire bed area. Each weed species was counted separately. Weed biomass (B) and cover (C) reductions were transformed to the percent of control using the following equation:

Weed
$$B/C = Treat/Control*100$$

where Treat is the weed biomass or cover value following the evaluated treatment, and Control is the evaluated value from the untreated control.

2.6. Statistical Analyses

A statistical analysis was performed using a JMP ver. 15.0 software (SAS Institute Inc., Cary, NC, USA). Each experiment was analyzed separately. Moringa fresh weight and weed biomass were subjected to ANOVA. For moringa crop safety, means were compared to the hand weeded control using the Dunnet test ($\alpha = 0.05$), and for weed control means were separated using the Tukey-HSD test ($\alpha = 0.05$). For the POST experiments the average of each sample was transformed by the arcsin \sqrt{x} transformation (0 < x < 1), and re-transformed for graphic visualization [28]. Box plots described the results dispersion, median, max, and min data points, and Q1 and Q3 percentile. Data visualization was conducted using the ggplot2 R studio 4.0.3 package (Rstudio, PBC, Boston, MA, USA).

3. Results

3.1. Pre-Emergence Herbicides

To test the crop safety of PRE herbicide application, we selected seven herbicides from four MOAs that represent widely used and common herbicides for summer field crops in Israel. In general, four active ingredients (s-metolachlor, diuron, flurochloridone, and saflufencil) were safe for moringa over two constitutive years of experiments as expressed in the non-significant effect on crop fresh weight. Wide screening of herbicides for crop safety during the 2018 season showed no significant differences (p = 0.77 and higher) between the moringa fresh weight of treated and untreated control plants, with values ranging from 82% to 106% (Figure S3A Supplemental Data). Fomesafen and fluometuron were more injurious to the crop, resulting in 26% and 24% fresh weight reduction, respectively, compared to the control (p = 0.38–0.47), and were not tested in the following year.

To validate these results, we conducted a similar experiment during the 2019 season and compared the crop safety of the candidate herbicide to the hand weeded control in addition to the untreated control. No treatment \times year interaction was found for all herbicides used (p = 0.68). The same four herbicides were also safe compared to the hand weeded control (p > 0.80), with moringa fresh weight ranging between 87% to 97% of the control (Figure 1A). In both years, saflufencil and diuron were the safest treatments with 106% and 97% moringa fresh weight compared to the hand weeded control, respectively. Additionally, all herbicide treatments did not result in significant differences in the moringa crop stand compared to the hand weeded control (as also expressed in low CV values), suggesting that all treatments did not affect the initial stand and crop early development (Table 1).



Figure 1. Effect of PRE herbicides application on moringa and weeds during the 2019 season. (**A**) Moringa fresh weight. Data are mean (n = 6). Differences between treatments were analyzed by the Dunnet test in comparison with hand weeded control. (**B**) Weed cover, and (**C**) weed biomass. Different letters indicate a significant difference according to the Tukey-HSD test. Treatments included: hand weeded control (hwc), untreated control (utc), S-metolachlor (met), diuron (diu), flurochloridone (flu), and saflufencil (saf).

Table 1. Pre-emergence herbicide application effect on moringa initial crop stand. Differences between treatments were analyzed by the Dunnet test in comparison with hand weeded control, and the coefficient of variance (CV).

Treatment	Crop Initial Stand (Plants Per Plot)	CV (%)
Hand-weeded	84.5	6.9
Untreated	78.6 ($p = 0.34$)	8.6
S-metolachlor	84.5 (p = 1)	5.0
Diuron	90.0 $(p = 0.39)$	9.0
Flurochloridone	$86.8 \ (p = 0.94)$	3.6
Saflufencil	87.3 ($p = 0.88$)	5.8

The main weed species in the experiments included: *Portulaca oleracea* L., *Amaranthus albus* L. AMAAL, *A. retroflexus exus* L. AMARE, and *A. blitoides* S. Watson, and only in 2019, *Cyperus rotundus* L. # CYPRO. To test the efficacy of the different treatments on weed control we used visual weed cover as the efficacy parameter. Generally, all PRE herbicides

were effective and resulted in a significant weed cover reduction compared to the untreated control (p < 0.001). The most effective treatments were diuron and fomesafen that resulted in ~1.5% weed cover compared to 76% in the untreated control (Figure S3B Supplemental Data). In the second year, we conducted a more *in-depth* characterization of four PRE treatments, using weed density (for each species and total) and weed biomass parameters in addition to weed cover, to evaluate the control efficacy. All four PRE treatments were effective and resulted in significant (p < 0.001) reductions in weed biomass and weed cover compared to the untreated control. Reductions in weed biomass values ranged between 80 and 98% compared to the untreated control, indicating the high efficacy of these treatments (Figure 1B,C). Generally, s-metolachlor was the most effective herbicide, providing a reduction of 98, 97, and 50% of weed cover, biomass, and density as compared to the untreated control, respectively. Weed density values at the first evaluation (30 days after treatment [DAT]) were significantly different between most treatments (expect flurochloridone) and the untreated control. However, at the following evaluation (45 and 60 DAT) there were no significant differences between weed density values of all treatments (Table S4, Supplemental Data). The control level of C. rotundus was partial, which increased the total weed density values (Table S5, Supplemental Data). This species is considered highly noxious, in Israel and many parts of the world, and the herbicides tested in our experiments did not control it effectively. The high weed density in the later evaluations (45 and 60 DAT) were most probably secondary emergence flashes. However, the significantly lower weed biomass values suggest that the development of the escaped weeds was highly inhibited by the treatments.

3.2. Post-Emergence Herbicides

The safety and efficacy of four POST herbicide treatments were evaluated in 2019 and 2020. In the 2019 experiment, no significant differences in moringa fresh weights were observed between all treatments and the hand weeded control. However, bentazon + imazamox and aclonifen were the safest treatments, with moringa fresh weight values of ~90% compared to the hand weeded control (Figure 2). Foramsulfuron + isoxadifen ethyl and rimsulfuron were more injurious to the crop and resulted in moringa fresh weight values of 80% and 73% compared to the hand weeded control, respectively. In the 2020 experiment, bentazon + imazamox, aclonifen and foramsulfuron + isoxadifen ethyl had a more injurious impact on moringa fresh weight. Significant reductions were observed in this experiment, and only 25-36% fresh weight values were recorded compared to the hand weeded control (Figure 2). Rimsulfuron was the safest herbicide and no significant decrease in moringa fresh weight was observed (p = 0.19). This treatment resulted in only 26% reduction compared to the hand weeded control. In both years, all treatments did not result in significant differences in moringa crop stand compared to the hand weeded control, suggesting that the treatments did only affect the crop growth of moringa but did not cause crop death (Table S6, Supplemental Data). In general, the field conditions in the 2020 experiment were less suitable for moringa growth, so there were significant differences between yield results in the two experiments. Except for rimsulfuron (p = 0.46), all treatments showed significant yield reduction in 2020 in comparison to 2019, with $p \le 0.01$ (Figure 2).



Figure 2. Effect of POST herbicides application on moringa fresh weight. Treatments included: untreated control (utc), bentazon + imazamox (ben + ima), aclonifen (acl), rimsulfuron (rim), and foramsulfuron (for). Comparisons between years within treatment were assessed using a *t*-test (n = 6).

The main weed species were cutleaf groundcherry (Physalis angulata), common purslane (P. oleracea), tumble pigweed (A. albus), redroot pigweed (A. retroflexus), and prostrate pigweed (A. blitoides). All treatments significantly reduced weed cover and biomass compared to the untreated control (p < 0.027). Weed cover reductions ranged between 57% and 98% and weed biomass reductions ranged from 63% to 95% compared to the untreated control, indicating the differences between treatments' efficacy (Figure 3A,B). Bentazon + imazamox and rimsulfuron were the most effective treatments, with weed control levels of 98% and 89–95% compared to the untreated control for weed cover and biomass, respectively. As for weed density, significant differences ($p \le 0.001$) between all treatments and the untreated control were observed in both evaluations (14 and 35 DAT) (Table S7, Supplemental Data). Except for *P. oleracea*, there were significant differences (p = 0.001-0.003) in the weed density between most treatments and the untreated control, including the overall number of weeds, in the final evaluation (Table S8, Supplemental Data). In the 2020 experiment, weed cover and biomass were evaluated, but different results were observed compared to 2019. All treatments significantly reduced weed cover and biomass compared to the untreated control ($p \le 0.001$). Weed cover reductions ranged between 56% and 84% and weed biomass reductions ranged between 77% and 95% compared to the untreated control (Figure 3). Foramsulfuron + isoxadifen ethyl provided the best results, with weed cover and biomass reductions of 84% and 95%, respectively, compared to the untreated control. Unlike the crop yield, there were no significant differences between most treatments in terms of weed control (Figure 3). The only significant differences were observed for rimsulfuron weed cover (p = 0.01).



Figure 3. Effect of POST herbicides application on (**A**) weed biomass, and (**B**) weed cover. Treatments included: bentazon + imazamox (ben + ima), aclonifen (acl), rimsulfuron (rim), and foramsulfuron (for). Comparisons between years within a treatment were assessed using a *t*-test (n = 6).

3.3. Cultivation

To test the crop safety and efficacy of the number of cultivation applications, three sequential finger weeder cultivation treatments were held. The moringa fresh weight showed a reduction of 8%, 29%, and 44%, for the single, double, and triple applications of the finger weeder, respectively (p = 0.93, 0.31, 0.08) (Figure S4 Supplemental Data). As for decline in the final crop stand, there was no significant difference between cultivation treatments (p = 0.55-0.99); however, the first treatment was the most important one. This treatment resulted in a 29–33% reduction in crop stand, compared to an additional 8% and 3% reduction for the second and the third applications, respectively (Table S9, Supplemental Data). These results suggest that the timing of the first application should be targeted to a safer time window.

In 2020, single and double treatments were applied and emphasis was given to the timing of the first treatment. All treatments resulted in a significant reduction of the moringa fresh weight ($p \le 0.012$) compared to the hand weeded control. The late and early double applications resulted in reductions of 45% and 53%, respectively. The single

treatments resulted in a higher fresh weight reduction (70%), compared to the hand weeded control (Figure 4A). The single treatment reduced the initial crop stand by ~5%, while the double treatment resulted in a ~16% reduction. The untreated control had little effect on the crop stand, with only a ~5% decrease (Figure 4B). Although the double treatment resulted in higher crop stand reduction compared to the single treatment, it had a lower effect on the final moringa fresh weight, probably due to better weed control.



Figure 4. Effect of finger weeder treatments on the moringa. (**A**) Fresh weight. Differences between treatments were analyzed by the Dunnet test in comparison with the hand weeded control (n = 6). (**B**) Crop stand reduction. Different letters indicate significant differences according to the Tukey-HSD test (n = 6). The treatments included: hand weeded (hwc), untreated control (utc), early single (ea sin), early double (ea dou), late single (la sin), and late double (la dou).

The main weed species in the cultivation plots were *C. rotundus*, *P. oleracea*, *A. albus*, *A. retroflexus*, and *A. blitoides*. In 2019, weed cover, biomass, and density were evaluated; nonetheless, no significant differences were observed between the cultivation treatments and the untreated control. The double treatments were the most effective, resulting in a weed cover and biomass reduction of 35% and 20% compared to the untreated control, respectively. The single and the triple treatments had a similar effect on the weed cover, and both treatments resulted in a non-significant reduction of ~25% compared to the untreated control (Figure S5 Supplemental Data). Similar results were revealed for the density of the weeds, where no significant differences were observed between all cultivation treatments. The double treatment reduced the total weed density by 51%, while the single and triple treatments had only a minor effect on this parameter (Table S10, Supplemental Data). The main weed species had no significant differences in their density between all treatments, including the overall count (Table S11, Supplemental Data).

In 2020, control results were favorable over 2019 and significant reductions in the weed biomass and cover were observed between all treatments and the untreated control. Similar to 2019, the double treatments were the most effective. Weed cover reductions were 80% and 55% for the early and the late applications, and weed biomass reductions were 85% and 75% compared to the untreated control, for the early and the late application, respectively (Figure 5). In comparison, the early and late single treatments resulted in weed cover reductions of 30% and 25%, and weed biomass reductions of 38% and 35%, respectively, compared to the untreated control (Figure 5).



Figure 5. Effect of finger weeder treatments on weed control efficacy. (A) Weed cover and (B) biomass in the 2020 season. The treatments included: untreated control (utc), early single (ea sin), early double (ea dou), late single (la sin), and late double (la dou). Different letters indicate a significant difference between treatments according to the Tukey-HSD test (n = 6).

4. Discussion

Weed control is an essential element for the transition of crop plants from traditional practices to an intensive cropping system. *Moringa oleifera* is a new super-forage crop in the Mediterranean basin, which has been mostly cultivated as a perennial tree in orchards. Not many studies have addressed the weed control issues of this crop. For a new crop, this task is more challenging as the data regarding the safety and efficacy of different treatments are scarce. Here, we evaluated the potential usage of different herbicides, from various MOAs, and cultivation applications for weed control in the moringa forage crop. We also aimed to set the foundations for the development of a sustainable IWM protocol that combines several weed control methods [26]. By doing so, long-term efficacy and environmental sustainability protocols can be maintained, while ensuring the farmer's profitability [29,30].

Several PRE herbicides, that represent the most commonly used MOA in the Mediterranean climate, were evaluated over two consecutive years and were found to be the most effective and safe weed control strategy. These treatments prevented the emergence of most weed species and indorsed a significant advantage for the moringa over the weeds. The most common weed species were *A. albus, A. retroflexus, A. blitoides.*, and *P. oleracea*; all have small seed sizes and mainly germinate from the upper layers of the soil [31], which can explain the high efficacy of the treatments. Five of these herbicides resulted in high yields with no impact on the moringa development and growth (82–106% of the control), suggesting that these treatments should be favored as the primary treatment during sowing. The relatively wide range of herbicides MOAs (i.e., inhibition of carotenoids, long-chain fatty acids, PPO, and PSII) that were found safe for moringa may be a consequence of its large seed size and other growth characteristics. Moringa has a large seed (7.6-9.6 mm length), allowing it to emerge from deep sowing (3 cm and deeper), thus escaping the treated layer. Furthermore, moringa has a rapid root system development at the early growth stages [32]. We may hypothesis that the moringa root system architecture enables it to elongate outside the treated layer before the absorption of the herbicide and avoid its toxic effect [33]. Similar growth characteristics are also typical for cotton (*Gossypium hirsutum*) which can be seeded at a depth of 3–5 cm and has a high resistance to some herbicides (e.g., trifluralin) [34,35]. However, the low sensitivity of moringa to flurochloridone, which is absorbed through the root and the stem, suggests that more than one tolerance mechanism may be involved in this crop. We assume that a high detoxification rate or low translocation rate of the herbicides in the plant might be the second tolerance mechanism [36].

Despite the high efficacy and crop safety for the moringa plants, the impact of the PRE treatments is reduced over time [37]. Moreover, moringa is a summer forage crop grown under the irrigated agro-system in the Mediterranean basin. Thus, several weeds species emerge later in the season in the secondary emergence flashes. One of these species, *C. rotundus*, was observed during the later growing seasons (Tables S4 and S5). Despite its fast development, it only had a minor effect on the moringa development, as expressed by non-significant yield reductions, compared to the hand weeded control (Figure 1A). These results suggest that *C. rotundus* was inhibited to some extent by the herbicides, which provided sufficient advantage for the moringa plants. However, the incomplete control of *C. rotundus* (and other weeds) throughout the growing season indicates the need for complementary treatments. In this regard, the POST treatments can complete the PRE application for season-long control. Nonetheless, the POST herbicides were less safe for the moringa plants. Furthermore, three of these herbicides had inconsistent crop safety results between both years, in terms of moringa fresh weight.

Temperature conditions during the experiments may impact the crop safety of the POST herbicide treatments and can explain the non-consistency in the results between the years, in terms of the moringa yield. In 2020, we had a drainage problem in the experimental plots. Consequently, the development of the moringa plants was significantly inhibited which may affect the plant's sensitivity to the application of the herbicides. Moreover, in 2019 the POST treatment was applied in early August, while in 2020 the treatment was applied in early June. Moringa is a fast-growing tree, especially on high-temperature regimes [32,38]. The higher temperatures in August (29 °C) compared to June (24 °C) may cause faster detoxification of the herbicide in the crop, and as a result a lower effect on the crop development. Recently, Matzrafi [39] showed that elevated temperatures can result in faster ACCase herbicide de-toxification (i.e., temperature-dependent resistance). Likewise, other environmental conditions have been shown to play a role in herbicide inconsistency (reviewed by [40,41]). These results suggest that complex G × E × M interaction may affect the level of crop safety of the various POST herbicide applications.

The inconstancy of the POST herbicides treatment suggests that incorporating another POST weed control method is needed. We decided to test the finger weeder cultivation system as a non-chemical weed control alternative based on its high effectiveness for Mediterranean summer field crops such as maize (*Zea mays*), cotton, sunflower (*Helianthus annuus*), and tomato (*Solanum lycopersicum*) [42]. The focus was given to cultivation repetition and timing; two main aspects affecting finger weeder crop safety and efficacy. The crop establishment before the first treatment is essential for the safe treatment, while applying the finger weeder on sensitive crop plants could inhibit their growth and cause yield loss. On the other hand, later application provides more time for the weed to emerge and

establish, nonetheless, results in lower weed control efficacy [18,43]. Owing to the earlier application in 2020 compared to 2019 (6 vs. 14 days after emergence) better weed control results were observed both in the single and double cultivation treatments, demonstrating the importance of the cultivation timing. In terms of crop safety, although higher fresh weight reduction was recorded in the 2020 experiment, there were lower crop stand reductions in comparison to 2019. These results can be explained by the differences between field conditions mentioned above. Application of the finger weeder in corn, soybean, and sunflower, at similar phenological stages, resulted in efficient weed control (~93% reduction in weed cover), with no damage to the crop [21]. These results emphasize the importance of the timing of the first cultivation treatment and the need for complementary treatment for the late application.

Multiple treatments of the finger weeder had no additional contribution for the weed control in 2019. Furthermore, these treatments resulted in higher damage to the crop than the single application, probably due to a reduction in the crop stand by uprooting. In contrast, in 2020 the multiple treatments had more than 80% weed cover reduction when applied at the early treatment (6 days after moringa emergence). Moreover, higher yields were obtained compared to the single application, despite the lower crop stand. These results emphasize the importance of the combination between the application timing and multiple applications, allowing safe crop establishment, and later growth afterward [18,44]. Despite the high fresh weight reduction in both years, the fact that the early cultivation treatments had little impact on the moringa crop stand and adequately controlled weeds suggests the potential of this control tool. Our results can set the basis for future research of cultivation treatment as a non-chemical method for this crop, which has a high interest in the organic food market.

5. Conclusions and Future Perspective

Our results offer several PRE and POST herbicides as effective safe weed control solutions for the new forage crop moringa. Additionally, they demonstrate the potential contribution of the finger weeder as an alternative non-chemical control means. We suggest that the best strategy for weed control in moringa should include PRE herbicides, followed by inter-row and intra-row cultivation, or POST application after the moringa establishment. The suggested multi-tactic approach is mainly essential for high weed infestation conditions where several weed emergence flashes are evident [44]. Furthermore, the non-chemical control should not be used as a sole treatment, and to achieve higher yields and weed control levels, few non-chemical methods should be considered for the non-chemical IWM procedure [42,45]. Our approach is compatible with the concept of IWM, thereby providing higher yields and longer periods of effective weed control [27]. Our findings indicate potential new weed control tools for the future development of IWMs for moringa. Thus, this study set the foundations for successful integration of this promising forage crop moringa into the semi-arid Mediterranean basin cropping systems, and for increasing the crop rotation options of farmers.

Supplementary Materials: The following are available online at https://www.mdpi.com/article/ 10.3390/agronomy11061055/s1. Figure S1. Field experiment documentation. Figure S2. Average daily temperature during growth periods. Figure S3. Effect of pre-emergence herbicides treatment on moringa and weeds during 2018 season. Figure S4. Effect of sequential finger weeder treatments on the moringa fresh weight. Figure S5. Effect of sequential finger weeder treatments on the moringa weed control efficacy. Table S1. Main operation dates in the field experiments. Table S2. List of herbicides used for the field experiments. Table S3. Number and timings of the cultivation treatments. Table S4. Weed density after pre-emergence herbicides treatment. Table S5. Main weed species following the pre-emergence herbicide treatments. Table S6. Moringa crop stand before the POST herbicide treatments. Table S7. Weed density after POST herbicides treatment. Table S8. Main weed species following the POST herbicide treatments. Table S9. The impact of sequential finger weeder treatments on moringa crop stand. Table S10. Weed density after finger weeder treatment. Table S11. Main weed species following the finger weeder treatment.

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References

- 1. Jahn, S. Using moringa seeds as coagulants in developing countries. Am. Water Work. Assoc. 1988, 80, 43–50. [CrossRef]
- 2. Olson, M.E.; Carlquist, S. Stem and root anatomical correlations with life form diversity, ecology, and systematics in Moringa (*Moringaceae*). *Bot. J. Linn. Soc.* **2001**, *135*, 315–348. [CrossRef]
- 3. Camacho, P.F.; Sousa, S.V.; Bergamasco, R.; Teixeirs, R.M. The use of *Moringa oleifera* as a natural coagulant in surface water treatment. *Chem. Eng. J.* 2017, 313, 222–237. [CrossRef]
- Falowo, A.B.; Mukumbo, F.E.; Idamokoro, E.M.; Lorenzo, J.M.; Afolayan, A.J.; Muchenje, V. Multi-functional application of Moringa oleifera Lam. in nutrition and animal food products: A review. Food Res. Int. 2018, 106, 317–334. [CrossRef] [PubMed]
- 5. Vaknin, Y.; Mishal, A. The potential of the tropical "miracle tree" *Moringa oleifera* and its desert relative *Moringa peregrina* as edible seed-oil and protein crops under Mediterranean conditions. *Sci. Hortic.* **2017**, 225, 431–437. [CrossRef]
- 6. Mridha, M.A.U. Prospects of moringa cultivation in Saudi Arabia. J. Appl. Environ. Biol. Sci. 2015, 5, 39-46.
- 7. Gopalakrishnan, L.; Doriya, K.; Santhosh, D. *Moringa oleifera*: A review on nutritive importance and its medicinal application. *Food Sci. Hum. Wellness* **2016**, *5*, 49–56. [CrossRef]
- 8. Makin, D.; Solowey, E.M. Observations on the adaptability of some species of moringa in Israel. *Acta Hortic.* **2017**, *1158*, 33–44. [CrossRef]
- Cohen-zinder, M.; Weinberg, Z.; Leibovich, H.; Chen, Y.; Rosen, M.; Sagi, G.; Orlov, A.; Agmon, R.; Yishay, M.; Miron, J.; et al. Ensiled *Moringa oleifera*: An antioxidant-rich feed that improves dairy cattle performance. *J. Agric. Sci.* 2017, 155, 1174–1186. [CrossRef]
- Cohen-zinder, M.; Orlov, A.; Tro, O.; Agmon, R.; Kabiya, R.; Shor-shimoni, E.; Wagner, E.K.; Hussey, K.; Leibovich, H.; Miron, J.; et al. Dietary supplementation of *Moringa oleifera* silage increases meat tenderness of Assaf lambs. *Small Rumin. Res.* 2017, 151, 110–116. [CrossRef]
- Cohen-Zinder, M.; Leibovich, H.; Vaknin, Y.; Sagi, G.; Shabtay, A.; Ben-Meir, Y.; Nikbachat, M.; Portnik, Y.; Yishay, M.; Miron, J. Effect of feeding lactating cows with ensiled mixture of *Moringa oleifera*, wheat hay and molasses, on digestibility and efficiency of milk production. *Anim. Feed Sci. Technol.* 2016, 211, 75–83. [CrossRef]
- 12. Oerke, E.C. Crop losses to pests. J. Agric. Sci. 2006, 144, 31-43. [CrossRef]
- 13. Puschner, B. Problem weeds in hay and forages for lifestock. In Proceedings of the 35th California Alfalfa & Forage Symposium, Visalia, CA, USA, 12–14 December 2005; pp. 12–14.
- 14. Casida, J.E.; Bryant, R.J. The ABCs of pesticide toxicology: Amounts, biology, and chemistry. *Toxicol. Res.* **2017**, *6*, 755–763. [CrossRef]
- 15. Monaco, T.J.; Weller, S.C.; Ashton, F.M. Weed Science: Principles and Practices; John Wiley & Sons: New York, NY, USA, 2002.
- 16. Hernández, A.F.; Parrón, T.; Tsatsakis, A.M.; Requena, M.; Alarcón, R.; López-guarnido, O. Toxic effects of pesticide mixtures at a molecular level: Their relevance to human health. *Toxicology* **2013**, *307*, 136–145. [CrossRef] [PubMed]
- 17. Peruzzi, A.; Martelloni, L.; Frasconi, C.; Fontanelli, M.; Pirchio, M.; Raffaelli, M. Machines for non-chemical intra-row weed control in narrow and wide-row crops: A review. *J. Agric. Eng.* **2017**, *583*, 57–70. [CrossRef]
- Pannacci, E.; Lattanzi, B.; Tei, F. Non-chemical weed management strategies in minor crops: A review. Crop Prot. 2017, 96, 44–58.
 [CrossRef]
- 19. Ascard, J.; Fogelberg, F. Mechanical in-row weed control in transplanted and direct-sown bulb onions. *Biol. Agric. Hortic.* 2008, 25, 235–251. [CrossRef]
- 20. Van der Weide, R.; Bleeker, P.; Achten, V.; Lotz, L. Innovation in mechanical weed control in crop rows. *Weed Res.* 2008, 48, 215–224. [CrossRef]
- 21. Pannacci, E.; Tei, F. Effects of mechanical and chemical methods on weed control, weed seed rain and crop yield in maize, sunflower and soyabean. *Crop Prot.* **2014**, *64*, 51–59. [CrossRef]
- 22. Pérez-Ruíz, M.; Slaughter, D.C.; Fathallah, F.A.; Gliever, C.J.; Miller, B.J. Co-robotic intra-row weed control system. *Biosyst. Eng.* **2014**, *126*, 45–55. [CrossRef]

- 23. Melander, B.; Lattanzi, B.; Pannacci, E. Intelligent versus non-intelligent mechanical intra-row weed control in transplanted onion and cabbage. *Crop Prot.* 2015, 72, 1–8. [CrossRef]
- Znova, L.; Melander, B.; Lisowski, A.; Klonowski, J.; Edwards, G.T.C.; Nielsen, S.K.; Green, O. A new hoe share design for weed control: Measurements of soil movement and draught forces during operation. *Acta Agric. Scand. Sect. B Soil Plant Sci.* 2018, 68, 139–148. [CrossRef]
- 25. Buhler, D.D.; Gunsolus, J.J.; Ralston, D.F. Integrated weed management techniques to reduce herbicide inputs in soybean. *Agron. J.* **1992**, *84*, 973–978. [CrossRef]
- 26. Swanton, C.J.; Weise, S.F. Integrated Weed Management: The rationale and approach. Weed Res. 1991, 5, 657–663. [CrossRef]
- 27. Buhler, D.D. 50th Anniversary-Invited Article: Challenges and opportunities for integrated weed management. *Weed Sci.* 2002, 50, 273–280. [CrossRef]
- 28. Sokal, R.R.; Rohlf, F.J. *Biometry: The Principles and Practice of Statistics in Biological Research*, 3rd ed.; W.H. Freeman and Company: New York, NY, USA, 1997; pp. 419–422.
- 29. Deytieux, V.; Nemecek, T.; Freiermuth, R.; Gaillard, G.; Munier-jolain, N.M. Is Integrated Weed Management efficient for reducing environmental impacts of cropping systems ? A case study based on life cycle assessment. *Eur. J. Agron.* **2012**, *36*, 55–65. [CrossRef]
- Liebman, M.; Gallandt, E.R. Many little hammers: Ecological management of crop-weed interactions. In *Ecology in Agriculture*; Academic Press: San Diego, CA, USA, 1997; pp. 291–343.
- 31. Patel, J.B.; Bhatiya, V.J.; Babariya, C.A.; Sondarva, J. Effect of seed size on seedling vigour, plant growth, seed yield and its parameters: A review. *Res. Environ. Life Sci.* 2016, *9*, 859–864.
- 32. Muhl, Q.E.; Du Toit, E.S.; Robbertse, P.J. Temperature effect on seed germination and seedling growth of *Moringa oleifera* Lam. *Seed Sci. Technol.* **2011**, *39*, 208–213. [CrossRef]
- Fakayode, O.A.; Akpan, D.E.; Ojoawo, O.O. Size characterization of moringa (*Moringa oleifera*) seeds and optimization of the dehulling process. J. Food Process Eng. 2019, 42, 1–17. [CrossRef]
- 34. Eshel, Y. Tolerance of cotton to diuron, fluometuron, norea, and prometryne. Weed Sci. 1969, 17, 492–496. [CrossRef]
- 35. Kleifeld, Y.; Rubin, B.; Blumenfeld, T.; Herzlinger, G.; Buxbaum, H.; Golan, S. Soil-incorporated trifluralin protects cotton (*Gossypium hirsutum*) from root-absorbed herbicides. *Weed Res.* **1994**, *34*, 461–469. [CrossRef]
- 36. Cole, D.J. Detoxification and activation of agrochemicals in plants. Pestic. Sci. 1994, 42, 209–222. [CrossRef]
- 37. Altland, J.E. Efficacy of preemergence herbicides over time. J. Environ. Hortic. 2019, 37, 55–62. [CrossRef]
- Muhl, Q.E.; du Toit, E.S.; Robbertse, P.J. Adaptability of *Moringa oleifera* Lam. (Horseradish) tree seedlings to three temperature regimes. *Am. J. Plant Sci.* 2011, 2, 776–780. [CrossRef]
- Matzrafi, M.; Seiwert, B.; Reemtsma, T.; Rubin, B.; Peleg, Z. Climate change increases the risk of herbicide-resistant weeds due to enhanced detoxification. *Planta* 2016, 244, 1217–1227. [CrossRef]
- 40. Baucom, R.S. Evolutionary and ecological insights from herbicide-resistant weeds: What have we learned about plant adaptation, and what is left to uncover? *New Phytol.* **2019**, *223*, 68–82. [CrossRef] [PubMed]
- 41. Ziska, L.H. Climate change and the herbicide paradigm: Visiting the future. Agronomy 2020, 10, 1953. [CrossRef]
- 42. Asaf, E. Mechanical weed control for row crop using finger weeder. Ph.D. Thesis, The Hebrew University of Jerusalem, Jerusalem, Israel, 2021.
- 43. Melander, B.; Rasmussen, I.A.; Bàrberi, P. Integrating physical and cultural methods of weed control—Examples from European research symposium integrating physical and cultural methods of weed control. *Weed Sci.* 2005, *53*, 369–381. [CrossRef]
- 44. Rask, A.M.; Larsen, S.U.; Andreasen, C.; Kristoffersen, P. Determining treatment frequency for controlling weeds on traffic islands using chemical and non-chemical weed control. *Weed Res.* 2013, *53*, 249–258. [CrossRef]
- Cirujeda, A.; Aibar, J.; Moreno, M.; Zaragoza, C. Effective mechanical weed control in processing tomato: Seven years of results. *Renew. Agric. Food Syst.* 2013, 30, 223–232. [CrossRef]