



## Article

# Practical Knowledge of Injuries Caused by Simulated Herbicide Drift in Young Tomato Plants

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**Abstract:** Tomatoes are often grown in proximity to other crops such as grain, which can increase their susceptibility to herbicide drift and subsequent crop. Therefore, the objective of this study was to evaluate the effect of simulated herbicide drift on tomato plants. Treatments were established in a  $10 \times 3 + 1$  factorial scheme using a completely randomized design with four replications. The first factor consisted of ten herbicides, while the second was composed by three subdoses (1/4, 1/16, and 1/32) along with an additional treatment without herbicide application. The herbicides 2,4-D, dicamba, glyphosate, saflufenacil, oxyfluorfen, and isoxaflutole caused injury levels greater than 20% or reductions in plant biomass greater than 30% at the lowest subdose. Increasing the subdose resulted in a corresponding increase in injury level and a reduction in biomass. Tomato exposed to hexazinone, diuron, nicosulfuron, and diquat at a subdose of 1/64 exhibited low injury levels and biomass reductions. However, at other subdoses, these herbicides caused significant plant damage. Among the herbicides tested, the auxinic herbicides, particularly dicamba, presented a higher risk for the tomato crop. The documentation and description of the visual symptoms caused by each herbicide applied to tomatoes will aid producers to identify drift problems in the field.

**Keywords:** ALS; glyphosate; injury levels; photosystem II inhibitors; PPO; subdoses; synthetic auxin



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## 1. Introduction

The tomato plant is native to the Andes and exhibits adaptability to diverse regions and significant genre variability. Tomatoes are part of the Solanaceae family, which also includes peppers and bell peppers [1].

Tomatoes are among the most consumed vegetables in Brazil, ranking ninth in global production and first among South American countries [2]. However, between 2016 and 2020, the area planted with tomatoes decreased by 18.94%, from 64,296 to 52,117 ha, and the annual productivity also declined by 11.16% between 2017 and 2020, from 4.225 to 3.753 million tons [3]. This reduction in tomato cultivation is primarily due to competition with the grain sector. Despite the recent decline, Hortifruti Brasil anticipates a recovery in 2022 compared to 2021, with a 9.7% increase in the planted area and 21.3% growth in the industry. This growth is driven by a shortage of pulp stocks at the start of the year [4]. The competition for cultivation areas between the tomato and grain sector implies that these crops are often grown in proximity to each other. This proximity can expose tomato crops to problems associated with herbicide drift from areas cultivated with grains.

Herbicide drift refers to the deviation of spray molecules from the target area due to wind action [5]. Drift is a significant problem in agriculture as it reduces the effectiveness of herbicide applications and puts crops in neighboring areas at risk [6]. For instance, the drift of 2,4-D and dicamba onto ‘Ponkan’ tangerine seedlings (*Citrus reticulata*) caused significant injury levels, leaf loss, and reduced chlorophyll content in the leaves [7]. Drift of fomesafen onto sugar beet crops resulted in reductions in morphological variables such as plant height, root length, and biomass, as well as physiological variables such as photosynthetic rate,

stomatal conductance, transpiration rate, and total leaf chlorophyll content [8]. Glyphosate drift in coffee crops caused damage characterized by chlorosis and narrowing of the leaf blade, in addition to a reduction in plant height, leaf area, and root dry matter [9]. Drift can decrease productivity and affect crop morphology [10].

According to Takeshita et al. [11], herbicides that reach the soil are subject to physical-chemical processes that regulate their environmental fate, including the persistence of herbicide molecules in the soil. Bioassays with plant species that are sensitive to the herbicide being studied, easy to cultivate, and fast growing can be used to evaluate this persistence [12]. Marchesan et al. [13] tested several species as bioindicators of the herbicide atrazine and found that tomatoes and radishes were the most sensitive species.

Tomato is a highly herbicide-susceptible crop, particularly to auxinics, and similar to the aforementioned crops, it is vulnerable to severe damage from herbicide drifts. Therefore, further studies are necessary to assess the impact of different herbicides on tomatoes in the context of drift. The aim of this study was to evaluate the effect of simulated herbicides drift at sublethal doses on tomato plants. The findings of this study will serve as a basis for agricultural producers and professionals to gain knowledge and identify the damage caused by herbicide drift, leading to improved management of these resources.

## 2. Materials and Methods

### 2.1. Site and Plant Material Description

The study was performed from April to May 2022 in a greenhouse located at the Department of Agronomy (DAA) and the Integrated Weed Management (IWM) Laboratory of the Federal University of Viçosa, Viçosa, MG, Brazil (altitude: 648 m; latitude: 20°45'144" S; longitude: 42°52'53" W).

The tomato cultivar utilized in the study was *Átila*, a hybrid of the saladette type. It has indeterminate growth, is rustic and vigorous, highly productive, and displays high sequential fruit set up to the pointer. These characteristics made it easy to handle in the greenhouse. Seedlings were acquired from the Semearte company (Coimbra, MG, Brazil), with approximately 3–4 fully expanded leaves.

The tomato seedlings were transplanted into 250 mL plastic pots filled with substrate Tropstrato HT Hortaliças (Composition: Pine Bark, Vermiculite, PG Mix 14.16.18, Potassium Nitrate, Simple Superphosphate, and Peat). Following transplanting, the seedlings were acclimatized for 15 days before treatment application.

### 2.2. Experimental Design and Treatments

The experimental design consisted of a randomized blocks with four replications. The treatments followed a  $10 \times 3 + 1$  factorial scheme. The first factor included ten herbicides with different modes of action (2,4-D, dicamba, glyphosate, saflufenacil, oxyfluorfen, hexazinone, diuron, diquat, nicosulfuron, and isoxaflutole), and the second factor comprised three levels of simulated drift, which were equivalent to 1/16D, 1/32D, and 1/64D of the highest label dose (D) recommended in the field, in addition a control treatment (Table 1).

The simulated herbicides drift was performed in an outdoor environment outside the greenhouse between 8 and 9:30 am using a backpack sprayer pressurized with CO<sub>2</sub>, operating at a constant pressure of 294.2 kPa, equipped with a bar containing two TT 110.02 nozzle models spaced 50 cm apart, adjust to deliver a spray volume 154 L ha<sup>-1</sup>. The application was carried out at a height of 50 cm above the plant canopy. At the time of application, the temperature was 21 °C, the relative air humidity was 94%, and the wind speed was 0.3 m/s, according to the National Institute of Meteorology, at station A510 located in Viçosa, MG, Brazil [14]. The seedlings used in the study had approximately 6–7 fully expanded leaves. After the application of treatments, the pots were placed on benches inside the greenhouse and left without irrigation for 6 h to ensure herbicide absorption. Daily irrigation was provided to maintain optimal soil moisture conditions and prevent substrate waterlogging.

**Table 1.** Herbicides and dose ratios derived from the highest label dose applied.

Modes of Action	Commercial Name	Name of a.i. or a.e.	Highest Commercial Dose	Highest Dose g of a.i or a.e./ha	Dose Ratio g of a.i or a.e./ha		
					1/16	1/32	1/64
Synthetic auxin	U46 BR Atectra	2,4-D	3.5 L/ha	2345	146.56	73.28	36.64
		Dicamba	1.5 L/ha	720	45	22.5	11.25
Enolpyruvylshikimate-3-phosphate synthase (EPSPs) inhibitor	Roundup Original	Glyphosate	6.0 L/ha	2220	138.75	69.38	34.69
Protoporphyrinogen oxidase (PPO) inhibitors	Heat Goal BR	Saflufenacil	200 g/ha	140	8.75	4.38	2.19
		Oxyfluorfen	6.0 L/ha	1440	90	45	22.5
Photosystem II (PSII) inhibitors	Hexazinona D NORTOX Diuron NORTOX 500 SC	Hexazinone	3.0 kg/ha	396	24.75	12.38	6.19
		Diuron	6.4 L/ha	3200	200	100	50
Photosystem I (PSI) inhibitors	Reglone	Diquat	3.5 L/ha	60	43.75	21.88	10.94
Acetolactate synthas (ALS) inhibitors	Nicosulfuron NORTOX 750WG	Nicosulfuron	80 g/ha	700	3.75	1.88	0.94
Carotenoid biosynthesis [4-hydroxyphenylpyruvate dioxygenase (HPPD)] inhibitors	Provence 750 WG	Isoxaflutole	350 g/ha	262.5	16.41	8.20	4.10

a.i. = active ingredient; a.e. = acid equivalent.

### 2.3. Assessments of Symptoms, Injury Level, and Dry Mass

The symptoms caused by simulated herbicide drift were monitored and documented using a photographic approach. A mini photographic studio was set up with black craft paper background and a digital camera (EOS Rebel T3i, with 18–135 mm IS optical kit, CANON, Canon, Huntington, NY, USA) mounted on a tripod. Photos were taken in daylight on the first day after application (DAA) and repeated daily until 7 DAA, and then at 14 and 21 DAA. The photographs were selected, edited, and organized according to the herbicide, mode of action, and dose ratio used.

At 21 DAA, the level of injury was assessed by visual observations, and scores were assigned on a scale ranging from 0 (normal plants) to 100 (dead plants) according to the modified EWRC [15] scale. For the evaluation of 2,4-D and dicamba, the scale by Wells et al. [16] was used since these herbicides have characteristic symptoms that differ from the other modes of action (as showing in Table 2).

**Table 2.** Scale used to evaluate the injury level caused by the simulated drift of 2,4-D and dicamba on the tomato crop.

Injury Level	Characteristics
0	No effect; normal plant
10	Slight wrinkling of terminal leaf leaflets
20	Curving of terminal leaflets, slight wrinkling of second-leaf leaflets, normal growth rate
30	Leaflets of two shell-shaped terminal leaves, terminal leaf expansion slightly suppressed
40	Malformation and growth suppression of two terminal leaves, terminal leaf size less than half that of the control. New axillary leaves develop at a substantially reduced rate
50	No terminal leaf expansion, size of second leaf half of control. Axillary leaf buds unable to open and develop
60	Small terminal growth, terminal leaf necrosis, and apparent axillary bud. Chlorosis and necrosis in clusters of axillary leaves
70	Terminal bud dead, substantial growth of heavily malformed axillary shoots
80	Limited axillary shoot growth, leaves present at the time of treatment chlorotic with slight necrosis
90	Plant dying, leaves predominantly necrotic
100	Dead plant

Source: Wells et al. [16].

At 21 DAA, the dry mass (DM) of tomato plants was determined by harvesting them and placed them in paper bags for drying in an oven (FANEM, model 320-SE, São Paulo, Brazil) for 72 h at 65 °C. The weight was measured on an analytical scale model AY220 (SHIMADZU, Kyoto, Japan) to determine the total DM. The total DM was calculated as a

percentage of the control DM (plants that did not receive the herbicide), with the control being assigned a value of 100%.

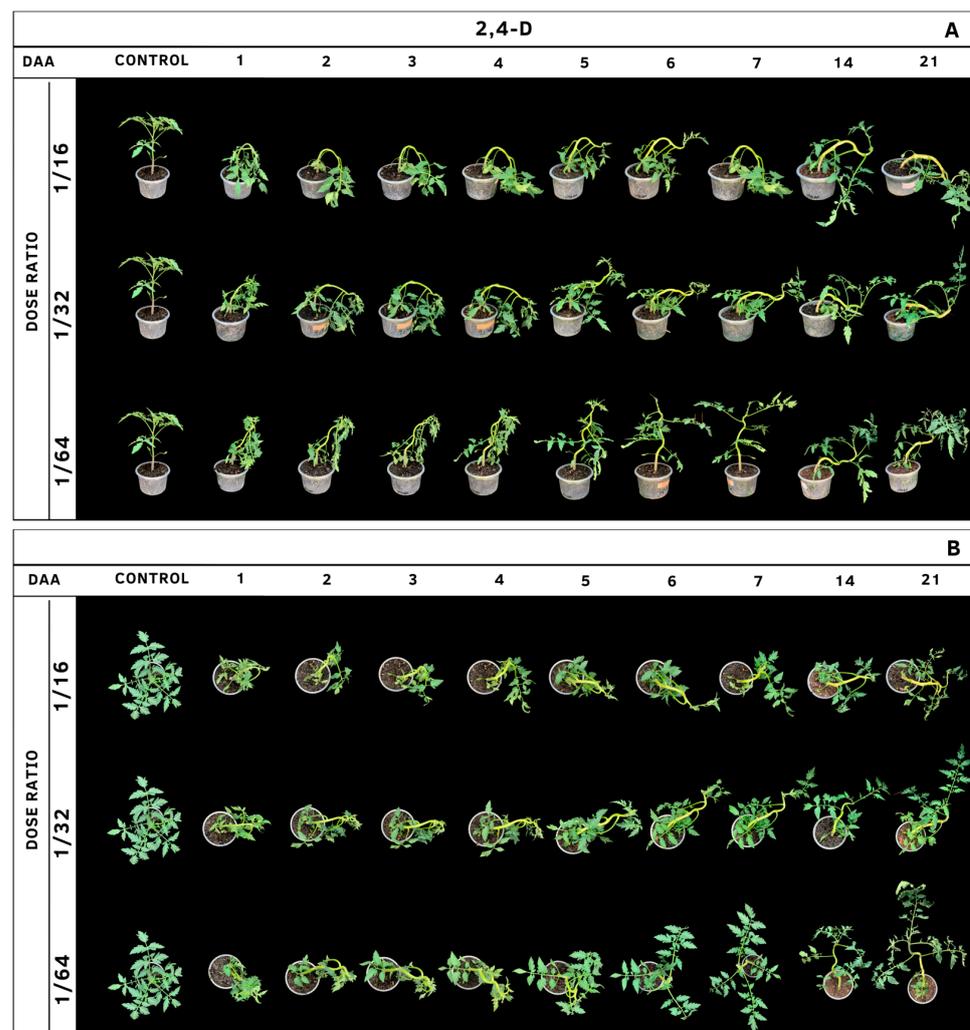
### 2.4. Statistical Analysis

Statistical analyses and graphs were performed using Sisvar software (version 5.6.) and SigmaPlot® (version 13.0 for Windows, Systat Software Inc., Point Richmond, CA, USA), respectively. The data obtained were analyzed using the F-test of variance analysis (ANOVA). When significant, the results were submitted to the Tukey’s test ( $p \leq 0.05$ ) to compare the means between the different simulated drifts.

## 3. Results

### 3.1. Effects of Simulated Drift Caused by Synthetic Auxin Herbicides

The simulated drift of 2,4-D and dicamba on tomato plants resulted in comparable symptoms, which included unusual plant growth, such as stems and petioles twisting, leaf wilting, epinasty, brittle stems that caused phloem blockage, and chlorosis. These symptoms were visible from 1 DAA of the simulated herbicides drift, and their severity increased with time, regardless of the dose ratio used. Examples of these symptoms are illustrated in Figures 1–4.



**Figure 1.** Symptoms evolution in tomato plants due to simulated 2,4-D drift at dose ratios 1/16D, 1/32D, and 1/64D (146.56, 73.28, and 36.64 g a.e. ha<sup>-1</sup>, respectively) of the recommended dose (D). Evaluations were made from 1 to 7, 14, and 21 days after application (DAA). Front view (A) and top view (B) of tomato plants affected by simulated drift.



Figure 2. The effects of simulated 2,4-D drift with a close-up view show injuries in tomato plants.

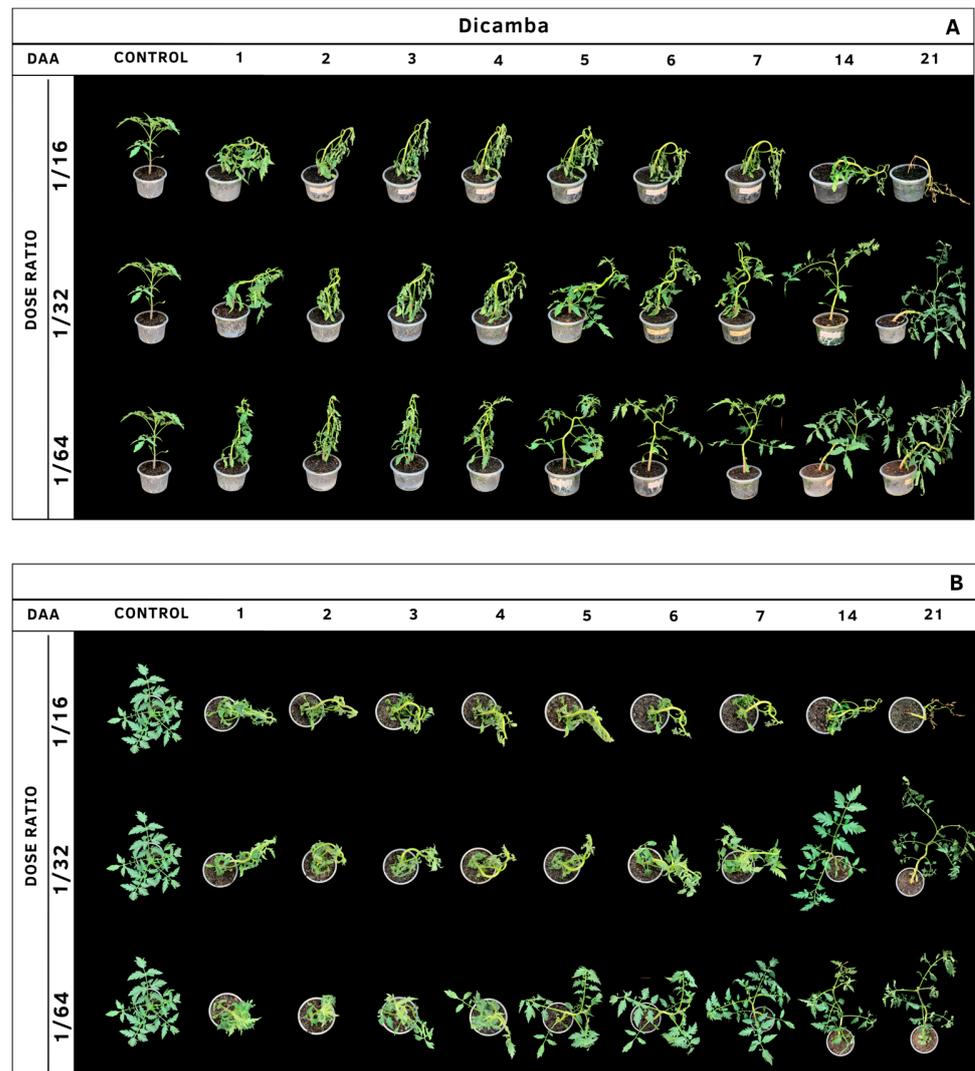


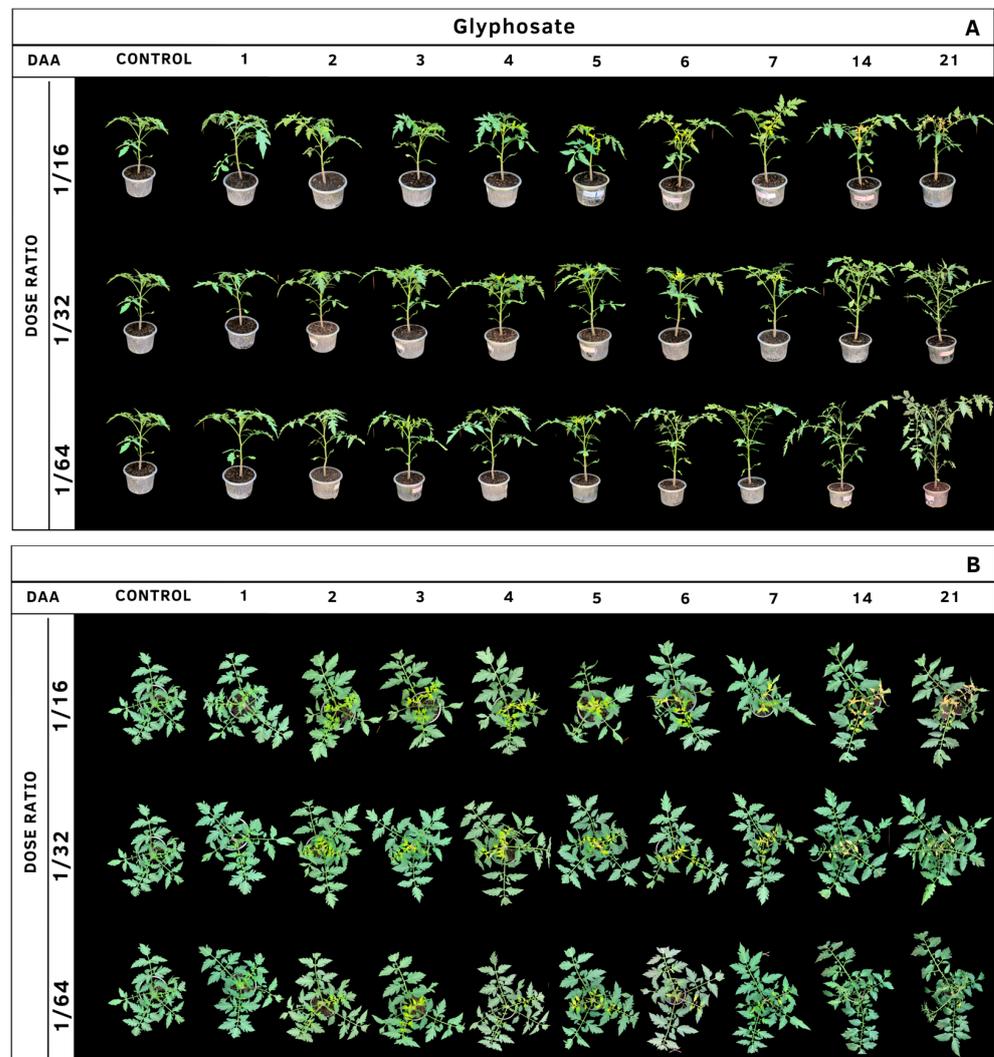
Figure 3. Symptoms evolution in tomato plants due to simulated dicamba drift at dose ratios 1/16D, 1/32D, and 1/64D (45, 22.5, and 11.25 g a.e. ha<sup>-1</sup>, respectively) of the recommended dose (D). Evaluations were made from 1 to 7, 14, and 21 days after application (DAA). Front view (A) and top view (B) of tomato plants affected by simulated drift.



**Figure 4.** The effects of simulated dicamba drift with a close-up view show injuries in tomato plants.

**3.2. Effects of Simulated Drift Caused by EPSPs-Inhibiting Herbicides**

The symptoms induced by the simulated glyphosate drift in tomatoes are characterized by chlorosis in young leaves, followed by necrosis (Figures 5 and 6). On the first day after exposure to the simulated herbicide drift at all applied dose ratios, no symptoms were observed, and they only began to appear from 2 DAA onwards.



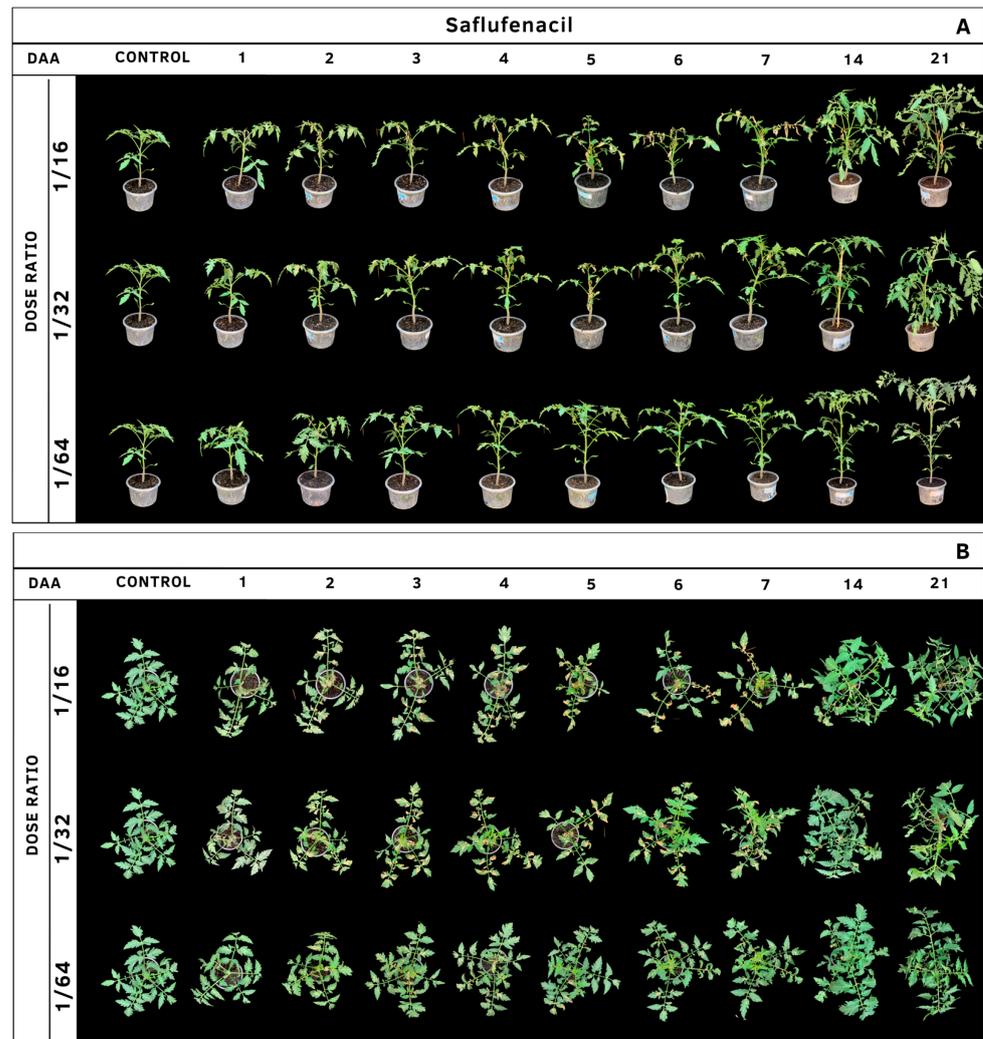
**Figure 5.** Symptoms evolution in tomato plants due to simulated glyphosate drift at dose ratios 1/16D, 1/32D, and 1/64D (138.75, 69.38, and 34.69 g a.e. ha<sup>-1</sup>, respectively) of the recommended dose (D). Evaluations were made from 1 to 7, 14, and 21 days after application (DAA). Front view (A) and top view (B) of tomato plants affected by simulated drift.



**Figure 6.** The effects of simulated glyphosate drift with a close-up view show injuries in tomato plants.

**3.3. Effects of Simulated Drift Caused by PPO-Inhibiting Herbicides**

The simulated drift of saflufenacil and oxyfluorfen in tomatoes resulted in similar symptoms characterized by the browning of young leaves, small white spots on leaves, and necrosis (Figures 7–10). These symptoms were observed on 1 DAA of the simulated herbicides drift, and they gradually worsened over time. All applied dose ratios showed the described symptoms.



**Figure 7.** Symptoms evolution in tomato plants due to simulated saflufenacil drift at dose ratios 1/16D, 1/32D, and 1/64D (8.75, 4.38, and 2.19 g a.i. ha<sup>-1</sup>, respectively) of the recommended dose (D). Evaluations were made from 1 to 7, 14, and 21 days after application (DAA). Front view (A) and top view (B) of tomato plants affected by simulated drift.



Figure 8. The effects of simulated saflufenacil drift with a close-up view show injuries in tomato plants.

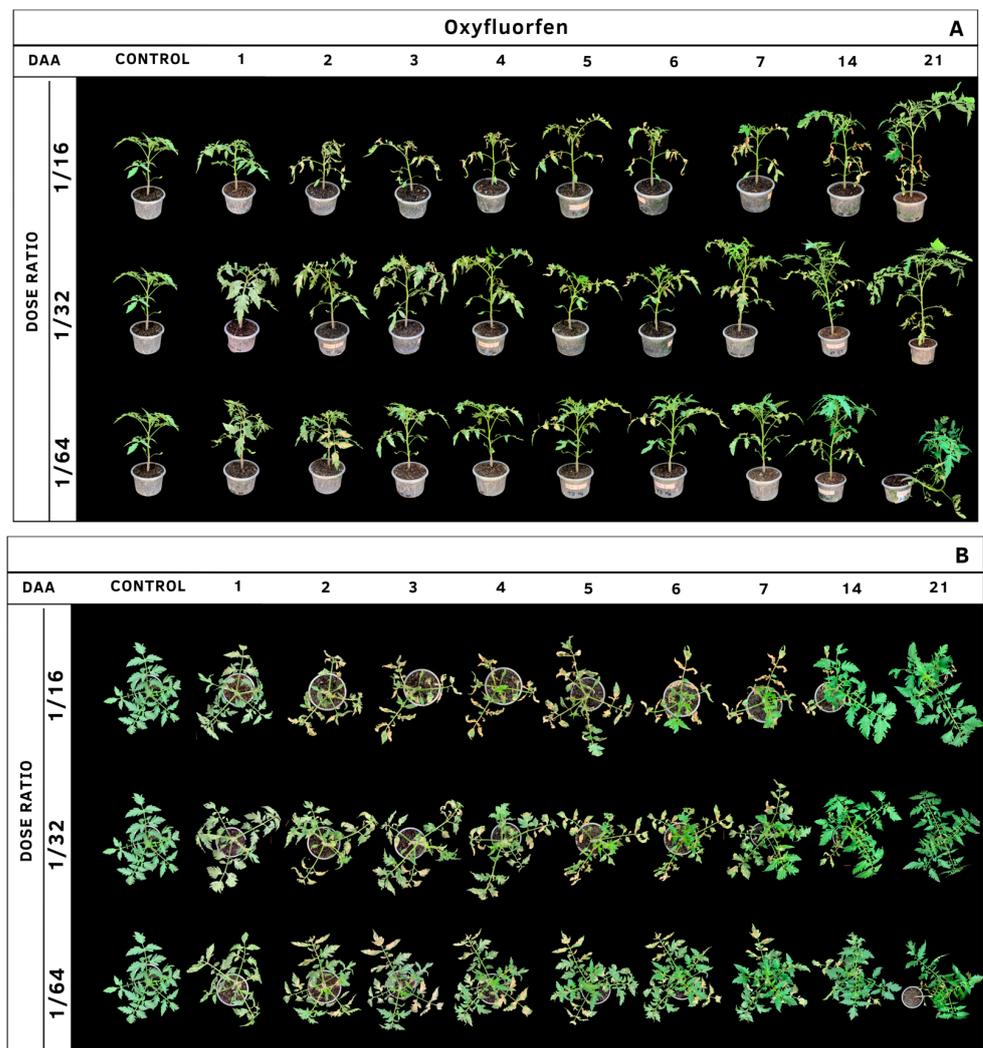


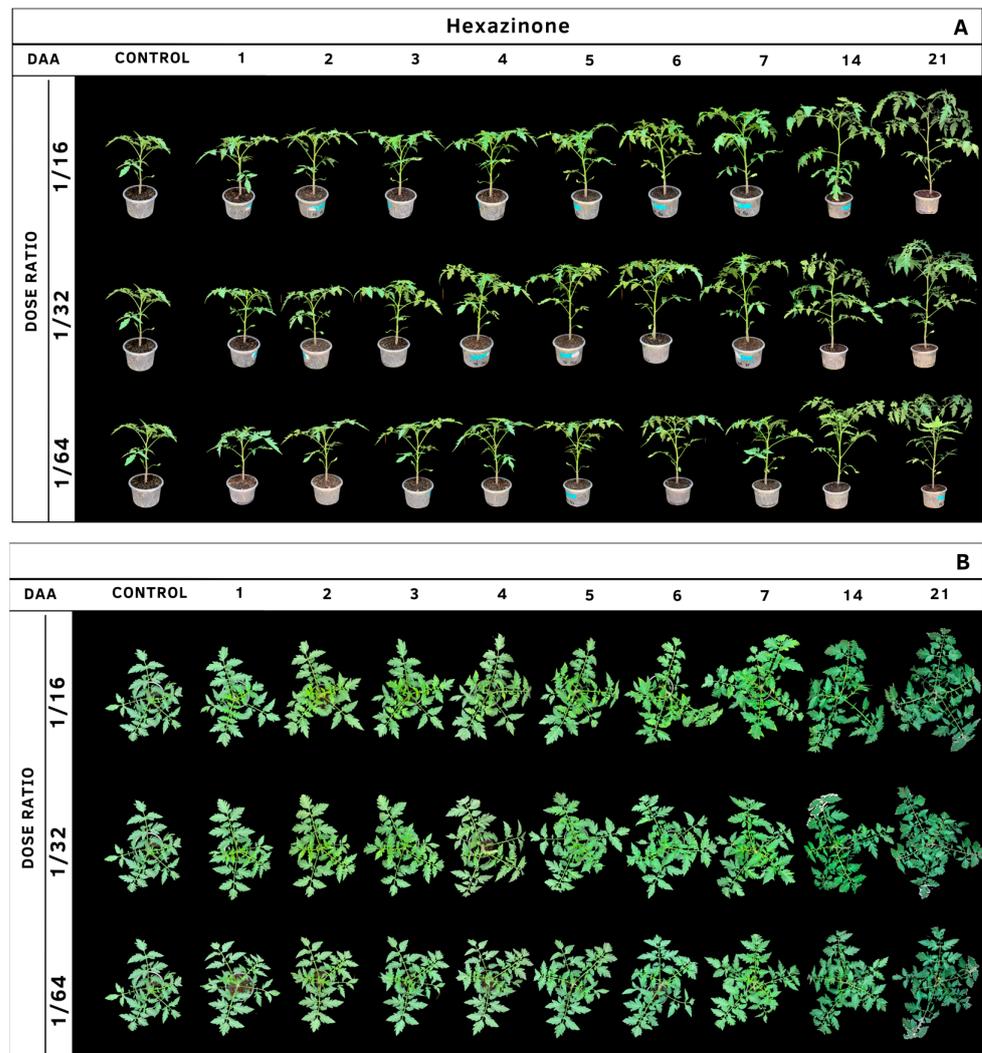
Figure 9. Symptoms evolution in tomato plants due to simulated oxyfluorfen drift at dose ratios 1/16D, 1/32D, and 1/64D (90, 45, and 22.5 g a.i. ha<sup>-1</sup>, respectively) of the recommended dose (D). Evaluations were made from 1 to 7, 14, and 21 days after application (DAA). Front view (A) and top view (B) of tomato plants affected by simulated drift.



**Figure 10.** The effects of simulated oxyfluorfen drift with a close-up view show injuries in tomato plants.

**3.4. Effects of Simulated Drift Caused by PSII-Inhibiting Herbicides**

The simulated drift of hexazinone and diuron in tomatoes causes similar symptoms characterized by chlorosis on leaf edges that progresses to the leaf center (Figures 11–14). Symptoms started appearing on the third day after exposure to the simulated herbicide drift in all applied dose ratios. The symptom was milder in the lowest simulated drift of 1/64.



**Figure 11.** Symptoms evolution in tomato plants due to simulated hexazinone drift at dose ratios 1/16D, 1/32D, and 1/64D (24.75, 12.38, and 6.9 g a.i. ha<sup>-1</sup>, respectively) of the recommended dose (D). Evaluations were made from 1 to 7, 14, and 21 days after application (DAA). Front view (A) and top view (B) of tomato plants affected by simulated drift.



Figure 12. The effects of simulated hexazinone drift with a close-up view show injuries in tomato plants.

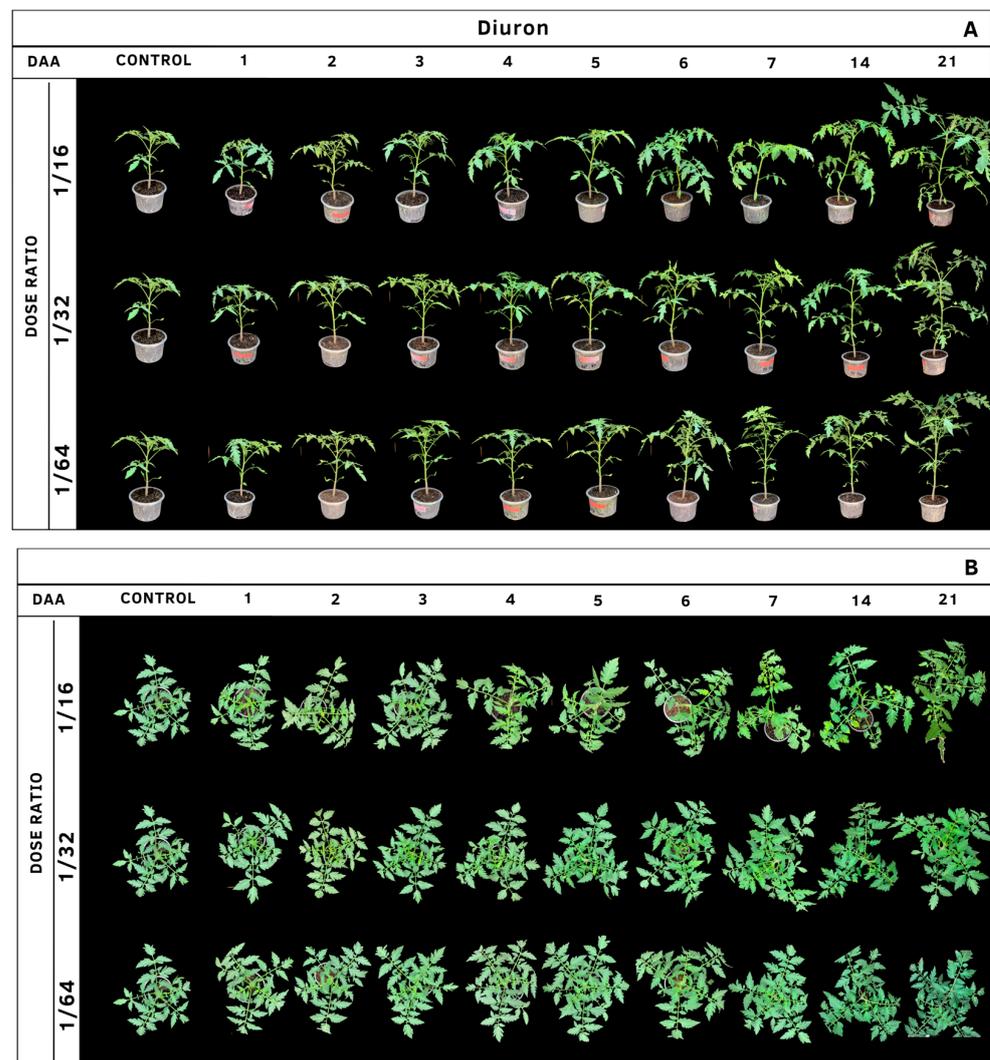


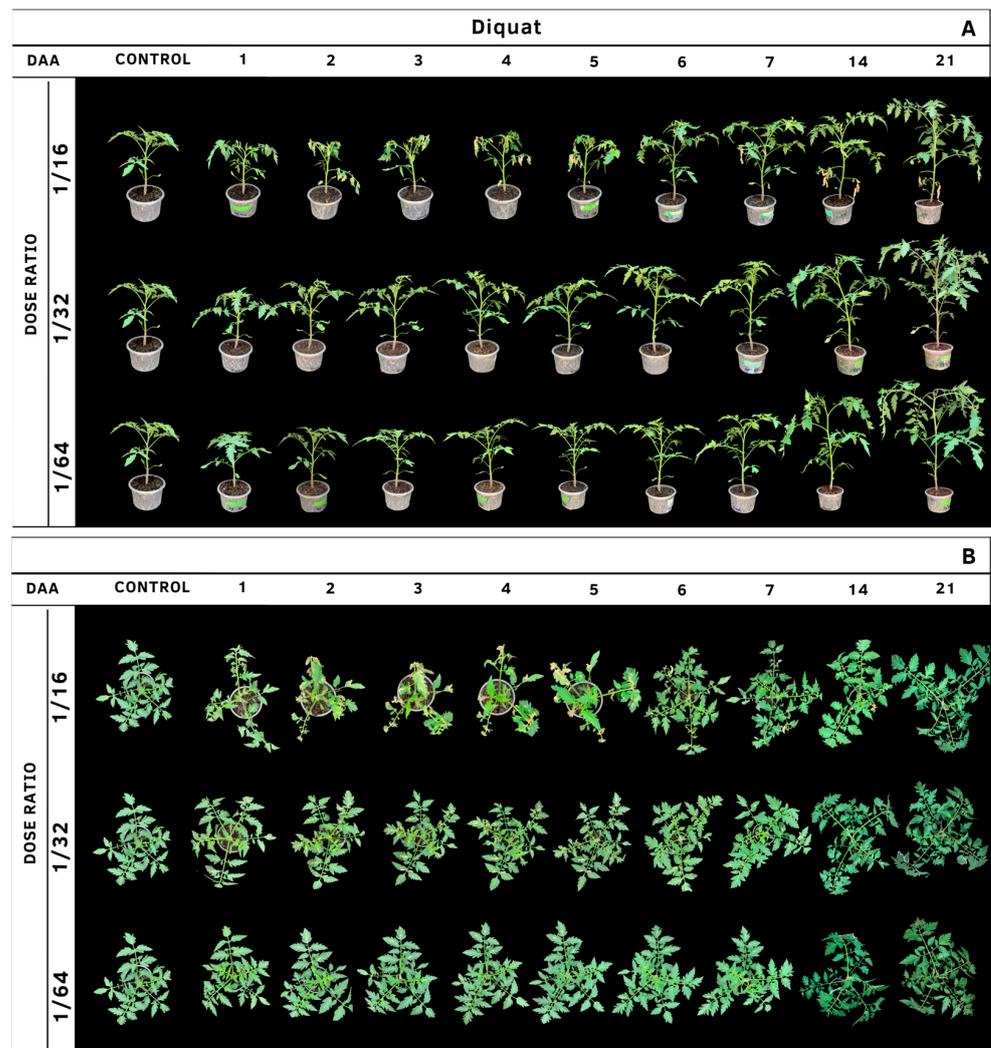
Figure 13. Symptoms evolution in tomato plants due to simulated diuron drift at dose ratios 1/16D, 1/32D, and 1/64D (200, 100, and 50 g a.i. ha<sup>-1</sup>, respectively) of the recommended dose (D). Evaluations were made from 1 to 7, 14, and 21 days after application (DAA). Front view (A) and top view (B) of tomato plants affected by simulated drift.



**Figure 14.** The effects of simulated diuron drift with a close-up view show injuries in tomato plants.

*3.5. Effects of Simulated Drift Caused by PSI-Inhibiting Herbicides*

The symptoms caused by simulated diquat drift in tomatoes are characterized by a wet leaf appearance, yellowing, and necrosis (Figures 15 and 16). It was already possible to observe these symptoms on 1 DAA of the simulated herbicide drift in all applied dose ratios. However, in the lowest simulated drift of 1/64, the symptoms were less severe.



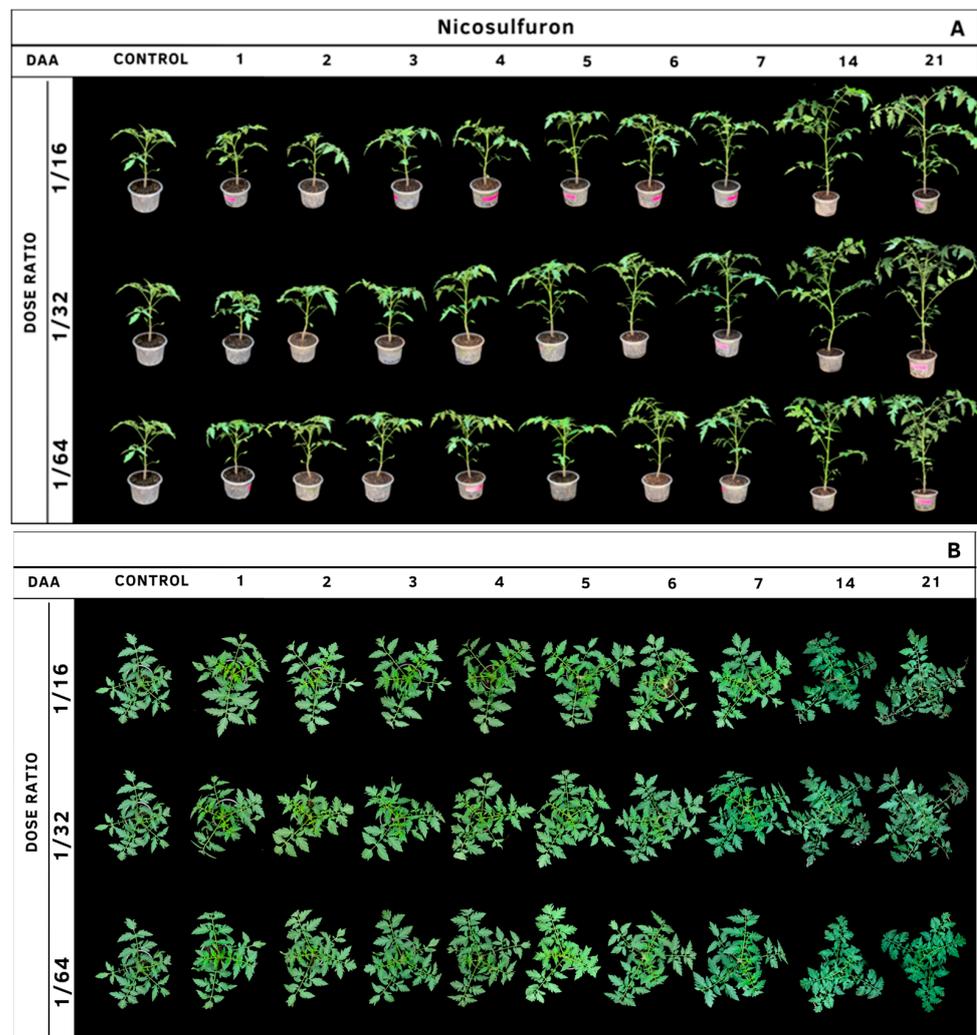
**Figure 15.** Symptoms evolution in tomato plants due to simulated diquat drift at dose ratios 1/16D, 1/32D, and 1/64D (43.75, 21.88, and 10.94 g a.i. ha<sup>-1</sup>, respectively) of the recommended dose (D). Evaluations were made from 1 to 7, 14, and 21 days after application (DAA). Front view (A) and top view (B) of tomato plants affected by simulated drift.



**Figure 16.** The effects of simulated diquat drift with a close-up view show injuries in tomato plants.

*3.6. Effects of Simulated Drift Caused by ALS-Inhibiting Herbicides*

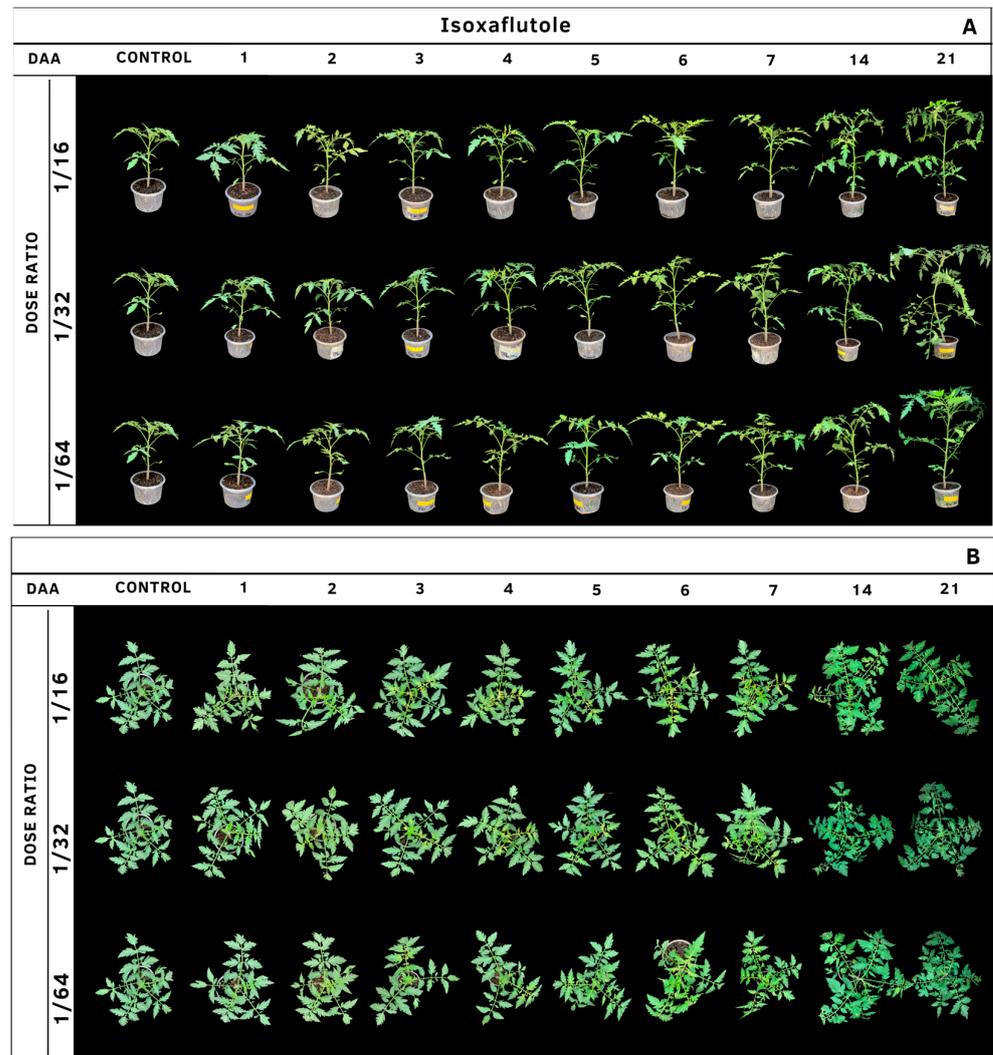
No symptoms were observed in all applied dose ratios due to the simulated nicosulfuron drift in tomato plants (Figure 17). The plants developed normally, similar to the control treatment, as shown in Figure 17.



**Figure 17.** Symptoms evolution in tomato plants due to simulated nicosulfuron drift at dose ratios 1/16D, 1/32D, and 1/64D (3.75, 1.88, and 0.94 g a.i. ha<sup>-1</sup>, respectively) of the recommended dose (D). Evaluations were made from 1 to 7, 14, and 21 days after application (DAA). Front view (A) and top view (B) of tomato plants affected by simulated drift.

### 3.7. Effects of Simulated Drift Caused by HPPD-Inhibiting Herbicides

The symptoms caused by simulated isoxaflutole drift in tomatoes are characterized by albino young leaves followed by necrosis (Figures 18 and 19). The onset of symptoms was observed 1 DAA of the simulated herbicide drift in all applied dose ratios.



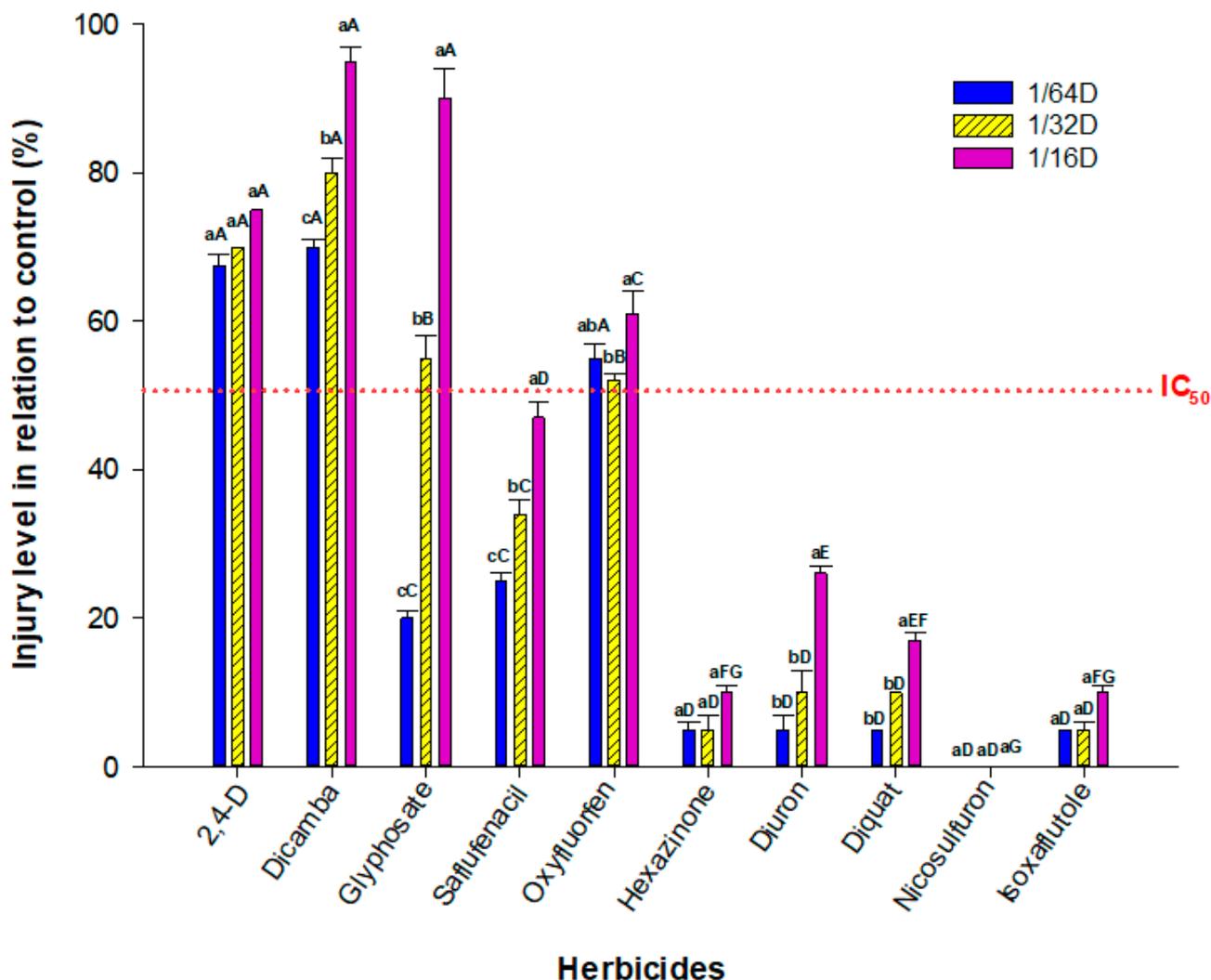
**Figure 18.** Symptoms evolution in tomato plants due to simulated isoxaflutole drift at dose ratios 1/16D, 1/32D, and 1/64D (16.41, 8.20, and 4.10 g a.i. ha<sup>-1</sup>, respectively) of the recommended dose (D). Evaluations were made from 1 to 7, 14, and 21 days after application (DAA). Front view (A) and top view (B) of tomato plants affected by simulated drift.



**Figure 19.** The effects of simulated isoxaflutole drift with a close-up view show injuries in tomato plants.

### 3.8. Effect of Simulated Herbicides Drift on Tomato Plants

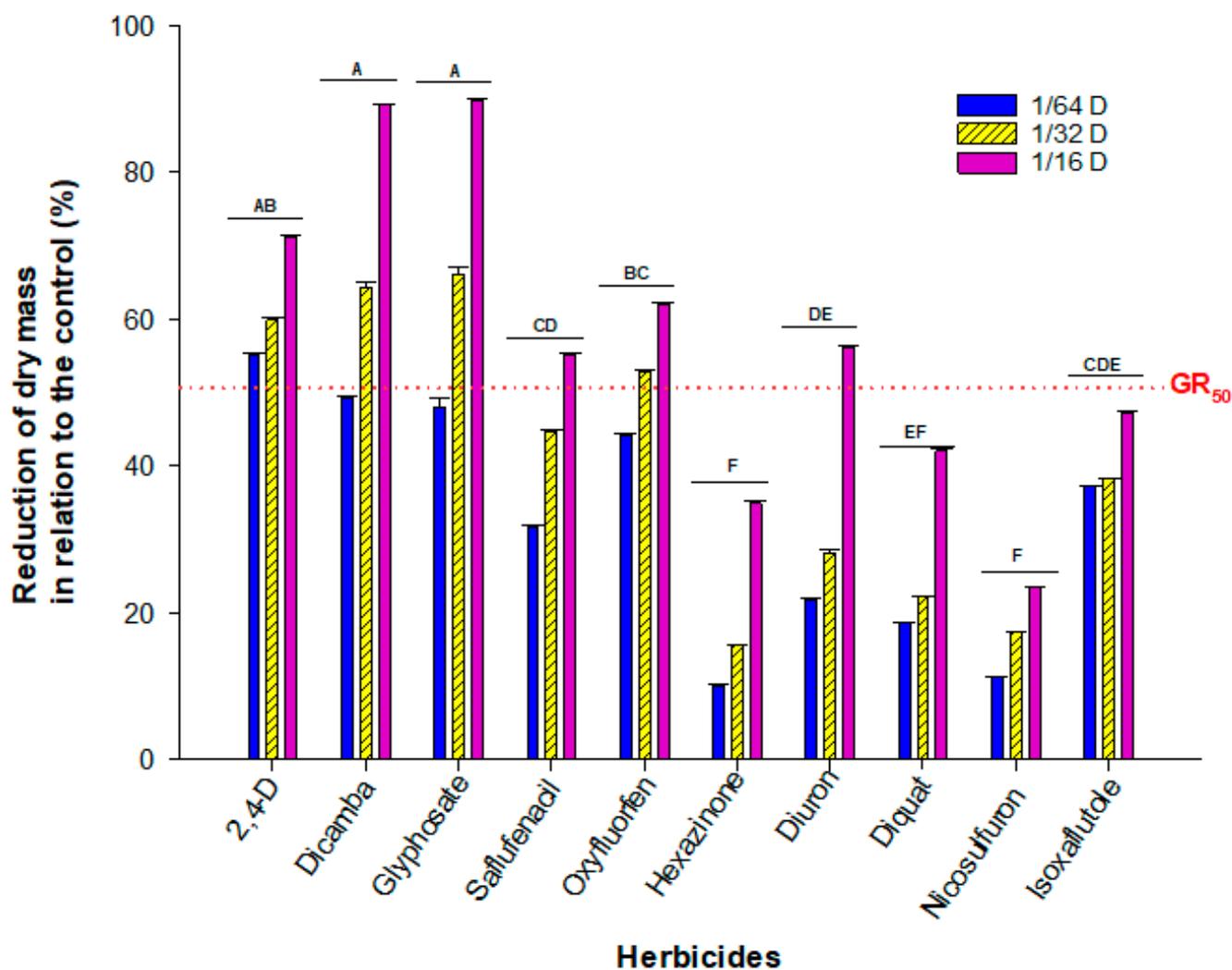
There was an interaction observed between the degree of tomato injury and the dose ratios of the same herbicide to the control at 21 DAA for the herbicides dicamba, glyphosate, saflufenacil, oxyfluorfen, diuron, and diquat. On the other hand, 2,4-D, hexazinone, nicosulfuron, and isoxaflutole showed no significant interaction or difference between the dose ratios (Figure 20).



**Figure 20.** Injury level at 21 days after application (DAA) in relation to the control treatment, under different herbicides subdoses in the simulated drift of 2,4-D (146.56, 73.28, and 36.64 g a.e. ha<sup>-1</sup>), dicamba (45, 22.5, and 11.25 g a.e. ha<sup>-1</sup>), glyphosate (138.75, 69.38, and 34.69 g a.e. ha<sup>-1</sup>), saflufenacil (8.75, 4.38, and 2.19 g a.i. ha<sup>-1</sup>), oxyfluorfen (90, 45, and 22.5 g a.i. ha<sup>-1</sup>), hexazinone (24.75, 12.38, and 6.19 g a.i. ha<sup>-1</sup>), diuron (200, 100, and 50 g a.i. ha<sup>-1</sup>), diquat (43.75, 21.88, and 10.94 g a.i. ha<sup>-1</sup>), nicosulfuron (3.75, 1.88, and 0.94 g a.i. ha<sup>-1</sup>) and isoxaflutole (16.41, 8.20, and 4.10 g a.i. ha<sup>-1</sup>). The subdoses correspond to the proportion of 1/16D, 1/32D, and 1/64D of the commercial dose, respectively. The simulated drift doses with equal lowercase letters between herbicides and equal uppercase letters between the simulated drift dose proportions do not differ among themselves by Tukey’s test ( $p < 0.05$ ). The IC<sub>50</sub> represents the dose that causes 50% injury.

At 21 DAA, there was no difference in injury level with increasing dose ratios in simulated 2,4-D drift, indicating a consistent injury level of approximately 70% regardless of subdose (Figure 20). However, with the dicamba herbicide, the injury level increased as the dose proportions applied increased, reaching approximately 95% control in the

simulated drift of 1/16D, suggesting greater tomato sensitivity to this herbicide (Figure 20). This result is evident when comparing the DM reduction with the control in the largest simulated 1/16D drift of 2,4-D and dicamba, which presenting values of ~70% and ~90%, respectively (Figure 21). Based on the applied subdoses of 2,4-D and dicamba, a safe exposure level for tomatoes could not be established. The injury levels caused by the lowest dose of 1/64 of both herbicides were >60%, which severely compromised crop development and could result in significant reductions in productivity. This impairment is evident since, at this dose, the DM reduction was ~50% (Figure 21).



**Figure 21.** Dry mass (DM) reduction (%) at 21 days after application (DAA) in relation to the control treatment, under different herbicide subdoses in the simulated drift of 2,4-D (146.56, 73.28, and 36.64 g a.e. ha<sup>-1</sup>), dicamba (45, 22.5, and 11.25 g a.e. ha<sup>-1</sup>), glyphosate (138.75, 69.38, and 34.69 g a.e. ha<sup>-1</sup>), saflufenacil (8.75, 4.38, and 2.19 g a.i. ha<sup>-1</sup>), oxyfluorfen (90, 45, and 22.5 g a.i. ha<sup>-1</sup>), hexazinone (24.75, 12.38, and 6.19 g a.i. ha<sup>-1</sup>), diuron (200, 100, and 50 g a.i. ha<sup>-1</sup>), diquat (43.75, 21.88, and 10.94 g a.i. ha<sup>-1</sup>), nicosulfuron (3.75, 1.88, and 0.94 g a.i. ha<sup>-1</sup>) and isoxaflutole (16.41, 8.20, and 4.10 g a.i. ha<sup>-1</sup>). The subdoses correspond to the proportion of 1/16D, 1/32D, and 1/64D of the commercial dose, respectively. The simulated drift doses with equal capital letters do not differ from each other by Tukey’s test ( $p < 0.05$ ). The GR<sub>50</sub> represents the dose that causes a 50% DM reduction.

There was an interaction observed between the level of tomato injury and glyphosate dose ratios, wherein there was a progressive increase in injury levels as the applied doses ratios increased, reaching ~90% injury at the 1/ 16D dose ratio, indicating a high sensitivity

of tomato plants to this herbicide (Figure 20). This finding is supported by Figure 21, which shows an ~90% reduction in DM at the 1/16D dose ratio. However, when evaluating the dose ratios, a significant reduction in both injury levels and DM reduction was observed. At the 1/32 dose ratio, injury levels were ~55% with a DM reduction of ~65%, and at the 1/64 dose ratio, injury levels were ~20% with a DM reduction of ~50% (Figures 20 and 21).

Saflufenacil showed a difference in the injury level observed at 21 DAA among the three-dose ratios. The injury level increased as the applied subdoses increased, although all values remained below 50%, and the reduction in DM was less than 60% (Figures 20 and 21). In contrast, oxyfluorfen did not show a difference in injury level between the subdoses of 1/32D and 1/64D, but there was a difference between these subdoses and simulated drift of 1/16D. At the 1/16 dose ratio, the injury level and DM reduction was ~60% (Figures 20 and 21). Thus, oxyfluorfen caused more damage than saflufenacil. This finding is supported by Figure 21, which shows that all oxyfluorfen applied dose ratios result in greater DM reduction compared to all saflufenacil dose ratios.

There was no difference in the injury level among the applied dose ratios of hexazinone at 21 DAA. The obtained values were <10% (Figure 20). However, diuron showed a difference in the injury level in the highest dose proportion of 1/16D, reaching values of ~25%. In the other subdoses, there was no difference in the injury level, presenting values < 10% (Figure 20). The highest simulated drift of 1/16D showed that diuron values were higher than hexazinone, indicating greater tomato sensitivity to this herbicide. This result is evident when comparing the DM reduction to the control, with hexazinone and diuron presenting values <40% and ~60%, respectively (Figure 21). For diquat, there was no difference in the injury level observed at 21 DAA in the dose ratios of 1/32D and 1/64D, which were close to 10% (Figure 20). However, when comparing these subdoses with the simulated 1/16 drift, there was a difference in the injury level, with a value > 20% (Figure 20). This difference in the injury level was also observed in the DM reduction, where the dose of 1/16D presented a value greater than 50%, while in the other subdoses, values were proximally to 25% (Figure 21).

Nicosulfuron did not cause any injury level, regardless of the applied dose ratio. However, all dose ratios of nicosulfuron caused a reduction in DM. The DM reduction values observed in the dose ratios of 1/16D, 1/32D, and 1/64D were >20%, ~20%, and ~10%, respectively (Figure 21). Although there was a reduction in DM, injury levels and DM reduction did not significantly affect the crop development.

The injury level of isoxaflutole did not differ among the three dose ratios, all of which were <20% (Figure 20). However, there was a reduction in DM in all applied dose ratios, with a reduction ~50% in the simulated drift of 1/16, and reductions of ~20% in the subdoses of 1/32D and 1/64D (Figure 21).

At 21 DAA, none of the applied herbicides at simulated drift doses caused 100% injury level to tomato plants. On the other hand, the herbicides that caused the most damage to tomato plants at the highest dose ratio of 1/16 were 2,4-D, dicamba, glyphosate, and oxyfluorfen, showing values higher than  $IC_{50}$  (herbicide dose that provides 50% control). Dicamba and glyphosate caused injury levels proximally to 90% of control in the simulated drift of 1/16 (Figure 20). However, saflufenacil, hexazinone, diuron, nicosulfuron, diquat, and isoxaflutole showed values lower than  $IC_{50}$  and caused less damage. Nicosulfuron did not cause any injury at any dose ratio, indicating that this herbicide did not affect tomato plants.

At 21 DAA, none of the applied herbicides at any dose ratio resulted in a 100% reduction in DM. However, dicamba and glyphosate had DM reduction values close to 90% (Figure 21).

The herbicides that caused the greatest reduction in tomato DM at the highest dose ratio of 1/16 were 2,4-D, dicamba, glyphosate, saflufenacil, oxyfluorfen, and diuron, with values exceeding the  $GR_{50}$  (herbicide dose that provides 50% reduction in DM) (Figure 21). Therefore, hexazinone, nicosulfuron, diquat, and isoxaflutole caused less damage with values below the  $GR_{50}$ . Among these herbicides, the synthetic auxin herbicides (2,4-D and

dicamba) and the EPSPs-inhibiting herbicide (glyphosate) caused the greatest reduction in DM.

#### 4. Discussion

Tomatoes are susceptible to herbicide drift. This susceptibility is superior for particularly to certain modes of action such as auxinic herbicides [4]. According to Xu et al. [17], synthetic auxin herbicides cause similar symptoms characterized by changes in plant growth, even in small amounts. The effects of these herbicides are divided into three stages in the plant: stimulation of abnormal growth and gene expression, inhibition of growth and physiological responses, and senescence and cell death [7,18]. The most common plant symptoms under the effects of these herbicides are epinasty, stem and branch curvature, and growth arrest with meristem chlorosis, followed by necrosis [19,20]. Our results obtained in this study are consistent with those found by Meyers et al. [21], where dicamba drift negatively affected tomato plants and compromised fruit productivity. In addition, Warmund et al. [4] reported that tomato cultivars treated with herbicide 2,4-D and dicamba can produce less total and marketable yield than their respective non-treated control. For most cultivars studied, dicamba-treated plants had less marketable yield than 2,4-D-treated plants in the greenhouse.

Glyphosate is another herbicide that can cause significant damage to tomato crops, even at low doses. According to Duke [22], glyphosate is a systemic herbicide that has a total action and is moderately absorbed by the cuticle, requiring approximately 6 h without rain after application. Symptoms of glyphosate drift on tomato plants include meristem yellowing and necrosis, and plant death may occur 7–30 days after application. However, with the advent of transgenic plants, glyphosate is no longer used in a non-selective way and has become an option for selective weed control in these crops [19]. Nevertheless, drift of this herbicide can still significantly reduce tomato crop yields, as observed in the study conducted by McNaughton et al. [23], which reported a reduction of almost 90% in tomato plant yields due to glyphosate drift.

PPO inhibitors are globally significant herbicides. They can penetrate young plants through their roots, stems, and leaves, and their activity relies on light availability [24]. These herbicides cause necrosis in leaves, which appear as droplets from the spray due to their limited movement within the plant [24]. At least one herbicide belonging to this mode of action is registered for use in all crops with large cultivated areas in Brazil, underscoring the significant importance of these herbicides in Brazilian agriculture [25]. The symptoms caused by these herbicides are characterized by chlorosis (browning) between the veins and at the leaf margins, which progresses from the margins towards the center of the leaf, leading to generalized necrosis [26]. The accumulated chlorosis is primarily observed at the leaf margins due to the herbicide's translocation via the xylem [27]. In addition to drift, it is essential to exercise caution with PPO inhibitor residues in the soil because these herbicides have residual activity and can cause damage to tomato plants even without direct spraying [28,29].

Symptoms from PSI-inhibiting herbicides develop within a few hours of treatment, with affected leaves appearing wet due to the degradation of the plasma membrane. Within two days, the affected areas become necrotic [30]. Unlike other herbicides, diquat does not have a residual effect on the soil and is a non-selective herbicide, allowing for its use primarily in crop desiccation and pre-harvest for grains [11]. It is important to emphasize that herbicide drift resulting from spraying in the late afternoon or at night may have an enhanced effect due to greater absorption of the herbicide in the absence of light [31].

ALS-inhibiting herbicides are active in the soil and are non-volatile due to their low vapor pressure. The death of treated plants can occur within two weeks, and these herbicides are effective against both narrow and broadleaf [32]. According to Zhou et al. [33], growth stops 7–10 days after herbicide application, and interveinal chlorosis develops or becomes purple in susceptible plants. The application of nicosulfuron alone did not result in any observed injury. However, it is worth noting that tank-mixing with other herbicides

can result in reduced tomato yields. For example, Boyd and Dittmar [34] reported reduced yields when nicosulfuron was tank-mixed with rimsulfuron.

Herbicides that inhibit carotenoid biosynthesis (HPPD) do not affect old leaves that were formed before application, as they do not affect pre-existing carotenoids, symptoms are observed only on young leaves [35]. The most visible symptoms of these herbicides are completely white foliage followed by necrosis. The tomato crop is very sensitive to isoxaflutole and can suffer more than 50% injury in the presence of subdoses starting at 15 g a.i. ha<sup>-1</sup> [36]. It is important to note that seed treatment with safeners is currently available to induce resistance to carotenoid-inhibiting herbicides, and it is an important tool for the safe use of these herbicides [37].

## 5. Conclusions

Among all the herbicides studied, auxinic (2,4-D and dicamba), glyphosate, and oxyfluorfen posed the highest risk to young tomato plants, with dicamba and glyphosate causing more damage at the highest simulated drift of 1/16. In contrast, saflufenacil, hexazinone, diuron, nicosulfuron, diquat, and isoxaflutole were found to cause less damage, with no direct impact on the initial vegetative phase of the crop.

Exposure of tomato crops to herbicide drift is a cause for concern as even lower doses of simulated drift can pose risks to the crop, which may directly impact productivity. The symptomatology data combined with a quantitative analysis of the injury levels and biomass accumulation in this study can provide valuable knowledge to producers. This can help them identify problems caused by simulated drift, as well as to assess the extent of impairment to their crop. This is an important parameter in property planning and helps to identify the herbicide responsible for the symptoms observed.

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