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# Comparative Study of Photosynthesis Performance of Herbicide-Treated Young Triticale Plants during Drought and Waterlogging Stress

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**Abstract:** Owing to global climate changes, periods of soil drought or waterlogging occur. Each of these factors causes negative effects on plant physiological processes and growth. Weeds are another factor that limits plant productivity. The main task of this study is to investigate the physiological reactions of triticale to herbicide treatment and subsequent drought or waterlogging. Young triticale plants were treated with Serrate® (selective herbicide produced by Syngenta) and exposed for 7 days to drought or waterlogging. Plant growth, chlorophyll and carotenoids content, the net photosynthesis rate and chlorophyll *a* fluorescence were measured during the stress period and after 4 days of plant recovery. Herbicide by itself did not induce considerable changes in the abovementioned parameters during the stress period. Serrate® did not affect strongly the efficiency of the photosynthetic machinery under harsh conditions. A significant reduction in fresh weight (85%), water content (93%), net photosynthesis rate, chlorophyll *a* fluorescence indices $F_v/F_m$ and $F_v/F_0$, and leaf pigments (58% for chlorophyll *a*, 53% for chlorophyll *b*, and 45% for carotenoids) was found because of drought. Waterlogging also influenced negatively these parameters but to a smaller extent. After resuming the normal irrigation, the photosynthesis and chlorophyll *a* fluorescence tended to increase and showed signs of recovery. The comparative analysis of growth and photosynthetic parameters demonstrated that triticale plants subjected to waterlogging could recover to a higher degree than those exposed to drought.

**Keywords:** herbicide; chlorophyll *a* fluorescence; waterlogging; *Triticosecale*; drought; gas exchange parameters



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## 1. Introduction

Crops grown in field conditions are often exposed to variety of unfavorable factors that disrupt plant physiological processes and limit growth and yield. Moreover, under the natural environment, not only individuals but also multiple environmental factors of biotic or abiotic origin can influence the plant's metabolism.

In the extensive agriculture, the use of herbicides is still an essential strategy for chemical weed control [1]. The preparation Serrate®, developed by Syngenta (Bazel, Switzerland), is a selective herbicide suitable for the effective control of annual grassy and broad-leaf weeds in the field areas sown with wheat, rye, and triticale. Its effectiveness results from its specific double-component formulation: Serrate consists of clodinafop-propargyl, an inhibitor of acetyl co-enzyme A carboxylase (enzyme of the fatty acids biosynthetic pathway), and pyroxsulam, which inhibits acetolactate synthase (a key enzyme of the branched-chain amino acids biosynthesis pathway) [2]. According to the producer's recommendations, Serrate® should be applied on healthy cereals, which have not faced preliminary environmental unfavorable issues. However, occasionally, stress threats can occur later after the herbicide application. Both water excess and water deficit are abiotic stress factors related to global climatic changes. They can disorder normal plant physiology and disturb vital metabolic processes [3,4]. An imbalance in water supply, deficit or excess, causes similar and fast physiological plant responses such as significant decline in the photosynthesis, resulting in biomass decrease, decreased crop yield and even plant death [5,6].

Recently, we assessed the alterations in photosynthesis-related traits of wheat plants preliminary treated with Serrate® and then exposed to water deficit or water excess [7]. We found that the herbicide applied alone did not alter significantly the photosynthesis-related parameters. On the other hand, drought and waterlogging provoked considerable changes in the physiological traits when applied individually or after herbicide treatment. Wheat plants successfully recovered the photosynthetic processes after cessation of the drought. Meanwhile, water excess irreparably impaired the photosynthesis, and the negative consequences on this key physiological process continued even after termination of the stress program. Here, we expand our research on triticale plants. Triticale (×*Triticosecale* Wittmack) is an intergeneric hybrid crop based on hybridization and experimental polyploidy, combining in one plant the genomes of wheat and rye. Triticale is believed to be more stress tolerant than wheat because of its rye ancestry [8–10].

In this study, we aimed to compare the physiological responses of seedlings pretreated with Serrate® triticale and then subjected to drought or waterlogging and to investigate how plants are capable of recovering photosynthesis after cessation of the stress. Our study provides a new information regarding the physiological reactions of triticale to herbicide treatment and subsequent drought or waterlogging by evaluation of the gas exchange parameters, the fluorescence of chlorophyll *a* and the content of leaf pigments.

## 2. Materials and Methods

### *2.1. Plant Material and Treatments*

A pot experiment was carried out under controlled growth conditions according to our earlier model scheme [7]. A leached meadow cinnamon soil (pH 6.2) and sand mixture with a ratio of 3:1 was used as a substrate. The parameters of the growth chamber were: 16/8 h day/night photoperiod, 22/19 °C (day/night temperature) (200 µmol $m^{-2}$ $s^{-1}$ photon flux density was provided by fluorescent lamps), and 60% relative air humidity. Seeds of triticale (×*Triticosecale Wittm.*) cv. Rozhen were obtained from the Institute of Plant Genetic Resources (Sadovo, Bulgaria). This cultivar is officially certified in the Republic of Bulgaria. The pots were divided into 6 groups. Each treatment group consisted of 6 pots, and each pot contained 20 plants. The herbicide Serrate® was applied by spraying on 17-day-old seedlings in a dose of 1 mg $mL^{-1}$ according to the instructions provided by Syngenta. The stresses were initiated 72 h after the treatment with herbicide and were realized for 7 days by withholding of water supply (for drought stress) or by keeping the pots in a bigger container filled with water, which was maintained 2 cm over the soil level. After the end of the stress programs, plants were left for recovery under the normal irrigation regime. The measurements were carried out on days 4 and 7 of the stress duration and on day 4 of recovery.

### *2.2. Biometric Parameters*

Immediately after harvesting the above-ground part of plants, the fresh weight (FW) was measured on a Precision Standard electronic balance (OHAUS®, Parsippany, NJ, USA), and the length was recorded using a ruler. The leaf material was heated in an oven at 80 °C until a constant dry weight (DW) was obtained. Leaf water content (WC) was calculated as follows: WC = (FW − DW)/DW [11].

### *2.3. Leaf Pigment Content*

Leaf pigments content was measured according to Arnon [12]. Approximately 50 mg leaf material was ground in 5 mL of 80% acetone, and the extracts were centrifuged for

5 min at 3000× *g* in a refrigerated centrifuge (Sigma 2–16 K, SciQuip, Wem, UK). The supernatants' absorbance was measured at 663, 645, and 460 nm on a spectrophotometer (Multiskan Spectrum, Thermo Electron Corporation, Vantaa, Finland).

### 2.4. Chlorophyll *a* Fluorescence

The fluorescence of chlorophyll *a* was measured on a Multi-Function Plant Efficiency Analyzer—Hendy PEA fluorimeter (Hansatech Instruments Ltd., Norfolk, UK). The device contains 3 red LEDs, which provide a peak wavelength of 650 nm and photon flux density of 3500 μmol $m^{-2}$ $s^{-1}$. The plants were initially dark adapted for 30 min, and then the chlorophyll *a* fluorescence was measured. A 4 mm diameter area of a fully developed leaf was illuminated by red light, and the quick chlorophyll *a* fluorescence was determined. The following parameters were recorded: $F_0$ (the minimal fluorescence when all reaction centers are open); $F_m$ (the maximal fluorescence when all reaction centers are closed), and $F_v$ (the variable fluorescence). These values were used for the mathematical expressions of the JIP test [13] of selected parameters specific for the light phase of the photosynthesis such as: $F_0/F_m$ (the quantum yield of the energy dissipation as heat); $F_v/F_0$ (the primary photochemical reactions efficiency); $F_v/F_m$ (the PSII maximal quantum yield, i.e., $\varphi P_0$); $\varphi E_0$ (the electron transport quantum yield between PSII and PSI); $\varphi R_0$ (the electron transport quantum yield from PQ to PSI terminal electron acceptors); $\psi E_0$ (exciton transfer efficiency to electron transport chain); $\delta R_0$ (probability that an electron is transported from reduced PQ to the electron acceptor side of PSI); $PI_{abs}$ (the performance index calculated on energy absorption basis); and $PI_{total}$ (the productivity of the photosynthetic apparatus, including PS II, PS I and the electron transport chain between them).

### 2.5. Gas Exchange Parameters

The leaf gas exchange parameters—net rate of photosynthesis ($A_n$, μmol $CO_2$ $m^{-2}$ $s^{-1}$), stomatal conductance ($g_s$, mmol $m^{-2}$ $s^{-1}$), and transpiration rate (E, mmol $H_2O$ $m^{-2}$ $s^{-1}$)—were recorded by an infrared gas analyzer system Li6800 (LI-COR Biosciences Inc., Lincoln, NE, USA) equipped with light sourced chamber (LI6800-02). Here, 10 L buffer was used to counterpoise the fluctuations of $CO_2$ and $H_2O$ in the air [14]. Two fully expanded leaves per plant were used to read leaf gas exchange parameters. The leaves were preliminary adapted to the surrounding environment. Records were completed between 11:00 h and 14:00 h under controlled conditions: temperature 25 °C; relative air humidity at 45 to 55%; air flow rate 200 μmol $s^{-1}$; actinic PAR 200 μmol $m^{-2}$ $s^{-1}$ photon flux density. Water use efficiency (WUE, μmol $CO_2$ $mmol^{-1}$ $H_2O$) was calculated as follows: WUE = $A_n/E$.

### 2.6. Recovery and Resilience Indices of Selected Parameters

The recovery and resilience indices were calculated using the following equations:

$$\text{Recovery} = (X_{4dR} - X_{7dS})/X_{7dS} \times 100 \tag{1}$$

$$\text{Resilience} = (X_{4dR} - X_c)/X_c \times 100 \tag{2}$$

where $X_{7dS}$ and $X_{4dR}$ are the values of the selected functional parameter measured after 7 days of drought or waterlogging and after 4 days of recovery; and $X_c$ is the control value of the functional parameter after 4 days of recovery [7].

### 2.7. Statistical Analysis

Two independent experiments were conducted, and each analysis consisted of 10 replicates. The data were statistically analyzed using one-way ANOVA. Duncan's multiple-range test ($p < 0.05$) was applied to evaluate the significance of the differences between the treatments in each sampling point. All data presented are average values ± standard error (SE).

## 3. Results

### 3.1. Biometric Parameters

The growth parameters' alterations of triticale caused by drought, waterlogging and herbicide application are presented in Figure 1. The herbicide treatment did not cause substantial variations in triticale length and fresh weight. In contrast, it provoked an accumulation of dry biomass, which was increased by 23% after 4 days of recovery. This reflected in lowered water content by 12% compared with the control.

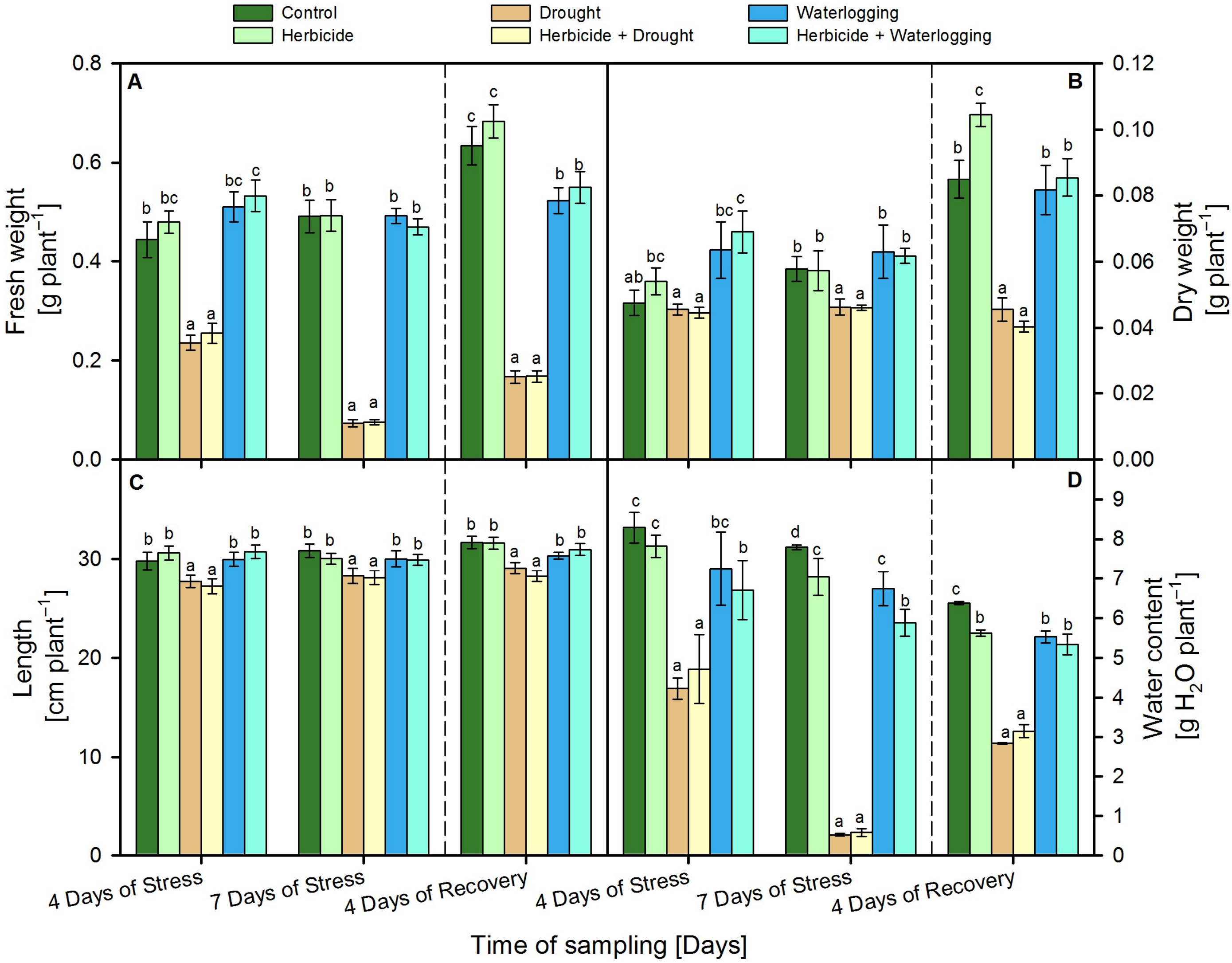


**Figure 1.** Growth parameters of triticale plants treated with Serrate and exposed to drought or waterlogging stress: (**A**) Fresh weight; (**B**) Dry weight; (**C**) Length; (**D**) Water content. Data are average values ± SE. The small letters represent significant differences between the treatments in the same group at $p < 0.05$.

The alterations caused by drought and herbicide + drought were comparable and depended on drought severity only. Significant inhibition was detected in fresh biomass (by 47%) and water content (by 49%) on the 4th day of stress. The negative stress effects intensified, and a decrease in fresh weight by 85% and water content by 93% was recorded after 7 days of drought. The plant length and dry biomass accumulation were arrested during the time course of the experiment. After resuming the normal water supply, the plants exposed to drought restarted their growth. The fresh weight was increased up to 26%, and the water content was raised up to 49% of the respective controls. No significant alterations in growth parameters were detected between triticale plants subjected to waterlogging or herbicide + waterlogging treatment. The fresh biomass (Figure 1A), the dry biomass (Figure 1B), and the length (Figure 1C) were nearly at the control levels during the whole experimental period. The water content (Figure 1D) decreased constantly even during the recovery period.

### *3.2. Recovery and Resilience Indices of Biometric Parameters*

The recovery index is expressed as the ratio of the parameters measured during stress and after recovery. The resilience index is represented by the ratio of the control and treatments during recovery.

Regarding FW and WC, the highest degree of recovery was found after drought and herbicide + drought treatments (Table 1). However, the resilience index of all parameters was negative. Unlike for drought, the recovery and resilience indices were not greatly influenced in plants exposed to either waterlogging or herbicide + waterlogging stress, indicating that these plants were capable of sustaining successfully the water excess. The resilience and recovery indices after herbicide individual application revealed that the FW and DW of triticale were not negatively influenced, and the plants even had vigorous fitness.

**Table 1.** Recovery and resilience indices of the biometric parameters after treatment with Serrate, drought and waterlogging (%).

| Trait | Treatment | Recovery | Resilience |
|---|---|---|---|
| FW | Herbicide | 39 ↑ | 8 ↔ |
| | Drought | 129 ↑↑↑↑ | −74 ↓↓↓ |
| | Herbicide + Drought | 123 ↑↑↑↑ | −74 ↓↓↓ |
| | Waterlogging | 6 ↔ | −18 ↓ |
| | Herbicide + Waterlogging | 17 ↑ | −13 ↓ |
| DW | Herbicide | 82 ↑↑↑ | 23 ↑ |
| | Drought | −2 ↔ | −46 ↓↓ |
| | Herbicide + Drought | −12 ↓ | −53 ↓↓ |
| | Waterlogging | 30 ↑ | −4 ↔ |
| | Herbicide + Waterlogging | 38 ↑ | 1 ↔ |
| WC | Herbicide | −20 ↓ | −12 ↓ |
| | Drought | 443 ↑↑↑↑ | −55 ↓↓ |
| | Herbicide + Drought | 446 ↑↑↑↑ | −51 ↓↓ |
| | Waterlogging | −18 ↓ | −13 ↓ |
| | Herbicide + Waterlogging | −9 ↔ | −16 ↓ |

Notes: FW—fresh weight; DW—dry weight; WC—water content. Average data of the traits were used to calculate the trends. Symbols: ↔—±10%; ↑↓—±11–40%; ↑↑↓↓—±41–70%; ↑↑↑↓↓↓—±71–100%; ↑↑↑↑↓↓↓↓—±>100%.

### *3.3. Leaf Pigment Content*

The leaf pigment content (Figure 2) was significantly decreased after all treatments except in the plants treated with Serrate on the 7th day of stress. The most significant decreases were detected in drought-treated triticale: 58% for chlorophyll *a*, 53% for chlorophyll *b*, and 45% for carotenoids.

Regarding the leaf pigment content, the gap between treated and control plants diminished after 4 days of recovery. The chlorophyll *a*/*b* ratio remained almost stable during the whole experimental period, although some fluctuations were detected.

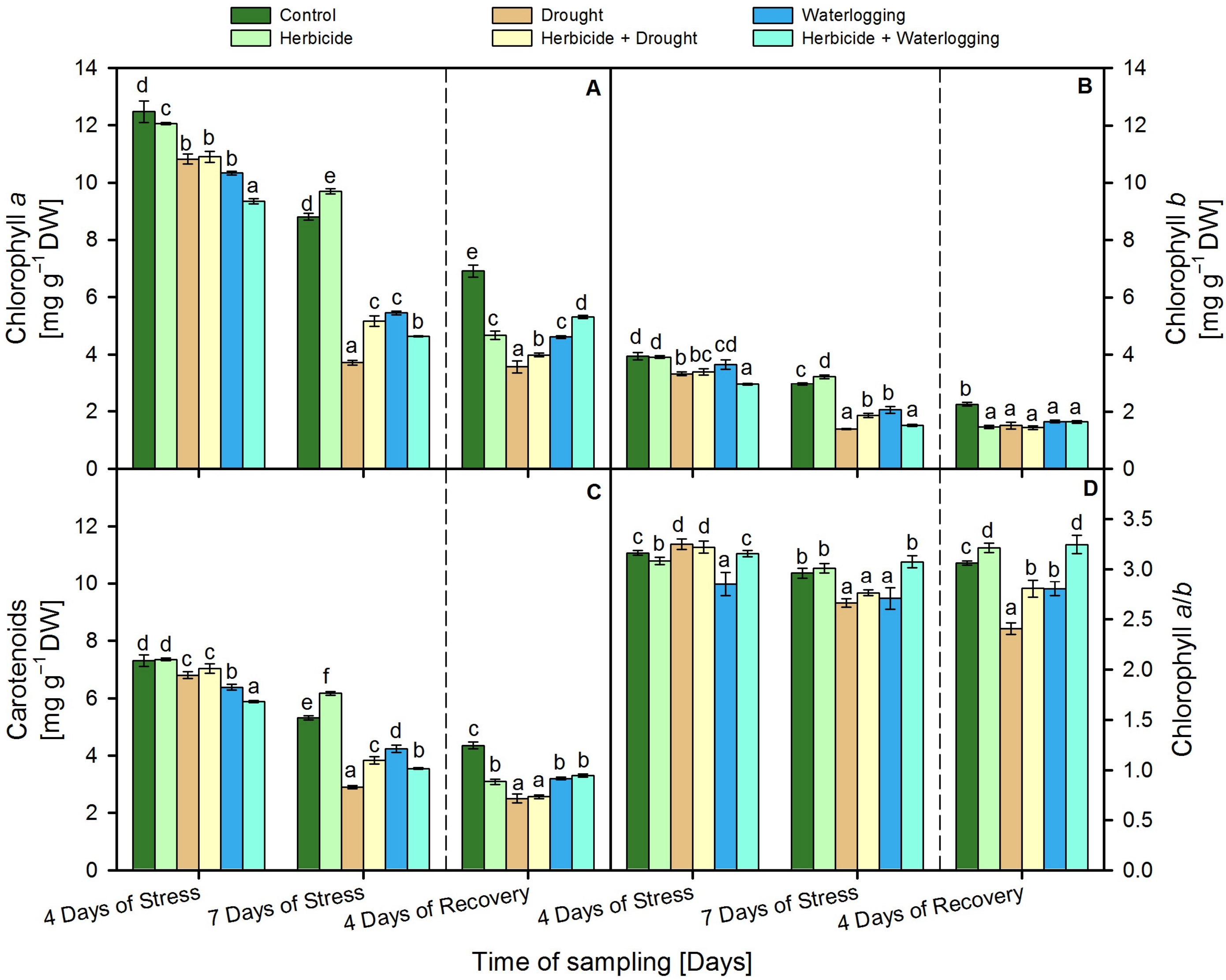


**Figure 2.** Leaf pigment content in triticale plants treated with Serrate and exposed to drought or waterlogging stress: (**A**) Chlorophyll *a*; (**B**) Chlorophyll *b*; (**C**) Carotenoids; (**D**) Chlorophyll *a*/*b*. Data are average values ± SE. The small letters represent significant differences between the treatments in the same group at $p < 0.05$.

### *3.4. Leaf Gas Exchange Parameters*

Serrate did not cause considerable changes in leaf gas exchange parameters, except for a 57% increase in the transpiration rate E (Figure 3B) and an 84% increase in the stomatal conductance $g_s$ (Figure 3C) during the recovery period.

A significant decline was detected in the transpiration, stomatal conductance and photosynthesis rate in plants exposed to drought and herbicide + drought; i.e., the photosynthesis was inhibited. Water use efficiency (WUE) was not calculated for these variants because of the negative values of $A_n$ (the respiration is higher than the photosynthesis) and too low values of the transpiration rate.

Waterlogging also provoked a time-dependent decline in the gas exchange parameters, but the alterations were not so strong as compared to drought. In relation to these parameters, no statistical differences were found between waterlogging and herbicide + waterlogging treatments. A substantial increase was detected in the photosynthesis-related parameters in both drought- and waterlogging-stressed plants after 4 days of recovery.

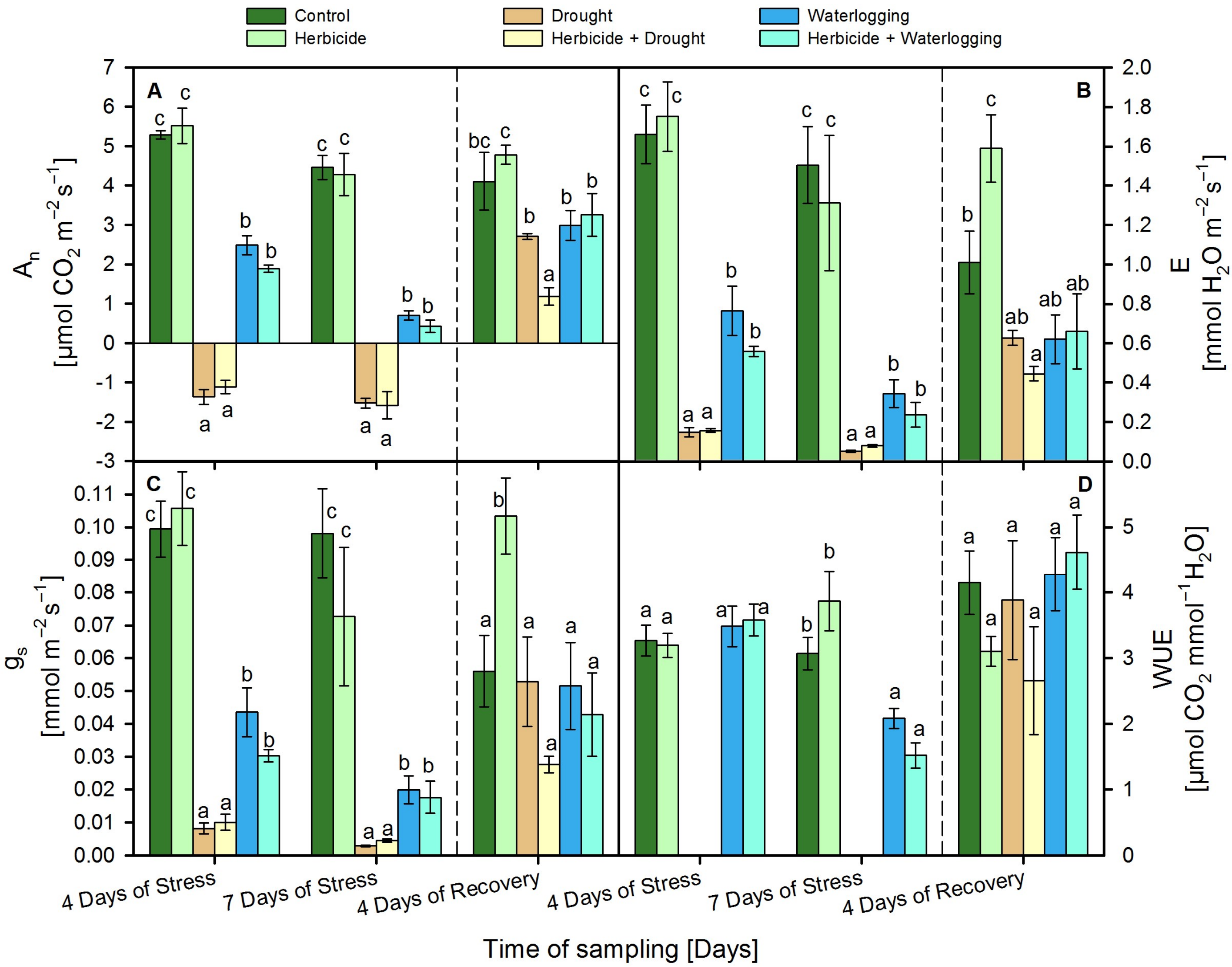


**Figure 3.** Leaf gas exchange parameters in triticale plants treated with Serrate and exposed to drought or waterlogging stress: (**A**) Net photosynthesis rate; (**B**) Transpiration rate; (**C**) Stomatal conductance; (**D**) Water use efficiency. Data are average values ± SE. The small letters represent significant differences between the treatments in the same group at $p < 0.05$.

### *3.5. Indices of Recovery and Resilience of Photosynthesis*

A positive recovery index of photosynthetic parameters was found in waterlogged and herbicide + waterlogged plants (Table 2). The resilience index had minimal negative values in plants subjected to water excess.

**Table 2.** Recovery and resilience indices of some photosynthesis parameters after treatment with Serrate, drought and waterlogging (%).

| Trait | Treatment | Recovery | Resilience |
| --- | --- | --- | --- |
| $A_n$ | Herbicide | 12 | 16 |
| | Drought | −277 | −34 |
| | Herbicide + Drought | −175 | −71 |
| | Waterlogging | 322 | −27 |
| | Herbicide + Waterlogging | 649 | −21 |

**Table 2.** *Cont.*

| Trait | Treatment | Recovery | Resilience |
|---|---|---|---|
| E | Herbicide | 21 ▲ | 57 ▲▲ |
| | Drought | 1133 ▲▲▲▲ | −38 ▼ |
| | Herbicide + Drought | 467 ▲▲▲▲ | −56 ▼▼ |
| | Waterlogging | 81 ▲▲▲ | −39 ▼ |
| | Herbicide + Waterlogging | 180 ▲▲▲▲ | −35 ▼ |
| $G_s$ | Herbicide | 42 ▲▲ | 84 ▲▲▲ |
| | Drought | 1705 ▲▲▲▲ | −6 ◀▶ |
| | Herbicide + Drought | 510 ▲▲▲▲ | −51 ▼▼ |
| | Waterlogging | 159 ▲▲▲▲ | −8 ◀▶ |
| | Herbicide + Waterlogging | 144 ▲▲▲▲ | −23 ▼ |

Notes: FW—fresh weight; DW—dry weight; WC—water content. Average data of the traits were used to calculate the trends. Symbols: ◀▶—±10%; ▲▼—±11–40%; ▲▲▼▼—±41–70%; ▲▲▲▼▼▼—±71–100%; ▲▲▲▲▼▼▼▼—±>100%.

Although the recovery indices of stomatal conductance and transpiration had their highest values in triticale exposed to drought or herbicide + drought stress, the recovery index of the net photosynthesis rate was negative. The resilience indices of plants subjected to water deficit maintained negative values.

The recovery and resilience indices for all photosynthesis-related traits had positive values in plants treated with herbicide only.

### *3.6. Parameters of Chlorophyll a Fluorescence*

The changes in the chlorophyll *a* fluorescence parameters are presented in Figure 4. $F_0$ (minimal fluorescence, Figure 4A), $F_m$ (maximal fluorescence, Figure 4C), and $F_v$ (variable fluorescence, Figure 4E) were substantially decreased by the drought, and this depended on the stress duration and severity. These parameters were not altered significantly by waterlogging. After 4 days of recovery, there were no statistically differences of these parameters, except for the $F_0$ (Figure 4A) of both waterlogged triticale where these values were higher than the control. After 4 days of stress, $F_v/F_0$ (Figure 4B) and $F_v/F_m$ (Figure 4D) ratios were slightly varied due to all treatments; then, a substantial decline was found due to drought. After 4 days of recovery, $F_v/F_0$ and $F_v/F_m$ maintained control levels with the exception of herbicide + waterlogging treatment, where they were lower. The herbicide by itself did not significantly affect the measured parameters.

The $PI_{abs}$ index (Figure 4F), which represents the fluorescence performance on the absorption base and relates to the general vitality of PSII, was decreased only by waterlogging during the first 4 days of stress. After 7 days of stress, the decrease was most obvious in the drought-treated triticale plants. After resuming the irrigation, no significant statistical variances were detected between control and stress-treated plants. After 4 days of recovery, the $PI_{abs}$ index was considerably higher in Serrate-treated plants compared to the control.

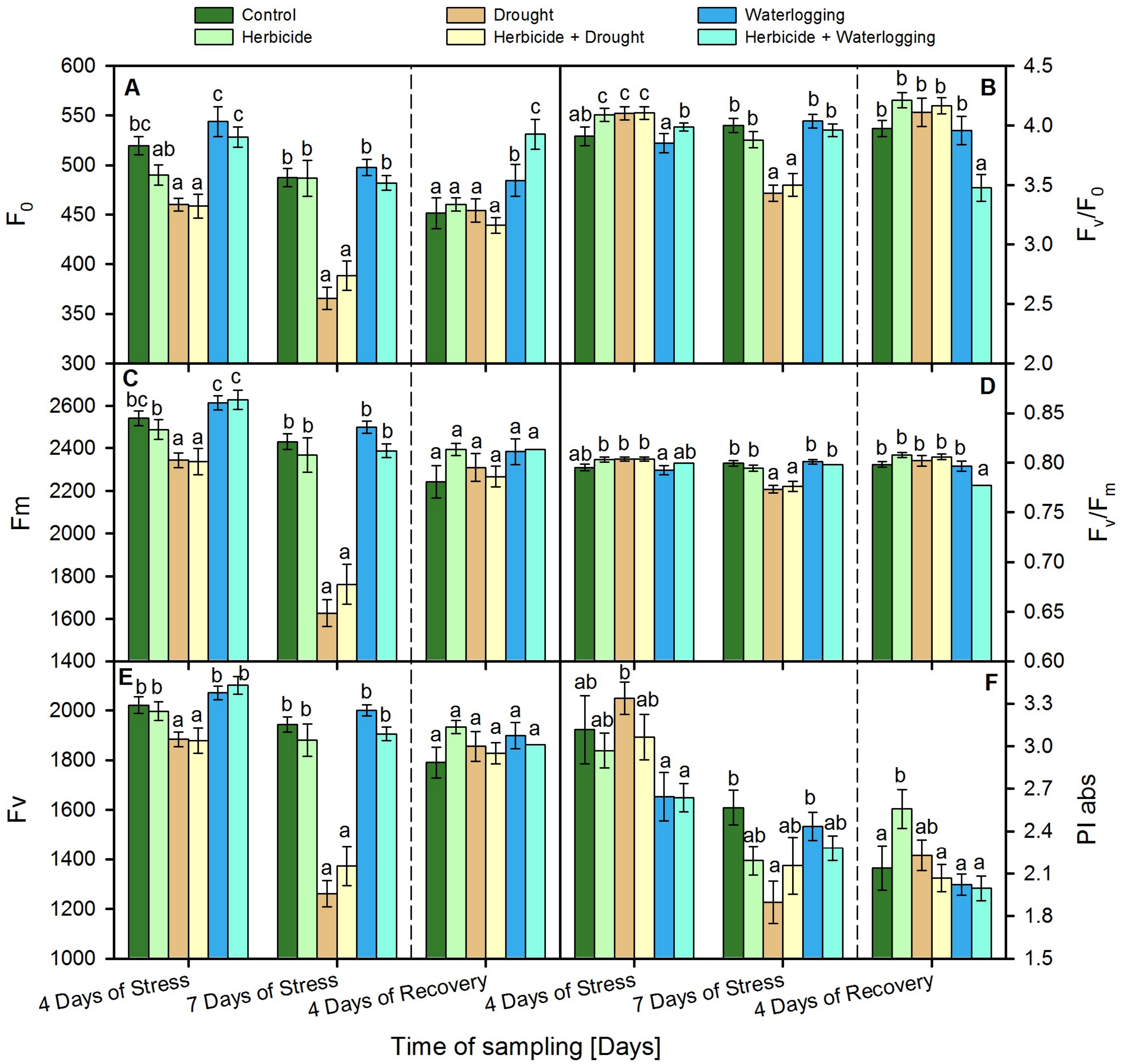


**Figure 4.** Parameters of chlorophyll *a* fluorescence in triticale plants treated with Serrate and exposed to drought or waterlogging stress: (**A**) Minimal fluorescence; (**B**) Primary photochemical reactions efficiency; (**C**) Maximal fluorescence; (**D**) Maximal quantum yield of PSII; (**E**) Variable fluorescence; (**F**) Fluorescence performance on the absorption base. Data are average values ± SE. The small letters represent significant differences between the treatments in the same group at $p < 0.05$.

### *3.7. Spider Plot Presentation of Parameters of Chlorophyll a Fluorescence*

Several biophysical functional parameters describing the photosynthesis performance during the light phase were considered according to the JIP test. Selected fluorescence parameters during the stress and recovery phases are shown in Figure 5 as spider plots.

During the first 4 days of the stress, $PI_{total}$, $\varphi R_o$ and $\delta R_o$ were decreased due to waterlogging (Figure 5A). In contrast, the same parameters were increased by drought. $PI_{total}$ provides information about the productivity of the entire photosynthetic apparatus, while $\varphi R_o$ and $\delta R_o$ reflect the electron transport in the PSI only.

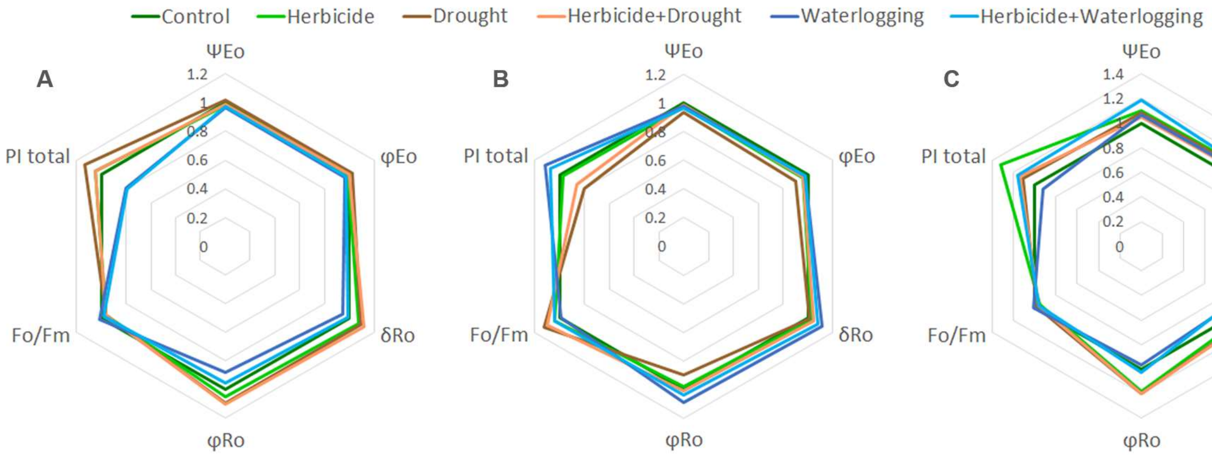


**Figure 5.** Plot of selected chlorophyll *a* fluorescence parameters in triticale plants on 4th day (**A**), 7th day (**B**) of drought or waterlogging and on 4th day of recovery (**C**) after drought or waterlogging.

Comparing the 4th day of stress (Figure 5A) and 7th day of stress (Figure 5B), these parameters showed a reversed trend. In addition, the plants exposed to drought and herbicide + drought had an increased $F_0/F_m$ ratio, which reflects the quantum yield of light energy dissipated by heat.

When the normal irrigation was resumed (Figure 5C), the most significant deviations were detected mainly in $\varphi R_o$ (herbicide, drought, and herbicide + drought treatments) and in $PI_{total}$ (herbicide alone treatment).

## 4. Discussion

The disturbance in water availability, both excess and deficit, imposes an important issue, because it is leading to the manifestation of water stress, which inhibits plant growth and distresses primary plant physiological processes [15–18]. There is limited information regarding the changes of plant growth and photosynthesis parameters due to treatment with selective herbicides, which principally are tolerated by crops, and following the exposure of plants to disturbed water availability such as drought or waterlogging. Our earlier findings [7] revealed that wheat tolerated Serrate® application in relation to its photosynthesis-related parameters. Principally, the mode of action of Serrate is not associated to photosynthesis, and its application did not lead to crucial alterations in the growth and photosynthesis-related traits neither alone applied nor in combination with stress factors. The changes in these parameters were mainly due to water stresses applied either alone or after herbicide treatment. Furthermore, wheat was capable of recovering photosynthesis after drought stress, while waterlogging worsened it even during the recovery period. This result was in agreement with the fact that wheat is susceptible to water excess and has relatively good tolerance to water deficiency [15].

In the current investigation, we also found that the treatment of triticale with Serrate® either alone or in combination with water stresses caused minor but inconsequential changes in the photosynthesis and growth traits (Figures 1–5), especially during the first days of its application. Some deviations were detected after the recovery period, such as a significant increase in DW, which reflected the decreased WC (Figure 1) and leaf pigment (Figure 2). A substantial intensification in $g_s$ and E (Figure 3) was accompanied by peak values of $PI_{abs}$ (Figure 4) and $PI_{total}$ (Figure 5C). The highest performance indices demonstrate the best utilization of light energy by the herbicide-treated plants for photosynthesis and better functional state of the entire photosynthetic apparatus, which returns as an accumulation of biomass and confirms the safe utilization of Serrate®. The observed higher values of DW in triticale after herbicide application are not unusual, as similar results were already reported [19,20]. For example, Clodinafop (one of the active substances of Serrate®) had increased DW, height and yield components of wheat compared to the control [19]. This observation was explained by the reduced weed infestation that might have enabled the wheat plants to augment more dry matter because of an absence of weed competition and better nutrient uptake [19]. This explanation could be valid (at least in part) in our model system.

Summarizing the data, we could approve that the treatment with Serrate® before exposure of triticale to drought or waterlogging did not provoke further negative alterations in growth and photosynthesis-related parameters, and the observed changes were mostly linked to the abiotic stresses applied.

Usually, a disturbance in water supply causes a disruption of plant growth [5,6]. Triticale plants' growth was affected mainly due to water deficit (Figure 1). Waterlogging did not cause substantial alterations in biometric parameters, and unlike our previous study on wheat, triticale sustained its growth after recovery. This observation is in agreement with the fact that the triticale is a relatively more waterlogging-tolerant crop than wheat [8,9].

The alterations in growth traits of triticale correlated with the changes in the leaf pigment content (Figure 2), parameters of photosynthesis (Figure 3), and chlorophyll fluorescence (Figure 4). The plants that have less chlorophyll and carotenoids will not be able to carry out as much photosynthesis because of their role as light-harvesting pigments and photoprotective agents, respectively [21]. We detected that the decrease in leaf pigments under water stress (Figure 2) occurred simultaneously with the decline of leaf gas exchange characteristics (Figure 3). The results showed that water deficit impaired to a greater degree the photosynthetic parameters $A_n$, E, and $g_s$ compared to the waterlogging stress (Figure 3). Net photosynthesis ($A_n$) was most affected by water shortage and even had negative values during the stress period, which suggested that photorespiration process occurred in these plants [22]. The deterioration of photosynthetic function caused by both water stresses intensified with time. In drought-treated plants, photosynthesis impairment occurred in parallel with the decrease in the photochemical efficiency of the photosynthetic apparatus (Figure 4). The chlorophyll *a* fluorescence is a widely used method for a quick non-destructive assessment of the efficiency of the photosynthetic machinery, the electron transport, and the physiological status of the plants [23]. Mainly, the maximal quantum yield of PSII ($F_v/F_m$) and the electron transport outside $Q_A^-$ ($F_v/F_0$) are used as important reliable indicators reflecting the performance of PSII under an unfavorable environment [24]. Under drought stress conditions, an overexcitation of PSII occurs, because the absorbed light energy exceeds the capabilities of the chloroplasts to utilize it, which in turn causes further photoinhibition and photodamage, as indicated by decreased maximal quantum yield of PSII ($F_v/F_m$) [25,26]. PSII photodamage can happen through photooxidative stress, either at the acceptor side or at the donor side via inactivation of the oxygen-evolving complex [27,28]. Under drought stress conditions, the availability of water for oxidation is limited, which reduces the efficiency of the oxygen-evolving complex at the donor side ($F_v/F_0$) [25,29]. Our results indicate that additional oxidative damage could take place because of limited photoprotection as a consequence of decreased carotenoids content in drought-treated plants (Figure 2). The data of chlorophyll *a* fluorescence (Figure 4) showed that drought caused a substantial decrease in the basic fluorescence parameters $F_0$, $F_v$, and $F_m$ during the stress period and especially after 7 days. This in turn led to a significant decline in $F_v/F_m$ and $F_v/F_0$, which is indicating damages to PSII due to water deficit (Figure 4). Similar changes in fluorescence parameters were observed in wheat plants subjected to drought [30,31]. In addition, the decrease in $PI_{abs}$ (Figure 4) and $PI_{total}$ (Figure 5B) indicated a worsened utilization of light energy because of dissipation through heat, which is in accordance with earlier reports [32]. This is evidenced by the increased $F_0/F_m$ ratio (Figure 5B) in the drought-treated plants during prolonged water deficit.

The chlorophyll *a* fluorescence parameters $F_v/F_m$ and $F_v/F_0$ did not show significant fluctuations in triticale exposed to waterlogging, which suggested that the electron transport was not impaired severely and was sustained close to the physiological state. Similar results were observed in winter wheat subjected to waterlogging [33]. The initial small decrease in the $PI_{abs}$ (Figure 4), accompanied by the reduced $\varphi R_o$ and $\delta R_o$ (which is suggesting a minor disruption in the electron transport of PSI) in waterlogged plants (Figure 5A) led to decreased $PI_{total}$ (Figure 5A). However, all these were fully recuperated in a short time, as it was noticed on 7th day of stress (Figures 4 and 5B).

Most of the growth, biochemical and biophysical parameters tended to reach the control levels after cessation of the stress (Figures 1–4). After restoration of the normal irrigation, an increase in all gas exchange indices $A_n$, E, $g_s$, and WUE, along with the chlorophyll *a* fluorescence parameters $F_0$, $F_v$, $F_m$, $F_v/F_m$ and $F_v/F_0$, indicated reparation in the electron transport chains and higher $CO_2$ assimilation. This is suggesting that the photosynthetic functions in the treated plants were nearly recovered. Our findings are in line with the observations of other researchers regarding photosynthesis performance of drought-treated and/or waterlogged crops during the recovery period [4,5,15,16,34–36]. In addition, the high values of $\varphi R_o$ and $\delta R_o$ of drought and herbicide + drought-treated triticale (Figure 5C) mean that more electrons have reached PSI to reduce final electron acceptors at the PSI acceptor side, which could signify an over-compensatory effect on quantum yields and efficiency at the PSI acceptor side [37].

The indices of recovery and resilience of the biometric parameters (Table 1) showed that although triticale was capable of recovering FW and WC after the termination of drought stress, their DW remained stunted; i.e., plants needed more time to accumulate newly developed biomass. On the other hand, the recovery and resilience indices in waterlogged triticale indicated that its growth continued even during the stress period, and after the recovery, plants tended to reach their control growth level more rapidly. Similarly, the recovery and resilience indices, calculated for photosynthesis-related parameters (Table 2), indicated that although the drought-treated triticale recovered transpiration and stomatal conductance at highest degree, $A_n$ still was negative, which probably indicates that plants needed more time to recover successfully their deprived photosynthesis rate. The recovery indices of waterlogged triticale had positive values, and resilience indices had minimal negative values, which confirmed again that the plants almost reached their initial physiological state (Table 2) and recovered better than those exposed to drought. The indices of recovery and resilience had positive values in triticale treated with Serrate® only (Tables 1 and 2), except for WC, which indicates that this herbicide could be used as a reliable implement even under unfavorable environment conditions such as water stress.

## 5. Conclusions

Our study confirms that the application of the selective herbicide Serrate® did not cause considerable variations in the growth and photosynthesis performance of triticale when applied alone or in combination with subsequent exposure of plants to drought or waterlogging. Drought and waterlogging decreased the efficiency of photosynthesis of triticale to a different extent during the stress period. We also found that waterlogging did not worsen significantly most of the biometric, chlorophyll *a* fluorescence, and photosynthesis-related parameters in both waterlogged and herbicide + waterlogging-treated plants. After the period of recovery, the photosynthesis of waterlogged plants was almost completely recovered, while drought-treated plants needed more time to repair the photosynthetic functions and to continue to grow. These findings are also supported by the indices of recovery and resilience of triticale growth and photosynthesis parameters. Our study provides new facts about the photosynthesis performance and chlorophyll *a* fluorescence responses of triticale associated to the application of Serrate® under unfavorable growth conditions, which extends the information in this particular topic to encourage upcoming exploration in the same area.

**Author Contributions:** I.S. and D.T. conceptualized and coordinated the research; D.T. grew and treated the plants; S.A., V.A., D.T. and I.S. performed the analyses, collected and interpreted the data; I.S. and V.A. prepared figures and photos; D.T. prepared the original draft of the manuscript; I.S. and D.T. reviewed and edited the manuscript; D.T. provided project administration and funding acquisition. All authors have read and agreed to the published version of the manuscript.

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