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Effects of Waterlogging, Drought and Their Combination on Yield and Water-Use Efficiency of Five Hungarian Winter Wheat Varieties

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Abstract: The effects of simulated waterlogging, drought stress and their combination were examined in a model experiment in Martonvásár, Hungary, in 2018. Four modern winter wheat varieties ('Mv Toborzó' (TOB), 'Mv Mambó' (MAM), 'Mv Karizma' (KAR), 'Mv Pálma' (PAL)) and one old Hungarian winter wheat cultivar ('Bánkúti 1201' (BKT)) were tested. Apart from the control treatment (C), the plants were exposed to two different abiotic stresses. To simulate waterlogging (WL), plants were flooded at four leaf stage, while in the WL + D treatment, they were stressed both by waterlogging and by simulated drought stress at the early stage of plant development and at the heading stage, respectively. The waterlogging treatment resulted in a significant decrease in plant biomass (BKT, TOB), number of spikes (TOB), grain yield (BKT, TOB), water use (BTK) and water-use efficiency (TOB, MAM, PAL) compared to the controls. The combined treatment (WL + D) led to a significant decrease in plant height (BTK, MAM, KAR), number of spikes (BTK, TOB, MAM, KAR), thousand kernel weight (TOB), harvest index (BTK), biomass, grain yield, water-use efficiency (in all varieties) and water use (BKT, TOB, MAM, KAR) of the plants. The best water-use efficiency was observed for MAM; therefore, this genotype could be recommended for cultivation at stress prone areas. The varieties MAM, KAR and PAL also showed good adaptability.

Keywords: water surplus; water deficiency; water uptake; small grain cereals; abiotic stress

1. Introduction

In the near future, agriculture, especially field crop production, will face various challenges. One of them will be to meet the food requirements of the growing population while the available freshwater reserves are in steady decline [1]. Wheat production plays a crucial role in the food supply, however, the industry is highly susceptible to climatic and environmental changes [2]. Extreme weather events are being experienced more frequently due to climate change in many parts of the world and this includes changes in the precipitation patterns as well. Consequently, dry areas become drier and wet areas become even wetter. As a result, cereals are now exposed to various water-related stress conditions within one growing season [3,4]. Increasing air temperature combined with limited water availability are the most important yield-limiting factors globally [5]. Drought is one of the most important stressors for cereals, including wheat [6]. It damages crop growth physiology and productivity [7], can reduce the amount of biomass by more than 25% and can cause large crop

failure [8]. Shifts in rainfall occurrence and increasingly frequent incidence of water deficiency will have a crucial role in the development and yield of winter cereals in Central Europe [9].

The climatic conditions of the Carpathian Basin are influenced by three climatic macro-regions (Continental, Ocean and Mediterranean). Based on climatological prediction, the dominance of the Mediterranean effect is expected to emerge. Using a long-term data series, it has been clearly determined that the winters are becoming wetter, and the amount of precipitation is in decrease during the spring and summer. Additionally, there is an increase in the intensity of rainfall; therefore, rainwater utilization becomes less efficient. It is very likely that waterlogging and drought occur within one growing season of the winter cereals (October–July) and the most exposed areas are the same in the case of both stressors [9]. When endeavouring to reach optimum yields with limited water supplies, farmers need to choose new water-saving technologies and such varieties that use water more efficiently [10]. However, not only the deficit of rainfall, but also the high amount of precipitation, can affect plant growth negatively: 25% of the global wheat plantation is affected by waterlogging [11]. Short-time waterlogging do not cause severe problems, but a constant water coverage affects plant physiology and productivity negatively, depending on the resistance of the harvested cultivars. Additionally, it is influenced by several other factors, such as plant growth stages or temperature [12]. Studies indicated that plants in the reproductive phase (i.e. the stages from stem elongation to anthesis) were relatively less tolerant to water stress [13,14]. Furthermore, the effects of water stress on cereals (especially on wheat) depend on the duration and severity of the stress. Ercoli [15] observed that grain yield, dry matter accumulation and remobilization were negatively affected by drought stress during the grain-filling stage. Mild water stress was found under the conditions of water limitation [16]. Waterlogging had relatively less effect on wheat, possibly because it does not depend on where surface drains occur [17]. Except for the mentioned growing stages and the severity of the stress, the effects of water shortage or surplus on wheat also depend on soil type and environmental conditions [14,18,19], grown cultivars [20,21] and cultivation technologies [22,23].

Water-use efficiency (WUE; kg·m^{-3}) is a key indicator of drought tolerance, because it reflects how the carbon and water cycles are related. WUE is an essential feature for assessing the responses of plants to climate change. The substantial differences between the WUE values of individual cereal species is a well-known phenomenon [24,25]. Maximising the water-use efficiency will become essential in those areas where water is the most limiting factor in wheat production [26]. The irrigation of small grain cereals, such as wheat, is not profitable in the Carpathian Basin; mainly because of the high energy costs. Therefore, the primary solution for minimizing yield losses could be the efficient use of the available water resources reserved in the soil. Plant breeding programmes in this region focus on the selection of genotypes with good adaptability and high water-use efficiency.

Our objectives were (1) to determine how waterlogging at tillering stage and waterlogging in combination with drought stress induced at the heading stage influence the plant phenological parameters and the yield of five Hungarian winter wheat varieties; (2) to highlight the changes that can be induced by these stresses in the water uptake and water-use efficiency of plants.

2. Materials and Methods

2.1. Experimental Design

Four winter wheat (*Triticum aestivum* L.) varieties bred in Martonvásár, Hungary ('Mv Toborzó' (TOB), 'Mv Mambó' (MAM), 'Mv Karizma' (KAR) and 'Mv Pálma' (PAL)) and one old Hungarian wheat cultivar ('Bánkúti 1201' (BKT)) were examined in a model experiment at the Agricultural Institute, Centre for Agricultural Research in Martonvásár. The study was carried out in a climate-controlled greenhouse chamber. The experiment was started on 2 February 2018 and ended at the end of June 2018, when the plants were harvested manually. 'Mv Toborzó' is an early-ripening variety; 'Mv Mambó' and 'Bánkúti 1201' are late-ripening genotypes; 'Mv Pálma' and 'Mv Karizma' are middle-ripening varieties. The experimental design consisted of three treatments and five genotypes. Control ('C')

plants were watered 2–3 times a week and the soil water content (SWC) was kept at 60%. The soil water content was monitored biweekly by 5TE soil moisture sensors (5TE soil moisture sensors, Decagon Devices Ltd., Pullman, WA, USA). The waterlogging simulation ('WL') was started when the plants reached the four leaf stage (Z14 stage) [27]. The pots were filled completely with water, and 5 cm waterlogging was kept above the soil surface for 12 days. In the combined stress treatment, one third of the plants were stressed by waterlogging at Z14 stage, and the irrigation was stopped completely for 15–17 days when they reached Z59 stage [27]. At the end of the treatment, the soil water content was determined by 5TE sensors (pots were rewatered when SWC dropped below 5 *v/v*%). In this way, the plants were continuously exposed to the same stress intensity.

Five vernalized plants (15 plants of each genotype) were planted in plastic pots (depth: 27 cm; diameter: 24 cm), as described by Varga et al. [28,29]. The pots were drilled at the end of the waterlogging treatment, and the amount of the leaching water was measured by using a glass measuring cylinder (accuracy: 5 mL). After reaching full maturity, plant height, dry weight of aboveground biomass (DW) and yields per pot were measured. The exact water uptake of plants/pot was monitored with a digital scale (ICS689g-A15, Mettler Toledo Ltd., Budapest, Hungary) from the time of the planting to the final harvest. The plants were watered two times a week until the tillering stage and three times a week after that. Plants were irrigated by tap water of which quality was summarized in Table A1. The quality of the irrigation water was determined in 2018, but the temporal changes were not recorded. The soil was covered with non-transparent foil to prevent soil evaporation. Grain yield and biomass were measured with a digital scale (440-45N, KERN & SOHN GmbH, Balingen, Germany). Repeated three times per each treatment, the weight of 100 seeds was determined and the measured weights were multiplied ten times. The harvested aboveground biomass was oven-dried for two days, at 70 °C. After that, the dry weight of the plant materials was measured.

Water-use efficiency (WUE) was calculated by using Equation (1).

$$\text{WUE} = \frac{\text{GY}}{\text{WU}} \tag{1}$$

where WUE is water-use efficiency (kg·m^{-3}), GY is grain yield (kg), and WU is water use (m^3).

The harvest index was calculated as described in Equation (2).

$$\text{HI} = \frac{\text{GY}}{\text{BM} \times 100} \tag{2}$$

where HI is harvest index (%), GY is grain yield (kg), and BM is the dry aboveground biomass (kg).

2.2. Plant Growth Conditions

Seeds of each variety were germinated on 15 December 2017. The seeds were kept at room temperature (22 °C) in a dark plastic box for two days; after that they were transferred into a vernalization chamber for 47 days (temperature: 4 °C). Five seedlings were planted into pots on 2 February 2018, which contained 10 litres 3:1:1 (*v/v*) homogenous mixture of soil, sand and humus. The pots were placed into a greenhouse chamber. The climatic conditions were automatically regulated by using the Spring-Summer program [30]. The air temperature was increased from the initial 10–12 °C to 24–26 °C during the growing period, while the relative humidity was kept between 60% and 80% and regulated by the ventilation of the chamber. In case it was required, the natural light intensity was enhanced by artificial illumination to a value of 500 μmol·m^{-2}·s^{-1} at the beginning of the vegetation period, which was gradually increased to 700 μmol·m^{-2}·s^{-1}. The CO_2 concentration was kept at ~400 ppm. Nutrient solution was provided once a week. To each pot, 22 mL water-soluble fertilizer (14% N, 7% P_2O_5, 21% K_2O, 1% Mg, 1% B, Cu, Mn, Fe, Zn; Volldünger Classic; Kwizda Agro Ltd., Vienna, Austria) was added, before the irrigation.

2.3. Statistical Processing

The experimental design involved five winter wheat genotypes and three watering treatments in three replicates. A two-way ANOVA was performed to determine the effects of the tested factors (genotype and water supply) and Tukey's post hoc test to compare the means. SPSS 16.0 program (IBM, Armonk, NY, USA), Microsoft Excel (Microsoft, Redmond, WA, USA) and ggplot package of R software (Free Software Foundation, Boston, MA, USA) were used for the statistical analysis and visualization. Significance level was set at $P = 0.05$. The assessment of uncertainty is shown in Table A2.

3. Results

The plant height of 'Bánkúti 1201' (BKT) was significantly greater in all treatments than that of the modern varieties. The combined stress treatment (WL + D), however, resulted in a significant decrease (12%) in plant height compared to the control plants. In the case of 'Mv Toborzó' ((TOB) and 'Mv Pálma' (PAL) genotypes, no changes were caused by the treatments in the plant height, but the WL + D treatment induced significant reduction in the plant heights of 'Mv Mambó' (MAM) and 'Mv Karizma' (KAR), compared with the controls. The observed reductions were 18.8% and 21.8%, respectively. Overall, waterlogging (WL) did not affect the plant height in itself, however, in the case of the combined stress treatment, a significant decrease in plant height was detected in BKT, MAM and KAR genotypes (Figure 1).
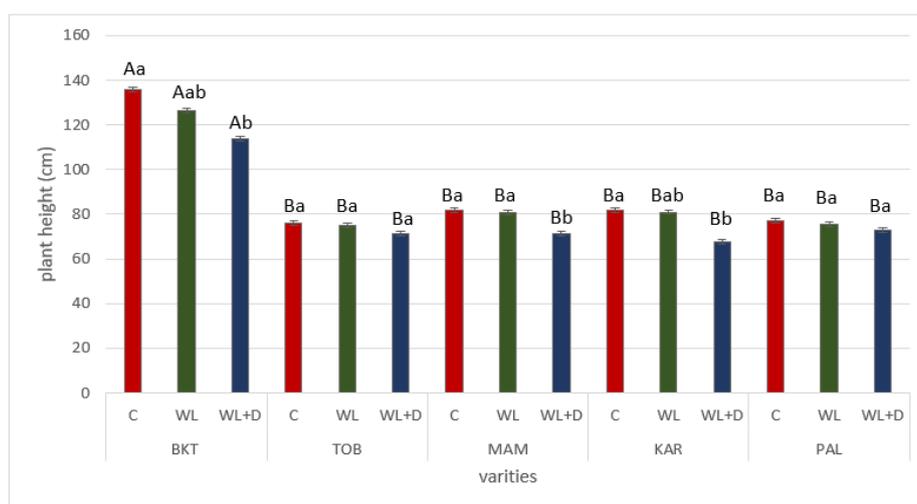

Figure 1. Plant heights of 'Bánkúti 1201' (BKT), 'Mv Toborzó' (TOB), 'Mv Mambó' (MAM), 'Mv Karizma' (KAR) and 'Mv Pálma' (PAL) varieties in the different treatments. 'C': control treatment; 'WL': waterlogging treatment; 'WL + D': combined treatment of waterlogging and drought. Capital letters indicate the statistical significance between the varieties at $p \leq 0.05$ level; lowercase letters indicate the statistical significance between the treatments at $p \leq 0.05$. Error bars represent honestly significant difference (HSD) 5% values (n = 3).

In contrast to plant height, water stresses caused significant changes in the aboveground biomass (Table 1). Based on the ANOVA test, the effect of the two factors (genotype and water supply) was significant, but no interaction could be confirmed. When plants were grown under optimum conditions, the highest biomass production was measured for BKT. The biomass production of MAM and KAR was similar and these genotypes were followed 'Bánkúti 1201'. Significantly lower biomass production was determined for TOB and PAL compared with the other varieties. In the case of BKT and TOB, significant differences could be observed between the control plants and the ones under waterlogging stress, but the water shortage did not reduce the biomass production further. The waterlogging treatment did not influence the biomass production of BKT, MAM and KAR statistically, but significantly lower values were measured for PAL and TOB. Under combined stress, only the biomass production of

the old Hungarian variety (BKT) was significantly higher compared with the other four genotypes, but among them, no considerable differences could be confirmed by the post-hoc test. There was a significant difference between the biomass production of the control and WL treatments, as well as between WL and WL + D treatments in KAR. In the case of PAL, a decreasing linear trend was observed, but the reduction in biomass production was found to be significant only between the C and WL + D treatments (Table 1).

Table 1. Biomass values of 'Bánkúti 1201' (BKT), 'Mv Toborzó' (TOB), 'Mv Mambó' (MAM), 'Mv Karizma' (KAR) and 'Mv Pálma' (PAL) varieties in the different treatments. 'C': control treatment; 'WL': waterlogging treatment; 'WL + D': combined treatment of waterlogging and drought; HSD: honestly significant difference. Capital letters indicate the statistical significance between the varieties at $p \leq 0.05$ level; lowercase letters indicate the statistical significance between the treatments at $p \leq 0.05$ (n = 3).

	Biomass Value (g)	Treatments			HSD 5%
		C	**WL**	**WL + D**	
	BKT	80.92 [Aa]	58.66 [Ab]	48.54 [Ab]	26.02
	TOB	41.72 [Ca]	28.11 [Cb]	22.52 [Bb]	15.52
Genotypes	MAM	56.24 [BCa]	47.54 [ABa]	31.90 [Bb]	19.38
	KAR	63.22 [Ba]	48.16 [Ab]	32.61 [Bc]	24.05
	PAL	41.30 [Ca]	33.63 [BCab]	25.55 [Bb]	12.38
	HSD 5%	16.74	12.44	10.20	

The number of spikes per pots are shown in Figure 2. Under optimum circumstances and in both treatments (W and WL + D), the highest number of spikes was observed for KAR. In the case of TOB, MAM and PAL, the numbers of spikes were significantly lower. The waterlogging treatment compared with the control caused a significant decrease only in the early-ripening genotype (TOB), but the drought did not reduce the number of spikes further. The combined treatment resulted in a significant decrease in the number of spikes of BKT, MAM and KAR varieties, compared with the control. Between the two stress treatments, a significant difference in the number of spikes was observed for BTK and KAR genotypes. The spike number remained stable even under stress condition for PAL.
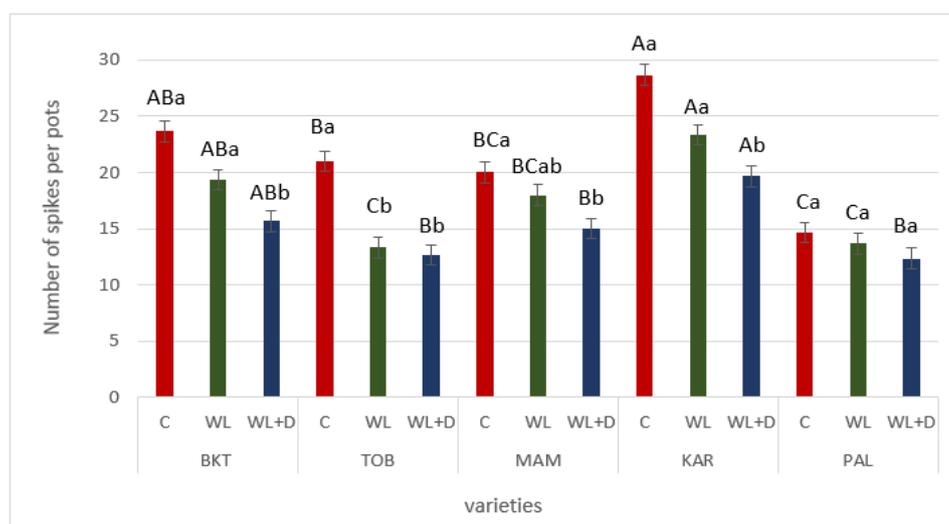

Figure 2. Number of spikes per pots of 'Bánkúti 1201' (BKT), 'Mv Toborzó' (TOB), 'Mv Mambó' (MAM), 'Mv Karizma' (KAR) and 'Mv Pálma' (PAL) varieties in the different treatments. 'C': control treatment; 'WL': waterlogging treatment; 'WL + D': combined treatment of waterlogging and drought. Capital letters indicate the statistical significance between the varieties at $p \leq 0.05$ level; lowercase letters indicate the statistical significance between the treatments at $p \leq 0.05$. Error bars represent honestly significant difference (HSD) 5% values (n = 3).

Thousand kernel weights (TKW) of the studied plants are shown in Table 2. The genotypes and water treatments themselves already had a significant impact on the grain yield. Under normal watering, MAM had the highest TKW, but no significant differences could be observed between MAM, BKT and TOB, however, PAL and KAR showed significantly lower TKWs. Waterlogging did not change the ranking of the varieties; the only difference was that the TKW of PAL was significantly lower than that of MAM and BKT. The treatments did not influence the TKW except for TOB, as the WL + D treatment induced a significant decrease in its TKW (32.5% compared with the control) (Table 2).

Table 2. Thousand kernel weights of 'Bánkúti 1201' (BKT), 'Mv Toborzó' (TOB), 'Mv Mambó' (MAM), 'Mv Karizma' (KAR) and 'Mv Pálma' (PAL) varieties in the different treatments. 'C': control treatment; 'WL': waterlogging treatment; 'WL+D': combined treatment of waterlogging and drought; HSD: honestly significant difference. Capital letters indicate the statistical significance between the varieties at $p \leq 0.05$ level; lowercase letters indicate the statistical significance between the treatments at $p \leq 0.05$ (n = 3).

	Thousand Kernel Weight (g)	**Treatments**			**HSD 5%**
		C	**WL**	**WL + D**	
Genotypes	BKT	38.72 ^{ABa}	41.13 ^{ABa}	39.39 ^{Aa}	1.95
	TOB	38.62 ^{ABa}	37.30 ^{BCa}	26.09 ^{Ab}	10.81
	MAM	45.68 ^{Aa}	44.77 ^{Aa}	41.21 ^{Aa}	3.71
	KAR	38.04 ^{Ba}	36.77 ^{BCa}	36.77 ^{Aa}	2.65
	PAL	34.19 ^{Ba}	31.48 ^{Ca}	27.48 ^{Aa}	5.298
	HSD 5%	4.21	5.06	7.51	

The differences in stress tolerance of the genotypes are indicated by the large variability that could be observed by comparing the harvested yields (Figure 3). Both factors (genotype and water supply) influenced the grain yield significantly, and their interactions were statistically significant too. The grain yield of the old Hungarian variety, BKT, was as high as that of MAM, but the other genotypes showed significantly lower yield production, even under optimum watering. Waterlogging resulted in a significant decrease in the grain yield of BKT and TOB. The observed reductions in their grain yield were 39.4% and 30.4%, respectively. While the combined stress did not reduce the grain yield of TOB further, a decreasing trend was detected even for BKT, and waterlogging combined with simulated drought caused a 72.9% reduction in the grain yield. Waterlogging did not influence the yield production of MAM, KAR and PAL significantly, however, compared with the control, the combined stress lowered the harvested grain yield by 41%, 56.6% and 37.6%, respectively.

The calculated values of the harvest index (HI) (Equation (2)) are presented in Figure 4. There was no significant decrease in the harvest index of BKT between the control and waterlogging treatment, but there was a significant decrease between the waterlogging and the combined stress treatment. The harvest index remained stable for the other varieties and no changes could be observed between the treatments. KAR showed significantly lower HI than TOB, MAM and PAL, but comparing the HI within all treatments, BKT and KAR did not differ from each other (Figure 4).

The changes caused by the induced stress treatments in the plants' water use (WU) and the differences between the varieties are shown in Figure 5. The individual factors (genotype and water supply) and their interactions had significant effects on the water uptake of the plants. The plant's habit (aboveground biomass, total leaf area, number of tillers per plant, etc.); the length of the vegetation period and many environmental parameters influence the water use; this variability can be seen in Figure 5 as well. Under the control treatment, the water use of BKT was significantly higher than that of all other examined varieties. The water use of KAR significantly exceeded that of the other modern varieties (41.8% higher water use compared with the average of MAM, TOB and PAL under C treatment). The two lowest water demands were detected for TOB and PAL under normal watering. Similar differences between the varieties were found under WL treatment, but no significant differences

were observed between MAM and KAR. Under WL + D treatment, the highest WU was measured for BKT and the lowest was observed for TOB. By comparing the effects of the stress treatments on water use, we found that waterlogging reduced WU significantly (10.3% lower compared with the control), and the combined treatment resulted in a significant decrease in BKT, TOB and KAR by 27%, 25% and 31%, respectively (Figure 5). Even the effects of water supply were confirmed by the ANOVA, and significant interactions could be detected.
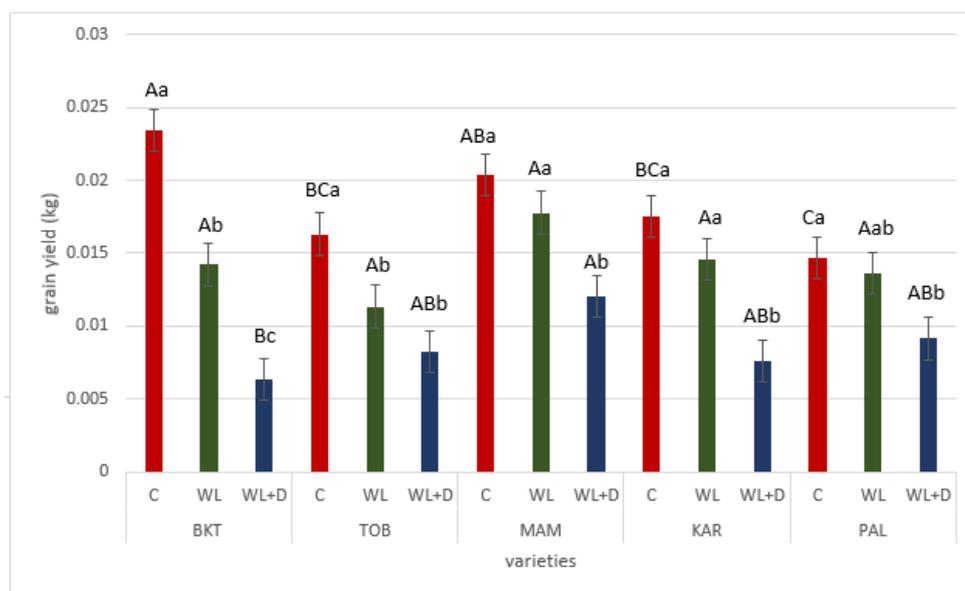

Figure 3. Grain yields of 'Bánkúti 1201' (BKT), 'Mv Toborzó' (TOB), 'Mv Mambó' (MAM), 'Mv Karizma' (KAR) and 'Mv Pálma' (PAL) varieties in the different treatments. 'C': control treatment; 'WL': waterlogging treatment; 'WL + D': combined treatment of waterlogging and drought. Capital letters indicate the statistical significance between the varieties at $p \leq 0.05$ level; lowercase letters indicate the statistical significance between the treatments at $p \leq 0.05$. Error bars represent honestly significant difference (HSD) 5% values (n = 3).
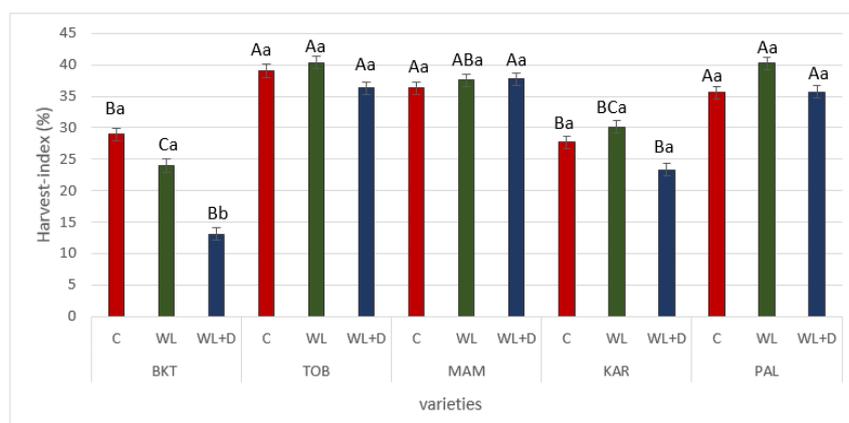

Figure 4. Harvest index values of 'Bánkúti 1201' (BKT), "Mv Toborzó" (TOB), 'Mv Mambó' (MAM), 'Mv Karizma' (KAR) and 'Mv Pálma' (PAL) varieties in the different treatments. 'C': control treatment; 'WL': waterlogging treatment; 'WL + D': combined treatment of waterlogging and drought. Capital letters indicate the statistical significance between the varieties at $p \leq 0.05$ level; lowercase letters indicate the statistical significance between the treatments at $p \leq 0.05$. Error bars represent honestly significant difference (HSD) 5% values (n = 3).
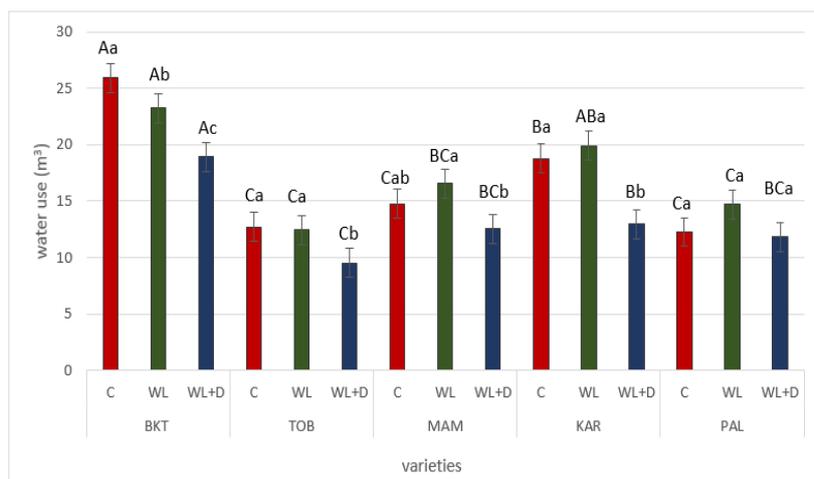
Figure 5. Water-use values of 'Bánkúti 1201' (BKT), 'Mv Toborzó' (TOB), 'Mv Mambó' (MAM), 'Mv Karizma' (KAR) and 'Mv Pálma' (PAL) varieties in the different treatments. 'C': control treatment; 'WL': waterlogging treatment; 'WL + D': combined treatment of waterlogging and drought. Capital letters indicate the statistical significance between the varieties at $p \leq 0.05$ level; lowercase letters indicate the statistical significance between the treatments at $p \leq 0.05$. Error bars represent honestly significant difference (HSD) 5% values (n = 3).

The water-use efficiency (WUE) values are shown in Figure 6. Typically, the highest WUE was observed under normal watering, while the stress treatments generally resulted in a decrease in WUE. Under well-watered conditions, the highest WUE was observed in MAM and TOB (1.37 and 1.28 kg·m^{-3}, respectively), whereas the lowest value was detected for BKT (0.9 kg·m^{-3}). The water coverage at the four leaf stage caused a reduction in WUE in TOB, MAM and PAL; they reached 29%, 22% and 22.8%, respectively. No changes of WUE in the WL treatment were observed for BKT and KAR compared with the controls. The combined treatment did not decrease the WUE significantly further for MAM, TOB and PAL, but the WUE values of all varieties were significantly lower than it was calculated for the control treatment. Under both stress conditions, the highest WUE values were observed for MAM, which indicates that, even under unfavourable conditions, this genotype used the available water resources the most efficiently (Figure 6).
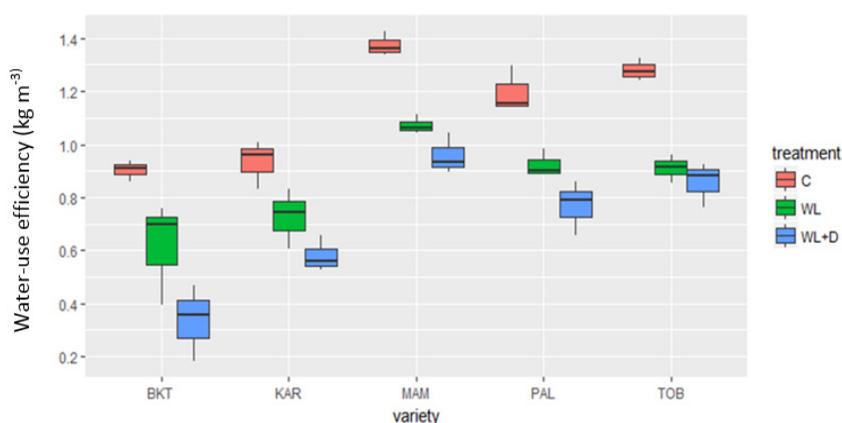

Figure 6. Water-use efficiency of 'Bánkúti 1201' (BKT), 'Mv Toborzó' (TOB), 'Mv Mambó' (MAM), 'Mv Karizma' (KAR) and 'Mv Pálma' (PAL) varieties in the different treatments. 'C': control treatment; 'WL': waterlogging treatment; 'WL + D': combined treatment of waterlogging and drought (n = 3).

4. Discussion

According to our results, in the case of the tested winter wheat genotypes, the waterlogging treatment caused no significant changes in the plant height compared with the control treatment. However, in contrast to our findings, Akhtar et al. [31] reported that the plant height was generally decreased in wheat by hypoxia. The effects of the combined waterlogging and drought stress resulted in a significant decrease in the plant height of 'Bánkúti 1201' (BKT), 'Mv Mambó' (MAM) and 'Mv Karizma' (KAR) varieties. Akhtar and Nazir [32] also found that plants under drought and waterlogging stresses suffered growth reduction. It is consistent with the observations of Varga et al. [29] that the length of the vegetation period could influence the stress tolerance of the cereal varieties.

Resulting from the waterlogging, a significant decrease in the biomass production of BKT, 'Mv Toborzó' (TOB) and KAR were observed. Corroborating our observations, De San Caledonio et al. [33] published that waterlogging reduced the aboveground biomass of wheat and barley plants. González et al. [34] also found that plants under drought and waterlogging decreased biomass production in quinoa plants. Ding et al. [12] reported that severe waterlogging stress at stem elongation and heading stages significantly reduced the total biomass. Our experiment confirmed that even waterlogging itself affects the plant biomass negatively, but the negative effects can be even more severe if waterlogging and drought occur within one growing season.

In relation to the thousand kernel weight (TKW), Wollmer et al. [35] found that this parameter of two winter wheat cultivars ('Kredo' and 'JB Asano') was not influenced by early waterlogging, but late waterlogging was associated with significantly decreased number of grains per spike and lowered thousand kernel weight. The results of Ding et al. [12] confirmed that mild and severe waterlogging stress at stem elongation stage significantly decreased the thousand kernel weight and the same was observed for severe waterlogging at heading stage. However, according to the findings of Ghobadi et al. [36], the thousand kernel weight did not drop significantly as a result of waterlogging at tillering and jointing stages. Our results highlighted that waterlogging itself did not affect the thousand kernel weight of the tested genotypes, but due to waterlogging combined with water shortage, the TKW decreased significantly by the early ripening cultivar (TOB).

According to our findings, the grain yield of the plants decreased for all genotypes, as a result of the induced stress treatments. Other authors also published that hypoxia and anoxia are responsible for yield reduction and a decrease in grain yields in most wheat varieties [31,32]. Ding et al. [12] claimed that yield losses increased with the growing degree of stress. Additionally, they reported that mild and severe waterlogging at stem elongation and heading stages significantly decreased grain yield. Confirming our findings, Araki et al. [13] also found that waterlogging stress at stem elongation stage decreased the grain yield, and claimed that this process was mainly due to the loss in the thousand kernel weight [13]. Another study revealed that both at tillering and at jointing stages of wheat, the grain yield of the plants was significantly decreased compared with the control plants [36].

Ding et al. [12] found that the harvest index was significantly lowered by mild and severe waterlogging stress at stem elongation stage, but at heading stage, no significant changes were observed. However, Ghobadi et al. [36] reported that no significant changes were witnessed in the harvest index, either at tillering or at jointing stage of the examined wheat varieties, as a result of waterlogging. In our experiment, the harvest index decreased significantly only in one genotype as a result of the combined treatment.

In the present study, the combined stress treatment reduced the water use significantly in three varieties (BKT, TOB and KAR), but similar trends were observed only for one genotype (BKT) as a result of waterlogging. Parent et al. [37] claimed that waterlogging-limited water use led to internal water deficit. Another study also stated that water coverage significantly decreased the water use of the winter wheat [32].

The water-use efficiency (WUE) of all varieties decreased significantly, resulting from the combined stress, but statistically confirmed changes were observed only for three genotypes as a result of the waterlogging treatment. According to Angus and van Herwaarden [38], the water-use efficiency

provides a simple means of assessing whether the yield is limited by water supply or other factors. Kang et al. [39] found that WUE showed a linear increase, together with the harvest index. Based on their data, Qiu et al. [40] stated that the seed-setting and milky ripe stages were the most critical regarding WUE. Several papers concluded that the variability in water-use efficiency may be substantial, even within one variety, primarily due to environmental effects [38,41,42].

5. Conclusions

Our results confirm that there is a difference in the adaptive capacity of various winter wheat genotypes, and the modern cultivars have far better environmental plasticity than the old landraces. Both waterlogging and drought can result in serious yield losses, although significant differences among the susceptibility of cultivars were observed.

The waterlogging treatment resulted in a significant decrease in the biomass production of the old Hungarian (BKT) variety and the early-ripening genotype. The fast development and the short tillering stage contribute to the reduction in the number of spikes of TOB under waterlogging. The sensitivity to stresses at the early phenological stages of BKT and TOB is indicated by the significant grain yield reduction resulting from waterlogging. The water use during the vegetation period of TOB did not change as a result of waterlogging, which means that the transpiration remained stable after the stress. However, the significant reduction in the grain yield indicates that the varieties with shorter vegetation periods could be more sensitive to abiotic stresses at the early stages of vegetation. The water use of BTK and the water-use efficiency of TOB, MAM, PAL decreased as a result of waterlogging compared to the controls. The combined treatment (waterlogging and drought) led to a significant decrease in plant height (BTK, MAM, KAR), number of spikes (BTK, TOB, MAM, KAR), thousand kernel weight (TOB), harvest index (BTK), biomass, grain yield and water-use efficiency (all varieties) and the water use (BKT, TOB, MAM, KAR) of plants. The best water-use efficiency was observed in each treatment for MAM. Therefore, this genotype could be recommended for cultivation at stress prone areas. Besides this, KAR and PAL varieties also showed good adaptability.

We can conclude that these kinds of investments and topic-oriented experiments could contribute to the fulfilment of food demand as well.

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Appendix A

Table A1. Water quality of the irrigation water. EC: Electrical conductivity; SO_4^{2-}: sulfate; NH_4^+: ammonium; COD: chemical oxygen demand; Fe: iron.

Parameter	Unit	Values
pH	-	7.8
EC	$dS \cdot m^{-1}$	0.71
SO_4^{2-}	$mg \cdot L^{-1}$	44
NH_4^+	$mg \cdot L^{-1}$	0.30
COD	$mg \cdot L^{-1}$	0.24
Fe	$mg \cdot L^{-1}$	0.63

Appendix B

Table A2. Assessment of uncertainty.

Assessment of Uncertainty		
Measurement	**Accuracy**	**Possibility of uncertainty**
Relative water content	Apparent dielectric permittivity: ±1 Ea from 1–40 cm, ±15% from 40–80 cmVWC: ±0.03 m^3/m^3 (±3% VWC) in mineral soils, ±0.01–0.02 m^3/m^3 (±1–2% VWC) in any porous medium	Small chance of error
Plant development	Made exactly according to the instructions of Zadoks scale	To minimize errors, this measurement was taken by one person.
Grain yield, biomass	Readability 0.1 g; accuracy 0.1 g	The measurements of these parameters were highly accurate.
Number of the grains	Man-made	To minimize the uncertainty, counting was repeated two times by one person.
Plant height	Made by a gauging-stick	Minor possibility of uncertainty
Water use	Readability 5 g; accuracy 1 g ≤ x ≥ 5 g	Possibility of uncertainty between 1 and 5 grams
Temperature of the cambers	For temperature measurements in the greenhouse chamber, sensors with accuracy of 0.1 °C were used. Temperature during the vegetation period was regulated by a standardized climatic program [28].	The same climatic program was used for the phytotron studies. Minor possibility of uncertainty.
Relative humidity	For the relative humidity measurements, sensors with accuracy of 1% were used. Temperature during the vegetation period was regulated by a standardized climatic program [28].	The same climatic program was used for the phytotron studies. Minor possibility of uncertainty.
Artificial illumination	Wavelength of the applied lamps covered PAR spectrum (380–720 nm). Artificial illumination was started at an intensity of 500 μmol·m^{-2}·s^{-1} and was increased to 700 μmol·m^{-2}·s^{-1} until the end of the vegetation.	The same climatic program was used for the phytotron studies. Minor possibility of uncertainty.
CO_2 concentration	CO_2 sensor (TIM10) with an accuracy of 3% was installed in the chamber. When the measured value reached 450 ppm, the windows opened automatically.	Very reliable system, minor possibility of uncertainty.
Adding of water-soluble fertilizer	Done with manual pipette (Pipet-Lite Pipette LTS L-20MLXLS, Mettler Toledo Ltd., Busapest, Hungary) Accuracy: ±0.6%/120 μL; Precision ±0.6%/32 μL	Minor possibility of uncertainty.

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