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Enhancing the Photosynthetic and Yield Performance of Rice in Saline Soil by Foliar-Applying Cost-Effective Compounds as Sources of Carbon Dioxide and Potassium

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Abstract: Although rice is highly sensitive to salinity, it is considered one of the best crops to grow in salt-affected mudflat soils to alleviate the salinity problem. Applying chemical compounds for an increase in leaf CO_2 and nutrient levels can help mitigate the negative impact of salinity on plants in a cost-effective manner. To identify the benefits of using lithovit (Liv), ethanol (Eth), and potassium carbonate (KC) as a source of CO_2 and K to enhance rice production in salt-affected soils, a field study was conducted to assess the effects of these compounds on the agro-physiological parameters of two rice genotypes (Giza178 and Giza179) in saline soils. The compounds were applied as a foliar spray at a concentration of 30 mM each before and after the heading growth stage. The results indicated that the genotype, application time, compounds, and their potential two-way interactions significantly influenced all agro-physiological parameters, with only a few exceptions. The genotype Giza 179 exhibited higher pigment contents, photosynthetic capacity, relative water content (RWC), grain yield, and most yield components compared to Giza 178, with increases ranging from 2.1% to 37.9%. Foliar application of different compounds resulted in a 9.7–37.9% increase in various parameters and a 34.6–43.2% decrease in the number of unfilled grains (NUFG) per panicle compared to untreated treatment. Foliar application of different compounds before heading resulted in an increase in various parameters by 4.8–16.1% and a decrease in the NUFG per panicle by 22.9% compared to those applied after heading. Heatmap clustering analysis revealed that foliar application of Liv before heading was the most effective treatment in enhancing various parameters for both genotypes and mitigating the negative effects of salinity stress on the NUFG. This was followed by Eth and KC before heading for Giza 179. Applying Eth and KC to the leaves after heading had a moderate positive impact on most parameters for Giza 179, outperforming the application after heading for Giza 178. Overall, our findings indicate that spraying readily available compounds that elevate CO_2 and K levels in rice leaves can help alleviate the negative impacts of salt stress and improve rice production in salt-affected soils in a cost-effective manner.

Keywords: ethanol; grain yield; lithovit; photosynthetic attributes; pigment contents; potassium carbonate; relative water content

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1. Introduction

Rice (*Oryza sativa* L.) is a vital staple food for about half of the world's population. It is grown on 11% of the world's cultivated land and contributes over 20% of the world's total calorie intake [1,2]. By 2030, rice production needs to increase by 20% to meet the growing population's demands and decrease in arable land [2]. Rice yield significantly increased in the latter half of the 20th century, but the growth rates of rice yields have decreased and even reached a plateau in key rice-producing countries like the USA, South Korea, China, and Indonesia [1,3,4]. This indicates that in the future, increasing rice production in these countries may necessitate expanding the cultivated land rather than boosting yields per unit area. However, many new areas are experiencing salinity stress, particularly in arid and semiarid regions, due to climate fluctuations, high temperatures, and limited freshwater resources. Over 331 million hectares (Mha) of land worldwide are affected by salinity stress, which is expected to increase by about 1.0–1.5 Mha each year [5,6].

Rice is a suitable option for enhancing salt-affected mudflat soil despite its sensitivity to salt stress (threshold EC around 3 $dS\,m^{-1}$). The water in paddy fields aids in salt leaching, mitigating the adverse impacts of salinity on rice growth and yield [7,8]. Applying proper agronomic techniques allows these soils to be utilized more effectively for rice production. Salt stress can limit crop growth and yield by limiting water availability to the plant (osmotic stress), increasing the uptake of harmful ions like Na^+ and Cl^- (specific ion toxicities), and inhibiting the uptake of essential nutrients like K^+ and Ca^{2+} (ion imbalance) [6,9,10]. These three components of salinity stress interact together to, directly and indirectly, distort various physiological and biochemical processes necessary for plant growth and production, such as photosynthetic machinery, CO_2 assimilation, respiration, photosystem II function, Rubisco activation, Calvin cycle patterns, stomatal conductance, mesophyll biochemical capacity, chloroplast structure, and plant water relations [11,12]. Most importantly, CO_2 assimilation and stomatal conductance are closely linked under saline conditions and tend to decrease together. Reduced stomatal conductance limits CO_2 entry into the leaf, leading to decreased photosynthetic capacity of the mesophyll, particularly in C_3 plants [12–14]. Therefore, increasing CO_2 concentration can boost plant growth and help counteract the harmful impacts of salinity stress on rice production. This is because a significant portion of a plant's dry weight is derived from CO_2 assimilation during photosynthesis. Previous studies have shown that higher CO_2 levels in C_3 plants enhance leaf photosynthesis and growth by increasing Rubisco enzyme activity, which is not fully saturated at current atmospheric CO_2 levels, and inhibiting leaf respiration rates [15,16]. Elevated CO_2 also reduces CO_2 losses from photorespiration by inhibiting oxygenation by Rubisco [17–19]. Zheng et al. [20] reported that elevated CO_2 concentrations could enhance the growth and grain yield of C_3 plants by stimulating the leaf photosynthesis rate and increasing non-structural carbohydrate levels. Furthermore, elevated CO_2 concentrations can mitigate the negative impacts of drought stress on the growth and production of C_3 plants by decreasing stomatal conductance and leaf transpiration rate and increasing leaf photosynthesis rate [21]. Kazemi et al. [22] found that increased CO_2 levels helped reduce the negative effects of salinity stress on rice growth and production. However, the extent of this benefit varied depending on the rice genotypes and salinity levels. Generally, elevated CO_2 concentrations have a greater positive impact on C_3 crops than C_4 crops. Previous studies have shown that elevated CO_2 concentrations can increase the production of C_3 plants by 15–41% and C_4 plants by 5–10% [23–25]. Intriguingly, increased CO_2 levels notably improved crop growth and yield in stressful conditions, with little effect in non-stressful conditions [26–28].

Fortunately, in open field environments, plant CO_2 levels can be boosted by applying nano-fertilizers or specific chemicals directly onto the leaves, known as CO_2 fertilization. One example of such a product is lithovit, a natural CO_2 foliar fertilizer derived from Nano-$CaCO_3$. When lithovit is sprayed onto leaves, it breaks down into calcium oxide (CaO) and releases CO_2 through leaf stomata, raising CO_2 levels within the leaves higher than in the atmosphere. The nanoparticles in lithovit can easily enter through the stomata of plant leaves and form a thin layer on the leaf surface. This layer is more effectively absorbed when the leaves are moistened with dew overnight [29–31]. Additionally, lithovit contains nano-iron (Fe) and nano-magnesium (Mg), essential for various physiological and biochemical functions in plants, whether in normal or stressful conditions. Nano-Fe is crucial for photosynthetic reactions as it activates enzymes in the photosynthetic system, aids in electron transport, and supports RNA synthesis. Nano-Mg is essential for photosynthesis as it is the core of the chlorophyll molecule. It also aids in starch translocation, sugar synthesis, and nutrient uptake [32,33]. Therefore, foliar application of lithovit may enhance rice growth and production under salinity stress conditions by boosting the photosynthesis rate and reducing the negative effects of this stress. Previous studies have shown that applying lithovit to leaves can greatly enhance the growth and yield of field crops compared to other growth stimulants. This is because lithovit releases CO_2, which improves photosynthesis efficiency. In some cases, lithovit has increased crop production by up to 50% and improved plants' resilience to environmental stressors [34–36].

Ethanol (C_2H_5OH) is a clear, odorless, and non-toxic solvent that can also serve as a source of CO_2. When applied to leaves, ethanol is quickly converted to formaldehyde and formic acid, which are then metabolized to CO_2 in the leaf mesophyll. This process results in higher CO_2 levels and a higher CO_2 to O_2 ratio, which boosts photosynthesis and reduces photorespiration rates, especially in C_3 plants [37–39]. Increasing the CO_2 to O_2 ratio in the leaf mesophyll reduces competition between O_2 and CO_2 for Rubisco enzymes and 1,5-bisphosphate (RuBP) substrates, favoring RuBP carboxylation over oxygenation. In addition, applying ethanol to the leaves can enhance the production and stability of photosynthetic pigments, leading to improved photosynthesis [40]. Considering these facts, treating plants with ethanol may improve rice growth and production under salinity stress conditions by enhancing photosynthesis and mitigating stress effects. Previous studies have shown that applying ethanol externally can effectively alleviate the negative effects of various abiotic stresses. For instance, ethanol has been found to help mitigate chilling stress in rice [41], drought stress in safflower [37], heat stress in tomato [42], and salt stress in soybean [38] and rice [43].

Potassium (K) is a crucial nutrient for plant photosynthesis because it helps regulate stomatal function, which in turn controls the opening and closing of stomata to balance the intake of CO_2 and the release of water vapor. A deficiency in K can reduce stomatal conductance, increase mesophyll resistance, decrease RuBPC/RuBPO activity, and lower photosynthetic CO_2 fixation, finally decreasing the net photosynthesis rate. Potassium also activates the ATP synthase enzyme, supporting photosynthesis [44,45]. Therefore, to improve plant photosynthesis efficiency and mitigate the negative impacts of salinity stress, the external application of K can be beneficial. Burbulis et al. [46] found that applying potassium bicarbonate ($KHCO_3$) to green foxtail leaves can help mitigate drought stress effects by improving photosynthetic parameters like gas exchange, effective quantum yield (ΦPSII), and maximum quantum yield (Fv/Fm) of photosystem II (PSII). Li and Hao [47] also found that applying exogenous $KHCO_3$ had a significant effect on the photosynthesis of eggplant under low CO_2 conditions.

Therefore, the aims of this research were to (1) examine the effects of foliar application of CO_2 supplementation compounds such as lithovit and ethanol, as well as compounds that enhance abiotic stress tolerance like potassium carbonate (K_2CO_3), on the photosynthetic efficiency and yield of rice grown in saline soil; and (2) identify the optimal timing for the application of these compounds to enhance various agro-physiological traits in two rice cultivars with different levels of salt tolerance.

2. Materials and Methods

2.1. Experimental Site and Agricultural Practices

The research was carried out at the El-Sirw Agricultural Research Station in Damietta Governorate, Egypt, in the summer of 2022 and 2023. The station is situated at coordinates 31°25′3.1440′′ N and 31°48′51.9984′′ E. The experimental area experiences an arid climate with no precipitation during the rice cultivation periods. The average minimum and maximum temperatures varied from 30.0 to 35.0 °C and 10.0 to 17.3 °C, respectively. The relative humidity in the morning and at midday ranged from 70.5 to 83.5% and 42.5 to 56.5%, respectively. The average evaporation ranged from 5.56 to 7.61 mm day^{-1}. Soil samples were collected from a depth of 0–30 cm before sowing for evaluation of physicochemical properties. The analysis results are presented in Table 1. Standard methods described by Cottenie et al. [48] and Burt [49] were employed for analyzing the soil's physiochemical properties.

Table 1. Physicochemical properties of the experimental saline soil during the 2022 and 2023 growing seasons.

Soil Properties		**2022**	**2023**	**Soil Properties**		**2022**	**2023**
Soil texture (%)	Clay	56.15	55.90	Cations content (meq L^{-1})	Ca^{2+}	8.3	8.5
	Silt	30.20	30.65		Mg^{2+}	10.10	10.2
	Sand	13.65	13.45		K^{+}	0.70	0.70
Organic matter (%)		1.00	0.90		Na^{+}	60.1	62.4
pH		8.20	8.30	Anions content (meq L^{-1})	HCO_3^{-}	9.24	9.74
ECe (dS m^{-1})		7.90	8.20		Cl^{-}	61.5	60.9
Available ion content (mg kg^{-1})	N	30.0	29.0		SO_4^{2-}	9.03	11.03
	P	11.0	10.2	Microelements content (ppm)	Fe^{2+}	5.23	5.15
	K	400	380		Zn^{2+}	0.90	0.82
					Mn^{2+}	4.60	4.50

The seeds of the salt-sensitive genotype Giza178 and the salt-tolerant genotype Giza179 were sown in the nursery bed on May 1st and 3rd in the first and second growing seasons, respectively, at a seeding rate of 170 kg ha^{-1}. The pre-germinated seeds were evenly spread in the nursery bed to encourage early growth. When the permanent field was prepared, 30-day-old seedlings were transplanted into 10 m^2 plots with a 20 × 20 cm spacing. Phosphorus, nitrogen, and potassium fertilizers were applied at rates of 75 kg P_2O_5 ha^{-1}, 165 kg N ha^{-1}, and 57 kg K_2O ha^{-1} using single superphosphate (15% P_2O_5), urea (46.0% N), and potassium sulfate (48% K_2O), respectively. The phosphorus and potassium fertilizers were applied as a basal dose during field preparation, while the nitrogen fertilizer was applied in three equal doses at different growth stages: tilling stage, panicle initiation stage, and booting stage. Weed and disease control practices followed the recommended guidelines for rice cultivation in salt-affected soil. In both seasons, the previous crop was Egyptian clover (*Trifolium alexandrinum* L.).

2.2. Experimental Design and Treatments

The experiments were conducted using a randomized complete block design with a split-split plot arrangement and three replications. The main plots were assigned two genotypes (Giza178 and Giza179), the sub-plots were designated for two foliar application times (before heading and after heading), and the sub-sub-plots were randomly allocated with four treatments of CO_2 + K source compounds. The treatments for CO_2 + K source compounds consisted of foliar application with distilled water (untreated treatment), potassium carbonate (KC), ethanol (Eth), and Lithovite (Liv). Each KC, Eth, and Liv treatment, which purchased from Sigma Aldrich, St. Louis, MO, USA, was applied at a concentration of 30 mM. The Lith compound consisted of nanoparticles of magnesium carbonate (4.62%), calcium carbonate (79.19%), iron (0.75%), zinc (0.005%), and selenium dioxide (11.41%). The treatments were applied using a 16 L pressurized backpack sprayer with 0.2% Tween-20 added to improve leaf adhesion. The sprayer nozzle was positioned 40 cm above the plants to ensure thorough coverage of all above-ground parts of the rice plant. The various solutions were carefully sprayed onto the rice plant leaves until they reached the point of run-off.

2.3. Measured Parameters

2.3.1. Physiological Parameters

After 10 days of foliar application of CO_2 + K source compounds post-heading treatment, we randomly selected five fully expanded uppermost leaves from each experimental unit to measure the contents of chlorophyll-a (Chl a), chlorophyll-b (Chl b), and carotenoids (Car). Fresh samples weighing 400 mg were collected from the leaves and immersed in 7 mL of 80% acetone in the dark at room temperature until the samples were decolorized. The resulting solution was then filtered through Whatman No. 4 filter paper, centrifuged, and adjusted to a final volume of 15 mL with 80% acetone. The absorbance of the extracted sample was measured at 470 nm for Car, and at 645 nm and 663 nm for Chl a and Chl b using a spectrophotometer (Spectro UV-2550, Shimadzu, Tokyo, Japan). The content of Chla, Chl b, and Car (mg g^{-1} fresh weight) was calculated using the formulas described by Arnon [50] and Lichtenthaler and Wellbura [51].

Photosynthetic parameters were measured on the second fully expanded leaf from the top of five plants per experimental unit using a handheld Photosynthesis System (LI-6400XT, LiCor Inc., Lincoln, NE, USA) between 10:00 a.m. and 12:30 p.m. The parameters measured included net photosynthetic rate (*A*), stomatal conductance (*gs*), transpiration rate (*E*), and intrinsic water use efficiency (*WUEi*). The leaf chamber settings included a CO_2 concentration of 400 μmol mol^{-1}, a leaf temperature of 25 °C, and a leaf-to-air vapor-pressure difference of 0.7 kPa.

The leaf relative water content percentage (RWC%) of the second fully expanded leaf from the top was determined in five plants per experimental unit using the formula:

$$\mathrm{RWC} = (\mathrm{FW} - \mathrm{DW})/(\mathrm{TW} - \mathrm{DW}) \times 100, \tag{1}$$

where FW represents the fresh weight (g) of a 10 cm^2 leaf area, TW is the turgid weight (g) after soaking in water for 24 h at room temperature, and DW is the dry weight (g) after drying at 75 °C for 72 h.

2.3.2. Agronomic Parameters

Following the maturity stage, measurements were conducted on 10 randomly chosen hills from each experimental unit. Plant height (PH), panicle length (PL), and panicle number (PN) per hill were documented. Furthermore, 10 panicles were randomly gathered from each experimental unit to measure panicle weight (PW), the number of filled grains per panicle (NFG), the number of unfilled grains per panicle (NUFG), and thousand-grain weight (TGW). Grain yield (GY) and biological yield (BY) were assessed in a 6 m^2 plot. BY was determined through manual harvesting of the area, air-drying the plants, and weighing them. The grains were separated from the plants, cleaned, dried to 14% moisture content, and weighed to determine GY. The harvest index (HI) was calculated by dividing GY by BY after converting both values to tons per hectare (t ha^{-1}).

2.4. Statistical Analysis

The data were initially assessed for normality to detect and eliminate any outliers. Subsequently, a homogeneity test of variance (Bartlett test) was performed for each parameter to determine if the results from both seasons could be merged for analysis. An analysis of variance (ANOVA) was carried out for each measured parameter at a significance level of 5%. The impacts of the season (S), genotype (G), foliar application timing (FT), and CO_2 + K source compounds (CO_2 + K) were examined individually, along with potential interactions between them, including two-way and three-way interactions. Subsequently, Tukey's post hoc test was employed to compare multiple treatment means with a significance level of $p \leq 0.05$. Pearson correlation analyses were carried out to examine relationships among all variables. Statistical analysis was conducted using SAS-statistical software packages-version 9.3 (SAS Institute, Inc., Cary, NC, USA). Principal component analyses (PCA) and heatmap clustering analyses were conducted to explore the relationships between different agro-physiological parameters and experimental treatments and explain the main sources of variation among variables. PCA and heatmap were conducted using R Studio 1.4.1717 software (RStudio, Boston, MA, USA; available at: http://www.rstudio.org/, accessed on 31 October 2022). Standard errors of the mean were calculated using Sigma Plot 13.0 (Systat Software, Inc., San Jose, CA, USA) and are represented in the Figures as error bars.

3. Results

3.1. Impacts of Experimental Factors on Different Parameters

3.1.1. Pigment Contents

The analysis of variance showed significant effects of individual factors (S, G, FT, and CO_2 + K) on pigment contents (Chla, Chlb, Chlt, and Car) at $p \leq 0.05$, 0.01, and 0.001. Two-way interactions between the main factors of study (G × FT, G × CO_2 + K, and FT × CO_2 + K) also significantly impacted pigment contents. However, interactions between season and the three main factors did not significantly impact pigment contents (Table S1).

Regarding genotypes, Giza 179 had higher pigment contents than Giza 178. On average, over two seasons, Chla, Chlb, Chlt, and Car were 7.9%, 13.9%, 10.1%, and 14.8% higher in Giza 179 than in Giza 178, respectively (Figure 1). Applying different CO_2 + K source compounds to the leaves before heading, regardless of the genotypes, led to a significant increase in all pigment contents compared to the application after heading. Over two seasons, foliar application of CO_2 + K source compounds before heading resulted in significantly higher levels of Chla, Chlb, Chlt, and Car contents by 6.1%, 16.1%, 9.9%, and 13.9%, respectively, compared to those applied after heading (Figure 1). The foliar application of different CO_2 + K source compounds significantly increased pigment contents in plants compared to untreated plants, regardless of genotypes and application time. The highest pigment values were observed with Lith application, followed by Eth and KC. Over two seasons, Chla, Chlb, Chlt, and Car contents increased by 25.1%, 19.6%, 23.1%, and 29.6% with Lith application, 20.6%, 14.4%, 18.3%, and 23.7% with Eth application, and 18.2%, 10.3%, 15.3%, and 18.7% with KC application, respectively, compared to the control treatment (Figure 1).

Figure 2 shows how various pigment contents respond to the interactions between the main study factors (G × FT, G × CO_2 + K, and FT × CO_2 + K). The highest pigment contents were observed in Giza 179 when treated with foliar CO_2 + K source compounds before heading, as well as when treated with Lith or Eth. Initially, Giza 178 had lower pigment content than Giza 179. However, the pigment content of Giza 178 could be improved by treating it with foliar CO_2 + K source compounds before heading (not after heading) and with Lith. With these treatments, the pigment content of Giza 178 became comparable to that of Giza 179. Moreover, applying different sources of CO_2 + K before heading was more effective in enhancing pigment content compared to applying them after heading. However, there was no significant difference in pigment content when using the three CO_2 + K sources (Lith, Eth, and KC) either before or after heading. The control treatment consistently showed the lowest pigment content values, regardless of genotype or timing of foliar application (Figure 2).

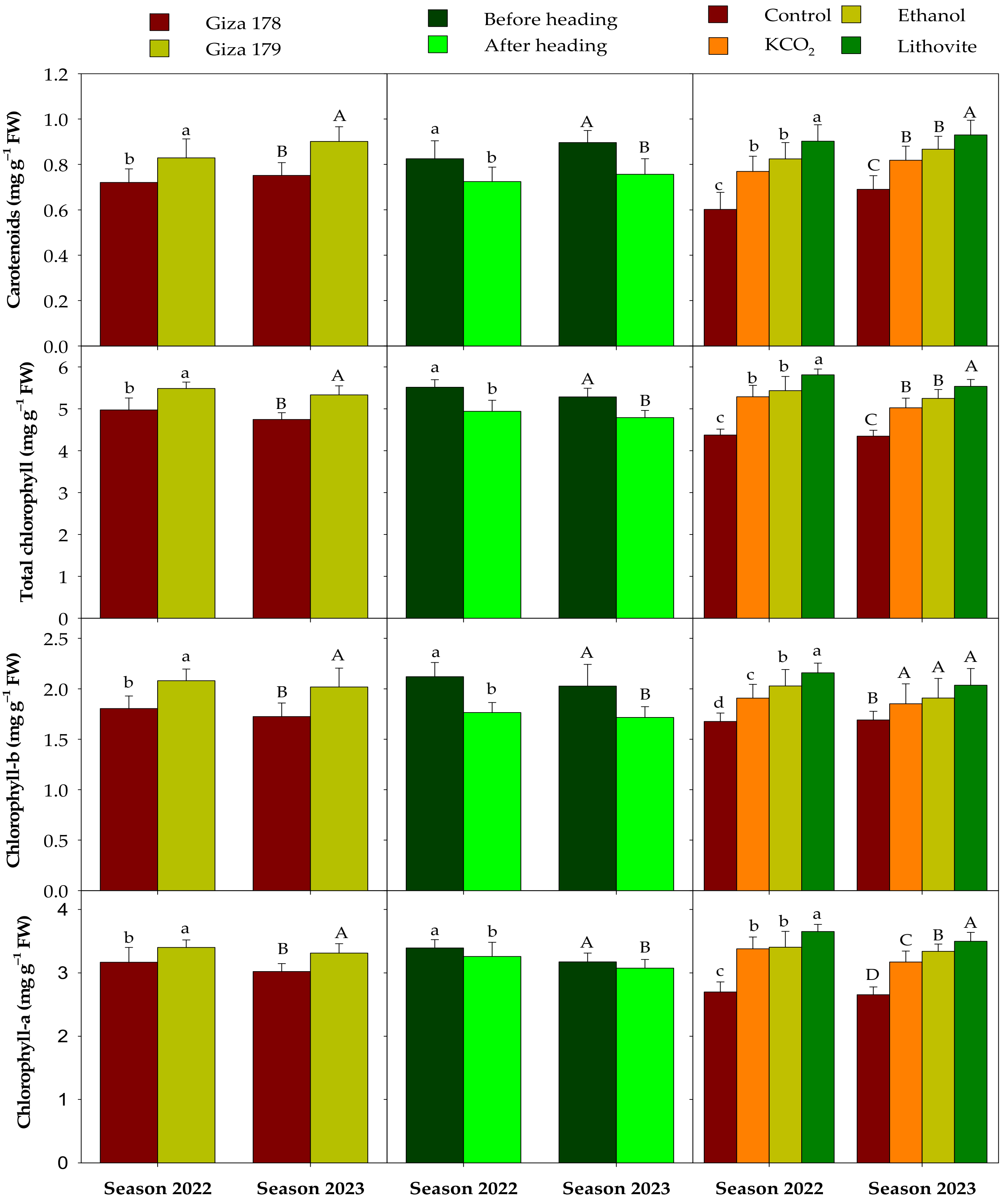

Figure 1. Influences of genotype, foliar application timing, and foliar CO_2 + K source compounds on different pigment contents in two seasons (n = 3 ± SE). The letters and bars on the top of each column represent significant differences at the 0.05 level based on Tukey's HSD test and standard error (SE), respectively. The lowercase letters belong to the first season, and the uppercase letters belong to the second season.

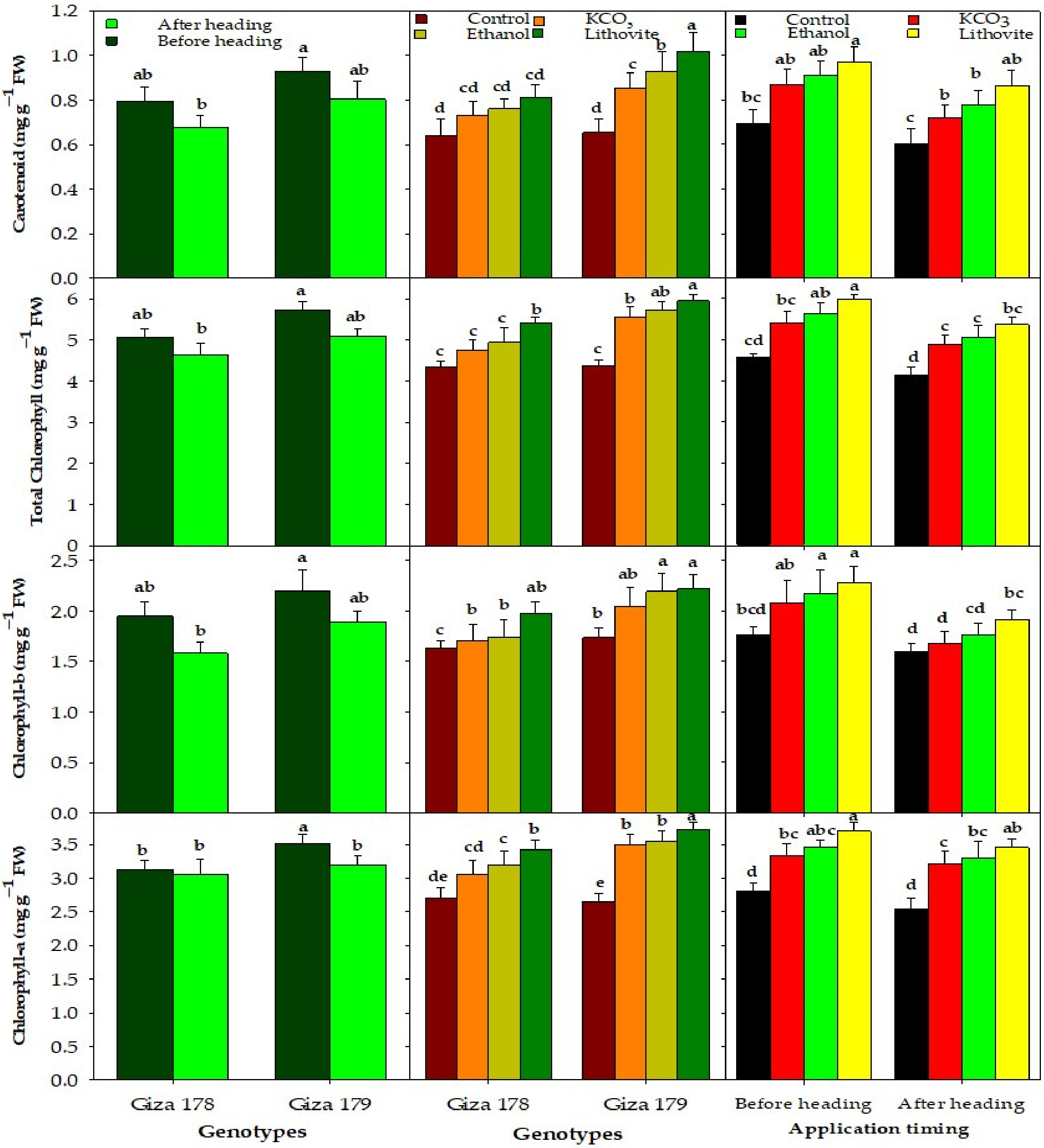

Figure 2. Influences of interaction between genotypes and foliar application timing, genotypes, and foliar CO_2 + K source compounds, as well as between foliar application timing and foliar CO_2 + K source compounds on different pigment contents (n = 6 ± SE). The letters and bars on the top of each column represent significant differences at the 0.05 level based on Tukey's HSD test and standard error (SE), respectively.

3.1.2. Photosynthetic and Relative Water Content

The combined analysis of variance showed that the individual factors (S, G, FT, and CO_2 + K) had different impacts on various photosynthetic parameters and RWC. The season had significant effects on all photosynthetic parameters except *A* and RWC. Only *A* showed significant differences among genotypes in terms of photosynthetic parameters. FT and CO_2 + K significantly affected all photosynthetic parameters and RWC, except for E, which did not vary significantly among FT treatments (Table S1). The interactions between seasons and various factors (G, FT, CO_2 + K) did not significantly affect photosynthetic parameters and RWC. However, the interactions of G and FT (excluding *A*), G and CO_2 + K, and FT and CO_2 + K (excluding *E*) significantly affected these parameters. Furthermore, the interaction of all three factors (G, FT, and CO_2 + K) significantly impacted only E, while the interaction of all four factors (S, G, FT, and CO_2 + K) significantly affected only *gs* (Table S1).

Net photosynthetic rate (*A*) was the only parameter that exhibited a notable difference between the two genotypes. More specifically, *A* was 7.8% higher in Giza 179 than in Giza 178 (Figure 3). When various CO_2 + K source compounds were applied before heading, there was a significant increase in *A*, *gs*, *WUEi*, and RWC by 8.9%, 8.7%, 8.9%, and 5.2%, respectively, compared to when applied after heading, irrespective of the genotypes (Figure 3). The use of different CO_2 + K source compounds significantly improved various photosynthetic and RWC parameters compared to the control treatments. Lith and Eth were the most effective, followed by KC. In contrast, the control treatment did not effectively alleviate salinity stress on photosynthesis and RWC parameters, as it yielded the lowest values for these parameters. Over two seasons, the Lith, Eth, and KC treatments increased *A* by 37.9%, 32.0%, and 20.1%, *gs* by 29.8%, 22.3%, and 13.0%, E by 12.5%, 10.9%, and 0.5%, *WUEi* by 29.0%, 23.7%, and 19.8%, and RWC by 17.9%, 16.9%, and 12.0%, respectively, compared to the control treatment (Figure 3).

Figure 4 illustrates the response of photosynthetic and RWC parameters to the interactions between the main study factors (G × FT, G × CO_2 + K, and FT × CO_2 + K). The application of CO_2 + K source compounds before heading did not differ from applying them after heading for Giza 179. However, for Giza 178, applying the compounds before heading improved *A*, *gs*, *IWUEi*, and RWC by 8.5%, 11.6%, 13.9%, and 7.7%, respectively, and decreased *E* by 5.9% compared to applying them after heading. The photosynthetic and RWC parameters of different CO_2 + K source compounds for each G or FT were ranked in the order of Lith > Eth > KC > control. In most cases, these source compounds showed better performance with Giza 179 compared to Giza 178 and when applied before heading rather than after heading (Figure 4).

3.1.3. Grain Yield and Yield Components

The combined analysis of variance showed that the main factors of study (G, FT, and CO_2 + K) had different effects on grain yield (GY) and its various components. The CO_2 + K source compounds and FT factors had a significant impact ($p \leq 0.05$ and 0.001) on GY and its components, except for PL and TGW, which did not show significant differences among FT treatments. Genotypes exhibited highly significant variations ($p \leq 0.001$) for the NUFG, TGW, GY, and HI, while the other yield components did not show significant differences among genotypes (Table S1). The interactions between the three main study factors (G × FT, G × CO_2 + K, and FT × CO_2 + K) significantly impacted GY and its yield components. However, PH, PN, PL, NFG, and HI were not significantly affected by the G × FT interaction. Additionally, PH was not significantly affected by the G × CO_2 + K interaction (Table S1). The TGW, GY, and HI values were higher in Giza 179 than in Giza 178, while the NUFG showed the opposite trend. The other yield components were similar in both genotypes. Over two seasons, TGW, GY, and HI were 16.8%, 11.0%, and 6.9% higher in Giza 179, respectively, while the NUFG was 28.6% higher in Giza 178 (Figure 5).

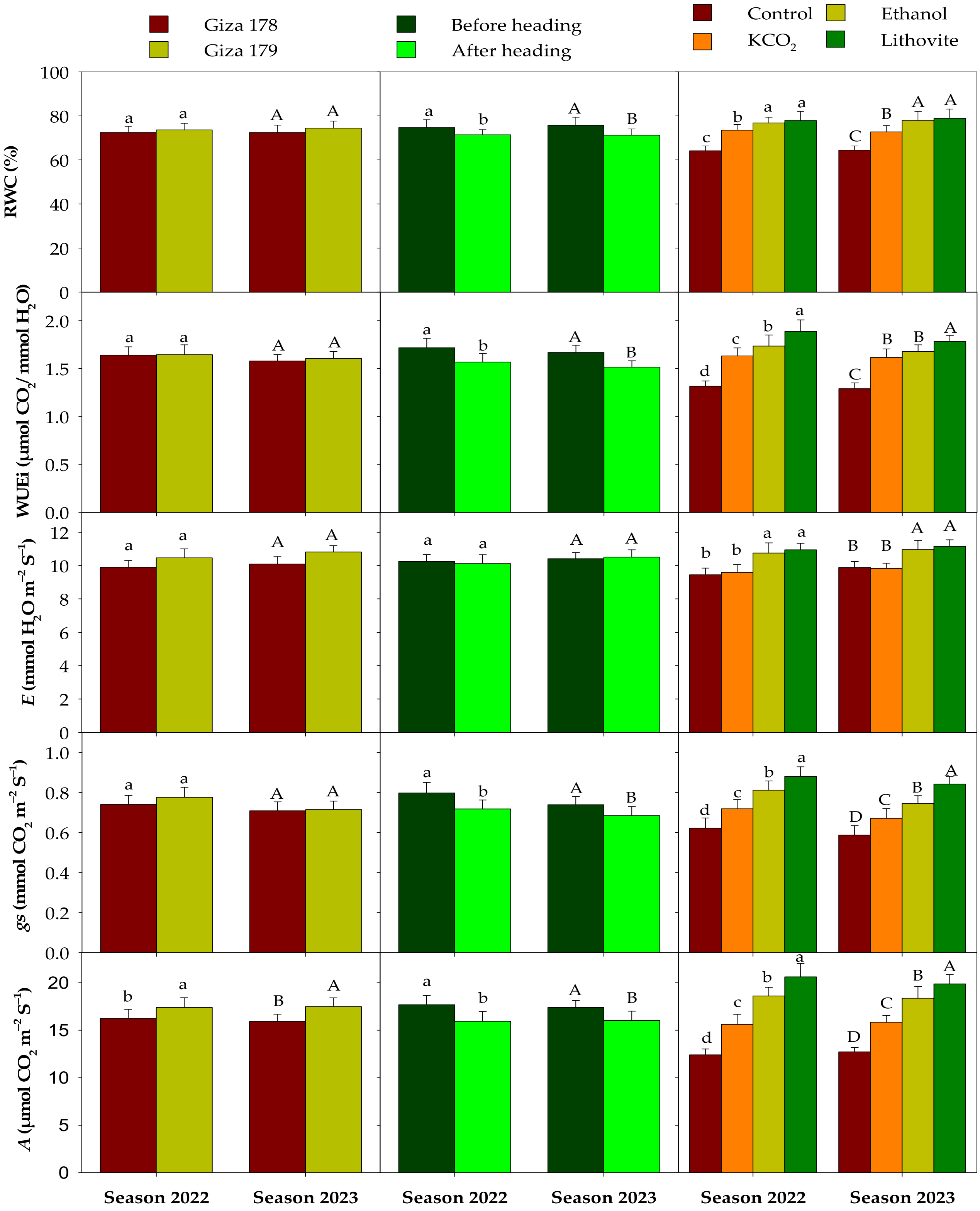

Figure 3. Influences of genotype, foliar application timing, and foliar CO_2 + K source compounds on net photosynthetic rate (*A*), stomatal conductance (*gs*), transpiration rate (*E*), intrinsic water use efficiency (*WUEi*), and relative water content (RWC) in two seasons (n = 3 ± SE). The letters and bars on the top of each column represent significant differences at the 0.05 level based on Tukey's HSD test and standard error (SE), respectively. The lowercase letters belong to the first season, and the uppercase letters belong to the second season.

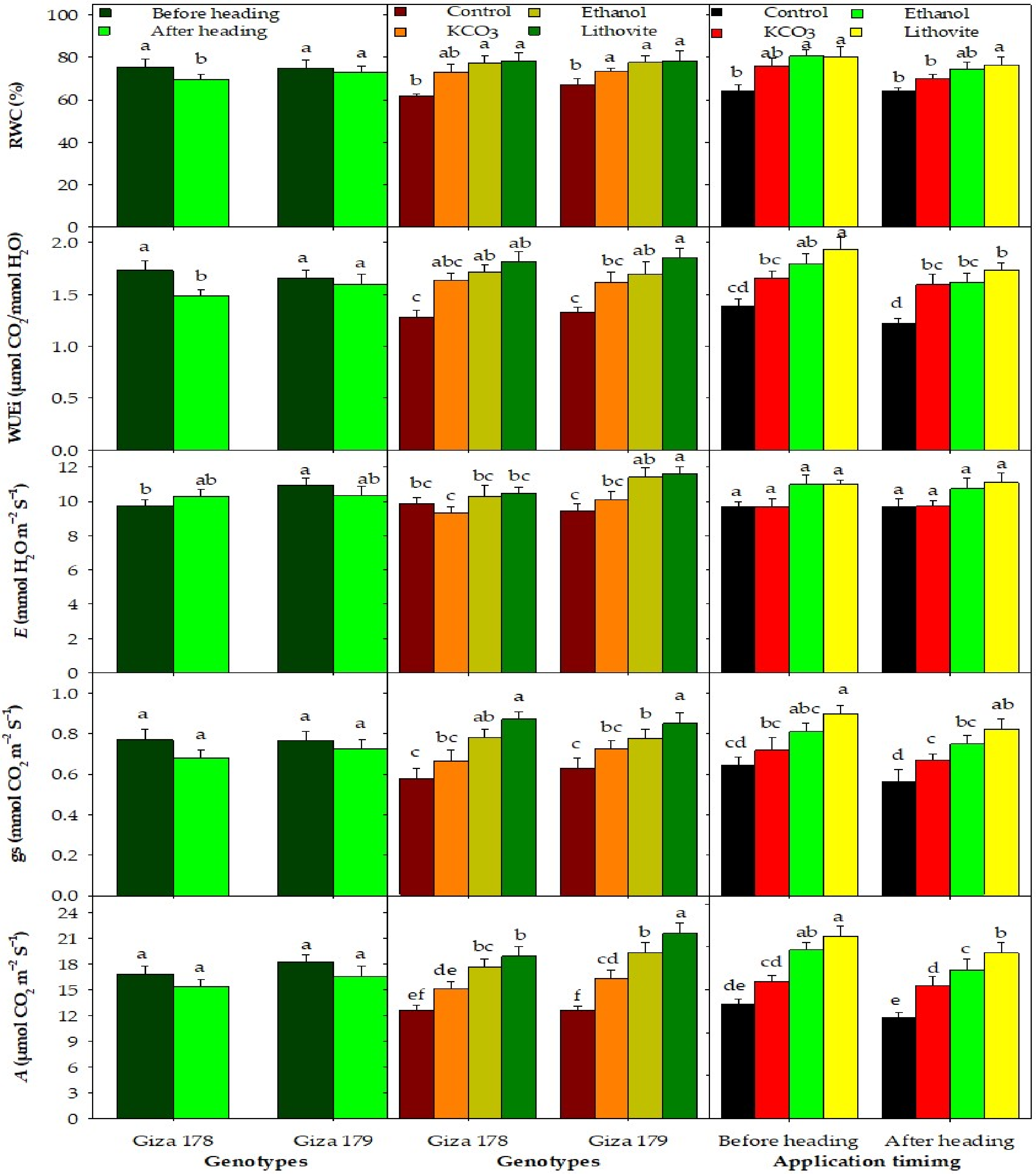

Figure 4. Influences of interaction between genotypes and foliar application timing, genotypes, and foliar CO_2 + K source compounds, as well as between foliar application timing and foliar CO_2 + K source compounds on net photosynthetic rate (*A*), stomatal conductance (*gs*), transpiration rate (*E*), intrinsic water use efficiency (*WUEi*), and relative water content (RWC) (n = 6 ± SE). The letters and bars on the top of each column represent significant differences at the 0.05 level based on Tukey's HSD test and standard error (SE), respectively.

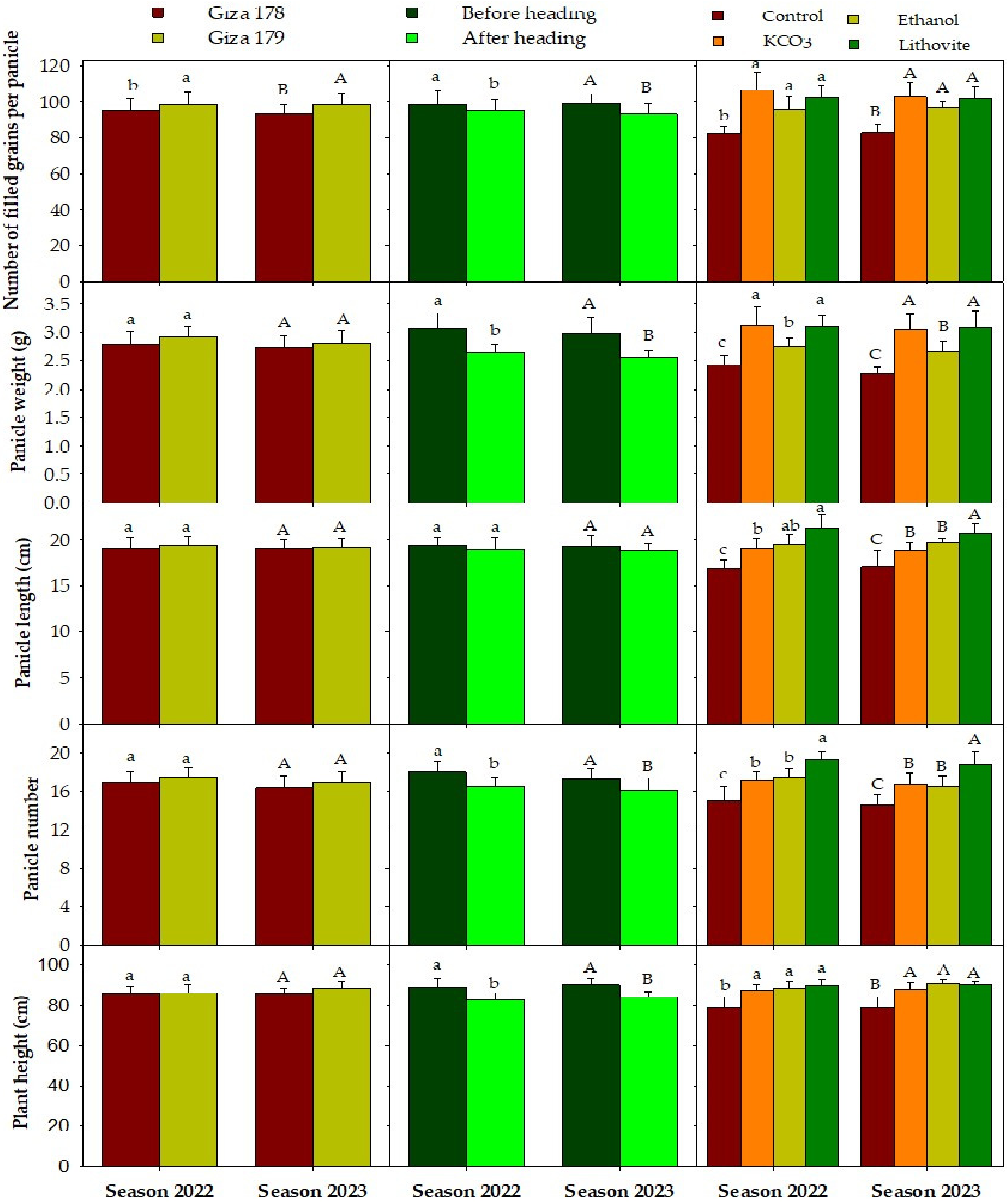

Figure 5. Influences of genotype, foliar application timing, and foliar CO_2 + K source compounds on different yield components in two seasons (n = 3 ± SE). The letters and bars on the top of each column represent significant differences at the 0.05 level based on Tukey's HSD test and standard error (SE), respectively. The lowercase letters belong to the first season, and the uppercase letters belong to the second season.

In terms of timing for applying CO_2 + K source compounds, it was found that the GY and yield components were higher when the compounds were applied before heading rather than after heading. However, the NUFG showed the opposite trend. Over two seasons, applying CO_2 + K source compounds before heading resulted in improvements in PH, PN, PW, NFG, GY, BY, and HI by 6.8%, 7.4%, 14.0%, 4.8%, 14.0%, 6.3%, and 8.0%, respectively, while decreasing the NUFG by 22.9% compared to applying them after heading (Figures 5 and 6). When comparing different CO_2 + K source compounds, all compounds improved GY and its components compared to the control treatment. Lith was the most effective among the compounds tested, followed by KC and Eth. Over two seasons, the foliar application of Lith, KC, and Eth significantly improved PH by 12.0%, 9.7%, and 11.6%, PN by 22.3%, 12.7%, and 13.0%, PL by 19.1%, 10.1%, and 13.4%, PW by 23.9%, 23.7%, and 13.1%, NFG by 19.0%, 21.2%, and 14.1%, TGW by 22.4%, 23.1%, and 15.5%, GY by 32.5%, 24.9%, and 23.5%, BY by 13.0%, 11.7%, and 11.2%, and HI by 22.3%, 15.2%, and 13.9%, while decreased the NUFG by 42.0%, 43.2%, and 34.6%, respectively, compared to control treatment (Figures 5 and 6).

Figures 7 and 8 show the response of GY and its components to the interactions between the main study factors (G × FT, G × CO_2 + K, and FT × CO_2 + K). Generally, applying CO_2 + K source compounds before heading had a more positive effect on GY and most of its components than applying them after heading in both genotypes. However, applying CO_2 + K source compounds before heading had a more positive effect on PH, PN, GY, BY, and HI in Giza 179 compared to Giza 178, while the NFG and NUFG showed the opposite trend. For example, in Giza 179, there was a significant increase in PH by 8.4%, PN by 8.5%, GY by 19.3%, BY by 7.6%, and HI by 12.5%. On the other hand, in Giza 178, the same treatment resulted in a smaller increase in PH (5.1%), PN (6.3%), GY (7.6%), BY (5.0%), and HI (2.9%). However, the NFG increased by 2.7% and 7.0%, while the NUFG decreased by 13.7% and 28.9% in Giza 179 and Giza 178, respectively (Figures 7 and 8). Over two seasons, different CO_2 + K source compounds led to significant increases in various parameters in both genotypes and application times. For instance, there was a 10.0–15.0% and 9.0–10.0% increase in PH, a 9.0–19.8% and 16.3–24.7% increase in PN, an 11.7–20.3% and 8.6–18.0% increase in PL, a 13.1–24.5% and 13.1–24.2% increase in PW, a 13.8–21.8% and 14.4–21.7% increase in the NFG, a 15.6–25.2% and 15.6–24.3% increase in TGW, a 25.6–35.4% and 19.3–30.0% increase in GY, a 10.0–12.4% and 12.0–13.6% increase in BY, and a 16.5–26.2% and 7.6–18.6% increase in HI. Additionally, there was a decrease in the NUFG by 30.7–44.8% and 40.1–41.0% in Giza 178 and Giza 179, respectively, compared to the control treatment. Similarly, the application of different CO_2 + K source compounds resulted in a 10.6–12.7% and 8.7–11.3% increase in PH, 14.3–23.8%, and 8.4–20.8% increase in PN, 11.9–22.4% and 8.4–15.8% increase in PL, 15.2–30.6% and 10.8–17.4% increase in PW, 12.4–19.9% and 15.8–22.2% increase in the NFG, 16.0–23.5% and 15.1–23.8% increase in TGW, 24.2–33.3% and 22.6–31.6% increase in GY, 12.3–14.5% and 10.0–11.4% increase in BY, and 12.7–21.9% and 14.0–22.8% increase in HI. Additionally, there was a decrease in the NUFG by 45.3–49.8% and 25.1–33.7% when applying these compounds before and after heading, respectively (Figures 7 and 8).

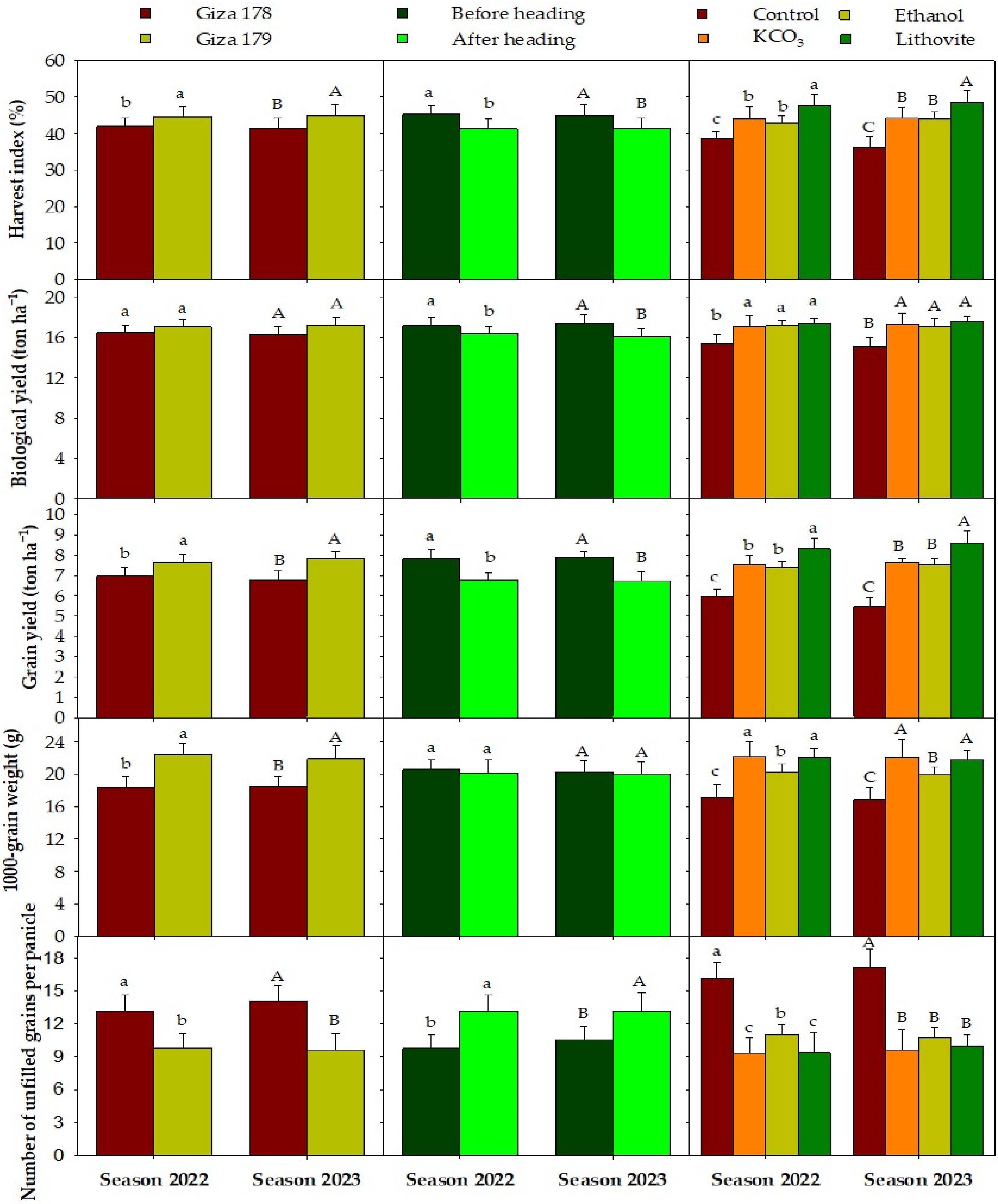

Figure 6. Influences of genotype, foliar application timing, and foliar CO_2 + K source compounds on grain yield and other yield components in two seasons (n = 3 ± SE). The letters and bars on the top of each column represent significant differences at the 0.05 level based on Tukey's HSD test and standard error (SE), respectively. The lowercase letters belong to the first season, and the uppercase letters belong to the second season.

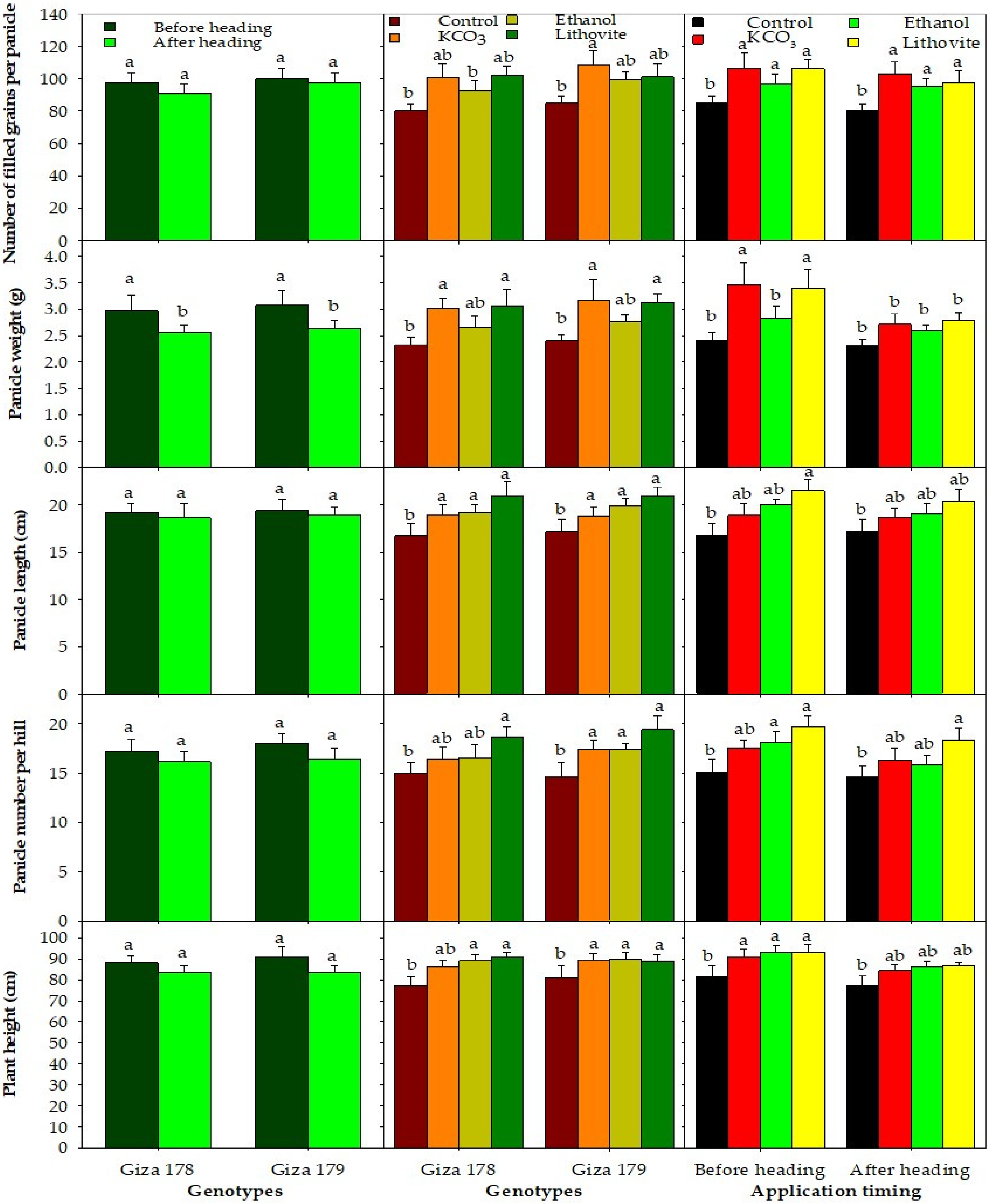

Figure 7. Influences of interaction between genotypes and foliar application timing, genotypes, and foliar CO_2 + K source compounds, as well as between foliar application timing and foliar CO_2 + K source compounds on different yield components (n = 6 ± SE). The letters and bars on the top of each column represent significant differences at the 0.05 level based on Tukey's HSD test and standard error (SE), respectively.

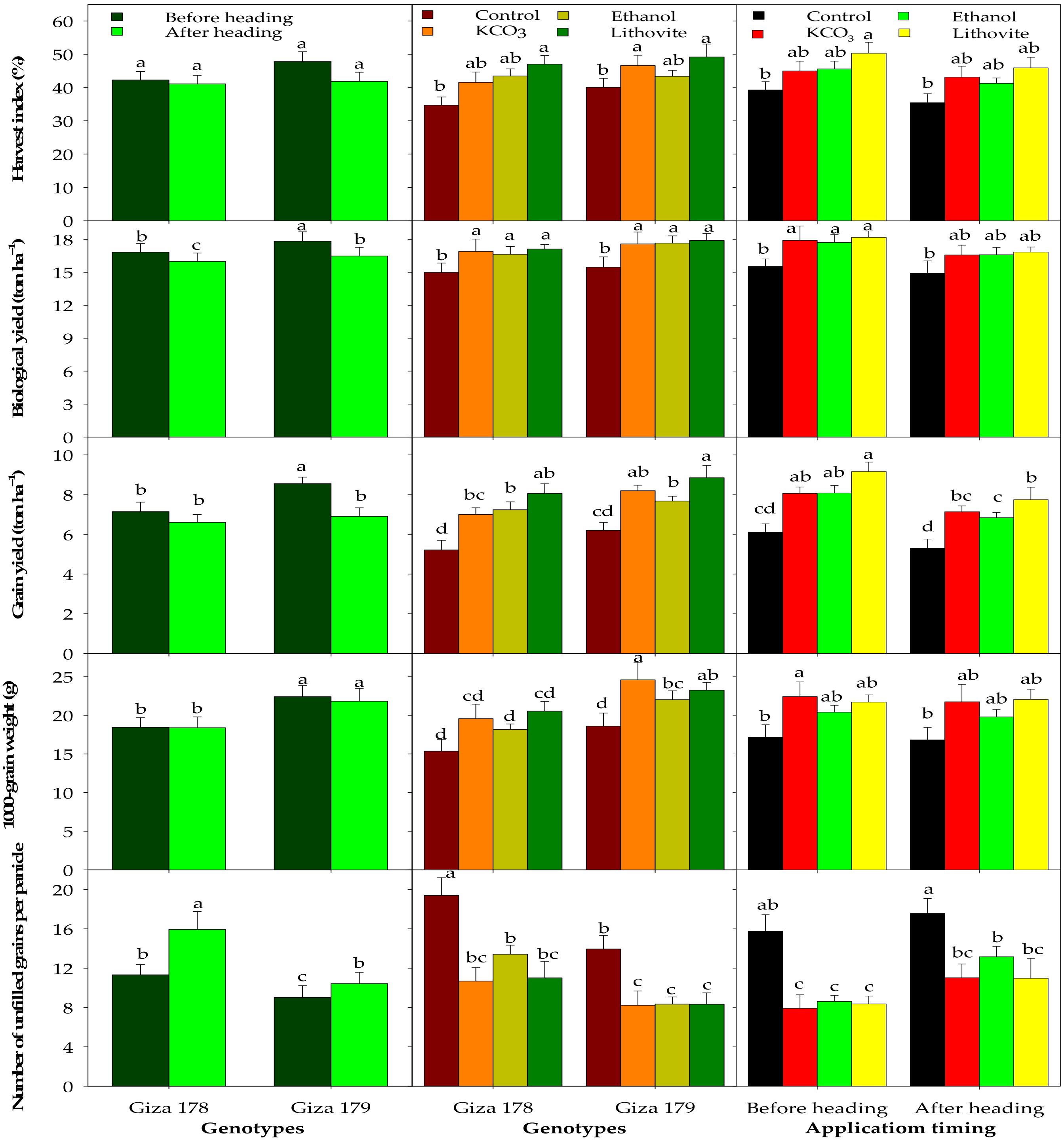

Figure 8. Influences of interaction between genotypes and foliar application timing, genotypes, and foliar CO_2 + K source compounds, as well as between foliar application timing and foliar CO_2 + K source compounds on grain yield and other yield components (n = 6 ± SE). The letters and bars on the top of each column represent significant differences at the 0.05 level based on Tukey's HSD test and standard error (SE), respectively.

3.2. Relationships Between Different Parameters for Each Experimental Factor

A Pearson correlation analysis was performed to investigate the relationships between all parameters for each experimental factor, as shown in Figures 9 and 10. In both genotypes, most parameters were strongly correlated with each other, except for *E*, which did not show significant correlations with other parameters in Giza 178. However, *E* showed strong correlations with pigment contents (r = 0.84–0.94), *A* and RWC parameters (r = 0.72–0.96), GY (0.74), BY (0.82), and certain yield components such as PH (0.72), PN (0.87), PL (0.92), and the NUFG (−0.76) in Giza 179. Additionally, in Giza 178, the NGF showed strong correlations with all parameters (r = 0.74–0.93) except Chl b and *E*. In Giza 179, it exhibited strong correlations only with Chla (0.84), Chlt (0.78), PN (0.72), PW (0.81), NUFG (−0.89), TGW (0.99), GY (0.74), and BY (0.78). Furthermore, in both genotypes, PW showed strong correlations with pigment contents (r = 0.76–0.84) and GY and its components (r = 0.75–0.93). However, it did not show a significant correlation with photosynthetic and RWC parameters, except for WUEi and RWC, which showed a significant correlation with PW only in the Giza 178 genotype (Figure 9).

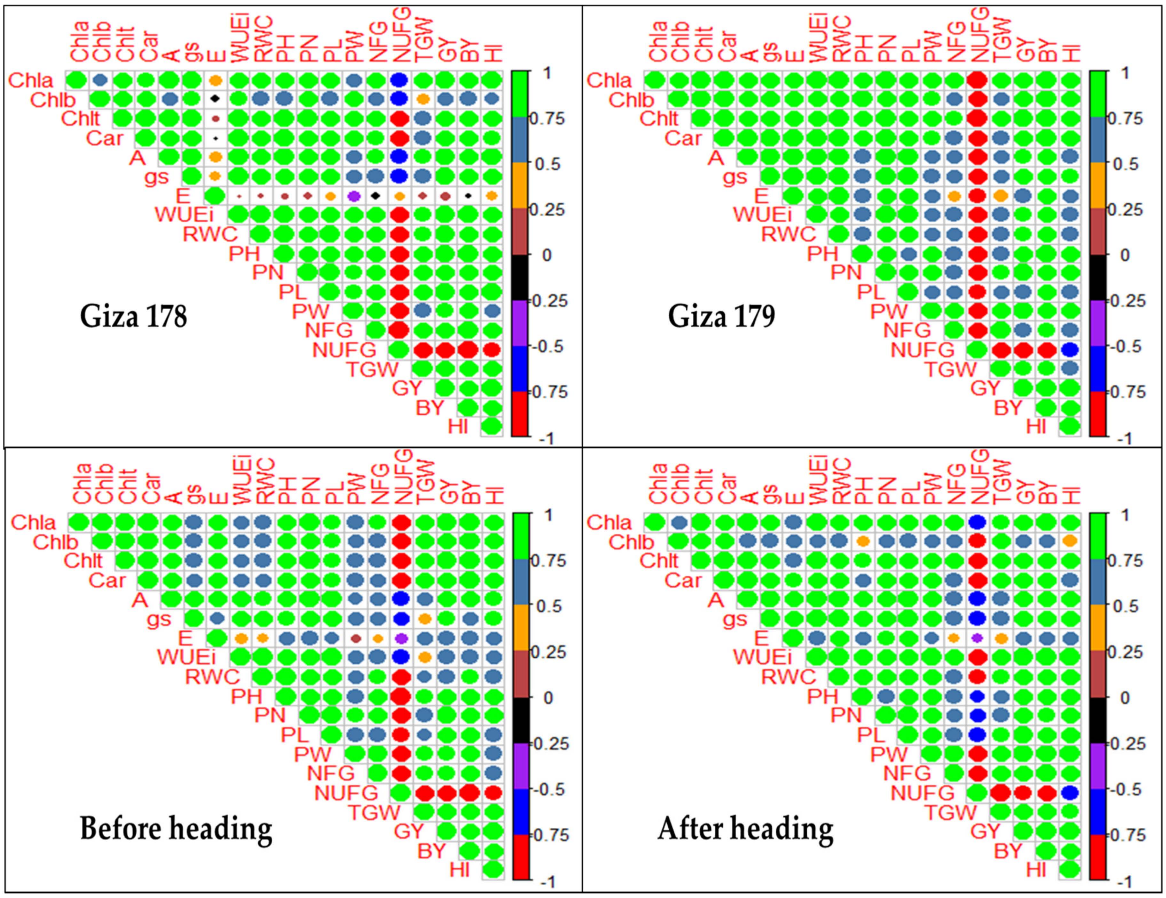

Figure 9. Pearson's correlation matrix between different parameters across two seasons in genotypes (Giza 178 and Giza 179) and application time of CO_2 + K source compounds (before and after heading) factors. The different abbreviations used in this figure are as follows: Chla = chlorophyll-a; Chlb = chlorophyll-b; Chlt = total chlorophyll; Car = carotenoid; *A* = Net photosynthetic rate; *gs* = Stomatal conductance; *E* = Transpiration rate; *WUEi* = Intrinsic water use efficiency; RWC = relative water content; PH = plant height; PN = panicle number per plant; PL = panicle length; PW = panicle weight; NFG = number of filled grains per panicle; NUFG = the number of unfilled grains per panicle; TGW = thousand-grain weight; GY = grain yield per ha; BY = biological yield per ha; and HI = harvest index.

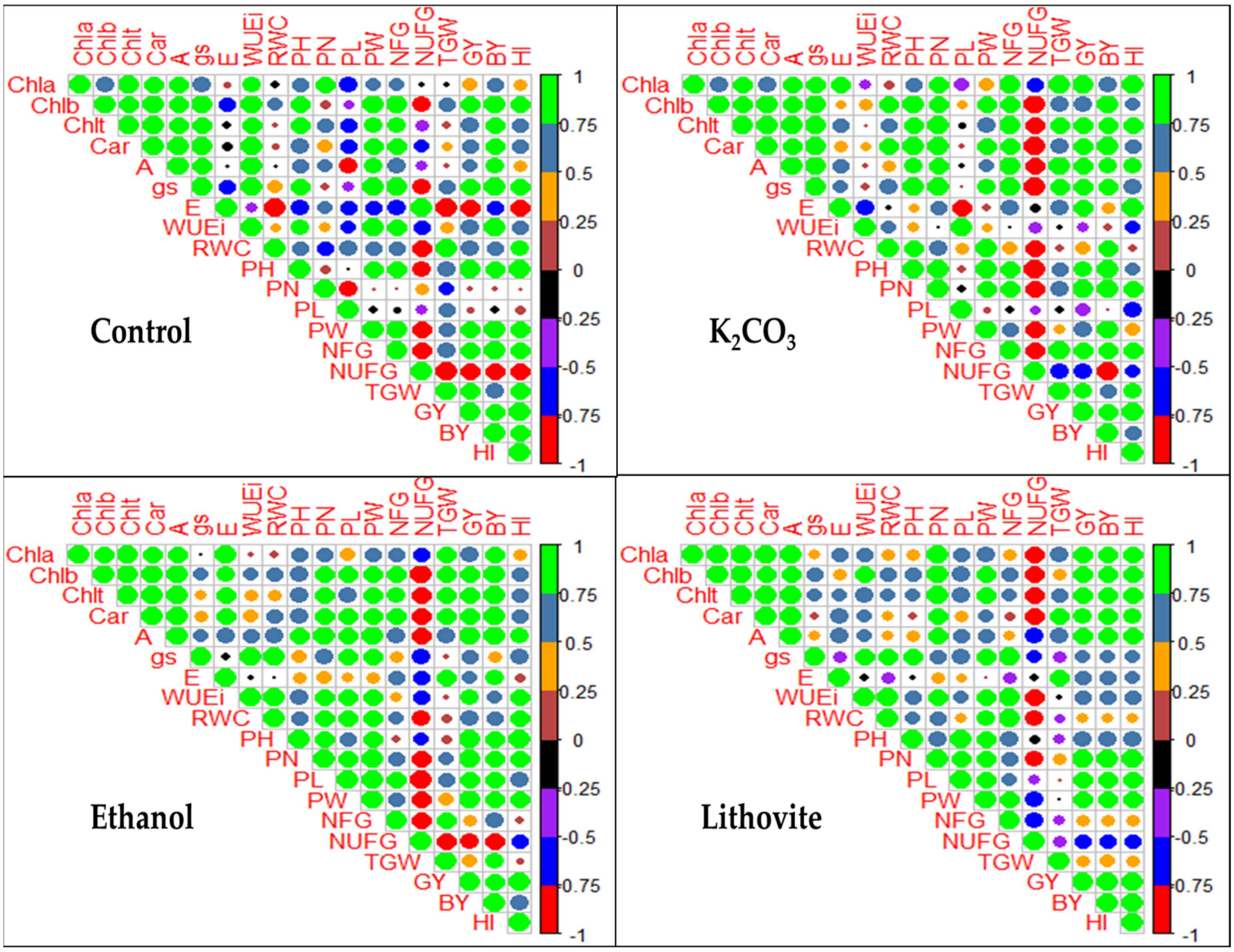

Figure 10. Pearson's correlation matrix between different parameters across two seasons in different CO_2 + K source compounds factor. The different abbreviations used in this figure are as follows: Chla = chlorophyll-a; Chlb = chlorophyll-b; Chlt = total chlorophyll; Car = carotenoid; *A* = Net photosynthetic rate; *gs* = Stomatal conductance; *E* = Transpiration rate; WUEi = Intrinsic water use efficiency; RWC = relative water content; PH = plant height; PN = panicle number per plant; PL = panicle length; PW = panicle weight; NFG = number of filled grains per panicle; NUFG = number of unfilled grains per panicle; TGW = thousand-grain weight; GY = grain yield per ha; BY = biological yield per ha; and HI = harvest index.

The results in Figure 9 indicate strong correlations among all parameters in both the application time of CO_2 + K source compounds (before and after heading), with a few exceptions. For instance, *E* did not show significant correlations with GY and its components in both application times, except for GY in the before heading application time and PN and PL in the after heading application time, which showed correlations with *E*. Additionally, *E* exhibited strong correlations with pigment contents and *A* in both application times and with gs and RWC only in the after-heading application time. The NUFG did not show significant correlations with *A*, *gs*, and *E* in both application times. At the same time, it displayed a strong negative correlation with PH (−0.97), PN (−0.82), and PL (−0.78) when CO_2 + K source compounds were applied before heading, not after heading. PW, NFG, and TGW did not show significant correlations with photosynthetic parameters (*A*, *gs*, *E*, and *WUWi*) when CO_2 + K source compounds were applied before heading. However, when applied after heading, they exhibited strong correlations with A (r = 0.73–0.84) and *WUEi* (r = 0.87–0.95) (Figure 9).

The strength correlations between different parameters also depended on the various CO_2 + K source compounds. Generally, the most studied parameters were strongly correlated with each other in Ethanol and KC treatments, followed by Lithovite. However, few parameters showed a strong correlation with each other in the control treatment (Figure 10). Various pigment contents and photosynthetic parameters were strongly correlated with GY and most yield components in treatments treated with different CO_2 + K source compounds. Only Chlb, *gs*, and *E* showed strong correlations with GY in the control treatment. In the control treatment, the various pigment contents and photosynthetic parameters were more strongly correlated with the NFG than the NUFG. In contrast, the opposite trend was observed in the Lithovite treatment. However, the Ethanol and KC treatments showed strong correlations with both the NGF and the NUFG (Figure 10).

3.3. An Overview of the Association Between Various Agro-Physiological Parameters and Experimental Factors

The performance of agro-physiological parameters under different interactions between three experimental factors (genotypes, application time, and CO_2 + K source compounds) was visualized through heatmap clustering (Figure 11a). The heatmap categorized the agro-physiological parameters into four clusters and the different experimental treatments into three clusters. *E* and the NUFG were in separate clusters (cluster C and cluster D, respectively). Pigment contents, except Chla, NGF, PW, and TGW, were grouped together in cluster B. The remaining parameters, including GY and most photosynthetic parameters and yield components, were grouped together in cluster A. The treatments involving Liv application before heading for both genotypes (Liv-B-V1 and Liv-B-V2) and Eth or KC application before heading for Giza 179 (Eth-B-V2 and KC-B-V2) were grouped together in group II. This group showed the highest values for most parameters of cluster A and cluster B and the lowest values for the NUFG. The value of *E* in this group was higher in Eth-B-V2 and Liv-B-V2 and moderate in Liv-B-V1 and KC-B-V2. On the other hand, the four control treatments (C-B-V1, C-B-V2, C-A-V1, and C-A-V2) and treatments involving Eth or KC application after heading for Giza 178 (Eth-A-V1 and KC-A-V1) were grouped in group I. Interestingly, group I displayed a reverse trend compared to group II, with the lowest values for cluster A, cluster B, and cluster C parameters and the highest values for the NUFG. The treatments that involved applying Liv after heading for both genotypes (Liv-A-V1 and Liv-A-V2) and applying Eth or KC before heading to Giza 178 (Eth-B-V1 and KC-B-V1) or after heading to Giza 179 (Eth-A-V2 and KC-A-V2) were combined in Group III. This group displayed intermediate values for all parameters compared to Group 1 and Group 2 treatments (Figure 11a).

The association between different agro-physiological parameters and experimental treatments was examined using principal component analysis (PCA) (Figure 11b). The PCA biplot shows that the first component (PC1) and a second component (PC2), which have eigenvalues greater than one, explain most of the total parameter variability, accounting for 80.1% and 6.5% of the total variability, respectively. Additionally, the PCA biplot clearly differentiates between all parameters and experimental treatments. PC1 is strongly and positively correlated with all parameters except the NUFG, which exhibits a negative relationship with PC2. Notably, the physiological parameters, such as chlorophyll contents and photosynthetic parameters, were closely associated with treatments involving Liv application before heading for Giza 179 (Liv-B-V2) or after heading for both genotypes (Liv-A-V1 and Liv-A-V2), as well as Eth application before heading for both genotypes (Eth-B-V1 and Eth-B-V2) and after heading for Giza 178 (Eth-A-V1). On the other hand, grain yield (GY) and its components, except the NUFG and RWC, were closely linked to the Liv application before heading for Giza 178 (Liv-B-V1) and KC application before heading for both genotypes (KC-B-V1 and KC-B-V2) and after heading for Giza 179 (KC-A-V2) (Figure 11b).

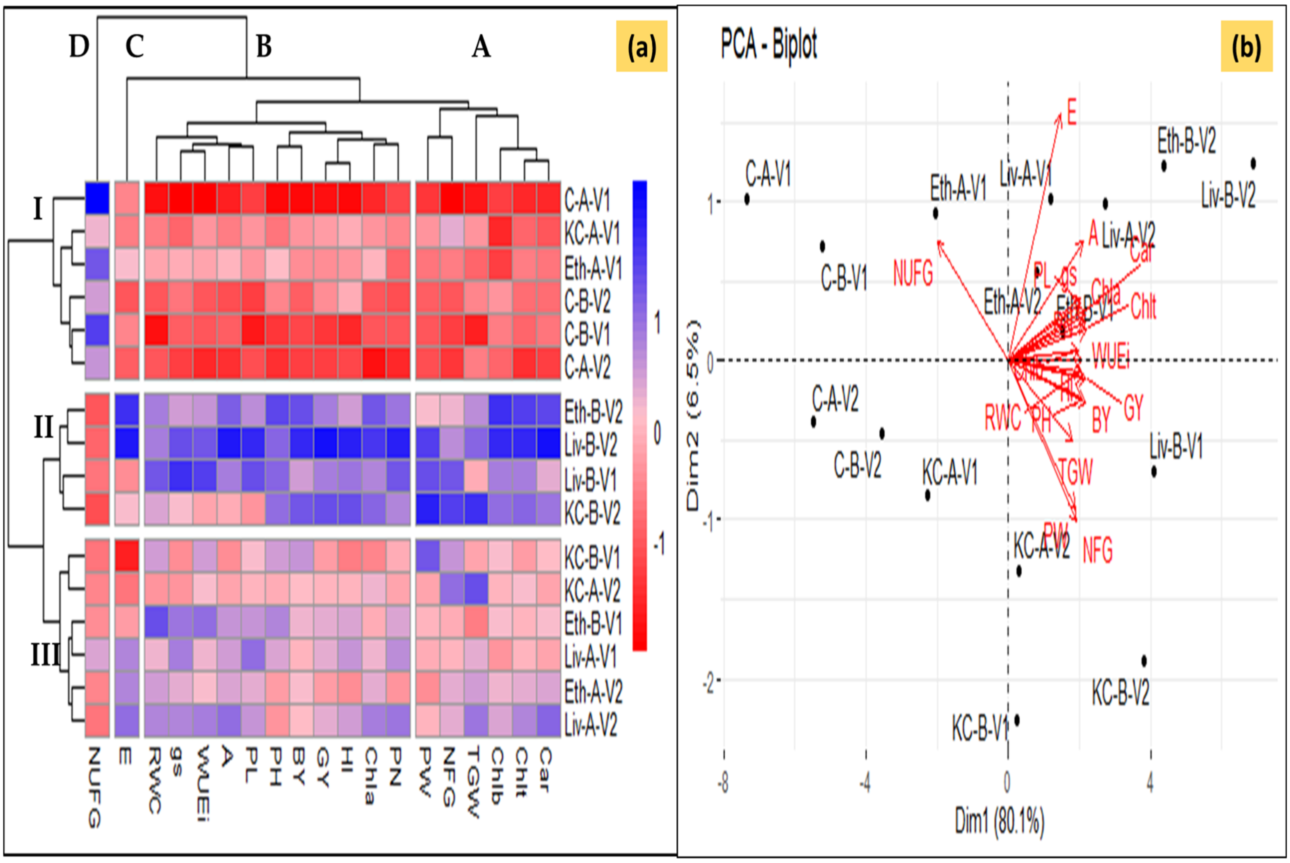

Figure 11. (**a**) Clustering heatmap categorizing different agro-physiological parameters and various experimental treatments for easy visualization. (**b**) Biplot of the first two components of principle component analysis shows the relationships between different agro-physiological parameters and experimental treatments. The different abbreviations used in this figure are as follows: Chla = chlorophyll-a; Chlb = chlorophyll-b; Chlt = total chlorophyll; Car = carotenoid; A = Net photosynthetic rate; gs = Stomatal conductance; E = Transpiration rate; WUEi = Intrinsic water use efficiency; RWC = relative water content; PH = plant height; PN = panicle number per plant; PL = panicle length; PW = panicle weight; NFG = number of filled grains per panicle; NUFG = number of unfilled grains per panicle; TGW = thousand-grain weight; GY = grain yield per ha; BY = biological yield per ha; and HI = harvest index. A, B, C, and D represent the trait groups, while I, II, and III represent the experimental treatment groups.

4. Discussion

Ensuring food security for a growing population requires enhancing rice production not only in normal soils but also in salt-affected soils, where rice is commonly used as a pioneer crop to rehabilitate these lands. However, rice is generally known to be salt-sensitive. Its yield tends to decrease by 12% for each unit increase in electrical conductivity of saturated soil extract (ECe) above 3 dS m^{-1} [52]. The ECe of 6 dS m^{-1} reduced the rice grain yield by 50% [53]. Additionally, moderate salinity stress (NaCl less than 50 mM) can result in a significant reduction in several plant growth and yield parameters of rice, such as biomass accumulation, tiller, and panicle numbers, spikelets per panicle, grain-filling percentage, and grain yield. On the other hand, severe stress (NaCl more than 100 mM) can lead to the death of rice plants before they reach maturity or a substantial decrease in grain yield, even in salt-tolerant genotypes [10]. For instance, Zayed et al. [54] found that the grain yield of salt-tolerant Giza 178 genotypes decreased from 9.45 ton ha^{-1} in non-saline soils to 4.35 ton ha^{-1} in saline soils with an EC of 8.4 dS m^{-1} (80 mM NaCl). In this study, various treatments were assessed in saline soil with an ECe ranging from 7.90 to 8.20 dS m^{-1} and a pH of 8.25 (Table 1). This indicates that this soil falls under the category of moderately saline (EC4—8 dS/m), which is roughly equivalent to 40 to 80 mM NaCl, as reported by Mondal et al. [55]. Therefore, with the application of proper agronomic techniques, these soils can be utilized more effectively for rice production.

Generally, salinity stress disrupts a variety of physiological processes at different plant levels, leading to a significant reduction in plant growth and yield. The closing of stomata and limitation of CO_2 uptake are significant adverse effects of salt stress, resulting in reduced photosynthesis efficiency and disruption of the Calvin cycle's carbon reduction process. This disruption hinders the oxidation of NADP+, an essential electron acceptor in photosynthesis [10,56,57]. Furthermore, salinity stress leads to plants accumulating high levels of sodium (Na^+) and chloride (Cl^-) ions while reducing the uptake and transport of potassium (K^+) and magnesium (Mg^{2+}) [6,9]. The excess Na^+ and Cl^- hinder chlorophyll production by lowering chlorophyll levels or speeding up its breakdown. Furthermore, Mg^{2+} is crucial for chlorophyll synthesis. Its deficiency can reduce the production of photosynthetic pigments by affecting enzyme activities and hindering chlorophyll biosynthesis in plants [10,57,58]. Potassium is essential for reducing oxidative stress and regulating stomatal function to maintain a balance between CO_2 absorption and water vapor release. Therefore, a deficiency in K^+ can negatively impact CO_2 fixation, assimilate transport, and cause damage to membranes and chlorophyll [44,45,57,59]. The abovementioned negative impacts of salinity stress on physiological and biochemical processes lead to a notable reduction in key agro-physiological parameters. This could explain why untreated plants in this study showed lower values for all studied parameters than those treated with compounds for CO_2 + K sources (Figures 1–8).

Because stomatal closure is a common response to salinity stress, which leads to a decrease in CO_2 entry into leaves and photosynthetic capacity, elevated CO_2 levels may mitigate these negative effects and enhance plant growth and productivity, especially in C_3 plants. Previous studies have shown that elevated CO_2 levels can improve leaf photosynthesis, plant growth, and yield in C_3 plants when they are stressed. This improvement is attributed to the increased activity of the Rubisco enzyme, which is not fully saturated at current atmospheric CO_2 levels [15,16,60]. Elevated CO_2 levels also reduce CO_2 losses from photorespiration by inhibiting oxygenation by Rubisco, leading to increased carbon assimilation for plant growth and development [17–19]. Elevated CO_2 levels reduce water loss through transpiration by decreasing stomatal conductance and transpiration rates. As a result, plants can use water more efficiently under stress conditions [61]. These various physiological adjustments lead to increased leaf photosynthesis rates, ultimately promoting better plant growth and higher crop yields in stressed plants. Studies have demonstrated that elevated levels of CO_2 result in a 15–41% increase in the production of C_3 plants, whereas C_4 plants only achieve a 5–10% enhancement [23–25]. Interestingly, elevated CO_2 levels generally led to increased growth and yield of several field crops. However, this enhancement was more pronounced under stressful conditions than under non-stressful conditions [26–28].

Raising ambient CO_2 levels in open fields on a large scale is costly and impractical. A more cost-effective approach involves foliar spraying compounds like lithovit and ethanol directly onto plant leaves. These compounds break down and release CO_2 inside the leaf intercellular, providing plants with the necessary CO_2 for photosynthesis. This process boosts light-saturated photosynthesis in C_3 plants, improving photosynthesis efficiency and ultimately enhancing crop growth and yield under stress conditions [34–40,62]. In this study, foliar spraying of Liv and Eth (a carbon foliar fertilizer) significantly enhanced various agro-physiological parameters, regardless of genotypes and application time, compared to the untreated treatment. The application of Liv increased pigment contents by 19.6–29.6% (Figure 1), photosynthetic parameters by 12.5–37.9% (Figure 3), and yield and its components by 12.0–32.5% (Figures 5 and 6), while it decreased the NUFG by 42.0% (Figure 6), when compared with untreated treatment. Similarly, Maswada and Abd El-Rahman [63] found that foliar spraying of Liv (a nano-$CaCO_3$) significantly increased total chlorophyll and carotenoids contents, ion contents, growth parameters, yield, and yield components of wheat under salinity stress. It also increased the total chlorophyll content, plant dry wheat, and leaf area of tomatoes under salinity conditions [64]. Sorour et al. [62] also found that the foliar application of Liv significantly enhanced various parameters related to growth, yield, and quality of sugar beet crops under salinity stress conditions. Additionally, Gomaa et al. [65] found that the growth of salt-stressed green bean plants, including plant dry weight and leaf area, was promoted by foliar spraying with Liv. Lithovit has been proven to boost crop production by up to 50% in certain crops by counteracting the harmful effects of abiotic stress on their physiological and chemical processes [34–36,66]. These findings indicate that using Liv through foliar spraying can be a cost-effective and sustainable way to enhance rice growth and yield in saline soils. Lithovit stays on the leaf surface and gets absorbed when dew moistens it, leading to increased CO_2 levels in the plant leaf. This boosts photosynthesis, plant physiology, growth, and yield, especially in stressful conditions.

This study also found that foliar spraying with lithovit was more effective in improving agro-physiological traits than Eth and KC (Figures 1, 3, 5 and 6). This could be attributed to the lithovit compound also containing various macro- and micro-nutrients such as Mg, Ca, Fe, Zn, and Si, which are involved in several physiological processes within plant cells. Therefore, these nutrients offer additional protection against the negative impacts of salinity stress on plant growth, physiology, and yield. These nutrients help mitigate the negative effects of salinity stress by preserving membrane integrity and stability, protecting chloroplast structure and functions, maintaining photosynthetic and enzyme activity, facilitating osmotic adjustment, regulating stomatal function, aiding in the transport of photoassimilates, promoting chlorophyll synthesis, and serving as functional, structural, or regulatory cofactors for numerous enzymes [67–70].

Ethanol is also a cost-effective compound, priced at $1.0 for 26.5 L of 20 mM ethanol, that can be used as a source of CO_2 to enhance photosynthesis efficiency under various abiotic stress conditions. When ethanol is sprayed on foliage, it breaks down into CO_2 within the leaf mesophyll, increasing the CO_2 to O_2 ratio. This improvement in ratio enhances the photosynthesis rate and decreases photorespiration, particularly in C_3 plants, ultimately improving plant growth, physiology, and yield under stress conditions [38–43]. In this study, Eth's foliar spraying significantly enhanced agro-physiological parameters, regardless of genotypes and application time, compared to the control. The application of Eth increased pigment contents, photosynthetic parameters, and yield and its components by 14.4–23.7% (Figure 1), 10.9–32.0% (Figure 3), 11.2–23.5% (Figures 5 and 6), respectively, while decreased the NUFG by 34.6% (Figure 6), when compared with control treatment. This finding indicates that exogenous application of Eth could be a cost-effective and sustainable approach to mitigate the harmful effects of salinity stress on rice's physiological and yield performance. Similarly, previous studies have shown that foliar spraying of Eth can effectively alleviate the negative impacts of various abiotic stressors by increasing CO_2 concentrations within the leaf and enhancing photosynthesis efficiency. It has been shown to mitigate the effects of salinity and drought stress in soybean [38,40], salinity and chilling stress in rice [41,43], heat stress in tomatoes [42], and drought stress in rice and wheat [71]. For instance, in a study by Das et al. [38], it was found that treating soybean plants with moderate (8 dS m^{-1}) and high (16 dS m^{-1}) salinity levels, along with 30 mM ethanol, resulted in enhanced photosynthetic pigment contents, ion content (K^+ and Mg^{2+}), net photosynthetic rate, RWC, water use efficiency, leaf area, and plant biomass. This ultimately led to improved growth performance of soybean plants, such as plant height and plant dry weight, by 12.35% and 32.38%, respectively, compared to untreated plants. Rahman et al. [40] also found that spraying ethanol (20 mM) on soybean plants improved their growth performance under drought conditions. This was accomplished by increasing photosynthetic pigment levels, gas exchange characteristics, RWC, plant biomass, and leaf area.

One effective way to help plants deal with abiotic stress is by providing essential nutrients through foliar spraying. Previous studies have demonstrated that foliar application of potassium can mitigate the adverse effects of salinity stress on various crops [72–75]. Potassium is crucial for various plant metabolic processes, and the K^+/Na^+ ratio is a reliable indicator of salinity tolerance. Salinity stress can trigger the production of ROS, leading to lipid peroxidation and potassium leakage from cells through potassium efflux channels [76]. Therefore, K is a vital macronutrient necessary for various plant functions under salinity stress, such as osmotic adjustment, maintaining turgor, stabilizing cell membranes, and regulating enzyme activation, protein synthesis, and cytoplasmic homeostasis. Previous studies have shown that foliar spraying of K in the form of K-carbonate (K_2CO_3) or K-bicarbonate ($KHCO_3$) can help alleviate the adverse effects of abiotic stress on various agro-physiological parameters [46,47,77]. Fortunately, these compounds are safe for both humans and the environment, making them suitable for eco-friendly farming practices. In this study, foliar spraying of KC resulted in improved pigment contents, photosynthetic parameters, and yield and its components by 10.3–18.7%, 12.0–20.1%, and 9.7–24.9%, respectively, compared to the control treatment (Figures 1, 3 and 5). KC was particularly effective in reducing the NUFG compared to Liv and Eth compounds, with a 43.2% decrease compared to the control (Figure 6). This indicates that K helps transfer photoassimilates for grain development, reducing the NUFG, especially under salinity stress. Previous studies have shown that maintaining sufficient K levels in stressed plants, which can be achieved rapidly and effectively by foliar spraying K directly onto the leaves, helps minimize the detrimental effects of stress, such as salinity and drought, on grain filling, yield, and yield components when compared to plants lacking adequate K levels [77–79]. Therefore, applying K foliar to rice crops in the form of KC, which is highly soluble compared to other forms of K [80], can be a key strategy to rapidly improve their salt tolerance.

The study's results also showed that the impact of CO_2 + K source compounds in enhancing the agro-physiological characteristics of rice in saline soils is influenced by the timing of application and the salt tolerance of the rice varieties under treatment. Overall, the compounds were more effective when applied before flowering rather than after, and they had better results on the salt-tolerant genotype (Giza 179) compared to the salt-sensitive genotype (Giza 178) (Figures 2, 4, 7 and 8). These results were confirmed by heatmap and PCA analysis (Figure 11a,b), showing that it is highly recommended to apply CO_2 + K source compounds to the leaves before heading to improve the physiological and yield performance of rice in saline soil conditions. According to a study by Mahmoodi et al. [81], foliar application of nutrients before the heading stage, i.e., "maximum tillering and panicle initiation", resulted in the highest values of several agro-physiological traits of rice plants. Similarly, Kumar et al. [82] found that the optimal crop growth rate in rice was achieved by applying nutrients foliar during the tillering and panicle initiation stages. Importantly, most rice varieties accumulate most of their total dry matter (65–70%) during the vegetative growth stages, with only 30–35% of dry matter accumulating after flowering [81]. These findings can be attributed to plants achieving their peak size during the exponential growth phase, and leaves have a high absorption capacity for foliar compounds. Therefore, administering compounds through foliar application in both the vegetative and pre-heading stages is more efficient and economical than after heading.

The higher values of various agro-physiological parameters in the salt-tolerant genotype Giza 179 compared to the salt-sensitive genotype may be attributed to differences in their mechanisms of salinity tolerance. Salt-tolerant genotypes usually have higher photosynthesis rates, stomatal conductance, RWC, photosynthetic pigments, osmotic adjustment ability, K^+/Na^+ ratio, lower Na^+ accumulation, increased productive tillers, larger root systems, minimal reduction in GY, higher levels of soluble carbohydrates, and enhanced activity of antioxidant enzymes [83–85]. These results indicate that using salt-tolerant rice varieties along with foliar application of CO_2 + K source compounds at the right timing also plays a vital role in enhancing the physiological and yield performance of rice in salt-affected soils. This finding was supported by heatmap and PCA analysis (Figure 11a,b).

5. Conclusions

While rice is highly susceptible to salinity stress, it is commonly planted as a pioneer crop to rehabilitate salt-affected mudflat soils. To improve the growth, physiology, and yield of rice plants in these soils, specific integrated farming techniques must be implemented to counteract the negative effects of salinity stress. The impact of various farming practices, such as foliar application of CO_2 + K source compounds at different stages (before and after heading) on two rice genotypes with varying salt tolerance, was assessed based on various agro-physiological parameters. The study results showed that foliar spraying with different CO_2 + K source compounds positively influenced pigment content, photosynthetic efficiency, yield, and its components. However, it also led to a reduction in the NUFG compared to untreated treatment. The efficacy of these compounds in achieving these results was Liv > Eth > KC, except for the NUFG, where KC was more effective in reducing it than Liv and Eth. The various CO_2 + K source compounds were more effective in improving the agro-physiological parameters of rice when applied before flowering rather than after, and they had a greater positive impact on the salt-tolerant genotype (Giza 179) compared to the salt-sensitive genotype (Giza 178). In conclusion, the study suggests that utilizing CO_2 + K source compounds at the appropriate timing and cultivating salt-tolerant rice varieties can help mitigate the negative effects of salinity stress on rice. Farmers can increase rice yield in salt-affected mudflat soils by adopting these integrated farming practices. Nevertheless, a more extensive field study that includes a variety of crop species and varying levels of salinity, as well as different application methods and concentrations of CO_2 + K source compounds, particularly for Liv and Eth, would be required to confirm the positive effects of these compounds in effectively addressing salinity problems. Additionally, other studies need to explore how CO_2 + K source compounds affect intracellular CO_2 levels, CO_2 diffusion, ion concentrations, and the function of antioxidant enzymes, which will be the main focus of our upcoming research.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/agronomy14122850/s1, Table S1: Combined analysis of variance (ANOVA) for the main effects of season (S), genotype (G), foliar application timing (FT), CO_2 + K source compounds (CO_2 + K), and their different interaction on all studied traits.

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