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Original Article

Trichoderma afroharzianum T22 induces tolerance to combined iron deficiency and drought stress in maize

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ABSTRACT

Combined iron (Fe) deficiency and drought stress frequently occur in agricultural soils and can severely impair crop growth, nutrient acquisition, and physiological performance. Beneficial microorganisms have emerged as promising tools for enhancing plant resilience under adverse environmental conditions. This study evaluated the effects of *Trichoderma afroharzianum* T22 on four maize genotypes exposed to combined Fe deficiency and drought stress. Plant growth, chlorophyll content, relative water content, rhizosphere siderophore production, and expression of the alternative oxidase-associated gene *ThAOX1* were assessed after six weeks of growth. Combined stress reduced root length, root biomass, shoot height, shoot biomass, chlorophyll content, and RWC across all genotypes. Inoculation with *T. afroharzianum* T22 partially mitigated these reductions, although responses varied among genotypes. Genotypes MSIRI3B and CHZM-07-134 exhibited the strongest responses to fungal inoculation, showing greater improvements in growth parameters and physiological traits than R243 and Hi-47. Expression of *ThAOX1* increased significantly in MSIRI3B and CHZM-07-134 under stress + TA and TA+ treatments, whereas no significant changes were detected in R243 and Hi-47. Rhizosphere siderophore production was also enhanced by fungal inoculation, particularly in MSIRI3B and CHZM-07-134, with the highest values observed under stress + TA treatment. The results demonstrate that *T. afroharzianum* T22 influences plant growth, water status, rhizosphere siderophore production, and alternative oxidase-associated gene expression under combined Fe deficiency and drought stress. The magnitude of these responses was genotype dependent, with MSIRI3B and CHZM-07-134 exhibiting greater responsiveness to fungal inoculation than R243 and Hi-47. These findings highlight the potential of *T. afroharzianum* T22 as a microbial biostimulant for improving maize performance under multiple environmental stress conditions.

Introduction

Maize (*Zea mays* L.) is one of the most important cereal crops worldwide, serving as a major source of food, feed, biofuel, and industrial raw materials. However, maize productivity is increasingly threatened by environmental stresses associated with climate change and soil degradation (Lesk et al., 2016; IPCC, 2023). Among these constraints, drought and nutrient deficiencies are considered major factors limiting crop productivity in both developed and developing agricultural systems (Farooq et al., 2009; Fahad et al., 2017). Importantly, plants growing under field conditions are frequently exposed to multiple stresses simultaneously rather than individually, and combined stresses often exert more severe effects than single stress factors because of complex interactions among physiological and molecular pathways (Mittler, 2006; Zandalinas et al., 2021). Iron (Fe) is an essential micronutrient required for numerous biochemical and physiological processes in plants, including photosynthesis, respiration, chlorophyll biosynthesis, nitrogen assimilation, and antioxidant metabolism (Marschner, 2012; Kobayashi and Nishizawa, 2012). Although iron is abundant in most soils, its bioavailability is often limited because ferric iron (Fe^{3+}) forms insoluble hydroxides and oxides, particularly in alkaline and calcareous soils (Morrissey and Guerinot, 2009). Consequently, iron deficiency is a widespread nutritional disorder that causes leaf chlorosis, reduced photosynthetic efficiency, impaired root development, and substantial yield losses in agricultural crops (Kobayashi and Nishizawa, 2012; Connorton et al., 2017). In graminaceous species such as maize, iron acquisition relies primarily on the secretion of phytosiderophores that chelate ferric iron and facilitate uptake through specialized transport systems (Curie and Mari, 2017). Drought stress represents another major limitation to crop productivity worldwide. Water deficit reduces cell expansion, stomatal conductance, photosynthetic carbon assimilation, nutrient transport, and overall biomass accumulation (Farooq et al., 2009; Osakabe et al., 2014). Moreover, drought promotes excessive generation of reactive oxygen species (ROS), including superoxide radicals, hydrogen peroxide, and hydroxyl radicals, resulting in oxidative damage to cellular structures and disruption of metabolic homeostasis (Apel and Hirt, 2004; Hasanuzzaman et al., 2020). The simultaneous occurrence of iron deficiency and drought can intensify these detrimental effects because both stresses impair photosynthetic electron transport, respiration, and antioxidant defense systems (Briat et al., 2015). Recent studies have demonstrated that combined stresses frequently trigger unique physiological and molecular responses that differ substantially from those induced by individual stresses (Rizhsky et al., 2004; Mittler, 2006; Suzuki et al., 2014). Therefore, understanding mechanisms that enhance plant adaptation to combined stress conditions has become a major objective of modern crop improvement programs. Among the promising approaches for improving crop resilience is the utilization of beneficial plant-associated microorganisms that can enhance nutrient acquisition, regulate phytohormone signaling, and activate stress-responsive pathways (Vurukonda et al., 2016; Backer et al., 2018). Species belonging to the genus *Trichoderma* are among the most extensively studied beneficial fungi because of their ability to promote plant growth and enhance tolerance against both biotic and abiotic stresses (Harman et al., 2004; Woo and Pepe, 2018). These fungi colonize plant roots and establish mutually beneficial associations that improve nutrient acquisition, stimulate root development, enhance antioxidant activity, and induce systemic resistance (Shoresh et al., 2010; Poveda et al., 2020). Numerous studies have reported positive effects of *Trichoderma* inoculation on plant performance under drought, salinity, nutrient deficiency, and temperature stress (Mastouri et al., 2010; Brotman et al., 2013; Contreras-Cornejo et al., 2014). Among various *Trichoderma* isolates, *Trichoderma afroharzianum* T22 has attracted considerable attention because of its broad-spectrum plant growth-promoting activities and consistent performance across diverse cropping systems (Lorito et al., 2010; Woo et al., 2014). Root colonization by T22 has been shown to alter plant transcriptomes, improve nutrient uptake, increase photosynthetic efficiency, and stimulate defense-related signaling pathways (Shoresh and Harman, 2008; Hermosa et al., 2012). Furthermore, T22-mediated enhancement of drought tolerance has been associated with improved root architecture, water-use efficiency, and antioxidant defense capacity (Bae et al., 2009; Mastouri et al., 2012). One important mechanism involved in plant stress adaptation is the alternative oxidase (AOX) pathway. Alternative oxidase is a terminal oxidase located in the mitochondrial inner membrane that provides an alternative route for electron transport by bypassing complexes III and IV of the cytochrome pathway (Vanlerberghe, 2013). Although this pathway generates less ATP, it prevents over-reduction of the electron transport chain and minimizes excessive ROS accumulation during environmental stress (Vanlerberghe, 2013; Del-Saz et al., 2018). Increasing evidence indicates that AOX plays critical roles in maintaining redox homeostasis and metabolic flexibility during drought, nutrient deficiency, salinity, and combined stress conditions (Clifton et al., 2006; Del-Saz et al., 2018). In addition to modulation of host physiological responses, beneficial microorganisms can influence rhizosphere processes that improve nutrient availability. Microbial siderophores are low-molecular-weight compounds with exceptionally high affinity for ferric iron and are considered important determinants of plant iron acquisition under iron-limiting conditions (Ahmed and Holmström, 2014). Siderophore-producing microorganisms can enhance iron solubilization, improve plant nutrition, and alleviate iron deficiency symptoms in several crop species (Rajkumar et al., 2010; Saha et al., 2016). However, the contribution of rhizosphere siderophore production to *Trichoderma*-mediated tolerance against combined drought and iron deficiency remains poorly understood. Therefore, the objectives of the present study were to investigate the effects of *T. afroharzianum* T22 on growth, physiological performance, alternative oxidase-associated gene expression, and rhizosphere siderophore production in maize subjected to combined iron deficiency and drought stress.

Materials and methods

Plant cultivation

Maize seeds were surface-sterilized and germinated in seedling trays containing a commercial potting substrate under controlled growth-room conditions. After two weeks of growth, uniform seedlings were transplanted into pots containing a 1:1 mixture of commercial potting mix and field soil. Following transplantation, plants were maintained under well-watered conditions for an additional two weeks to ensure uniform establishment before the initiation of treatments.

Four treatment groups were established: (i) Control, (ii) Dual Stress (combined iron deficiency and drought stress), (iii) Dual Stress + *T. afroharzianum* T22 (Stress+TA), and (iv) *T. afroharzianum* T22 alone (TA+). Dual stress was imposed two weeks after transplantation by simultaneously applying iron-deficient conditions (15 mM $NaHCO_3$) and drought stress. Drought stress was maintained by providing approximately 70% less water than the well-watered control plants throughout the treatment period. Plants assigned to the TA treatments were inoculated with 1 ml of *T. afroharzianum* T22 (1×10^7 CFU mL^{-1}) on the day of transplantation into the rhizosphere. Plants were grown for a total of six weeks after transplantation under a 16 h light/8 h dark photoperiod at 25 ± 2 °C. The experiment was arranged in a completely randomized design with multiple biological replicates per treatment. At the end of the experimental period, plants were harvested for physiological, biochemical, and molecular analyses.

Morphological and biomass measurements

Morphological traits were evaluated at the end of the six-week experimental period. Shoot height, stem diameter, root length, shoot dry weight, and root dry weight were recorded for each plant. Shoot height and root length were measured using a ruler, while stem diameter was determined using a digital caliper. Following harvest, shoots and roots were separated and dried in a forced-air oven at 70°C for 72 h to obtain constant weight. Dry biomass was then determined using an analytical balance.

Chlorophyll and relative water content

Leaf chlorophyll content was assessed using a portable SPAD chlorophyll meter (SPAD-502, Konica Minolta, Japan). Measurements were taken from fully expanded leaves, and the average SPAD value per plant was used for subsequent analyses. Relative water content (RWC) was determined as an indicator of plant water status. Fresh leaf samples were weighed immediately after collection to obtain fresh weight (FW), then immersed in distilled water for 24 h at room temperature to determine turgid weight (TW). Samples were subsequently oven-dried at 70°C for 72 h to obtain dry weight (DW). Relative water content was calculated using the following equation: $\text{RWC} = [(\text{FW} - \text{DW})/(\text{TW} - \text{DW})] \times 100$, where FW represents fresh weight, TW represents turgid weight, and DW represents dry weight.

Determination of siderophore production

Rhizosphere siderophore production was quantified using the chrome azurol S (CAS) assay. Briefly, rhizosphere soil samples were suspended in sterile distilled water and vigorously mixed to extract soluble siderophores. The suspensions were centrifuged at 10,000 × g for 10 min, and the resulting supernatants were collected for analysis. An aliquot of each extract was mixed with CAS reagent and incubated at room temperature for 30 min to allow color development. Absorbance was measured at 630 nm using a UV–Vis spectrophotometer. Siderophore production was expressed as percent siderophore units (SU) based on the decrease in blue color intensity of the CAS reagent based on formula (Himpsl and Mobley, 2019).

Results

Relative abundance of *ThAOX1*

The relative abundance of *ThAOX1* varied among maize genotypes and treatments (Figure 1). In genotype MSIRI3B, *ThAOX1* expression was significantly higher in both the Stress + TA and TA+ treatments compared with the Control and Stress treatments ($P < 0.05$). A similar pattern was observed in genotype CHZM-07-134, where Stress + TA and TA+ treatments resulted in significantly greater expression than Control and Stress treatments. In contrast, no significant differences among treatments were detected for genotypes R243 and Hi-47. Expression levels in these genotypes remained relatively stable across all treatments. Overall, the highest *ThAOX1* expression levels were observed in MSIRI3B and CHZM-07-134 under Stress + TA and TA+ treatments (Fig. 1).

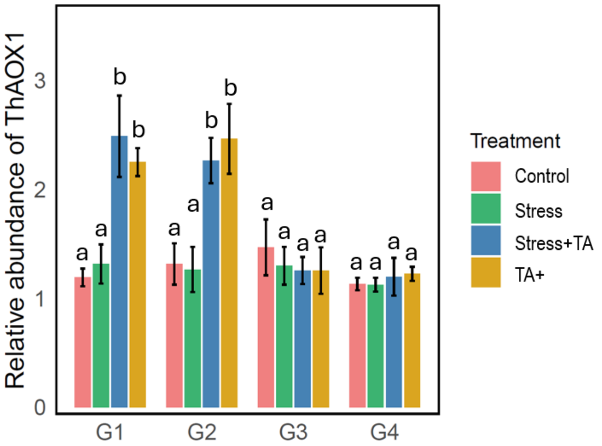

Fig. 1. Relative abundance of ThAOX1 in four maize genotypes (G1: MSIRI3B, G2: CHZM-07-134, G3: R243 and G4: Hi-47) under Control, Stress, Stress + *T. afroharzianum* (TA), and TA-only treatments. Bars represent mean ± SD. Different letters indicate significant differences among treatments within each genotype ($P < 0.05$).

Phenotypic responses of maize genotypes

Visual differences among treatments were observed after six weeks of growth (Fig. 2). In all four genotypes, plants subjected to the Stress treatment exhibited reduced plant size and increased leaf senescence compared with control plants. Plants receiving Stress + TA treatment generally displayed greater shoot development and larger canopy size than plants exposed to Stress alone. Plants receiving the TA+ treatment exhibited growth characteristics comparable to or greater than those of Control plants in most genotypes (Fig. 2).

Effects of *T. afroharzianum* on root growth

Root length differed significantly among treatments in all four maize genotypes (Fig. 3A). In MSIRI3B, root length decreased compared to control treatment under stress. Root length increased stressed plants inoculated with TA relative to the plants solely cultivated under stress (Fig. 3A). Similar trends were observed in CHZM-07-134, where root length was reduced under Stress and increased under Stress + TA and TA+ treatments. In R243, control and TA+ plants exhibited root lengths exceeding 40 cm, whereas stress and stress + TA plants displayed lower values. In Hi-47, root length was approximately 29 cm in both control and TA+ plants but decreased to approximately 13 cm and 11 cm under stress and stress + TA treatments, respectively (Fig. 3A). Root biomass also differed significantly among treatments (Fig. 3B). In MSIRI3B and CHZM-07-134, root biomass was highest in control and TA+ plants, intermediate in stress + TA plants, and lowest in stressed plants. In R243 and Hi-47, root biomass was reduced under stress and stress + TA treatments relative to control and TA+ treatments.

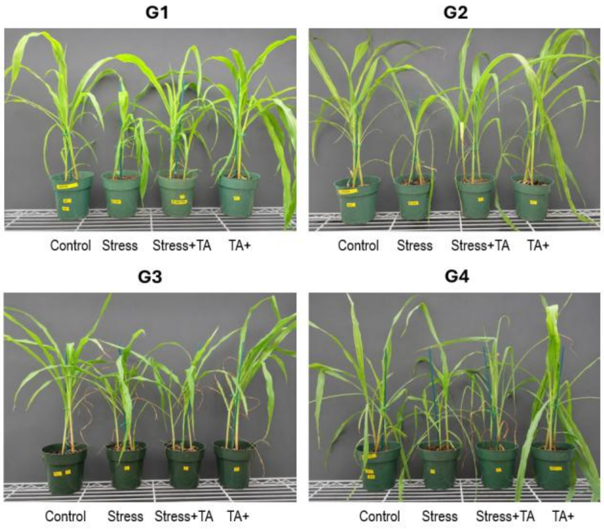

Fig. 2. Phenotypes of four maize genotypes (G1: MSIRI3B, G2: CHZM-07-134, G3: R243 and G4: Hi-47) grown under Control, Stress, Stress + T. afroharzianum (TA), and TA-only (TA+) treatments cultivated in the greenhouse for 6 weeks.

Effects of *T. afroharzianum* on shoot growth

Shoot height varied significantly among treatments within each genotype (Fig. 4A). In MSIRI3B, shoot height decreased from approximately 53 cm in control plants to approximately 34 cm under Stress. Stress + TA plants exhibited shoot heights of approximately 50 cm, while TA+ plants reached approximately 51 cm. Similar trends were observed in CHZM-07-134. In R243, shoot height was approximately 47 cm in control plants, 39 cm under Stress, 38 cm under stress + TA, and 53 cm under TA+ treatment. Genotype G4 exhibited lower shoot height than the other genotypes, ranging from approximately 17–21 cm across treatments (Fig. 4A).

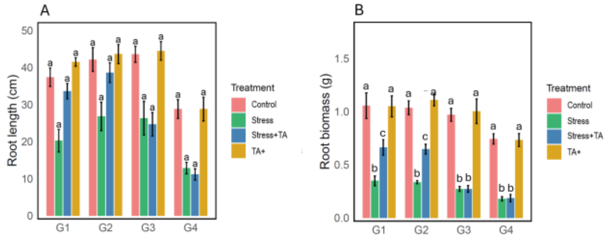

Fig. 3. Effects of stress and *T. afroharzianum* (TA) on (A) root length and (B) root biomass of four maize genotypes (G1: MSIRI3B, G2: CHZM-07-134, G3: R243 and G4: Hi-47). Treatments included Control, Stress, Stress + TA, and TA-only (TA+). Bars represent mean ± SD. Different letters indicate significant differences among treatments within each genotype (P < 0.05).

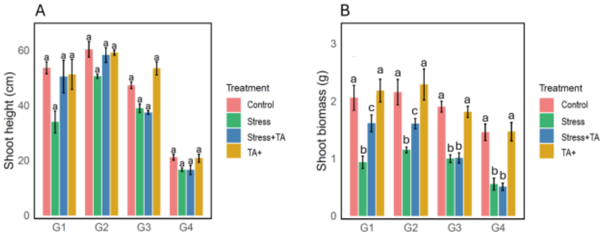

Fig. 4. Effects of stress and *T. afroharzianum* (TA) on (A) shoot height and (B) shoot biomass of four maize genotypes (G1: MSIRI3B, G2: CHZM-07-134, G3: R243 and G4: Hi-47). Treatments included Control, Stress, Stress + TA, and TA-only (TA+). Bars represent mean ± SD. Different letters indicate significant differences among treatments within each genotype (P < 0.05).

Shoot biomass followed patterns similar to those observed for shoot height (Fig. 4B). In MSIRI3B and CHZM-07-134, shoot biomass decreased under stress however, increased under stress + TA and TA+ treatments. In R243, shoot biomass remained relatively similar among treatments, although lower values were observed in stress and stress + TA plants compared with Control and TA+ plants. Genotype G4 exhibited the lowest shoot biomass among all genotypes, with values ranging from approximately 0.5 to 1.5 g (Fig. 4B).

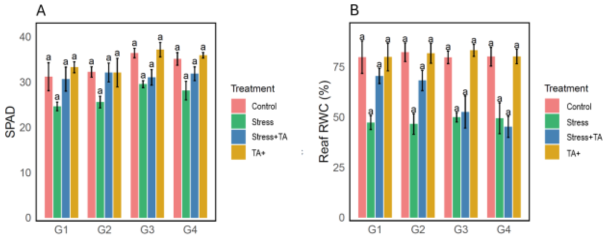

Fig. 5. Effects of stress and *T. afroharzianum* (TA) on (A) chlorophyll content (SPAD values) and (B) relative water content (RWC) in four maize genotypes (G1: MSIRI3B, G2: CHZM-07-134, G3: R243 and G4: Hi-47). Treatments included Control, Stress, Stress + TA, and TA-only (TA+). Bars represent mean ± SD. Different letters indicate significant differences among treatments within each genotype ($P < 0.05$).

Chlorophyll content and relative water content

SPAD values varied among genotypes and treatments (Fig. 5A). In MSIRI3B, chlorophyll content decreased from approximately 31 SPAD units in control plants to approximately 24 SPAD units under stress. Stress + TA and TA+ treatments resulted in SPAD values of approximately 30 and 33 units, respectively. In CHZM-07-134, SPAD values ranged from approximately 25 to 33 units across treatments. In R243, the highest SPAD values were observed under the TA+ treatment, while lower values were recorded under stress and stress + TA treatments. Similar trends were observed in Hi-47, where stress plants exhibited lower SPAD values than control and TA+ plants (Fig. 5A).

Relative water content also differed among treatments (Fig. 5B). In MSIRI3B and CHZM-07-134, RWC was approximately 80–83% in control and TA+ plants and approximately 46–47% under stress. Interestingly, stress + TA plants exhibited intermediate RWC values. In R243, RWC values ranged from approximately 50% under stress to approximately 83% under TA+ treatment. In Hi-47, Stress and stress + TA treatments resulted in lower RWC values than control and TA+ treatments. Overall, the highest RWC values were observed under TA+ treatment, whereas stress treatments generally exhibited the lowest values (Fig. 5B).

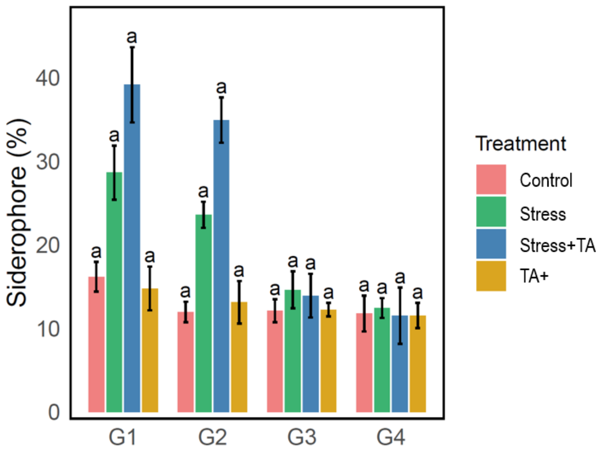

Fig. 6. Rhizosphere siderophore production (%) of four maize genotypes (G1: MSIRI3B, G2: CHZM-07-134, G3: R243 and G4: Hi-47) under control, Stress, Stress + *T. afroharzianum* (TA), and TA-only (TA+) treatments. Bars represent mean ± SD. Different letters indicate significant differences among treatments within each genotype ($P < 0.05$).

Rhizosphere siderophore production

Rhizosphere siderophore production differed among genotypes and treatments (Fig. 6). In MSIRI3B, siderophore production increased from approximately 16% in control plants to approximately 29% under stress and approximately 39% under stress + TA treatment. The TA+ treatment resulted in values of approximately 15%. A similar pattern was observed in CHZM-07-134, where siderophore production increased from approximately 12% in control plants to approximately 24% under stress and approximately 35% under stress + TA treatment. In contrast, genotypes R243 and Hi-47 exhibited smaller differences among treatments, with values ranging from approximately 11–15%. Among all treatment combinations, the highest siderophore production was observed in genotype MSIRI3B under the stress + TA treatment, followed by genotype CHZM-07-134 under the same treatment (Fig. 6).

Discussion

Combined abiotic stress frequently exerts more severe effects on crop growth and productivity than individual stress because plants must simultaneously coordinate multiple physiological and metabolic responses. In the present study, combined iron deficiency and drought stress markedly affected growth, physiological performance, and rhizosphere characteristics in maize. Inoculation with *Trichoderma afroharzianum* T22 modified several of these responses, including root and shoot growth, chlorophyll content, relative water content, rhizosphere siderophore production, and expression of the alternative oxidase-associated gene *ThAOX1*. The magnitude of these responses varied among genotypes, suggesting differential responsiveness to fungal inoculation.

One of the most notable observations was the reduction in plant growth under combined iron deficiency and drought stress. Root length, root biomass, shoot height, and shoot biomass were reduced in most genotypes exposed to stress. These responses are consistent with previous reports showing that both drought and iron deficiency restrict cell expansion, nutrient acquisition, and carbon assimilation, ultimately leading to reduced biomass accumulation (Farooq et al., 2009; Kobayashi & Nishizawa, 2012). Iron deficiency impairs chlorophyll biosynthesis and electron transport, whereas drought reduces water availability and nutrient mobility. The simultaneous occurrence of these stresses can therefore produce substantial limitations to plant growth and development (Mittler, 2006; Suzuki et al., 2014).

Inoculation with *T. afroharzianum* T22 partially or completely alleviated growth reductions in several genotypes. Improvements in root growth were particularly evident in MSIRI3B and CHZM-07-134, where Stress + TA plants exhibited greater root length and biomass than plants exposed to stress alone. Enhanced root development is among the most frequently reported benefits of *Trichoderma* colonization and is often associated with increased nutrient acquisition and improved adaptation to adverse environmental conditions (Contreras-Cornejo et al., 2009; Harman et al., 2004). Root system expansion may provide greater access to water and nutrients, thereby supporting improved plant performance under stress conditions. The observed differences among genotypes indicate that host genetic background strongly influences responsiveness to microbial inoculation. Genotype-dependent responses to beneficial microorganisms have been reported in maize, wheat, soybean, and other crops, where variations in root architecture, root exudation profiles, and signaling pathways affect microbial recruitment and colonization success (Backer et al., 2018). In the present study, MSIRI3B and CHZM-07-134 generally exhibited stronger responses to *Trichoderma* treatment than R243 and Hi-47, suggesting that host–microbe compatibility may contribute significantly to the effectiveness of fungal inoculation under combined stress conditions.

Maintenance of chlorophyll content represents an important indicator of stress tolerance because chlorophyll degradation is a common consequence of both drought and iron deficiency. Iron is required for chlorophyll biosynthesis and photosynthetic electron transport, while drought can accelerate chlorophyll degradation through oxidative stress and disruption of cellular metabolism (Briat et al., 2015; Farooq et al., 2009). In the present study, stress generally reduced SPAD values across genotypes, whereas *Trichoderma* inoculation increased chlorophyll content relative to stress-treated plants. Similar observations have been reported in tomato, wheat, maize, and sorghum, where *Trichoderma* treatment preserved photosynthetic pigments and improved photosynthetic performance under environmental stress (Mastouri et al., 2010; Bae et al., 2009).

Relative water content is widely used as an indicator of plant water status and drought tolerance. The substantial reduction in RWC under stress conditions observed in this study confirms that the imposed treatment effectively limited plant water availability. Plants inoculated with *T. afroharzianum* generally maintained higher RWC than stressed plants without fungal inoculation. Previous studies have shown that *Trichoderma* can improve water-use efficiency, promote root development, and enhance osmotic adjustment under drought conditions (Mastouri et al., 2012; Brotman et al., 2013). Maintenance of tissue hydration is particularly important under combined stress because adequate water status supports nutrient transport, photosynthesis, and metabolic activity.

Rhizosphere siderophore production was also strongly affected by stress and fungal inoculation. The highest siderophore levels were observed in MSIRI3B and CHZM-07-134 under Stress + TA treatment. Siderophores are high-affinity iron-chelating compounds that facilitate iron mobilization and acquisition under iron-limiting conditions (Ahmed & Holmström, 2014). Enhanced siderophore production may increase the availability of iron in the rhizosphere and improve nutrient acquisition by plants experiencing iron deficiency. Because iron availability is often severely restricted in alkaline or nutrient-deficient soils, microbial siderophore production represents an important mechanism for improving plant nutritional status. Previous studies have demonstrated that beneficial microorganisms can contribute to plant iron nutrition through production of siderophores and modification of rhizosphere chemistry (Rajkumar et al., 2010; Saha et al., 2016). The substantial increase in siderophore production observed in MSIRI3B and CHZM-07-134 under Stress + TA treatment suggests that *T. afroharzianum* may stimulate microbial processes associated with iron mobilization. This response may be particularly important under combined iron deficiency and drought stress, where nutrient availability and transport are simultaneously restricted.

The differential responses among genotypes highlight the complexity of plant–microbe interactions under combined stress conditions. While MSIRI3B and CHZM-07-134 displayed strong responses in terms of AOX expression, siderophore production, and growth characteristics, R243 and Hi-47 generally exhibited smaller responses. Such variation is consistent with previous studies showing that plant genotype can strongly influence microbial colonization, signaling, and downstream physiological responses (Backer et al., 2018). These findings emphasize the importance of considering host genotype when developing microbial inoculants for agricultural applications.

Taken together, the present study demonstrates that *T. afroharzianum* T22 influences multiple physiological and molecular processes associated with adaptation to combined iron deficiency and drought stress. Enhanced root growth, maintenance of chlorophyll content and plant water status, increased rhizosphere siderophore production, and elevated expression of *ThAOX1* collectively indicate that fungal inoculation affects both plant and rhizosphere responses to environmental stress. The stronger responses observed in MSIRI3B and CHZM-07-134 further suggest that host genetic background is an important determinant of the effectiveness of microbial-mediated stress mitigation.

Conclusion

Combined iron deficiency and drought stress negatively affected growth, chlorophyll content, relative water content, and rhizosphere characteristics in maize. Inoculation with *Trichoderma afroharzianum* T22 improved plant performance under dual stress by enhancing root and shoot growth, maintaining chlorophyll content and plant water status, increasing rhizosphere siderophore production, and stimulating *ThAOX1* expression. The response to fungal inoculation was genotype dependent, with MSIRI3B and CHZM-07-134 exhibiting greater responsiveness than R243 and Hi-47. These findings indicate that *T. afroharzianum* T22 can contribute to improved maize adaptation to combined iron deficiency and drought stress through coordinated physiological, molecular, and rhizosphere-mediated responses. The results support the potential use of *Trichoderma*-based biostimulants as a sustainable strategy for enhancing crop performance under multiple environmental stresses.

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Conflict of interest

The authors declare they have no conflicts of interest.

Author contributions

M.R.H. conceived and performed the study, conducted the bioinformatic analyses, interpreted the data, and prepared the original manuscript draft. A.H.K. supervised the research, contributed to study design and data interpretation and critically revised the manuscript.

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