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RESPONSE OF TOMATO PLANT TO ABIOTIC (HEAT STRESS) AND BIOTIC (TOMATO LEAF CURL NEW DELHI VIRUS, TOLCNDV) FACTORS

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ABSTRACT

Tomato (*Lycopersicon esculentum*) is a vital fruit-bearing plant renowned for its nutritional advantages. Subjected to various environmental stresses, its yield is significantly impacted. Plants, confronting combined biotic and abiotic stresses, exhibit diverse responses through the evolution of cellular defense mechanisms. Sensitivity to extreme temperatures and pathogenic threats, such as the white fly-transmitted tomato leaf curl New Delhi virus (ToLCNDV), a *Begomovirus*, underscores the global susceptibility of tomato crops. This study aimed to assess tomato plant responses to the compounding stress of heat and agroinfiltrated ToLCNDV, analyzing four key physiological parameters viz., photosynthesis rate, stomatal conductance, transpiration rates, and Water use efficiency. Results indicated reduced photosynthesis rate of 5.1 ± 0.54 % mol CO_2 $m^{-2}s^{-1}$, diminished stomatal conductance of 89.84±1.27% mmol.$m^{-2}s^{-1}$, and transpiration rates of 0.978 ± 0.148 % mmol.$m^{-2}s^{-1}$ as compared to control group as 8.56 ± 0.55 % mol CO_2 $m^{-2}s^{-1}$, 189.08±19.13% mmol.$m^{-2}s^{-1}$and 2.586 ± 0.415 % mmol.$m^{-2}s^{-1}$respectively. Water use efficiency increased in double-stressed plants at 9.1 ± 0.96 % mmol CO_2mol^{-1} H_2O with respect to control 5.84 ± 0.48 % mmol CO_2mol^{-1} H_2O. The combined stressors manifested a net adverse impact, with neither ToLCNDV mitigating heat stress effects nor vice versa. Contrarily, their combined consequences were significantly amplified compared to individual stressors, emphasizing the intricate interplay of heat stress and ToLCNDV on tomato plants. This research marks a pioneering effort to uncover the physiological responses of a vulnerable tomato cultivar to the combined pressures of ToLCNDV and heat stress. Furthermore, it signifies a novel pathway towards the development of transgenic cultivars capable of effectively managing these stresses.

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INTRODUCTION

Tomato *(Lycopersicon esculentum)* stands as the preeminent globally consumed vegetable, distinguished by its exceptional enrichment of essential vitamins A and C (Mellidou *et al.*, 2021). In 2022, tomatoes were identified as the predominantly cultivated vegetable, boasting a production volume of 186 million tonnes, whereas Pakistan generates a quantity of 4.2 million tonnes of tomatoes annually (FAO, 2022). However, tomato productivity in Pakistan remains suboptimal when juxtaposed with other global counterparts, frequently attributed to pest infestations and abiotic stresses (He *et al.*, 2020). In the context of field environments, plants frequently encounter concurrent exposure to diverse abiotic stresses, exerting profound influences on both crop yield and quality. Among the most severe abiotic stresses are drought and heat, particularly prevalent in nations characterized by hot climates (Prasad *et al.*, 2022).

The optimum temperature for tomato plant growth is 26 ± 2 °C in day time and 16 ± 2 °C at night (Hosseini *et al.*, 2018; Raja *et al.*, 2020). The mean temperature of Earth has been observed to elevate at a rate of 0.2°C per annum, with a projected increase ranging from 1.8°C to 4°C by the conclusion of the year 2100 (Tiwari *et al.*, 2020). Consequently, temperature emerges as a paramount and potentially deleterious environmental stressor. Cultivated plants such as tomatoes exhibit heightened susceptibility to thermal stress particularly during the reproductive stage, which manifests in phenomena such as flower abscission and constrained fruit development, consequently resulting in a significant diminishment of overall yield output (Thole *et al.*, 2021). The conjunction of drought and heat typically catalyzes the activation of plant pathogens, encompassing viruses, bacteria, fungi, and insects (Manacorda *et al.*, 2021). Also, heat stress induces oxidative stress in plants by triggering the generation of reactive oxygen species, including singlet oxygen ($^1O_2$), superoxide ion (O_2), hydrogen peroxide (H_2O_2), and hydroxyl radicals (OH^{-1}), leading to significant impairments in plant growth and production (Mazzeo *et al.*, 2018). The intricate interplay between the plant's environmental conditions and the presence of pathogens intricately regulates the plant's defense mechanisms (Mishra *et al.*, 2022), either attenuating or augmenting their attributes (Atkinson and Urwin, 2012; Raja *et al.*, 2020).

A growing body of research underscores the capacity of plant viruses to intricately regulate host responses in the face of environmental perturbations, including but not limited to factors such as wounding, heightened salinity, elevated temperature, and atmospheric CO_2 levels. Concurrently, these environmental shifts are concomitant with modifications in the biological attributes of the viruses, encompassing changes in titer, virulence, and transmission efficiency (Berges *et al.*, 2020).

Abiotic stresses have the potential to impact both the life cycle of viruses and the intricate interactions between host susceptibility factors and viruses. Reciprocally, viruses can exert influence over the plant's response to abiotic stresses (González *et al.*, 2021). For instance, in plants infected with turnip mosaic virus (TuMV), there is an observed heightened expression of defense genes. However, this enhanced defense gene expression is nullified in plants subjected to abiotic stresses. The deactivation of defense responses consequently renders plants more susceptible to viral infections (Manacorda *et al.*, 2021).

Although viruses are harmful to plants, they may prove beneficial under certain circumstances. A growing body of literature has demonstrated that viral infections possess the capacity to mitigate the adverse impact of certain abiotic stresses on plants (Hily *et al.*, 2016). The transmission of plant viruses, a pivotal epidemiological parameter, is predominantly facilitated by arthropod vectors (Bragard *et al.*, 2013). Research on the impact of abiotic stresses on virus dissemination has historically concentrated on vector biology (e.g., developmental time, longevity, fertility, migration) and ecology (Davis *et al.*, 2015). While numerous studies have postulated the potential effects of environmental alterations on virus transmission rates, empirical validation has only recently emerged (Van Munster *et al.*, 2017; Yvon *et al.*, 2017). Given the suggested positive correlation between transmission and virulence, as both are linked to viral accumulation (Froissart *et al.*, 2010), alterations in the environment may influence the interplay of these epidemiological parameters. Regrettably, studies frequently overlook the plant's ambient environment when investigating such epidemiological associations (Fraile and García-Arenal, 2016).

In this study, we monitored the cumulative effect of heat shock and ToLCNDV on different physiological parameters of tomato plants, like photosynthesis rate, stomatal conductance, transpiration rate, and water use efficiency. To study them, we chose a ToLCNDV-susceptible Panbahar cultivar of tomato from four agroinfiltrated cultivars. This cultivar was agroinfiltrated and given heat shock at different temperatures, then above-mentioned parameters were analyzed for further analysis.

MATERIALS AND METHODS

Plant Material and Growth Conditions

Four cultivars (Panbahar, Riogrande, Roma, Pakit) of tomato plants were grown in controlled laboratory conditions of 26 ± 2 °C temperature, 16/8 hrs light of 4500 lux, and relative humidity of 50-60% in pots.

ToLCNDV susceptible Panbahar cultivar was partitioned into four groups with six plants each. Group I and II are negative and positive controls whereas, III and IV are experimental groups.

Agroinoculation of Plants

The *Agrobacterium tumefaciens* LBA 4404 strain transformed with the infectious clone of ToLCNDV was cultivated in LB broth medium at 28°C and pellets from cultures exhibiting optimal growth with OD_{600} nearing 0.5 in fresh medium were resuspended in an infiltration buffer (Zheng *et al.*, 2021). The lower epidermis of 4-5 weeks old tomato plant leaves was subjected to infiltration using a needleless 1 ml syringe containing the ToLCNDV clone of both genome A (Accession # OQ190948) and genome B (Accession # LN854628) as shown in figure-1. The control plant underwent infiltration with a mock Agrobacterium suspension. The experimental procedure was executed in triplicate.

Figure 1. (A-C) Positive control plants; (D-F) Plants agroinfiltrated with ToLCNDV infectious clone; (G-I) Plants treated with heat at 40°C, 45°C, and 50°C for a duration of two hours; (J-L) Agroinfiltrated plants with ToLCNDV and treated with heat at 40°C, 45°C, and 50°C for a duration of two hours.

Heat Treatment

Three plants from the positive control group II underwent exposure to diverse temperature regimens within the laboratory setting. Elevated temperature stress condition above the physiological value was imposed at 45±2°C for two hours in controlled laboratory conditions, while the control plants were maintained at ambient room temperature.

Biochemical Assays

Various physiological parameters were assessed via biochemical assays, encompassing measurements of photosynthetic and transpiration rates, stomatal conductance, and water use efficiency in both healthy and stressed tomato plants. The experimental procedures were conducted in triplicate for each of the four distinct groups.

Photosynthetic Rate

Floating disk assay was used to determine the rate of photosynthesis (Zemedkun *et al.*, 2019). In a 500 ml beaker, a bicarbonate solution was meticulously prepared by combining 0.5 g of sodium bicarbonate and 1 droplet of surfactant in 300 ml of tap water, ensuring the prevention of bubble formation. Utilizing a hole puncher, leaf discs devoid of prominent veins were precisely excised from the leaves. A sterile syringe, devoid of the plunger, was selected. The leaf discs were introduced into the syringe, and the plunger was reinserted and gradually compressed to expel air without causing damage to the discs. Subsequently, they were immersed in 3 ml of the bicarbonate solution drawn into the syringe. The establishment of a vacuum was achieved by obstructing the syringe tip with a finger, concurrently retracting the plunger, and swirling the syringe. After a ten-second interval, the vacuum was released by lifting the finger, repeating this procedure 3-4 times. The duration for the leaf discs to surface is directly proportional to the rate of photosynthesis occurring within the disks.

Estimation of Stomatal Conductance

The assessment of stomatal conductance involved the implementation of a straightforward nail polish imprint technique. This method encompassed the application of a fine layer of nail polish onto the lower leaf surface, spanning a dimension of one inch in length and 0.3 inches in width, following the protocol established by a prior research consortium (Millstead *et al.*, 2020).

Transpiration Rate

The experimental procedure involved saturation of the soil through watering until optimal moisture levels were achieved. Subsequently, the plants, inclusive of their respective pots, underwent weighing. A hermetic enclosure was established by enveloping the entire plant with an impermeable plastic bag, meticulously sealed at the terminal end of the stem using adhesive tape, with deliberate exclusion of the pot. The plants were then exposed to direct sunlight for one hour, followed by a second weighing session. The plastic bag functioned as a receptacle for the water droplets exuded via transpiration (Zhu *et al.*, 2022). Repeated this several times to determine how much water was lost by plants.

Measurement of Water Use Efficiency

Well-irrigated plants were sustained at water stress for three weeks and the pots were weighed regularly. The total water consumed was calculated by

$$TWC = \frac{(WPot - WPot(min)}{(WPot(max) - WPot(min)} \times 100$$

WPOT is the actual weight of the empty pot, WPOT (min) is the minimum weight of the pot with soil and WPOT (max) is the maximum weight of the pot with soil. The dry weight of the initial biomass and final biomass was determined by drying 1 plant before and 1 plant after the experiment in an incubator at 60° C. The water use efficiency of the whole plant was measured by

$$WUE = \frac{Weight\ of\ final\ dry\ biomass - Weight\ of\ final\ dry\ biomass}{Total\ water\ consumed}$$

The WUE was evaluated for healthy and stressed tomato plants.

The data was evaluated by One-way Analysis of Variance (ANOVA) followed by Dunnett's test for the comparison of means (Adhikari *et al.*, 2020).

RESULTS

Photosynthetic Rate

The rate was slightly low in ToLCNDV group (8.26 ± 0.69 %) as compared to the control group (8.56 ± 0.55 %) with (P=0.768) determining it non-significant. While in the case of ToLCNDV and heat shock comparison, the photosynthetic rate (7 ± 0.58 %) was reduced in heat shock group with a P value of 0.011 which is a significant value. In the combined effect of ToLCNDV and heat shock, the rate was observed to be (5.1 ± 0.54 %) as compared to the control group with P value < .001. This shows a higher negative impact of both stresses on tomato plants, figure-2(a).

Estimation of Stomatal Conductance

Stomatal conductance in control group was the highest (189.08±19.13%) as compared to ToLCNDV (178.88±9.3%, P= 0.676) and heat-treated groups (134.9 ±4.02% P = .001). ToLCNDV and heat shock treated plants showed stomatal conductance of (89.84±1.27% P <.001) as compared to control group plants, showing more reduction in stomatal conductance. Hence, plants treated with heat stress and ToLCNDV showed detrimental effects, figure-2(b).

Transpiration Rate

The transpiration rate was slightly low in ToLCNDV group (2.44 ± 0.27 %) as compared to the control group (2.586 ± 0.415 %) with (P=0.881) proving it non-significant. While in the case of ToLCNDV and heat shock comparison, the photosynthetic rate (1.42 ± 0.577 %) was reduced in the heat shock group with a P value of 0.002 which is a significant value. In the combined effect of ToLCNDV and heat shock, the rate was observed to be (0.978 ± 0.148 %) as compared to the control group with P value < .001. This shows a higher loss in the ability of transpiration because of both stresses on tomato plants, figure-3(a).

Measurement of Water use Efficiency (WUE)

The control group showed the least WUE as compared to the other three groups. Comparison of ToLCNDV and control group showed no notable difference in WUE with (5.78 ± 0.57 %) and (5.84 ± 0.48 %) respectively with a non-significant P = 0.998. Heat shock treated and heat shock + ToLCNDV treated groups showed WUE of (7.02 ± 0.54 %, P = 0.026) and (9.1 ± 0.96 %) respectively, with P< .001 indicating that plants subjected to heat stress showed high WUE and the group facing combined stresses showed higher WUE, figure-3(b).

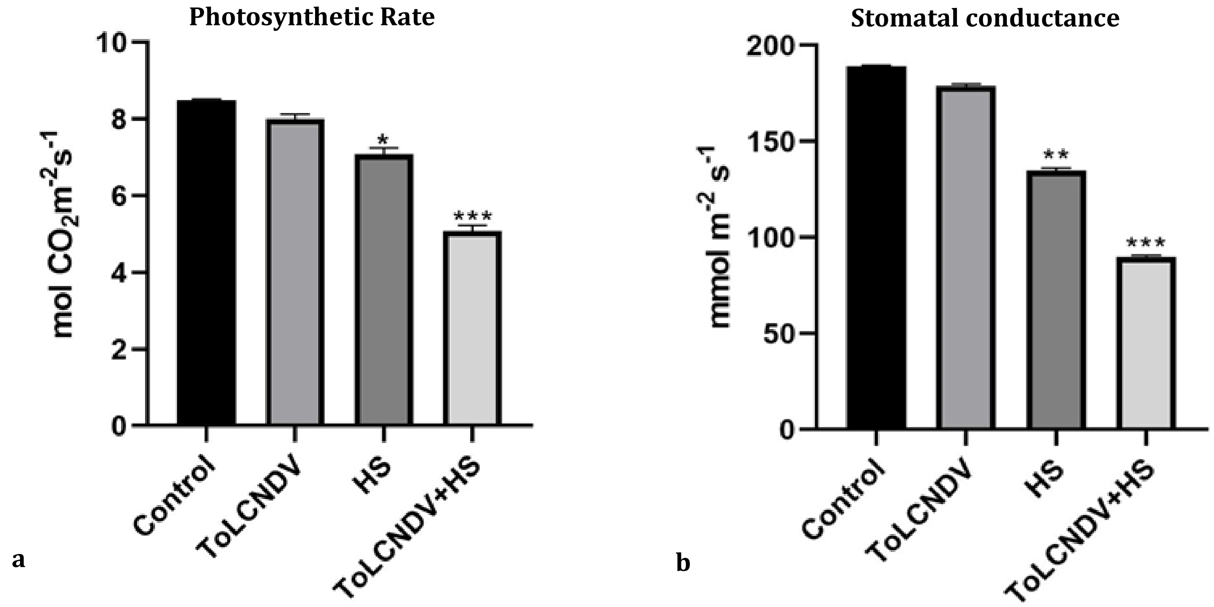

Figure 2. (a) Photosynthetic rates in three experimental groups at different conditions with respect to control plants (*P< 0.05, **P< 0.01, ***P< 0.001), the lowest rate was observed in cumulative stresses (b) Stomatal conductance is also the lowest in case of plants facing both biotic and abiotic stresses.

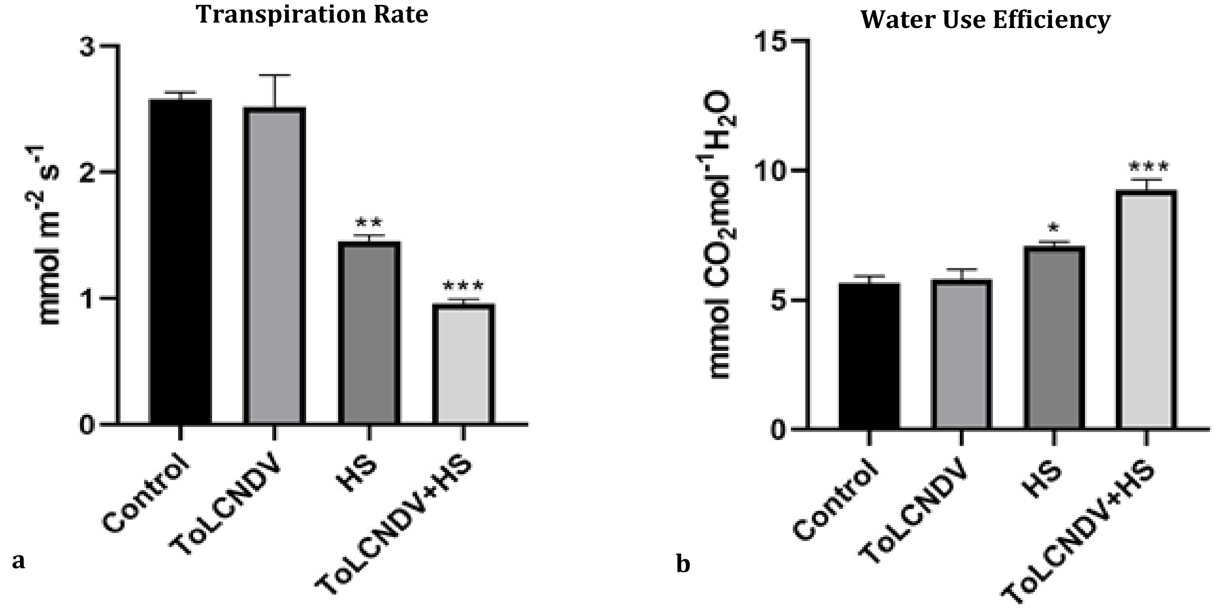

Figure 3. (a) Transpiration rates in three experimental groups at different conditions with respect to control plants (*P< 0.05, **P< 0.01, ***P< 0.001), the lowest rate was observed in cumulative stresses (b) Water use efficiency was the highest in case of HS and ToLCNDV affected plants.

DISCUSSION

The results of the present study shed light on the intricate interplay between tomato leaf curl New Delhi virus (ToLCNDV) infection and heat shock on various physiological parameters of tomato plants. Plants possess inherent adaptations for optimal performance within an environment characterized by favorable thermal conditions. Any departure from the optimum temperature exerts a discernible impact on their physiological growth and developmental processes

(Fraile and García-Arenal, 2016), intricately influencing cellular architecture and molecular signaling pathways (Hatfield and Prueger, 2015). Xu *et al.* (2008) demonstrated the capacity of various RNA viruses, including cucumber mosaic virus (CMV), tobacco mosaic virus (TMV), and bromo mosaic virus (BMV), infecting diverse crop species such as beet, tobacco, and rice, to significantly prolong the onset of drought-related symptoms in their respective hosts after water deprivation (Xu *et al.*, 2008). This phenomenon has also been observed in woody crops, exemplified by grapevine infected with grapevine fanleaf virus (GFLV) (Jež-Krebelj *et al.*, 2022). Furthermore, infection with a DNA virus, namely tomato yellow leaf curl virus (TYLCV), has been identified as conferring both thermotolerance and drought resistance in tomato plants (Mishra *et al.*, 2022; Corrales-Gutierrez *et al.*, 2020). Instances of the contrasting scenario, wherein viral infection induces a diminished tolerance to drought and temperature, though infrequent, have been documented. This is exemplified in the interactions between *Arabidopsis thaliana* (hereinafter referred to as Arabidopsis) and cauliflower mosaic virus (CaMV, a DNA virus) (Bergès *et al.*, 2018) or turnip mosaic virus (TuMV, an RNA virus) (Manacorda *et al.*, 2021). Our findings reveal significant alterations in photosynthetic rate, stomatal conductance, transpiration rate, and water use efficiency (WUE) under different stress conditions.

Firstly, the photosynthetic rate, a key indicator of plant health and productivity, was found to be slightly reduced in ToLCNDV-infected plants compared to the control group. However, the reduction became more pronounced when plants were subjected to heat shock, with a significant decrease observed in the photosynthetic rate. Notably, the combined effect of ToLCNDV infection and heat shock resulted in a substantially lower photosynthetic rate, indicating a synergistic negative impact of both stresses on tomato plants. In a similar study, tomato yellow leaf curl China virus (TYLCCNV) infection promptly led to a reduction in leaf surface area in *Nicotiana benthamiana*, subsequently resulting in a decrease in photosynthetic activity (Farooq *et al.*, 2019). The heat-sensitive photosystem II activity experiences a significant reduction in the face of high-temperature stress. Concurrently, Rubisco activity is notably diminished during heat shock, leading to an increased generation of reactive oxygen species (ROS). The upregulation of Heat Shock Proteins (HSPs) occurs as a regulatory response when plants encounter high temperatures, thereby playing a crucial role in supporting the maintenance of physiological and cellular processes within leaves (Hasanuzzaman *et al.*, 2013). At temperatures exceeding 35 °C, a reduction in the rate of carbon dioxide (CO_2) assimilation occurred, with a more pronounced decline observed during light exposure compared to dark conditions consequently decreasing the rate of photosynthesis (Yin *et al.*, 2010).

Similarly, stomatal conductance, which regulates the exchange of gases and water vapor between plants and the atmosphere, exhibited a significant reduction in ToLCNDV-infected plants and further decreased in response to heat shock. The combined stress of ToLCNDV infection and heat shock resulted in the most substantial reduction in stomatal conductance, highlighting the synergistic detrimental effects of both stressors on plant physiology. In a study, Chick pea plants subjected to elevated temperatures exhibited a decline in moisture content (Prasad *et al.*, 2022), leading to a concomitant reduction in the aperture of stomata (Khetrapal *et al.*, 2009). The tobacco mosaic virus (TMV) infestation disrupts stomatal density regulation, leading to a diminished stomatal conductance in susceptible *N. benthamiana* due to 9.8% reduction in stomatal indices (Murray *et al.*, 2016).

Furthermore, the transpiration rate, crucial for nutrient uptake and cooling of plants, showed a similar trend. While ToLCNDV infection alone did not significantly affect the transpiration rate compared to the control group, heat shock led to a significant reduction. The combined effect of ToLCNDV infection and heat shock resulted in a further decrease in transpiration rate, indicating a cumulative negative impact on the plant's ability to transpire under combined stress conditions. In *Arabidopsis thaliana* with TVCV (Turnip vein-clearing virus) infestation, a disruption in stomatal density of 12.3% reduction was recorded, leading to a diminished stomatal conductance and transpiration rate (Murray *et al.*, 2016).

Interestingly, the measurement of water use efficiency (WUE) revealed that plants subjected to heat stress, either alone or in combination with ToLCNDV infection, exhibited higher WUE compared to the control group. This suggests an adaptive response of plants to heat stress, where they utilize water more efficiently to cope with the environmental challenge. Notably, the group facing combined stresses showed the highest WUE, indicating a potential synergistic effect on water use efficiency under dual stress conditions. In a similar study, plants under combined HS and ToLCNDV stress express higher WUE (Davis *et al.*, 2015). Utilizing cDNA arrays in conjunction with physiological assessments, a group of researchers investigated the impact of concurrent drought and heat shock stress on tobacco plants. The concurrent exposure to drought and heat shock conditions elicited stomatal closure (Farooq *et al.*, 2019), inhibited photosynthetic processes (Vile *et al.*, 2012), heightened respiratory activity, and elevated leaf temperature (Rizhsky *et al.*, 2002).

These findings are consistent with previous studies demonstrating the complex interactions between biotic and abiotic stresses on plant physiology (Atkinson and Urwin, 2012; Adhikari *et al.*, 2020). The synergistic effects observed in this study underscore the importance of considering multiple stressors when assessing plant responses and developing strategies for crop improvement in the face of changing environmental conditions. Our results highlight the significant impact of ToLCNDV infection and heat shock on various physiological parameters of tomato plants, emphasizing the need for integrated approaches to mitigate the effects of multiple stresses on crop productivity and resilience. These investigations are paving the way for groundbreaking possibilities in the systematic engineering of sustained virus resistance in crops, consequently augmenting crop yield and overall productivity.

CONCLUSION

This study facilitates the comprehension of the responses exhibited by tomato plants under the concurrent influence of elevated temperatures and stress induced by ToLCNDV infection. Individual exposure to high temperature or ToLCNDV infestation elicited relatively modest effects on the rates of photosynthesis and transpiration. However, when both stresses were simultaneously applied, a substantial reduction in the rates of photosynthesis and transpiration was observed, accompanied by an augmentation in water uptake. The insights gained through the analysis of diverse physiological trait variations under stressful conditions hold promise for the development of stress-tolerant tomato cultivars. Furthermore, the acquired data can be extrapolated to other members of the Solanum genus to unravel the impacts of singular or combined biotic and abiotic stresses. Plants experiencing multiple stressors exhibit heightened susceptibility to damage. The ongoing escalation of Earth's temperature precipitates profound environmental transformations, necessitating the introduction of biotechnologically engineered stress-tolerant crop varieties to counteract the escalating threats posed by biotic and abiotic stresses in the environment.

CONFLICT OF INTEREST

No authors had any conflicts regarding the research paper.

AUTHORS CONTRIBUTION

All authors contributed equally to the research and manuscript writing.

REFERENCES

Adhikari, B., M. Adhikari, B. Ghimire, B. C. Adhikari, G. Park and E. H. Choi. 2020. Cold plasma seed priming modulates growth, redox homeostasis and stress response by inducing reactive species in tomato (*Solanum lycopersicum*). Free Radical Biology and Medicine, 156: 57-69.

Atkinson, N. J. and P. E. Urwin. 2012. The interaction of plant biotic and abiotic stresses: From genes to the field. Journal of Experimental Botany, 63: 3523-43.

Berges, S. E., F. Vasseur, A. Bediee, G. Rolland, D. Masclef, M. Dauzat, M. van Munster and D. Vile. 2020. Natural variation of *Arabidopsis thaliana* responses to Cauliflower mosaic virus infection upon water deficit. PLoS Pathogens, 16: e1008557.

Bergès, S. E., D. Vile, C. Vazquez-Rovere, S. Blanc, M. Yvon and M. van Munster. 2018. Interactions between drought and plant genotype change epidemiological traits of Cauliflower mosaic virus. Frontiers in Plant Science, 9: 363661.

Bragard, C., P. Caciagli, O. Lemaire, J. Lopez-Moya, S. MacFarlane, D. Peters, P. Susi and L. Torrance. 2013. Status and prospects of plant virus control through interference with vector transmission. Annual Review of Phytopathology, 51: 177-201.

Corrales-Gutierrez, M., L. Medina-Puche, Y. Yu, L. Wang, X. Ding, A. P. Luna, E. R. Bejarano, A. G. Castillo and R. Lozano-Duran. 2020. The C4 protein from the geminivirus Tomato yellow leaf curl virus confers drought tolerance in *Arabidopsis* through an ABA-independent mechanism. Plant Biotechnology Journal, 18: 1121.

Davis, T. S., N. A. Bosque-Pérez, N. E. Foote, T. Magney and S. D. Eigenbrode. 2015. Environmentally dependent host–pathogen and vector–pathogen interactions in the Barley yellow dwarf virus pathosystem. Journal of Applied Ecology, 52: 1392-401.

Farooq, T., D. Liu, X. Zhou and Q. Yang. 2019. Tomato yellow leaf curl china virus impairs photosynthesis in the infected *Nicotiana benthamiana* with βC1 as an aggravating factor. The Plant Pathology Journal, 35: 521.

Fraile, A. and F. García-Arenal. 2016. Environment and evolution modulate plant virus pathogenesis. Current Opinion in Virology, 17: 50-56.

Froissart, R., J. Doumayrou, F. Vuillaume, S. Alizon and Y. Michalakis. 2010. The virulence–transmission trade-off in vector-borne plant viruses: A review of (non-) existing studies. Philosophical Transactions of the Royal Society B: Biological Sciences, 365: 1907-18.

González, R., A. Butković, F. J. Escaray, J. Martínez-Latorre, Í. Melero, E. Pérez-Parets, A. Gómez-Cadenas, P. Carrasco and S. F. Elena. 2021. Plant virus evolution under strong drought conditions results in a transition from parasitism to mutualism. Proceedings of the National Academy of Sciences, 118: e2020990118.

Hatfield, J. L. and J. H. Prueger. 2015. Temperature extremes: Effect on plant growth and development. Weather and Climate Extremes, 10: 4-10.

He, Y.-Z., Y.-M. Wang, T.-Y. Yin, E. Fiallo-Olivé, Y.-Q. Liu, L. Hanley-Bowdoin and X.-W. Wang. 2020. A plant DNA virus replicates in the salivary glands of its insect vector via recruitment of host DNA synthesis machinery. Proceedings of the National Academy of Sciences, 117: 16928-37.

Hily, J. M., N. Poulicard, M. Á. Mora, I. Pagán and F. García-Arenal. 2016. Environment and host genotype determine the outcome of a plant–virus interaction: From antagonism to mutualism. New Phytologist, 209: 812-22.

Hosseini, S. A., G. R. Zamani, Z. M. Yaghub and M. Khayyat. 2018. Effects of Cucumber Mosaic Virus infection and drought tolerance of tomato plants under greenhouse conditions: Preliminary results. Journal of Berry Research, 8: 129-36.

Jež-Krebelj, A., M. Rupnik-Cigoj, M. Stele, M. Chersicola, M. Pompe-Novak and P. Sivilotti. 2022. The physiological impact of GFLV virus infection on grapevine water status: First observations. Plants, 11: 161.

Khetrapal, S., M. Pal and S. Lata. 2009. Effect of elevated temperature on growth and physiological characteristics in chickpea cultivars. Indian Journal of Plant Physiology, 14: 377-83.

Manacorda, C. A., G. Gudesblat, M. Sutka, S. Alemano, F. Peluso, P. Oricchio, I. Baroli and S. Asurmendi. 2021. TuMV triggers stomatal closure but reduces drought tolerance in *Arabidopsis*. Plant, Cell and Environment, 44: 1399-416.

Mazzeo, M. F., G. Cacace, P. Iovieno, I. Massarelli, S. Grillo and R. A. Siciliano. 2018. Response mechanisms induced by exposure to high temperature in anthers from thermo-tolerant and thermo-sensitive tomato plants: A proteomic perspective. Plos One, 13: e0201027.

Mellidou, I., A. Koukounaras, S. Kostas, E. Patelou and A. K. Kanellis. 2021. Regulation of vitamin C accumulation for improved tomato fruit quality and alleviation of abiotic stress. Genes, 12: 694.

Millstead, L., H. Jayakody, H. Patel, V. Kaura, P. R. Petrie, F. Tomasetig and M. Whitty. 2020. Accelerating automated stomata analysis through simplified sample collection and imaging techniques. Frontiers in Plant Science, 11: 580389.

Mishra, R., M. Shteinberg, D. Shkolnik, G. Anfoka, H. Czosnek and R. Gorovits. 2022. Interplay between abiotic (drought) and biotic (virus) stresses in tomato plants. Molecular Plant Pathology, 23: 475-88.

Murray, R. R., M. S. Emblow, A. M. Hetherington and G. D. Foster. 2016. Plant virus infections control stomatal development. Scientific Reports, 6: 34507.

Prasad, A., S. Sett and M. Prasad. 2022. Plant-virus-abiotic stress interactions: A complex interplay. Environmental and Experimental Botany, 199: 104869.

Raja, V., S. U. Qadir, M. N. Alyemeni and P. Ahmad. 2020. Impact of drought and heat stress individually and in combination on physio-biochemical parameters, antioxidant responses, and gene expression in *Solanum lycopersicum*. 3 Biotech, 10: 1-18.

Rizhsky, L., H. Liang and R. Mittler. 2002. The combined effect of drought stress and heat shock on gene expression in tobacco. Plant Physiology, 130: 1143-51.

Thole, V., P. Vain and C. Martin. 2021. Effect of elevated temperature on tomato post-harvest properties. Plants, 10: 2359.

Tiwari, S., A. Patel, M. Singh and S. M. Prasad. 2020. Regulation of temperature stress in plants. In: Durgesh Kumar Tripathi, Vijay Pratap Singh, Devendra Kumar Chauhan, Shivesh Sharma, Sheo Mohan Prasad, Nawal Kishore Dubey and Naleeni Ramawat (eds.), Plant Life Under Changing Environment Academic Press.

Van Munster, M., M. Yvon, D. Vile, B. Dader, A. Fereres and S. Blanc. 2017. Water deficit enhances the transmission of plant viruses by insect vectors. Plos One, 12: e0174398.

Vile, D., M. Pervent, M. Belluau, F. Vasseur, J. Bresson, B. Muller, C. Granier and T. Simonneau. 2012. *Arabidopsis* growth under prolonged high temperature and water deficit: Independent or interactive effects? Plant, Cell and Environment, 35: 702-18.

Xu, P., F. Chen, J. P. Mannas, T. Feldman, L. W. Sumner and M. J. Roossinck. 2008. Virus infection improves drought tolerance. New Phytologist, 180: 911-21.

Yin, Y., S. Li, W. Liao, Q. Lu, X. Wen and C. Lu. 2010. Photosystem II photochemistry, photoinhibition, and the xanthophyll cycle in heat-stressed rice leaves. Journal of Plant Physiology, 167: 959-66.

Yvon, M., D. Vile, V. Brault, S. Blanc and M. van Munster. 2017. Drought reduces transmission of Turnip yellows virus, an insect-vectored circulative virus. Virus Research, 241: 131-36.

Zemedkun, D., H. Alaparmak and S. Apte. 2019. The effect of herbicides on the rates of photosynthesis and respiration in spinach leaves. Journal of High School Science, 12: 7518.

Zheng, L., J. Yang, Y. Chen, L. Ding, J. Wei and H. Wang. 2021. An improved and efficient method of *Agrobacterium syringe* infiltration for transient transformation and its application in the elucidation of gene function in poplar. BMC Plant Biology, 21: 1-19.

Zhu, Y., Z. Cheng, K. Feng, Z. Chen, C. Cao, J. Huang, H. Ye and Y. Gao. 2022. Influencing factors for transpiration rate: A numerical simulation of an individual leaf system. Thermal Science and Engineering Progress, 27: 101110.