Uluslararası Tarım Araştırmalarında Yenilikçi Yaklaşımlar Dergisi
International Journal of Innovative Approaches in Agricultural Research 2024, Vol. 8 (4), 289-299
https://doi.org/10.29329/ijiaar.2024.1109.2

Original article

The Effect of Different Nitrogen Forms on Tomato Spotted Wilt Virus Infection in Pepper Plants Grown in Full and Deficient-Water Conditions

Zeynep Aslan [a, *], Miray Arlı-Sökmen [a] & Coşkun Gülser [a]

[a] Department of Plant Protection, Faculty of Agriculture, Ondokuz Mayis University, Samsun, Türkiye
[b] Department of Soil Science and Plant Nutrition, Faculty of Agriculture, Ondokuz Mayis University, Samsun, Türkiye

Abstract

Abiotic stress factors have an impact on both plants and their pathogens. Water and nutrient deficiencies are among the major abiotic stress factors in agricultural systems. Viruses are obligate parasite pathogens causing detrimental yield reductions on crop plants worldwide. However, there have been limited studies on the impact of abiotic factors on plant-virus interactions. In this study, the effects of different forms of nitrogen (N) fertilizers on Tomato spotted wilt virus (TSWV) infection in pepper plants grown under two different irrigation regimes [water deficient (30%) and full irrigation] were investigated in a growth room condition. Fertilizer applications consisted of five individual treatments with three replications. The pepper plants were supplied with 12 kg of N/da using different sources of nitrogen [urea (CH4N2O), NH4NO3, Mg(NO3)2 and (NH4)2SO4]. MgSO4 was also added to the experiment for comparison of the effects of Mg2+ and SO42- in the fertilizers. Each treatment consisted of TSWV-inoculated plants, as well as non-inoculated (healthy) and non-fertilized plants as controls. Every plant in all treatments was tested by enzyme-linked immunosorbent assay (ELISA) to confirm virus infection four and eight weeks after mechanical inoculation. The results showed that the virus-inoculated plants treated with urea and NH4NO3 had lower ELISA absorbance values in both deficient and full-irrigated conditions ($p<0.01$), suggesting greater tolerance to TSWV infection with these fertilizers. Additionally, the plants treated with Mg(NO3)2-containing fertilizer were relatively more affected by the virus under full irrigation than in water-deficient conditions. Conversely, the plants treated with (NH4)2SO4 had high virus content in deficient irrigation conditions. Despite better canopy development in full-irrigated conditions, TSWV symptoms in pepper plants were less prominent in water-deficient conditions than in full irrigation across all nutrient treatments. This study suggests that urea, NH4NO3, and Mg(NO3)2 may provide a positive contribution to plant fitness and virus suppression under water-deficient conditions. It is necessary to conduct further research to determine the impact of different nutrients and water levels on plant-virus interactions in field conditions.

Keywords: TSWV, Plant Nutrients, Water Stress, Disease Tolerance.

Received: 02 August 2024 * **Accepted:** 17 September 2024 * **DOI:** https://doi.org/10.29329/ijiaar.2024.1109.2

* **Corresponding author:**

Aslan Zeynep is a research assistant at the Department of Plant Protection in Ondokuz Mayis University, Samsun. Her research interests include agriculture, phytopathology and plant virology. She completed her undergraduate education at Ankara University. She studied on virus-plant interactions during her MSc thesis at Ondokuz Mayis University.
Email: zeynep.aslan@omu.edu.tr

INTRODUCTION

Interactions between plants, pathogens, and abiotic stress factors involve complex mechanisms that are not completely understood. Nutrient deficiencies and water shortage are among the major abiotic stressors in agricultural systems. The amount and type of nutrients significantly impact plant growth as well as the reproduction and development of microorganisms surrounding them. It has been found that plant disease development is connected with nutrients and soil water content (Agrios, 2005). Resistance or tolerance of plants to pathogenic microorganisms is mainly controlled by specific genes. However, the ability of a plant to express these genes, which can enhance the plant's fitness against pathogens, is affected by some environmental factors, such as mineral nutrition (Walters and Bingham, 2007; Marschner and Marschner, 2012). Nutrients play a crucial role in activating the production of enzymes that influence the synthesis of defense-related metabolites in plants (Datnoff et al., 2007). Seventeen mineral nutrients are essential for the survival of plants (Bolat and Kara, 2017), with nitrogen (N) being particularly important for various biochemical and physiological processes (Fageria, 2009). Adequate uptake of nitrogen is vital for the production of various structures and biochemical substances necessary for growth (Elmer ve Datnoff, 2014). Nitrogen may also affect plant-pathogen interactions (Dordas, 2008). However, different forms of nitrogen have varying effects on plant diseases (Bolton and Thomma, 2008; Mur et al., 2017). For instance, Fusarium wilt problem increased in plants such as asparagus, basil, sugar beet, tobacco, tomato, and watermelon when grown in soils with nitrate (NO_3^-) fertilization, while it decreased in soils with ammonium (NH_4^+) fertilization. On the other hand, *Gaeumannomyces* spp., *Thielaviopsis* spp., and *Verticillium* spp. infections were supressed by NH_4-N, but not by NO_3-N (Elmer ve Datnoff, 2014).

Viruses are among the most challenging pathogens to control, causing significant yield reductions in crop production. Tomato spotted wilt virus (TSWV), is one of the most devastating plant viruses, infecting over 1000 species worldwide. It has been ranked second in the top ten list of plant viruses in terms of scientific and economic significance (Scholthof et al., 2011). Tomato, pepper, tobacco, peanut, lettuce, and ornamental plants are among the most affected crops (Pataky, 1991; Rosello et al., 1996). The virus is a member of the *Orthotospovirus* genus in the *Tospoviridae* family (Abudurexiti et al., 2019), and the quasispherical enveloped virus particles (80-120 nm in diameter) contain three negative sense or ambisense single-strand RNA components (Kormelink, 2005). In nature, TSWV is efficiently transmitted by thrips *(Thysanoptera: Thripidae*) in a persistent-propagative manner, with *Thrips tabaci* Lindeman and *Frankliniella occidentalis* Pergande being the most important vectors (Krishna-Kumar et al., 1993).

There have been fewer studies on the impact of abiotic factors on the interactions between plants and viruses compared to fungal and bacterial pathogens. This paper presents the findings of a study that looked at how different nitrogen fertilizers affect TSWV infection in pepper plants grown under two different irrigation regimes, water deficient (30% of maximum water holding capacity) and full irrigation (70% of maximum water holding capacity), in a growth room condition.

MATERIALS and METHODS

Propagation of the virus isolate

The TSWV isolate-*Tsw* RB (kindly provided by Dr. İlyas Deligoz, The Black Sea Research Institute, Samsun, Türkiye), which is known to overcome the *Tsw*-resistance gene in pepper, was used in this study. Six week-old seedlings of pepper cv. Kandil Dolma (Arzuman), susceptible to TSWV, were inoculated with the TSWV isolate.

The inoculum was prepared by grinding 1 g of infected leaves in 10 ml of phosphate buffer [1% K_2HPO_4 (w/v) containing 0.1% sodium sulfite (w/v) and 0.2 M 2-mercapto ethanol] in a sterilized ice-cold mortar with a pestle (Mandal et al., 2008). The inoculum was then mixed with 1% carborundum powder (400 mesh; w/v) and rubbed onto the leaves of pepper plants (Mandal et al., 2001). The inoculated plants were maintained in growth room with 16 hours of photoperiod and temperatures of 25ºC (light) and 20ºC (dark) for virus propagation.

Nutrient applications

Four-week-old pepper seedlings were planted in 20-cm-diameter plastic pots containing a growth media mixture of perlite and peat in a ratio 1:4 (w: w). The study aimed to assess the impact of different nitrogen sources on TSWV infection in pepper plants. Four different N fertilizers [urea (CH_4N_2O), NH_4NO_3, $Mg(NO_3)_2$, $(NH_4)_2SO_4$ were used in the study. Additionally, $MgSO_4$ was used as a fertilizer source to compare the effects of Mg^{2+} and SO_4^{2-} having in the fertilizers besides nitrogen. The pepper plants received 12 kg/da dose of N from each fertilizer. The experiment was arranged in a randomized complete-plot design with three replicates. Each irrigation treatment contained virus-inoculated, non-inoculated (control 1) and non-fertilized (control 2) plants (Table 1). The experiment involved a total of 72 plants, half of which were mechanically inoculated with TSWV as described above 2 weeks after nutrient application. Throughout the experiment, the plants were observed for virus symptoms and vigor twice a week.

Irrigation and determination of the maximum water-holding capacity of growth medium

Two different irrigation levels consisting of 30% (deficient irrigation) and 70% (full irrigation) of the maximum water-holding capacity were used to investigate the impact of water stress on plant virus-nutrient interactions and disease severity in a pot experiment. The maximum water-holding capacity of the growth medium was determined based on previous reports linking it to the total porosity of the growth medium (Reynolds et al., 2002). The determination of the maximum water-holding capacity of the growth medium was carried out following the method outlined by Labuschagne et al.

(1995). Throughout the nine-week experiment, each plant in all treatment groups was watered at two-day intervals to ensure that the moisture content of the growth medium was maintained.

Table 1. Treatments used to investigate the effects of different sources of nitrogen fertilizers on Tomato spotted wilt virus infection in pepper under two irrigation regimes

Treatments				
	Full irrigation (70%)	**Replication**	**Deficient irrigation (30%)**	**Replication**
TSWV-inoculated	NH_4NO_3	3	NH_4NO_3	3
	$(NH_4)_2SO_4$	3	$(NH_4)_2SO_4$	3
	$Mg(NO_3)_2$	3	$Mg(NO_3)_2$	3
	$MgSO_4$	3	$MgSO_4$	3
	Urea	3	Urea	3
	Non-fertilized	3	Non-fertilized	3
Non-inoculated	NH_4NO_3	3	NH_4NO_3	3
	$(NH_4)_2SO_4$	3	$(NH_4)_2SO_4$	3
	$Mg(NO_3)_2$	3	$Mg(NO_3)_2$	3
	$MgSO_4$	3	$MgSO_4$	3
	Urea	3	Urea	3
	Non-fertilized	3	Non-fertilized	3
Total		36		36

Phenotypic assessment of pepper plants

Pepper plants in all treatments were individually assessed for virus symptoms and plant vigor on a weekly basis. Virus symptoms and general plant characteristics were then recorded and photographed four and six weeks after virus inoculation.

Serological testing of pepper plants

The upper leaves from each plant in all treatments were collected and subjected to double antibody sandwich-enzyme linked immunosorbent assay (DAS-ELISA) following the method described by Clark and Adams (1972) and the instructions provided by the antiserum manufacturer (Bioreba, Switzerland). This testing was performed at the fourth and eighth weeks after virus inoculation. Each plant sample was tested in duplicate wells, and a sample with mean absorbance values at 405 nm two-fold higher than that of non-inoculated controls was considered positive for the virus infection (Uyemeto et al., 1989).

Statistical Analysis

ANOVA test for the data obtained from this study was conducted using the randomized complete plot design in the SPSS program (Version 17). Statistical differences among the mean values were determined using the Duncan multiple comparisons test.

RESULTS and DISCUSSION

Symptoms of the Tomato spotted wilt virus-*Tsw* RB isolate on pepper

Two weeks after inoculation, inoculated pepper cv. Kandil dolma plants to propagate the TSWV-*Tsw* RB isolate displayed chlorotic ring spots on older leaves and yellow patches and mosaic on upper leaves (Fig. 1). The ELISA method applied for testing these leaves confirmed TSWV infection in the pepper plants.

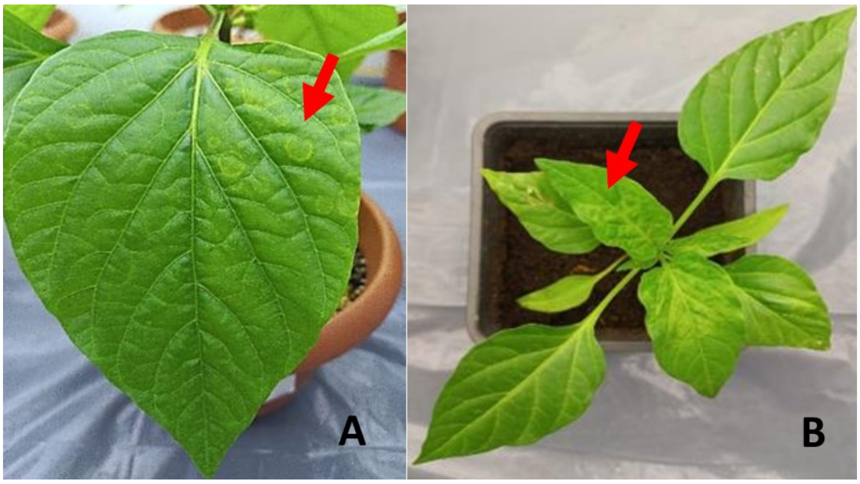

Figure 1. Symptoms of Tomato spotted wilt virus-*Tsw* RB isolate on older leaf (A) and upper leaves of pepper cv. Kandil Dolma

Effects of fertilizer applications on TSWV infection in pepper under full and deficient-irrigation conditions

Comparing non-inoculated pepper plants treated with five different fertilizers [urea, NH_4NO_3, $Mg(NO_3)_2$, $(NH_4)_2SO_4$, $MgSO_4$] to non-fertilized control plants showed that the fertilized plants, regardless of the irrigation regime, exhibited larger canopy development compared to the non-fertilized plants. Additionally, the leaves of the non-fertilized plants displayed a yellowish appearance by the end of the experiment. Most of these plants produced fewer flowers and fruit, as expected. The plants that were fertilized with urea, ammonium nitrate, and magnesium nitrate displayed better vegetative growth and had abundant flowers and fruits, unlike the plants fertilized with magnesium sulfate (data not shown). Additionally, pepper plants that were inoculated with TSWV and non-inoculated plants under 30% water deficit in all nutrient treatments showed reduced leaf size and plant height compared to those under full irrigation (70%). Despite better canopy development in well-watered conditions, virus symptoms in pepper plants were less noticeable in water-deficient conditions across all nutrient treatments (Fig. 2). Similarly, in their 2016 study, Putra and Damayanti found that Sugarcane streak mosaic virus (SCSMV) infections were more severe when soil water content was higher. This confirmed their previous field observations showing that the incidence of SCSMV was higher under irrigated conditions than under rain-fed conditions (Damayanti and Putra, 2011). Hosseini et al. (2018) demonstrated that the severity of Cucumber mosaic virus (CMV) symptoms in tomato plants decreased in dry conditions. While most previous studies have focused on the interactions between plants and viruses concerning water content or nutrients (Putra and Damayanti, 2016; Kendig et al., 2020; Easterday et al., 2022; Mutebi and Moranga, 2022), this study reports the results of multiple interactions involving plant, virus, nutrient, and water content.

When testing pepper plants using DAS-ELISA four weeks after virus inoculation, it was observed that the virus-inoculated-non-fertilized plants had relatively higher mean ELISA absorbance values in low irrigation conditions compared to plants in fully irrigated conditions. Interestingly, plants treated with either urea or ammonium nitrate displayed lower ELISA absorbance values in both deficient and fully irrigated conditions. There was no significant difference between the mean absorbance values of infected and healthy (non-inoculated) plants in these fertilizer treatments in both irrigation regimes ($p<0.01$), suggesting increased tolerance to TSWV infection with these fertilizers (Fig. 3). Moreover, virus-infected plants treated with magnesium nitrate fertilizer exhibited lower mean ELISA absorbance values under water-deficit (30%) conditions compared to fully irrigated (70%) conditions (Fig. 3). Virus-infected plants treated with ammonium sulfate fertilizer gave higher ELISA values (Fig. 3), indicating increased virus concentration under full irrigation and water-deficient conditions. Additionally, magnesium sulfate fertilization may have a negative effect on pepper plants' health, as evidenced by the less vigorous appearance of plants in both irrigation conditions (Fig. 2). These findings suggest that Mg^{++} in magnesium nitrate fertilizer and SO_4^{2-} in ammonium sulfate fertilizer may not significantly contribute to pepper plant’s tolerance to TSWV infection.

In a 1998 study, it was found that the incidence of TSWV infection in field-grown plants using 100 kg N/ha in full water conditions was 19.1%, while it was 69.1% in plants grown under water stress (Camele et al., 2000). The difference between the results of this study and our study regarding the relationship between virus supression and water conditions could be attributed to the form of nitrogen in the fertilizer used and the involvement of vectors in virus transmission in the field. It has been reported that nitrate and ammonium forms of nitrogen have different effects on plant's tolerance to pathogen attacks (Sun et al., 2020). The role of nitrogen in the mechanism of plant defense has not been well understood. While a high dose of nitrogen increased the disease incidence of some obligate parasite fungi, it decreased the incidence of some facultative parasite fungi (Dordas, 2008). The effect of different doses of nitrogen on TSWV infection was not investigated in our study; however, future investigations about the impact of different doses of nitrogen in pepper-TSWV interaction are necessary to be completed in the field conditions with various water contents.

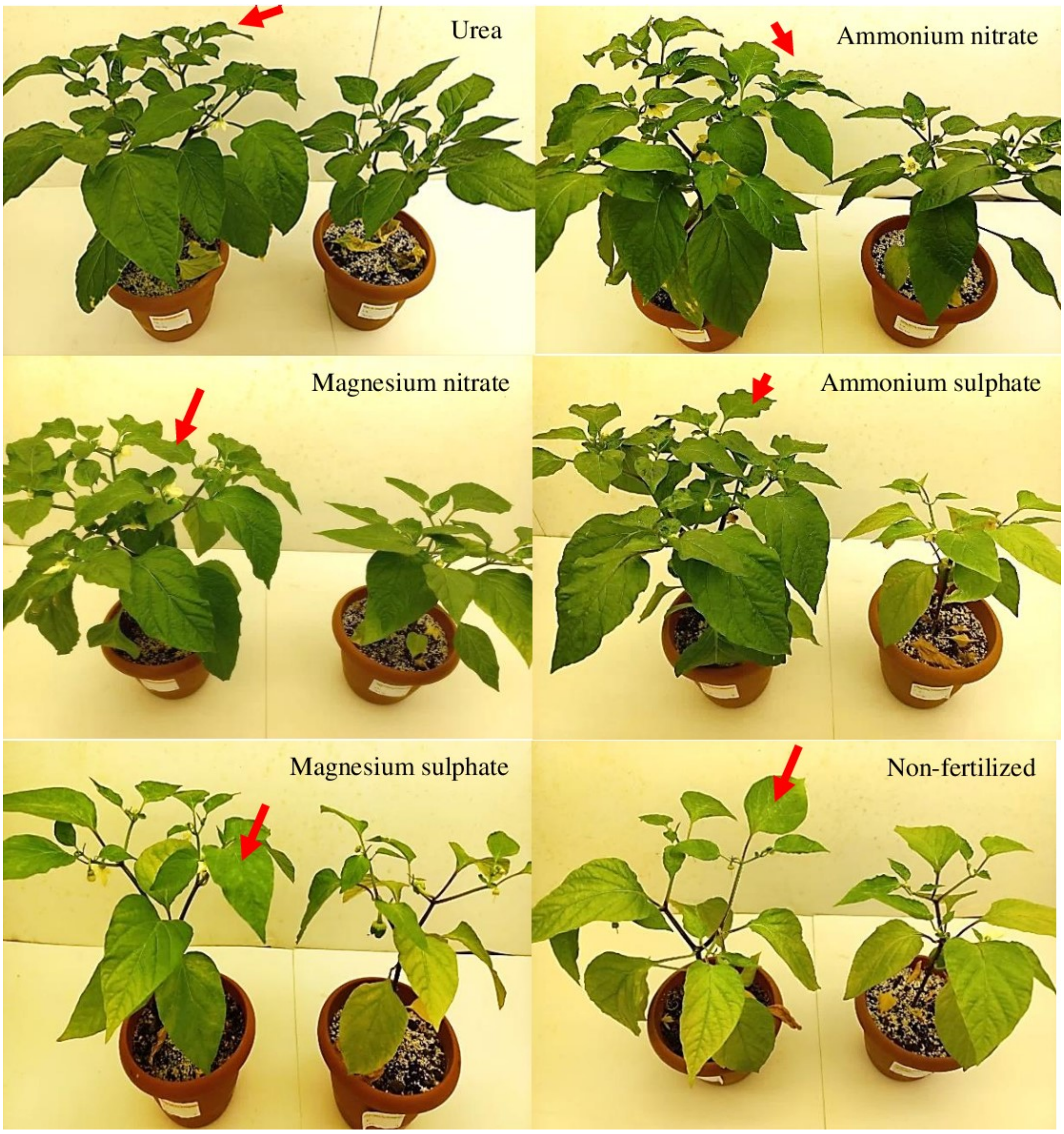

Figure 2. Symptoms of Tomato spotted wilt virus in pepper cv. Kandil Dolma under full (plants on the left) and deficient irrigation (plants on the right) conditions six weeks after fertilization/four weeks after virus inoculation. Chlorotic spots and leaf deformations were indicated with red arrow.

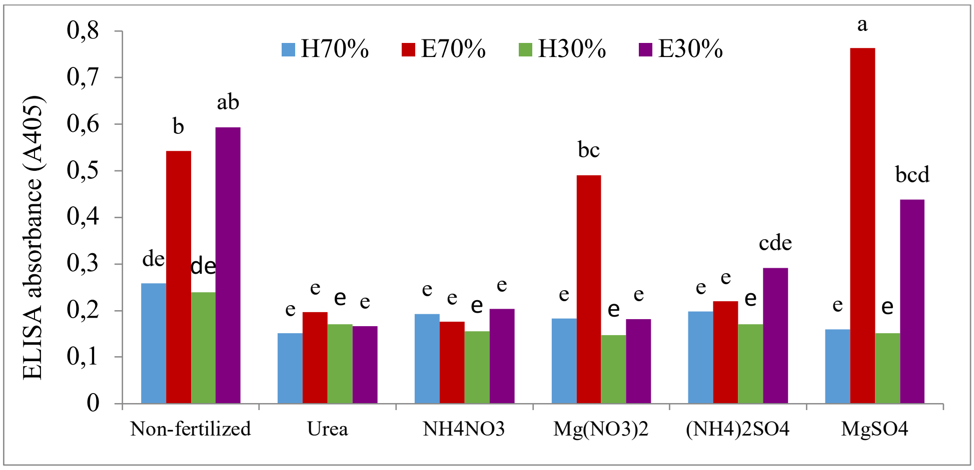

Figure 3. The mean ELISA absorbance values of virus-inoculated and non-inoculated pepper plants grown under full (70%) and deficient water (30%) conditions and treated with different nutrient sources. H: non-inoculated plants, E: virus-inoculated plants. The same letters indicate the absence of significant statistical differences between treatments ($p < 0.01$)

The results of this study indicated that the nitrogenous fertilizers containing urea and ammonium nitrate had a beneficial effect on pepper plants infected with TSWV, both under full irrigation and water-deficient conditions. Ammonium sulfate showed a similar impact under full irrigation conditions, while magnesium nitrate was effective under water-deficient conditions (Fig.3). This study did not explain why these plants exhibited milder virus symptoms and had lower ELISA absorbance values, especially in water-deficient conditions, but it is known that plant hormones can influence plant tolerance to biotic, and abiotic factors such as drought (Takahashi et al., 2020). Water shortage can be detected by plant root systems, which leads to the production and transportation of some signal molecules from roots to shoots, ultimately resulting in abscisic acid (ABA) synthesis in plant leaves. ABA may have positive and negative effects on the plant defense system against viruses. In the early stages of infection, it regulates the stomata closure and causes the accumulation of callose that limits cell-to-cell movement of viruses such as Tobacco mosaic virus and Tobacco necrosis virus. Application of ABA from external sources has been shown to reduce virus symptoms and virus levels (Iriti and Faoro, 2008; Alazem et al., 2014). Furthermore, ABA may have positive impact on other resistance mechanisms such as RNA interference (Li et al., 2012). On the other hand, activated ABA during the late stages of infection may potentially have an antagonistic effect on salicylic acid, which plays a crucial role in the local and systemic resistance of plants to certain pathogens (Alazem and Lin, 2015). Additionally, interactions between nitrogen and other plant nutrients may impact disease suppression. For instance, the NO_3^- induces potassium (K) uptake of the plant, but NH_4^+ has a controversial role in K uptake (Marschner, 1995). It is well known that K is one of the key elements in disease protection.

Conclusion

Plant-pathogen-environment interactions are based on various complex biological events. Although disease resistance to a specific pathogen is controlled by the interaction of both plant and pathogen genes, properly managing nutrition supply to plants can reduce disease severity and symptoms. Nitrogen is an essential nutrient for plants and crucial for many metabolic and physiological processes. In the current study conducted in controlled growth room conditions, pepper plants were treated with different sources of nitrogen [urea (CH4N2O), NH4NO3, Mg(NO3)2 and (NH4)2SO4] and exposed to TSWV infection and different irrigation levels. The results of the study indicated that CH4N2O and NH4NO3 may provide a positive contribution to plant fitness and virus supression under both full irrigation and water-deficient conditions. Additionally, Mg(NO3)2 had a similar positive effect under water-deficient conditions. In contrast, the application of (NH4)2SO4 fertilizer resulted in a lower virus level under full irrigation conditions. Despite better canopy development in full-irrigated conditions, TSWV symptoms were less pronounced in pepper plants in water-deficient conditions than in full irrigation, regardless of the nutrient treatments. It is necessary to conduct further research to understand the impact of different nutrients and water levels on plant-virus interactions in field conditions.

Acknowledgment

This study was supported by by Ondokuz Mayis University, Scientific Research Projects Unit under Grant Number PYO.ZRT.1908.22.012.

Conflict of Interest

There is no conflict of interest between the authors of the article.

REFERENCES

Abudurexiti, A., Adkins, S., Alioto, D., Alkhovsky, S. V., Avšič-Županc, T., et al. (2019). Taxonomy of the order Bunyavirales. *Arch. Vir.*, 164:1949-1965. https://doi.org/10.1007/s00705-019-04253-6.

Agrios, G. (2005). *Plant Pathology*. 5th Edition, Elsevier Academic Press, Amsterdam. 922 pp.

Alazem, M., Lin, K.Y. and Lin, N.S. (2014). The abscisic acid pathway has multifaceted effects on the accumulation of bamboo mosaic virus. *Molecular Plant-Microbe Interact*, 27: 177-189. http://dx.doi.org/10.1094/MPMI-08-13-0216-R.

Alazem, M, Lin, N. S. (2015). Roles of plant hormones in the regulation of host–virus interactions. *Molecular Plant Pathology*. 16 (5): 529-540. doi: 10.1111/mpp.12204.

Bolat, İ. ve Kara, Ö. (2017). *Bitki besin elementleri: Kaynakları, işlevleri, eksik ve fazlalıkları*. *Bartın Orman Fakültesi Dergisi,* 19(1): 218-228. Doi: 10.24011/barofd.251313.

Bolton, M. D. and Thomma, B. P. H. J. (2008). The complexity of nitrogen metabolism and nitrogen-regulated gene expression in plant pathogenic fungi. *Physiol. Mol. Plant Path.*, 72: 104-110. https://doi.org/10.1016/j.pmpp.2008.07.001.

Camele, I., Candido, V., Miccolis, V. and Rana, G. L. (2000). Influence of nitrogen fertilization and water stress on incidence of virus infections in sweet pepper culture under glasshouse. Atti, *Giornate Fitopatologiche*, Perugia, Vol. Primo: 51-54.

Clark, M.F. and Adams, A.N. (1977). Characteristics of the microplate method of enzyme linked immunosorbent assay for the detection of plant viruses. *J. Gen. Virol.*, 34(3): 475-483.

Damayanti, T. A., Putra, L. K. (2011). First occurrence of Sugarcane streak mosaic virus infecting sugarcane in Indonesia. *J. Gen. Plant Path.*, 77: 72-74. Doi 10.1007/s10327-010-0285-7.

Datnoff, L.E., Elmer, W.H., and Huber, D.M. (2007). Mineral nutrition and plant disease-American Phytopathological Society, *J. Plant Prot. Res.*, 48(1): 106. Doi: 10.2478/v10045-008-0016-7.

Dordas, C. (2008). Role of nutrients in controlling plant diseases in sustainable agriculture. A review. *Agron. Sustain. Dev.*, 28: 33-46. Doi: 10.1051/agro:2007051.

Easterday, C.A., Kendig, A.E., Lacroix, C., Seabloom, E.W., Borer, E.T. (2022). Long-term nitrogen enrichment mediates the effects of nitrogen supply and co-inoculation on a viral pathogen. *Ecol. Evol.*, 12(1): e8450. https://doi.org/10.1002/ece3.8450.

Elmer, W.H., Datnoff, L.E. (2014). Mineral Nutrition and Suppression of Plant Disease. In: Neal VA. *Encyclopedia of Agriculture and Food Systems*, (s. 231-244). San Diego. Elsevier.

Fageria, N.K. (2009). *The Use of Nutrients in Crop Plants*. New York: CRC Press

Hosseini, S.A., Zamani, G.R., Yaghub, Z.M. and Khayyat, M. (2018). Effects of Cucumber Mosaic Virus infection and drought tolerance of tomato plants under greenhouse conditions: Preliminary results. *J. Berry Res.*, 8(2): 129-136. Doi: 10.3233/JBR-170285.

Iriti, M. and Faoro, F. (2008). Abscisic acid is involved in chitosan-induced resistance to tobacco necrosis virus (TNV). *Plant Physiol. Biochem.*, 46: 1106-1111. https://doi.org/10.1016/j.plaphy.2008.08.002.

Kendig, A. E., Borer, E. T., Boak, E. N., Picard, T. C., Seabloom, E. W. (2020). Host nutrition mediates interactions between plant viruses, altering transmission and predicted disease spread. *Ecology*, *101*(11): e03155. https://doi.org/10.1002/ecy.3155.

Kormelink, R. (2005) Tomato spotted wilt virus. In: Description of plant viruses (DPV) no. 412, https://www.dpvweb.net/dpv/showdpv/?dpvno=412/ (accessed on 5 September 2024).

Krisha-Kumar, N.K., Ullman, D.E., and Cho, J.J. (1993). Evaluation of lycopersicon germ plasm for Tomato sotted wilt tospovirus resistance by mechanical and trips transmission. *Plant Dis.*, 77(9): 938-941.

Labuschagne, P.M. (1995). Casing media for Agaricus bisporus cultivation in South Africa (Doctoral dissertation, University of Pretoria).

Li, W., Cui, X., Meng, Z., Huang, X., Xie, Q., Wu, H., et al. (2012). Transcriptional regulation of Arabidopsis MIR168a and argonaute1 homeostasis in abscisic acid and abiotic stress responses. *Plant Physiol.*, 158:1279-1292. https://doi.org/10.1104/pp.111.188789.

Mandal, B., Pappu, H. R., Culbreath, A. K. (2001). Factors affecting mechanical transmission of Tomato spotted wilt virus to peanut (Arachis hypogaea). *Plant Dis.*, 85(12): 1259-1263. https://doi.org/10.1094/PDIS.2001.85.12.1259.

Mandal, B., Csinos, A.S., Martinez-Ochoa, N. and Pappu, H.R. (2008). A rapid and efficient inoculation method for Tomato spotted wilt tospovirus. *J. Virol. Methods*, 149(1): 195-198. Doi: 10.1016/j.jviromet.2007.12.007.

Marschner, H. (1995). *Mineral Nutrition of Higher Plants*. London: Academic Press. 889 pp.

Marschner, H., and Marschner, P. (2012). *Marschner's Mineral Nutrition of Higher Plants*, San Diego: Elsevier Academic Press. 1–651.

Mur, L.A., Simpson, C., Kumari, A., Gupta, A.K. and Gupta, K.J. (2017). Moving nitrogen to the centre of plant defence against pathogens. *Ann. Bot.*, 119(5): 703-709. Doi: 10.1093/aob/mcw179.

Mutebi, C.M. and Moranga, V. (2022). Effect of Different Soil Fertilities on Cowpea Mosaic Virus Disease Incidence. *Int. J. Hortic. Sci. Technol.,* 9(3): 355-362. https://DOI: 10.22059/IJHST.2021.331384.507.

Pataky, N.R. (1991). Tomato spotted wilt virus. *Report on Plant Disease*, RPD No. 665, Department of Crop Sciences, University of Illinois, Urbana Champaign.

Putra, L.K. and Damayanti, T.A. (2016). Effect of nitrogen and potassium fertilizers, and soil water content on incidence and severity of Sugarcane streak mosaic virus (SCSMV) infecting sugarcane. In *Proceedings of the International Society of Sugar Cane Technologists*, 29: 1620-1626.

Reynolds, W. D., Bowman, B. T., Drury, C. F., Tan, C. S., & Lu, X. (2002). Indicators of good soil physical quality: density and storage parameters. *Geoderma*, 110(1-2): 131-146. https://doi.org/10.1016/S0016-7061(02)00228-8.

Roselló, S., Díez, M.J. and Nuez, F. (1996). Viral diseases causing the greatest economic losses to the tomato crop. I. The Tomato spotted wilt virus-a review. *Sci. Hortic.*, 67(3-4): 117-150. https://doi.org/10.1016/S0304-4238(96)00946-6.

Scholthof, K.B.G., Adkins, S., Czosnek, H., Palukaitis, P., Jacquot, E., Hohn, T., et al. (2011). Top 10 plant viruses in molecular plant pathology. *Mol. Plant Path.*, 12(9): 938-954. Doi: 10.1111/j.1364-3703.2011.00752.x.

Sun, Y., Wang, M., Mur, L. A. J., Shen, Q. and Guo, S. (2020). Unravelling the roles of nitrogen nutrition in plant disease defences. *Int. J. Mol. Sci.*, 21(2): 572. https://doi.org/10.3390/ijms21020572.

Suzuki, N., Rivero, R.M., Shulaev, V., Blumwald, E., Mittler, R. (2014). Abiotic and biotic stress combinations. *New Phytol.*, 203: 32-43. https://doi.org/10.1111/nph.12797.

Takahashi, F., Kuromori, T., Urano, K., Yamaguchi-Shinozaki, K., Shinozaki, K. (2020). Drought Stress Responses and Resistance in Plants: From Cellular Responses to Long-Distance Intercellular Communication. *Front. Plant Sci.*, 11. https://doi.org/10.3389/FPLS.2020.556972.

Uyemoto, J.K., Luhn, C.F., Asai, W.K., Beede, R., Beutel, J.A., Fenton, R. (1989). Incidence of ilarviruses in young peach trees in California. *Plant Dis.*, 73(3): 217-20. Doi:10.1094/PD-73-0217, 0191-2917.

Walters, D.R., and Bingham, I.J. (2007). Influence of nutrition on disease development caused by fungal pathogens implications for plant disease control. *Ann. App. Biol.*, 151(3): 307-324. https://doi.org/10.1111/j.1744-7348.2007.00176.x.