THE INFLUENCE OF THE TREATMENT WITH FUNGICIDES ON THE PHYSIOLOGICAL PROCESSES IN *LYCOPERSICON ESCULENTUM* MILL. PLANTS ATTACKED BY THE *ALTERNARIA SOLANI* SORAUER

Ion NICOLAE , Luminița BUȘE-DRAGOMIR

University of Craiova, email:ionnicolaebio@yahoo.com
University of Craiova, email: luminita25dragomir@yahoo.com

Corresponding author email: *luminita25dragomir@yahoo.com*

Abstract
The influence of the treatment with fungicides on the physiological processes were observed in Lycopersicon esculentum Mill. plants cultivated in the climatic conditions in Oltenia region.The physiological researches were performed on August 23th 2021, both for plants treated with Dithane M-45 (0,2%), in four phases, at 7 days interval and also for the plants attacked by Alternaria solani Sorauer, in which treatments have not been performed. In the leaves of the plants attacked by the pathogen it was observed that the physiological processes' intensity (photosynthesis and transpiration intensity) is lower as a result of the effects produced by the pathogen manifested by the appearance of dark brown spots with closed concentric rings. The content in chlorophyll is higher at the analysed leaves after performing fungicide treatment, thus existing a positive correlation between this content and the photosynthesis intensity.

Key words: *attacked leaves, fungicide, pathogen, photosynthesis, transpiration.*

INTRODUCTION

Tomato (*Lycopersicon esculentum* Mill.) belongs to the family *Solanaceae*. Tomato is considered one of the world's most important and popular vegetables (Pritesh and Subramanian, 2011). It is native to South America and is widely cultivated in 140 countries of the worldwith.

Tomatoes have great importance for its nutritive value in both energy and protein content. Tomatoes are filled with a substance called lycopene. It gives the fruit red color, tomatoes are also rich in potassium, vitamins and other nutrients, lycopene is an antioxidant, which actively fights with cancer forming cells, lycopene also lowers bad cholesterol as well as blood pressure tomatoes have substances called lutein and zeaxanthin that may protect your eyes (Girish PS, 2021).

Tomato contains 95.3% of water, 0.07% calcium and niacin, and other compounds which have great importance in metabolic activities of humans. It is also good source of vitamin A, C and E (Gondal et al 2012).

Tomato is highly sensitive to abiotic stresses especially extreme temperature, salinity, drought, excessive moisture and environmental pollution and biotic stresses. Tomato plants are suffered with large number of biotic stresses including insect pests and diseases from the time of emergence to harvest (Bais et al., 2019).

Among the fungal diseases, early blight, also known as target spot, caused by *Alternaria solani*, is one of the most serious diseases. Early blight produces a wide range of symptoms at all stages of plant growth. The disease appears on leaves, stems, petioles, twig and fruits under favorable conditions resulting in defoliation, drying off of twigs and premature fruit drop (Mathur and Shekhawat, 1986).

Early blight (*Alternaria solani*) is a potential disease of tomato that reduces its production globally both in conventional and tunnel cultivations. Due to variability in pathogenic isolates, prolonged active disease cycle phase and broad host range early blight is very difficult to manage (Chohan et al., 2015).

Alternaria species are known as major plant pathogens, which are air borne and soil inhabiting, the inter-and intra-cellular mycelium of *A. solani* consists of septate and branched, light brown hyphae which become darker with age (Singh, 2009).

Symptoms of early blight are small, dark, necrotic lesions that usually appear on the older leaves and spread upward of the plants. As lesions enlarge, they commonly have concentric rings with a target-like appearance, and they are often surrounded by a yellowing zone (Sherf and MacNab, 1986).

The characteristic feature of genus is the production of beaked, pigmented conidia with relatively thin transverse and longitudinal septa.

The pathogen *Alternaria* has septate, dark coloured mycelium and produce short, simple, erect conidiophores that bear single and branched chains of conidia in acropetal chains (Zghair et al., 2014).

The leaf blight phase, commonly referred to as early blight, is the most important phase of the disease and can result in complete loss of the crop when incidence is severe (Ugwuja et al., 2014).

The disease, if favoured by high temperature and humidity (crowded plantation, high rainfall and extended period of leaf wetness from dew) and plants are more susceptible to the blight infection during fruiting period (Momel and Pemezny, 2006).

Alternaria solani is major damaging among all the fungal diseases. Hence there is need to control this disease with effective control strategy (Pawar et al., 2016).

The most common method for controlling effectively and extensively early blight tomato disease is the use of fungicides. Though they are costly, fungicide treatments are generally the most effective control measures. However, they are not only costly but also capable of creating problems on the environment, human health in all areas of the world and may lead to the development of resistance in pathogenic fungi to common fungicides (Karanwal et. al., 2021).

Preventive fungicides and botanicals inhibit the spore germination and penetration but pathogen can derive resistance against fungicide application so repeated application of fungicides at proper dose and interval of time is mandatory (Hossain and Hossain, 2009; Kumara et al., 2010).

Fungicides treatments are generally the most effective control measures for early blight disease (Chaerani and Voorrips, 2006).

After the application of the treatments with fungicides in the tomato plants attacked by *Phytophthora infestans* (Mont.) de Bary it has been noticed the significantly reduced degree of diseases, the reduction of the area affected by the pathogen; the applied treatments have also influenced the plant growth and the further development of the plants (Nicolae and Buşe-Dragomir, 2014).

Alternaria blight affect plant by reducing photosynthetic area. This disease is very difficult to control (Pasche et al. 2005).

The present study has therefore been undertaken with the objective to determine the influence some fungicides on the phisyological processes in tomato plants attacked by *Alternaria solani*.

MATERIALS AND METHODS

The physiological analyses were performed in tomato plants *Buzău 1600* variety cultivated in the climatic conditions in Oltenia region.

The tomato plants *Buzău 1600* variety have indeterminate growth and large, round-flattened fruits with 3-4 seminal lobes, intense red in color, fleshy, and very tasty. Their average weight is around 200-250 grams.

The treatments were carried out starting on July 26th 2021 and consisted of the application of four treatments with *Dithane* M-45 (0.2%) fungicide at an interval of 7 days (July 26th 2021, August 2th 2021, August 9th 2021, August 16th 2021). The physiological analyses were carried out according to the climatic conditions in the Oltenia area on August 23th 2021, in leaves treated with fungicide and in leaves attacked by pathogen in which treatments have not been performed.

The physiological processes intensity (photosynthesis intensity and transpiration intensity), photosynthetic active radiations, leaf temperature, stomatal conductance were determined with the ultra compact photosynthesis measurement system LCi. The water content and the dry substance content were determined by the gravimetric method and the chlorophyll content were analysed with the Minolta SPAD 502 chlorophyllmeter.

The estimation of the attack was made using the calculation formulae elaborate by Săvescu and Rafailă (1978).

Figure 1. The leaves of *Lycopersicon esculentum* attacked by *Alternaria solani* Sorauer (Original).

RESULTS AND DISCUSSIONS

Early blight in tomato is caused by the fungus *Alternaria solani* Sorauer. The most frequent and obvious symptoms manifest on the leaves of the plants, the attack starting on the mature basic leaves and extending to the top of the plants. Initial symptoms on leaves appear as small 1-2 mm black or brown lesions and under favourable environmental conditions the lesions will enlarge and are often surrounded by a yellow halo. Lesions greater than 10 mm in diameter often have dark pigmented concentric rings. This so-called “bullseye” type lesion is highly characteristic of early blight (Figure 1). As lesions expand and new lesions develop entire leaves may turn chlorotic and dehisce, leading to significant defoliation. Lesions occurring on stems are often sunken and lens-shaped with a light center, and have the typical concentric rings (Gleason and Edmonds, 2006; Kemmitt, 2002).

The fungus occasionally attacks fruit at the stem end, causing large, sunken areas with concentric rings and a black, velvety appearance. In the attacked tissues, a brown mycelium develops, on which simple, brown, short and septate conidiophores are formed, at the tip of which appear bottle-shaped conidia, provided with transverse and longitudinal septa (Figure 2).

Figure 2. *Alternaria solani* Sorauer - conidia (oc. 10 x ob. 40) - Original.

A. solani bears smooth, dark olive or olive-brown, muriform solitary conidia, with a long filiform beak ended with a small pore (Ellis, 1976).

The physiological analyses were performed out according to the climatic conditions, in tomato leaves treated with *Dithane* M-45 fungicide and tomato leaves attacked by pathogen in which treatments have not been performed.

The estimation of the attack (frequency, intensity and degree of attack) produced by *Alternaria solani* Sorauer in tomato leaves is presented in Figure 3.

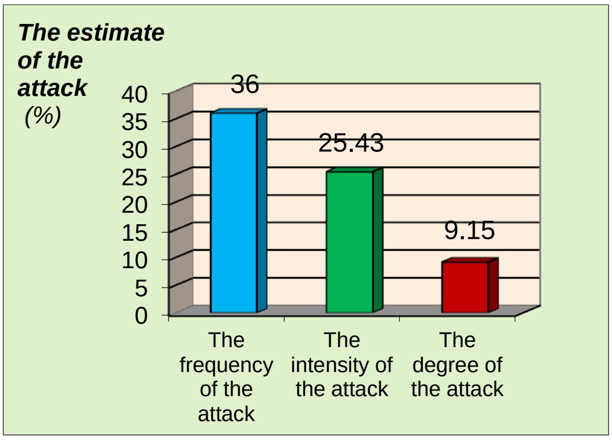

Figure 3. The estimation of the attack produced by *Alternaria solani* Sorauer in *Lycopersicon esculentum.*

The photosynthesis and transpiration's intensity has a lower value in the attacked plants as a result of the reduction of the assimilation surface due to the dark brown spots with dark concentric rings that appear (Figure 4 and Figure 5).

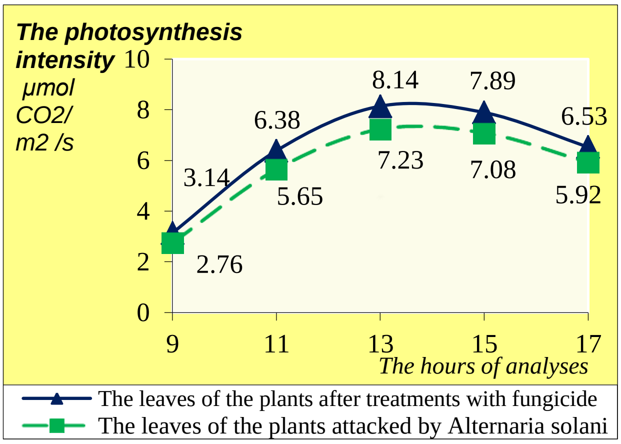

Figure 4. The photosynthesis intensity in the leaves of *Lycopersicon esculentum.*

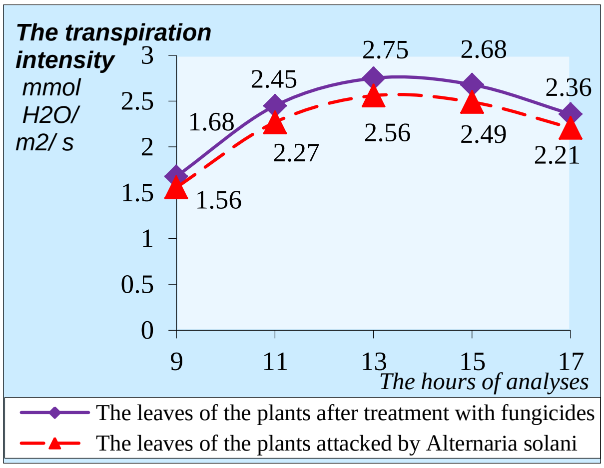

Figure 5. The transpiration intensity in the leaves of *Lycopersicon esculentum.*

The photosynthesis and transpiration's intensity are correlated with the physiological parameters (photosynthetic active radiation, leaf temperature and stomatal conductance).

The photosynthetic active radiation in the tomato plants increases starting with the morning (9 a.m.) when values are 1248 µmol/m^2/s in the plants treated with fungicides and 1236 µmol/m^2/s in the attacked plants by pathogen, they grow up until afternoon (1 p.m.) when values are 1623 µmol/m^2/s in the treated plants and 1598 µmol/m^2/s in the attacked plants and decrease towards evening (5 p.m.) when values are 1516 µmol/m^2/s in the plants treated with fungicides and 1487 µmol/m^2/s in the attacked plants.

Linear regression made between the photosynthesis intensity and photosynthetic active radiations shows a positive correlation between these, the coefficient of determination (R^2) was 0.91 for the tomato plants after treatments with fungicide and 0.88 for the attacked tomato plants; linear regression made between the transpiration intensity and photosynthetic active radiations shows a positive correlation, the coefficient of determination R^2 was 0.85 for the tomato plants after treatments with fungicide and 0.81 for the tomato plants attacked by pathogen (Figure 6 and Figure 7).

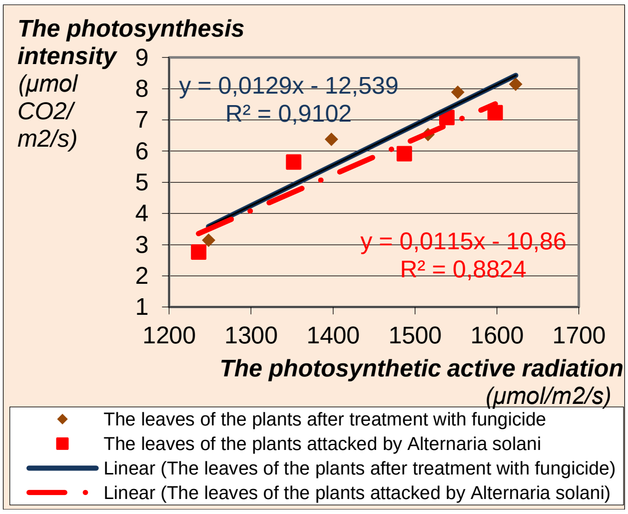

Figure 6. The correlation between the intensity of photosynthesis and the photosynthetic active radiation in *Lycopersicon esculentum.*

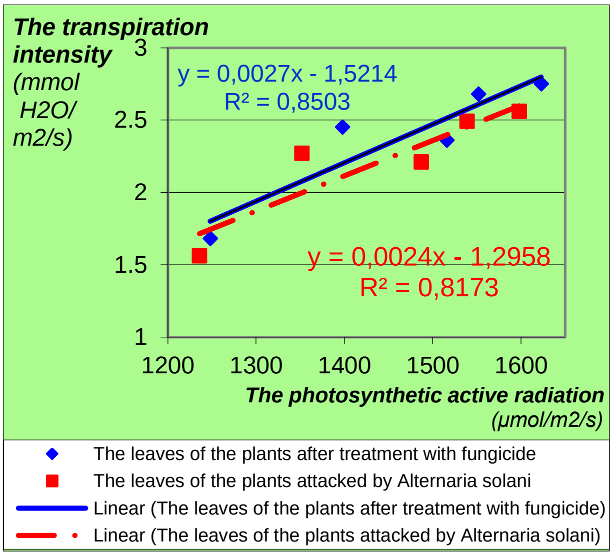

Figure 7. The correlation between the intensity of transpiration and the photosynthetic active radiation in *Lycopersicon esculentum.*

In tomato plants an increase of the leaf temperature can be noticed starting with the morning (9 a.m.) when values are 27.7 ^{o}C in the plants treated with fungicides and 27.9 ^{o}C in the attacked plants, they grow up until afternoon (1 p.m.) when values are 33.6 ^{o}C in the treated plants and 33.8 ^{o}C in the attacked plants and decrease towards evening (5 p.m.) when values are 32.9 ^{o}C in the plants treated with fungicides and 33.4 ^{o}C in the attacked plants by pathogen.

Linear regression made between the photosynthesis intensity and leaf temperature shows a positive correlation, the coefficient of determination (R^2) was 0.94 for the plants after treatments with fungicide and 0.93 for the plants attacked by pathogen; linear regression made between the transpiration intensity and leaf temperature shows a positive correlation, the coefficient of determination R^2 was 0.90 for the plants after treatments with fungicide and 0.88 for the attacked plants (Figure 8 and Figure 9).

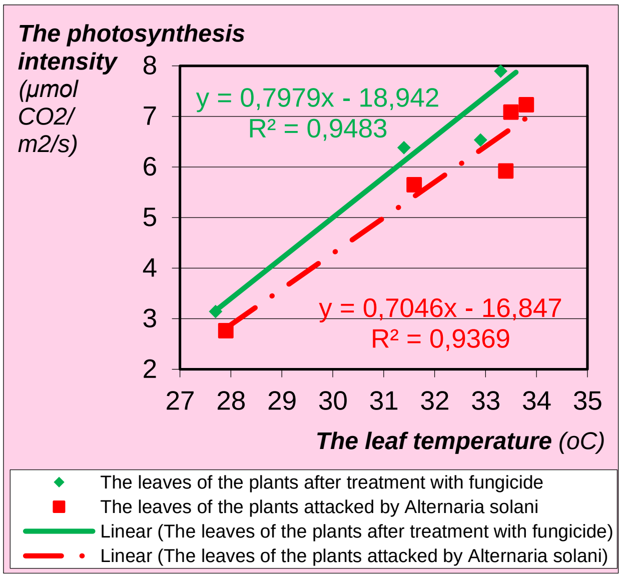

Figure 8. The correlation between the intensity of photosynthesis and the leaf temperature in *Lycopersicon esculentum.*

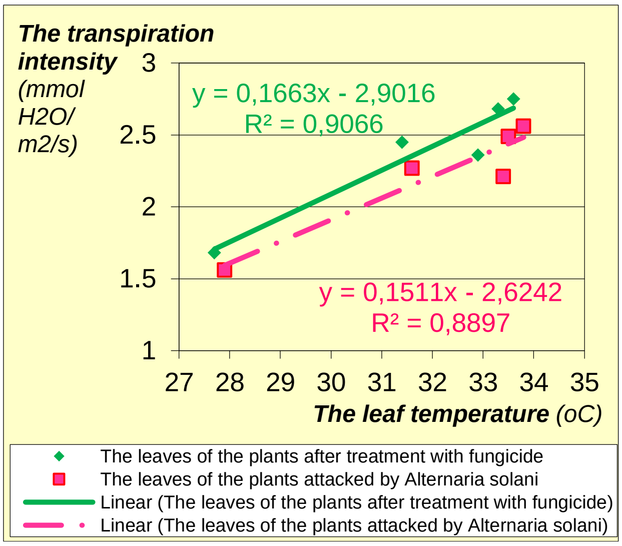

Figure 9. The correlation between the intensity of transpiration and the leaf temperature in *Lycopersicon esculentum.*

The stomatal conductance in the tomato plants increases starting with the morning (9 a.m.) when values are 0.04 mol/m^2/s in the plants treated with fungicides and 0.03 mol/m^2/s in the attacked plants, they grow up until afternoon (1 p.m.) when values are 0.11 mol/m^2/s in the treated plants and 0.08 mol/m^2/s in the attacked plants and decrease towards evening (5 p.m.) when values are 0.07 mol/m^2/s in the plants treated with fungicides and 0.05 mol/m^2/s in the attacked plants by pathogen.

The photosynthesis intensity and stomatal conductance show a positive correlation, the coefficient of determination (R^2) was 0.90 for the tomato plants after treatments with fungicide and 0.89 for the tomato plants attacked by pathogen; the transpiration intensity and stomatal conductance show a positive correlation, the coefficient of determination R^2 was 0.92 for the tomato plants after treatments with fungicide and 0.93 for the tomato plants attacked by pathogen (Figure 10 and Figure 11).

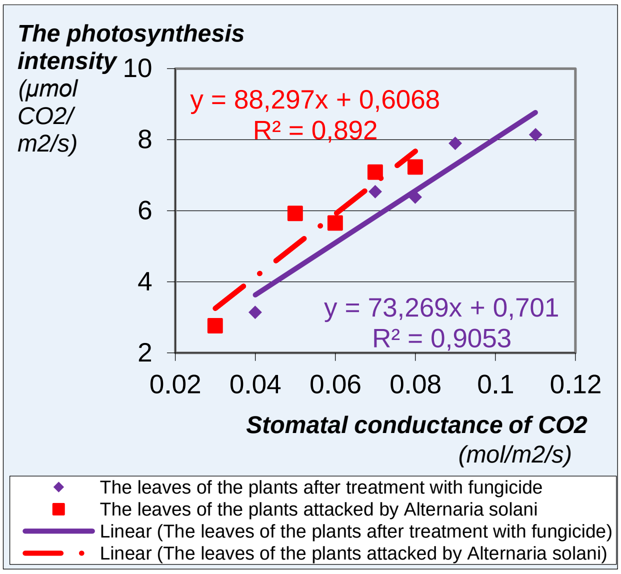

Figure 10. The correlation between the intensity of photosynthesis and the stomatal conductance in *Lycopersicon esculentum.*

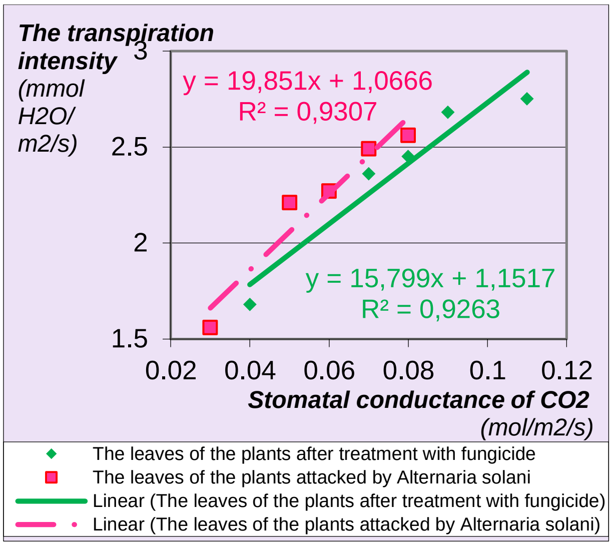

Figure 11. The correlation between the intensity of transpiration and the stomatal conductance in *Lycopersicon esculentum.*

In the attacked plants by *Alternaria solani* Sorauer it was registered a lower water content and a higher dry substance content in comparison with the analysed plants after performing treatments with fungicide (Figure 12).

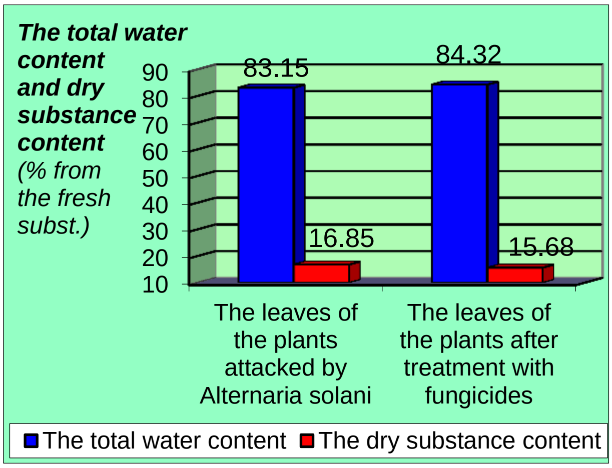

Figure 12. The water content and the dry substance content in *Lycopersicon esculentum*.

In the attacked tomato plants by the pathogen a lower chlorophyll content is recorded in comparison with the tomato plants treated with fungicide (Figure 13).

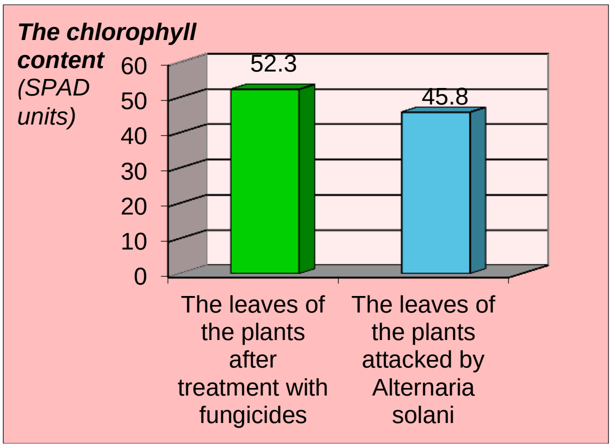

Figure 13. The chlorophyll content in *Lycopersicon esculentum.*

CONCLUSIONS

In tomato plants attacked by the pathogen one can observe that the intensity of the physiological processes (photosynthesis and transpiration intensity) is lower as a result of the appearance of dark brown spots with dark concentric rings, in comparison with the plants treated with fungicide. The photosynthesis and transpiration's intensity are positively correlated with the photosynthetic active radiation, leaf temperature and stomatal conductance, but present different values in the plants attacked by pathogen.

In the plants attacked by the pathogen one can observe the decrease of the water content and chlorophyll content, fact that causes withering and drying of the leaves, with implications on the growth and development plants.

REFERENCES

Bais, R.K., Ratan, V., Kumar, S., Tiwari, A.P., Somesh (2019). *Comparative analysis of various strategies for management of early blight of tomato incited by Alternaria solani (Ellis and Martin) Jones and Grout*, The Pharma Innovation Journal, 8, 15-20.

Chaerani, R., Voorrips, R.E. (2006). *Tomato early blight (Alternaria solani): the pathogen, genetics and breeding for resistance*, Journal of General Plant Pathology, 72, 335-347.

Chohan, S., Perveen, R., Mehmood, M.A., Naz, S., Akram, N. (2015). *Morpho-physiological studies, management and screening of tomato germplasm against Alternaria solani, the causal agent of tomato early blight*, Int. J. Agric. Biol., 17, 111-118.

Ellis, M.B. (1976). *More Dematiaceous Hyphomycetes*, Commonwealth Mycological Institute, Kew, Surrey, England, p 507.

Girish, P.S., Prasad, Rajendra, Kumar, Sandeep, Singh, Mahipal (2021). *In vitro evaluation of fungicides on Alternaria solani causing early blight in tomato,* Journal of Pharmacognosy and Phytochemistry, 10(4), 374-376.

Gleason, M.L. & Edmonds, B.A. (2006). *Tomato diseases and disorders*, University Extension PM 1266, Iowa State University, Ames,1266-1277.

Gondal, A.S., Ijaz, M., Riaz, K., Khan, A.R. (2012). *Effect of different doses of fungicide (Mancozeb) against Alternaria leaf blight of tomato in Tunnel*, Plant Pathology and Microbiology, 3(3), 1-3.

Hossain, M. S. & Hossain, M. M. (2009). *Efficacy of foliar fungicides in controlling early blight of tomato*, Bangladesh J. Plant Pathol., 25, 51-55.

Karanwal, Rajshree, Simon, Sobita, Lal, Abhilasha (2021). *Efficacy of Essential oil Against Early Blight (Alternaria solani) of Tomato (Lycopersicon esculentum Mill.)*, Biological Forum-An International Journal, 13(3a), 631-634.

Kemmitt, G. (2002). *Early blight of potato and tomato,* The Plant Health Instructor. DOI: 10.1094/PHI-I-2002-0809-01.

Kumara, A., Kulkarni, K.T., Thammaiah, M.S., Hegde, N. (2010). *Fungicidal management of early blight of tomato*, Indian Phytopathol., 63 (1), 96-97.

Mathur, K. & Shekhawat, K.S. (1986). *Chemical control of early blight in Kharif* *sown tomato*, Indian Journal of Mycology and Plant Pathology, 16, 235-238.

Momel, T.M., & Pemezny, K.L. (2006). *Florida plant disease management guide: Tomato. Florida Cooperation Extensive Service*, Institute of Food and Agriculture Sciences, 134.

Nicolae, I. & Buşe-Dragomir, Luminiţa (2014). *Research regarding the effects of the treatment with fungicides on the physiological processes in Lycopersicon esculentum Mill. plants attacked by Phytophthora infestans (Mont.) de Bary,* Oltenia. Studii şi comunicări. Ştiinţele Naturii, 30(1), 35-40.

Pasche, J.S., Piche, L.M., Gudmestad, N.C. (2005). *Effect of the F129L mutation in Alternaria solani on fungicides affecting mitochondrial respiration*, Plant Dis. 89, 269-278.

Pawar, P.R, Bhosale, A.M., Lolage Y.P. (2016). *Alternaria Blight of Tomato (Lycopersicon Esculentum Mill)*, International Journal of Advanced Technology and Innovative Research, 8(9),1727-1728.

Pritesh, P. & Subramanian, R.B. (2011). *PCR based method for testing Fusarium wilt resistance of tomato*, African J. Basic &mApplied Sci., 3(5), 222-227.

Săvescu, A. & Rafailă, C. (1978). *Prognoza în protectia plantelor*, Edit. Ceres, București, p 354.

Sherf, A.F., MacNab, A.A. (1986). *Vegetable diseases and their control*, John Wiley and Sons, New York, 634-640.

Singh, R.S. (2009). *Plant Diseases*, Ninth edition, Oxford and IBH Publishing Pvt. Ltd., New Delhi, 388-389.

Ugwuja, F.N., Maduewesi, J.N.C, Ugwoke, K.I., Mbadianya, J.I. (2014). *Invitro inhibition of radial growth, sporulation and germination of Alternaria solani by some fungicides*, Journal of Tropical Agriculture, Food, Environment and Extension, 13(1), 31-36.

Zghair, Qayssar Nadhim, Lal, Abhilashaa, Mane, Musadaq Mnsoor, Simon Sobita, (2014). *Effect of Bioagents and Fungicide against Earlyblight Disease of Tomato (Lycopersicon esculentum L*., International Journal of Plant Protection, 7(2), 330-333.