International Journal of Plant & Soil Science

Volume 35, Issue 19, Page 578-586, 2023; Article no.IJPSS.104105
ISSN: 2320-7035

Efficacy of Non-Conventional Chemical Inducers against *Fusarium oxysporum* Inciting Wilt in Chilli

Priyanka Gupta [a*], Pooja Sangwan [a] and Manjeet Singh [a]

[a] *Department of Plant Pathology, CCS Haryana Agricultural University, Hisar-125004, Haryana, India.*

Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/IJPSS/2023/v35i193587

Open Peer Review History:
This journal follows the Advanced Open Peer Review policy. Identity of the Reviewers, Editor(s) and additional Reviewers, peer review comments, different versions of the manuscript, comments of the editors, etc are available here: https://www.sdiarticle5.com/review-history/104105

Original Research Article

Received: 01/06/2023
Accepted: 05/08/2023
Published: 23/08/2023

ABSTRACT

Chilli wilt incited by *Fusarium oxysporum* is the most devastating disease, causing substantial yield losses throughout chilli growing areas of the world. *Fusarium oxysporum* is a soil-borne fungus which survives for several years in soil. The disease is favoured by high temperature and high moisture, this condition plays significant role in development of wilt symptoms. The disease management using chemicals is economically not feasible and is also unsafe for the environment. Due to growing public awareness to environment and human health, need of the hour is to find alternate approaches for disease management. Therefore, to investigate induction of systemic resistance against *Fusarium oxysporum* in chilli. Different non-conventional chemical inducers like salicylic acid (SA), jasmonic acid (JA), Indole acetic acid (IAA), Indole butyric acid (IBA), chitosan, salicylic acid nano formulation and chitosan nano formulation were used in an experiment conducted in Department of plant pathology, Chaudhary Charan Singh Haryana Agricultural University, Hisar, Haryana. These non-conventional chemicals with four concentrations 25, 50, 75 and 100 µg/ml were evaluated for their antifungal activity against *Fusarium oxysporum* under *in vitro* condition using poison food technique

**Corresponding author: E-mail: priyankagupta@hau.ac.in;*

with four replications. It was found that the highest mycelial growth inhibition was observed in salicylic acid nano formulation (79.73%) at 100 µg/ml concentration followed by Jasmonic acid (77.12%) at 100 µg/ml concentration and the lowest growth inhibition was recorded in Indole acetic acid (27.15%) at 100 µg/ml concentration. The same trend was observed when chilli varieties Pusa Jwala (susceptible) and Pusa Sadabahar (moderately resistant) were tested with the two best and the least effective non-conventional chemical inducer obtained *in vitro* for measuring the disease severity in screen house. This insight clarifies the scope of using non-conventional chemical inducers as an effective disease management strategy against *Fusarium* wilt of chilli.

Keywords: Chilli wilt; Fusarium oxysporum; disease management; non-conventional chemicals; growth inhibition.

1. INTRODUCTION

Chilli, scientifically known as *Capsicum annuum* L. (2n=24) belongs to family *Solanaceae* is an important vegetable and spice crop grown in the world. It was originally native to South Americans from where it spread to rest of the world [1]. India is leading producer and consumer of chilli in the world with total production of about 4221 billion tonnes. In India, Andhra Pradesh, Karnataka, Tamil Nadu and Maharashtra contributes to about 75% of total cultivated area under chilli. The crop is also cultivated in Haryana and is estimated to cover 13.290 thousand hectares with a total production of 141.650 thousand tonne [2].

Chilli grows well in warm and humid weather [3]. The maturity of plant is favoured by clean and dry conditions. The optimum mean temperature for optimal plant growth during the day is between 20-25°C and below 20°C at night. This particular crop grows best in sandy loam soil with a pH range of 5.5 to 8.5, however, it is sensitive to waterlogging and can better tolerate water stress than standing water which often brings about soil-borne diseases [4].

Chilli remains prone to many diseases; among them *Fusarium* wilt is economically more important. It causes significant yield losses around the world that varies from 10-80 per cent [5]. *Fusarium oxysporum* f. sp. *capsici*, an incitant of *Fusarium* wilt of chilli is a soil-borne fungus belonging to phylum *Ascomycota*, subphylum *Pezizomycotina*, class *Sordariomycetes*, order *Hypocreales* and family *Nectriaceae*. The pathogen survives in soil for several years as chlamydospores. Chlamydospores remain inactive in the residues of decomposed plants, and start germinating after acquiring nutrients from the roots of host plants. After germination, conidia and chlamydospores emerge from thalli within 8 h and 3 days respectively, under optimum conditions. High temperature and high moisture favours the disease development. The characteristic symptoms of the disease are initial yellowing of the lower leaves, wilting of leaves, brown vascular discoloration, epinasty, chlorosis and necrosis which ultimately results in death of the plant [6,7].

Although several methods have been developed to control and manage the disease. Chemical control (fungicides) is fast in action and easy to apply but there exist problems associated with them. These fungicides have residual toxicity in soil, the pathogen may show fungicide resistance, cause ground water pollution, risk to non-targeted flora, death of beneficial microorganisms and adverse effect on human health [8]. Use of alternate and eco-friendly approaches to disease management is, therefore, a high-priority research area. Non-conventional chemicals like Jasmonic acid, salicylic acid, chitosan, Indole acetic acid and Indole butyric acid play significant role in plant defense by stimulating the physiological processes which results in improved vegetative growth of plants [9]. These activators move through the plants systemically which lead to the expression of defense genes [10]. Salicylic acid is known to activate various enzymes like phenylalanine ammonia lyase (PAL), tyrosine ammonia lyase (TAL), chitinase, and Polyphenol oxidase (PPO) which results in the development of resistance [11]. Therefore, the present study was carried out with an objective to study induction of systemic resistance by using nano formulations and non-conventional chemical inducers on *Fusarium* wilt of chilli under *in vitro* and screen house conditions.

2. METHODOLOGY

2.1 Evaluation of Non-Conventional Chemical Inducers under *in vitro* Conditions

The inhibitory effect of seven non-conventional chemicals *viz.*, Salicylic acid, Jasmonic acid, Salicylic acid (nano formulation), Indole Acetic acid, Indole Butyric acid, Chitosan and Chitosan (nano formulation) against *Fusarium oxysporum* was examined using poisoned food technique [12]. For *in vitro* evaluation, various concentrations of 50, 100, 150 and 200 µg/ml were prepared by adding required quantity of sterile distilled water to the stock solution of each non-conventional chemical. The solutions of these concentrations were added in equal volume of double strength potato dextrose agar (PDA) medium in petri plates to get the final concentration of 25, 50, 75 and 100 µg/ml. Petri plates without test chemical in the medium served as control and each treatment was replicated four times. The small bits of 5 mm from 5-7 days old culture of the test fungus *i.e.*, *Fusarium oxysporum* were cut by using sterile cork borer and were transferred into the centre of each petri plate with the help of sterile inoculation needle under aseptic conditions (laminar air flow) and incubated at 28±1 °C. The radial growth of mycelium was measured when the growth in the control plates reached the periphery of petri plates and the percent mycelial growth inhibition was calculated, using the formula given by Vincent [13] as follows:

$$I (\%) = (C-T)/C$$

Where,

I is the percent mycelial growth inhibition, C is the colony diameter in control (cm) and T is the colony diameter in treatment (cm).

2.2 Evaluation of Non-Conventional Chemical Inducers under Screen House Conditions

Pots were filled with sterilized sandy loam soil and inoculum was added by mixing it with upper layer of soil and allowed for 7 days to infest the soil. Plants of two chilli varieties *i.e.*, Pusa Jwala (susceptible) [14] and Pusa Sadabahar (moderately resistant) were transplanted after 30-35 days of nursery sowing and raised in already inoculated pots under screen house conditions. Completely randomized design consisting of five treatments in all, each replicated four times was laid. Each replication consisted of 5 plants per pot. Two best (Salicylic acid nano formulation and Jasmonic acid) and the least effective (Indole acetic acid) chemical inducers obtained *in vitro* were applied through seedling dip and soil drench method as per the following treatment details at the time of transplanting. The observation on disease severity of each treatment was recorded after 60 days of transplanting.

Treatments:

- T1 : Control (No inoculation)
- T2 : Inoculation with *Fusarium oxysporum*
- T3 : T2 + Seedling dip and soil drenching with Salicylic acid nano formulation @ 100 µg/ml
- T4 : T2 + Seedling dip and soil drenching with Jasmonic acid (JA) @ 100 µg/ml
- T5 : T2 + Seedling dip and soil drenching with Indole Acetic Acid (IAA) @ 100 µg/ml

Disease severity was calculated by using 0-5 disease rating scale given by Jamil et al*.* [15]

Disease severity (%) = [Sum of all disease ratings] / [(Total number of plants) × (maximal disease grade)] × 100

Table 1. List of non- conventional chemicals used against *Fusarium oxysporum* under *in vitro* conditions

S. No.	Non- conventional chemical inducer	Concentration (µg/ml)			
1.	Salicylic acid	25	50	75	100
2.	Jasmonic acid	25	50	75	100
3.	Salicylic acid (Nano formulations)	25	50	75	100
4.	Indole Acetic Acid	25	50	75	100
5.	Indole Butyric Acid	25	50	75	100
6.	Chitosan	25	50	75	100
7.	Chitosan (Nano formulations)	25	50	75	100

Table 2. Scale for rating *Fusarium* wilt disease

Grade	Description
0	0% infection or no wilt
1	1-20% infection
2	21-40% infection
3	41-60 % infection
4	61-80% infection
5	81-100% infection

3. RESULTS AND DISCUSSION

3.1 Evaluation of Non-Conventional Chemical Inducers Under *in vitro* Conditions

Antifungal activities of the seven non-conventional chemical inducers assayed against *Fusarium oxysporum* is presented in Table 3, Fig. 1 and plate 1. The data revealed that significant reduction in mycelial growth of *Fusarium oxysporum* was observed in respect of all non-conventional chemical inducers tested. However, the maximum inhibition of the mycelial growth of test pathogen was observed in Salicylic acid (nano formulation) @ 100 µg/ml (79.73%) followed by Jasmonic acid @ 100 µg/ml (77.12%). The minimum inhibition of mycelial growth was recorded in treatment 6, Indole Acetic Acid (IAA) at 100 µg/ml (27.15%). Disease resistance in plants depends upon the initiation of defenses after contact to pathogens. Plants have evolved an effective basal defense system to detect and minimize the pathogen growth. The non-conventional chemical inducers act as signal molecule in plant defense against the pathogen infection. Ellis et al. [16] also reported that the use of chemical inducers such as salicylic acid and jasmonic acid help in managing fungal and bacterial diseases. Ojha and Chatterjee [17] treated tomato plants with different concentrations of salicylic acid and observed significant increase in the activities of both peroxidase and polyphenol oxidase imparting resistance against *Fusarium* wilt. Prominent enhancement was observed at 1.5 mM concentration of salicylic acid. Nasir et al. [18] also evaluated four plants activators *i.e.*, salicylic acid, KH_2PO_4, benzoic acid and ascorbic acid @ 0.5, 0.75 and 1% concentration for the management of *Fusarium* wilt disease. Among four plant activators, minimum disease incidence was expressed by salicylic acid (26.68%) followed by KH_2PO_4 (30.72%), Ascorbic acid (33.38%) and Benzoic acid (38.74%) as compared to the control. Salicylic acid @ 1% concentration gave statistically significant results. Similarly, Divya et al. [19] studied the antifungal and antioxidant activity of chitosan nanoparticle (ChNP) against *Rhizoctonia solani, Fusarium oxysporum, Collectotrichum acutatum* and *Phytophthora infestans* and found that ChNP inhibited the growth of *R. solani, C. acutatum* and *P. infestans* by 84.72%, 76.72% and 32.16% at a concentration of 50 mg/ml and 63.88% and *F. oxysporum* at a concentration of 40 mg/ml.

3.2 Evaluation of Non-Conventional Chemical Inducers under Screen House Conditions

Efficacy of two best and one least effective non-conventional chemicals *viz.*, Salicylic acid nano formulation, Jasmonic acid and Indole Acetic Acid (IAA) @ 100 µg/ml were tested under screen house conditions for their possible defense against *fusarium* wilt on two chilli genotypes *viz.*, Pusa Jwala and Pusa Sadabahar. In Pusa Jwala, the percent disease severity was maximum (87.50 %) in pots inoculated with *Fusarium oxysporum*. The minimum disease severity (37.50%) was recorded in pots inoculated with *Fusarium oxysporum* + seedling dip and soil drenching with Salicylic acid nano formulation @ 100 µg/ml followed by 53.12 per cent disease severity in pots inoculated with *Fusarium oxysporum* + seedling dip and soil drenching with Jasmonic acid @ 100 µg/ml. In Pusa Sadabahar, the percent disease severity was maximum (75%) in pots inoculated with *Fusarium oxysporum*. The minimum disease severity (31.25%) was recorded in pots inoculated with *Fusarium oxysporum* + seedling dip and soil drenching with Salicylic acid nano formulation @ 100 µg/ml followed by 43.75% disease severity in pots inoculated with *Fusarium oxysporum* + seedling dip and soil drenching with Jasmonic acid @ 100 µg/ml (Table 4, Fig. 2 and Plate 2). Mandal et al. [20] studied the effect of salicylic acid on tomato wilt and reported that the percent disease incidence was markedly reduced in plants treated with 200 mM salicylic acid (SA) by root dip and foliar spray whereas untreated plants exhibited typical symptoms of vascular browning, leaf yellowing and wilting. Similarly, Nasir et al. [18] evaluated four plants activators and the minimum disease incidence was expressed by salicylic acid (26.68%).

Table 3. Efficacy of non-conventional chemicals against *Fusarium oxysporum* under *in vitro* conditions

Treatments	Mycelial Growth inhibition (%)			
	Concentrations (µg/ml)			
	25	50	75	100
T_1: Salicylic acid (SA)	27.10 (31.35)*	31.01 (33.82)	40.78 (39.67)	48.69 (44.23)
T_2: Jasmonic acid (JA)	56.84 (52.23)	65.07 (53.49)	69.79 (56.85)	77.12(63.24)
T_3: Salicylic acid (Nano formulation)	62.51 (48.91)	64.65 (53.75)	70.12 (56.63)	79.73 (61.41)
T_4: Chitosan	65.08 (53.75)	67.17 (55.02)	70.18 (56.88)	72.40 (58.29)
T_5: Chitosan (Nano formulation)	63.79 (52.98)	65.51 (54.01)	69.19 (56.26)	73.70 (59.13)
T_6: Indole Acetic Acid (IAA)	12.93 (21.05)	18.96 (25.79)	20.25 (26.72)	27.15 (31.38)
T_7: Indole Butyric Acid (IBA)	28.01 (31.93)	35.77 (36.71)	40.06 (39.25)	45.25 (42.25)
T_8: Control (Check)	0 (0.57)	0 (0.57)	0 (0.57)	0 (0.57)
CD (p=0.01)	Treatment			0.64
	Concentration			0.45
	Treatment × Concentration			1.28

** Figures in parentheses represent angular transformed values*

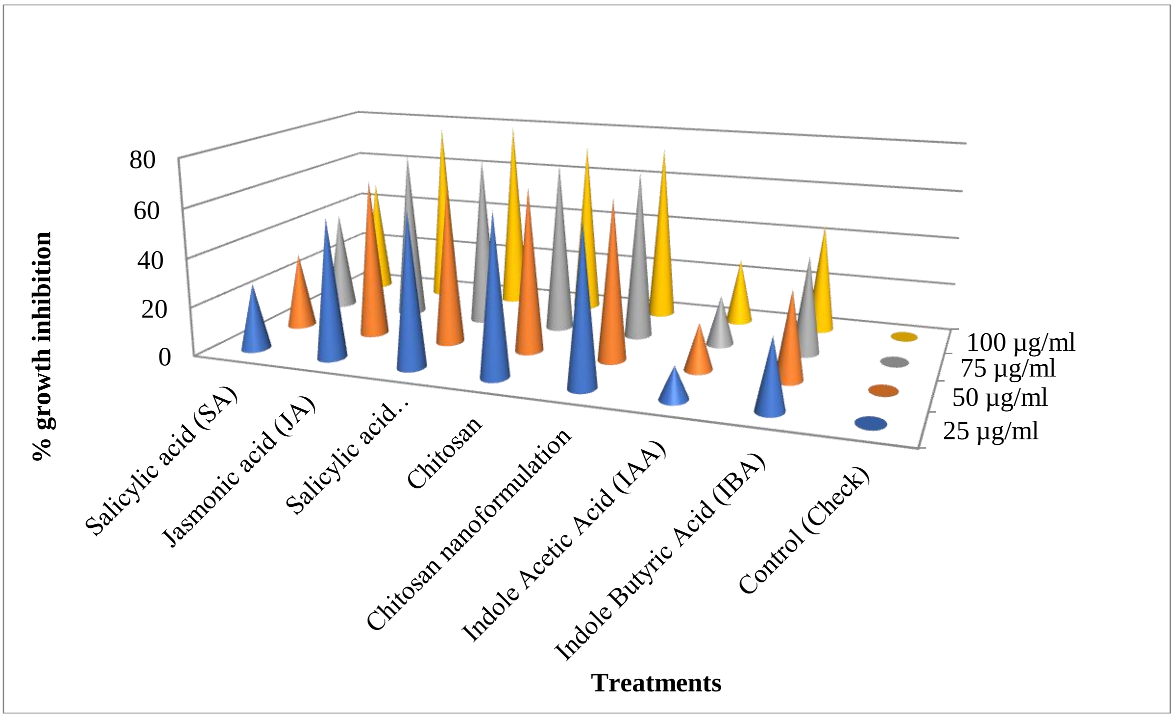

Fig. 1. Efficacy of non-conventional chemicals against *Fusarium oxysporum* under *in vitro* conditions

Plate 1. *In vitro* evaluation of non- conventional chemicals against *Fusarium oxysporum*

Plate 2. Evaluation of non- conventional chemicals against *Fusarium oxysporum* under screen house conditions

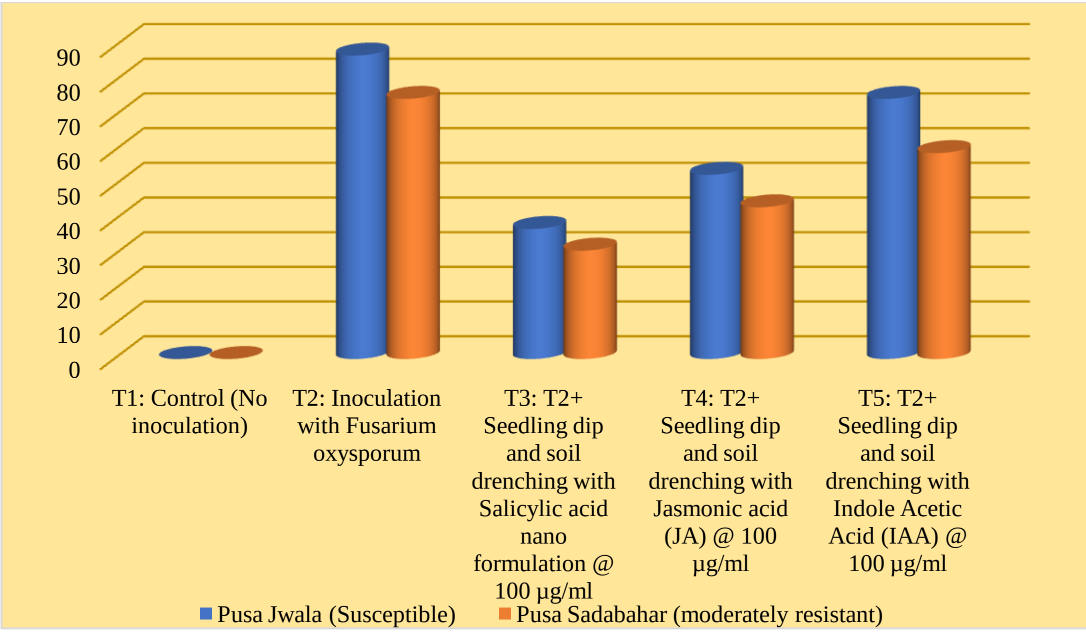

Fig. 2. Per cent disease severity in susceptible and resistant genotypes under different treatments in screen house

Table 4. Effect of two best and one least effective non-conventional chemicals on *Fusarium* wilt in susceptible and resistant genotypes of chilli under screen house conditions

Treatments	Disease severity (%)	
	Pusa Jwala (Susceptible)	**Pusa Sadabahar (Moderately Resistant)**
T_1: Control (No inoculation)	0.00 (0.57)	0.00 (0.57)
T_2: Inoculation with *Fusarium oxysporum*	87.50 (69.26)	75.00 (59.97)
T_3: T_2+ Seedling dip and soil drenching with Salicylic acid nano formulation @ 100 µg/ml	37.50 (37.61)	31.25 (33.86)
T_4: T_2+ Seedling dip and soil drenching with Jasmonic acid (JA) @ 100 µg/ml	53.12 (46.79)	43.75 (41.23)
T_5: T_2+ Seedling dip and soil drenching with Indole Acetic Acid (IAA) @ 100 µg/ml	75.00 (60.33)	59.37 (50.53)
CD (p=0.05)	7.87	8.66
C.V.	12.06	15.30

4. CONCLUSION

Use of non-conventional chemicals for induction of systemic resistance in chilli genotypes can prove to be an effective approach for management of the disease as all the non-conventional chemical inducers used in the study have shown significant results in inducing systemic resistance against *Fusarium oxysporum*, an incitant of *Fusarium* wilt of chilli. However, Salicylic acid nano formulation and Jasmonic acid gave the best results and therefore, are the best for inducing resistance against *Fusarium* wilt and development of resistant genotypes. The results of the study can be useful to control fungal diseases in plants as these non-conventional chemical inducers are eco-friendly and can be easily used as foliar or root application to plants.

ACKNOWLEDGEMENTS

We are grateful to the Department of Plant Pathology, Chaudhary Charan Singh Haryana Agricultural University, Hisar (Haryana), India for providing us all the available necessary facilities required to complete this research work smoothly.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Anonymous. Indian Horticulture Database. National Horticulture Board, Ministry of Agriculture, Government of India; 2016.
2. NHB. Advance estimates of area and production of horticultural crops; 2021-2022.
3. Asati BS Yadav, DS Diversity of horticultural crops in north eastern region. ENVIS Bulletin: Himalayan Ecology. 2004; 12(2):1-10.
4. Arin S. Scenario of chilli production and hindrances faced by the growers of Sindh province of Pakistan. Modern Concepts & Developments in Agronomy. 2019;4(3): 436-442.
5. M Loganathan, Venkataravanappa V, Saha, Sharma S, Tirupathi BK, S Verma, M.K. Morphological, cultural and molecular characterizations of *Fusarium* wilt infecting tomato and chilli. Indian Society of Vegetable Science, IIVR, Varanasi; 2013.
6. Roncero MIG, Hera, C, Ruiz-Rubio M, Maceira FIG, Madrid MP, Caracuel Z, Calero F, Delgado-Jarana J, Roldán-Rodrıguez, R, Martınez-Rocha, AL Velasco C. *Fusarium* as a model for studying virulence in soil-borne pathogens. Physiological and Molecular Plant Pathology. 2003;62(2):87-98.
7. Agrios GN Plant Pathology. 5th Edition. Academic Press, San Diego, USA; 2005;332.
8. Goswami SK, Singh V, Chakdar H, Choudhary P. Harmful effects of fungicides-Current status. International Journal of Agriculture, Environment and Biotechnology. 2018;11:1011-1019.
9. Agrios GN Plant Pathology. 4th Edition. Academic Press, San Diego, USA. 1997; 93-114.
10. Lu ZX, Gaudet D, Puchalski B, Despins T, Frick M. Laroche A. Inducers of resistance reduce common bunt infection in wheat seedlings while differentially regulating defense-gene expression. Physiological and Molecular Plant Pathology. 2005;67(3-5):138-148.
11. Wen PF, Chen JY, Kong WF, Pan QH, Wan SB, Huang WD. Salicylic acid induced the expression of phenylalanine ammonia-lyase gene in grape berry. Plant Science. 2005;169:928-934.
12. Nene YL, Thapliyal PN. Fungicides in plant disease control, 1979;413.
13. Vincent, J.M. Distortion of fungal hyphae in the presence of certain inhibitors. Nature. 1947;159(4051):850.
14. Nishani, S, Prashanthi, SK, Kulkarni MS, Moger N, Sridevi O, Desai S. Screening of the chilli cultivars for resistance to fusarium wilt in Northern Karnataka. The Pharma Innovation Journal. 2021;SP-10(10):638-643.
15. Jamil A, Musheer N, and Ashraf S. Antagonistic potential of *Trichoderma harzianum* and *Azadirachta indica* against *Fusarium oxysporum f. sp. capsici* for the management of chilli wilt. Journal of Plant Diseases and Protection. 2021;128(1):161-172.
16. Ellis C, Karafyllidis L, Turner JG Constitutive activation of jamonate signalling in an Arabidopsis mutant correlates with enhanced resistance to *Erysiphe cichoracearum*, *Pseudomonas syrinagae* and *Myzus persicae*. Molecular Plant-Microbe Interactions. 2002;15:1025-1030.

17. Ojha S, Chatterjee NC. Induction of resistance in tomato plants against *Fusarium oxysporum f. sp. lycopersici* mediated through salicylic acid and *Trichoderma harzianum*. Journal of Plant Protection Research. 2012;52(2):220- 225.
18. Nasir F, Atiq M, Sahi, ST, Bashir MR, Rajput, N.A., Ahmed, I., Sajid, M, Jabbar A. Assessment of plant activators against *Fusarium* wilt of chilli under field conditions. International Journal of Biosciences. 2017;10(5):66-74.
19. Divya K, Smitha V, Jisha MS. Antifungal, antioxidant and cytotoxic activities of chitosan nanoparticles and its use as an edible coating on vegetables. International Journal of Biological Macromolecules. 2018; 114:572-577.
20. Mandal S, Mallick N, Mitra A. Salicylic acid-induced resistance to *Fusarium oxysporum f. sp. lycopersici* in tomato. Plant Physiology and Biochemistry. 2009;47: 642-649.

Peer-review history:
The peer review history for this paper can be accessed here:
https://www.sdiarticle5.com/review-history/104105