



Plant Archives

Journal homepage: <http://www.plantarchives.org>

DOI Url : <https://doi.org/10.51470/PLANTARCHIVES.2025.v25.no.2.287>

IN-VITRO EVALUATION OF DIFFERENT BRASSICA CROPS AGAINST THE PATHOGEN CAUSING FUNGAL WILT DISEASE IN TOMATO

Lakhimi Saikia¹, Budha Bora^{1*}, Nirmal Mazumder¹, Ranima Mishra¹, Hemendra Choudhury² and Nayanmoni Buragohain³

¹Department of Plant Pathology, Biswanath College of Agriculture, AAU, Biswanath Chariali - 784 176, Assam, India.

²Department of Plant Physiology, Biswanath College of Agriculture, AAU, Biswanath Chariali - 784 176, Assam, India.

³Department of Horticulture, Assam Agricultural University, Jorhat - 782 013, Assam, India.

*Corresponding author E-mail : budha.bora@aau.ac.in

(Date of Receiving-18-06-2025; Date of Acceptance-02-09-2025)

ABSTRACT

Tomato is one of the world's most popular vegetable crops and is widely cultivated throughout the world. Its production is, however, challenged by various constraints, among which the wilt disease caused by *Fusarium oxysporum* f.sp. *lycopersici* (FOL) is one of the serious diseases observed regularly in tomato-growing areas. The disease causes substantial yield losses of tomato both in the field and greenhouse-grown tomatoes worldwide. Considering the importance of the disease, the present investigation was undertaken in a Completely Randomized Block Design (CRD) to screen different brassicaceous plants against the pathogen causing wilt disease of tomato *in vitro*. Eighteen Brassica crops were selected for preliminary screening at 25 % concentration to evaluate their antifungal efficacy *in vitro* against *Fusarium oxysporum* f.sp. *lycopersici* (FOL). Amongst all, three promising brassica crops selected based on preliminary screening were further evaluated at different concentrations (10, 20, 30, and 40%) to find out the minimum inhibitory concentration (MIC) against the pathogen. All the brassica crops were effective in inhibiting the mycelial growth of FOL. However, the efficacy of the aqueous extracts of brassica crops increases with the increase in concentration. Among all the brassica crops, Rapeseed variety TS-38 was found to be the most effective in inhibiting the maximum mean (49.85%) radial mycelial growth of FOL followed by Rapeseed varieties M-27 and TS-67 with 41.68% and 39.53% inhibition of radial mycelial growth, respectively.

Key words : Tomato, Wilt, FOL, Brassica crops, Radial mycelial growth, MIC.

Introduction

Tomato (*Lycopersicon esculentum* L.) is one of the most important and popular vegetables in the world. Tomato is an herbaceous plant belonging to the Solanaceae family. It is classified as a vegetable due to its nutritional content and is a good source of vitamin C and the phytochemical lycopene. It is universally regarded as “protective food” and provides a substantial amount of almost all vitamins and minerals. It is a good source of income for small and marginal farmers (Parmar *et al.*, 2019).

According to FAOSTAT, Tomatoes ranked as the most-produced vegetable with 189 million tonnes from 5,167,388 hectares in 2021, achieving an average yield of 37.1 metric tonnes per hectare (Anonymous, 2021a).

India is the second largest producer of tomatoes, accounting for 10.51% of total production after China, followed by Turkey and the United States and occupies an area of about 854 thousand ha producing over 21,181,000 tonnes in 2021 and an estimated 20.34 million metric tonnes in 2022 with a productivity of 25.0 MT per hectare (Anonymous, 2022 a). In India, Madhya Pradesh is the leading producer of tomatoes, producing 29,70,000 tonnes, followed by Andhra Pradesh and Karnataka (Anon, 2022b). Tomato is cultivated over 18.28 thousand ha with a total production of 396.24 thousand MT in Assam (Anonymous, 2022 b).

India is the third largest exporter of tomatoes, following Italy and Turkey. It is an important crop having significant economic value; however, there are many constraints in

its production, causing severe yield loss. Tomato crop faces a yield loss of up to 25-55% percent due to fungal infections caused by *Fusarium oxysporum* all over the world (Devi *et al.*, 2016). *Fusarium* wilt of tomato is one of the most destructive diseases of tomatoes caused by *FOL*. This disease can result in a yield loss of up to 80% when it occurs in a severe form.

Farmers mostly depend on chemical fungicides to manage the disease. Fungicides, however, have been widely recognized to have adverse environmental effects, resulting in the withdrawal of such chemicals from the market. Some synthetic fungicides, such as carbendazim and benomyl, have been observed to be effective in controlling the disease, but these chemicals are very expensive and not environmentally safe, necessitating alternative solutions for sustainable control of this important and destructive disease.

Biofumigation is a sustainable method of soil management that can increase soil organic matter, moderate soil pH, suppress weeds and soil-borne pathogens through glucosinolates and increase water infiltration (Rudolph *et al.*, 2015). Bio-fumigation greatly reduced pesticide application, making farming cheaper and safer as it added organic matter to the soil, leading to increased soil aeration, water infiltration rates, and soil water holding capacity (Karavina and Mandumbu, 2012). This present study aims to evaluate the bioefficacy of Brassica crops against the pathogen inciting wilt disease of tomato under *in vitro* conditions.

Materials and Methods

The present experiment was carried out in the Department of Plant Pathology, Biswanath College of Agriculture, Assam Agricultural University, Biswanath Chariali, during 2023-2024.

Isolation and purification of the pathogen

Disease specimens showing typical symptoms of wilt disease in tomatoes were collected from the Horticultural Orchard at Biswanath College of Agriculture, Biswanath Chariali. The samples were brought to the laboratory for critical observation and investigation, such as symptoms, isolation, and description of the pathogen for further studies. The pathogen was isolated and purified using a tissue isolation technique (Ricker and Ricker, 1936). The pure culture of the pathogen was maintained on Potato Dextrose Agar (PDA) slants by routine sub-culturing at regular intervals and preserved at 4°C in a refrigerator, and the stored culture was used when needed after restoring it to an active state by keeping it at room temperature for all subsequent studies. The pathogenicity

of the pathogen causing fungal wilt of tomato was confirmed by Koch's postulates.

Identification of the isolated pathogen

The pathogen was identified based on the cultural and morphological characteristics. The pathogen was identified with the available relevant literature, a key and monograph (Tamuro, 2013), and the CMI description. Identification of *Fusarium oxysporum* f.sp. *lycopersici* was done based on the spore morphology and colony characteristics of the fungus and by referring to the "Illustrated genera of Imperfect fungi" (Barnett and Hunter, 1972).

Preparation of aqueous extract

The cold-water extract method was used for the preparation of aqueous extracts following the procedure described by Shekhawat and Prasad (1971) with certain modifications.

Preliminary Screening of Brassicaceous crops against *FOL*

Aqueous extracts of eighteen selected brassicaceous crops were prepared and evaluated against the pathogen at a 25% concentration *in vitro* for preliminary screening. For the preliminary screening test, 25 ml of 100 percent basic stock solution of aqueous extract was aseptically added into an Erlenmeyer flask containing 75 ml molten PDA to get the final concentration of 25 percent of the extract in the medium. The Erlenmeyer flask containing PDA without any added extract served as a control. The molten and cooled PDA was poured into 9 cm petri plates, @ 20 ml per plate. After solidification, a fungal culture disc (5mm diameter) obtained from a 7-day-old culture was taken using a cork borer and inoculated in the center of the petri plate under aseptic conditions and incubated at 25±1°C for 7 to 10 days.

Evaluation of different brassicaceous crops against the pathogen

Out of eighteen brassicaceous crops, three promising crops selected based on the results of preliminary screening were further evaluated against the pathogen at 10%, 20%, 30% and 40% concentration by following the Poison Food Technique described by Nene and Thapliyal (2000). The experiment was conducted in a Completely Randomized Block Design (CRD) with five replications.

The diameter of the pathogen colony was assessed upon complete coverage of the petri plates by the mycelium in the control plates. The percent inhibition of the mycelial growth was calculated by following the formula given by Vincent (1947).

Table 1 : List of different Brassicaceous crops used in the experiment.

Scientific name	Common name	Variety
<i>Brassica juncea</i>	Mustard	NRCHB101
		PM-25
		PM-26
		PM-27
		TM-2
		Binoy
<i>Brassica napus</i>	Rapeseed	TS -36
		TS-46
		TS-38
		TS-67
		M-27
<i>Brassica rapa</i> subsp. <i>pekinensis</i>	Mustard green	Local
<i>Raphanus sativus</i>	Radish	
<i>Brassica rapa</i> subsp. <i>rapa</i>	Turnip	
<i>Brassica oleracea</i> var. <i>capitata</i>	Cabbage	
<i>Brassica oleracea</i> var. <i>botrytis</i>	Cauliflower	
<i>Brassica oleracea</i> var. <i>italica</i>	Broccoli	
<i>Brassica oleracea</i> var. <i>gonylodes</i>	Knol-khol	

$$I = (C-T/C) \times 100$$

Where, I = Inhibition of mycelial growth (%)

C = Growth in control (mm)

T = Growth in treatment (mm)

The best effective concentration of brassica crops will be screened out by calculating the average radial mycelial growth and by comparing the percent inhibition of radial mycelial growth of the fungus over the control.

Statistical analysis

The experimental data collected were analyzed statistically for their significance difference by the normal statistical procedure adopted for a completely randomized block design, and interpretation of data was carried out in accordance with Gomez and Gomez (1984). The observed data were analyzed by the OPSTAT package of programs (Sheoran, 2006) after angular transformation. The treatment means were compared by Duncan’s Multiple Range Test (DMRT).

Results

Identification of the isolated pathogen

The cultural and morphological characteristics of the associated fungal isolates were studied for the identification of the pathogen.

Morphological identification of the pathogen

In culture, the colony of the fungus appears white and cottony in colour, then it gradually turns pink in colour, and in later stages, the reverse view of the culture turns reddish in colour. Conidial masses were observed in the cottony portion of the culture, which were oval to kidney-shaped, tapering, and septate in three cells. The growth rate of the pathogen was slow to moderate. Conidia are hyaline, oval to kidney-shaped, measuring 15.00 to 20.00 µm in length and 4-7 µm in width. Chlamydo spores are formed in chains. Oval to kidney-shaped microconidia have false heads on short monophialides and macroconidia are sickle-shaped, thin-walled and delicate.

The fungal isolates were identified as *Fusarium oxysporum* f.sp. *lycopersici* after comparing with standard literature and by referring to the “Illustrated

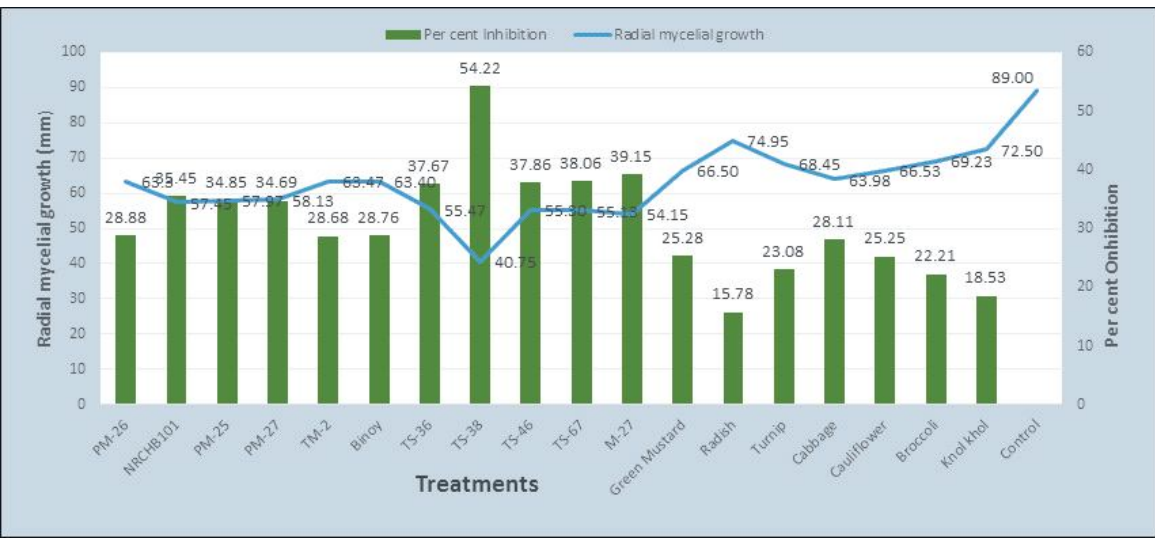


Fig. 1 : Preliminary Screening of Aqueous extract of Brassicaceous crops against *Fol* *in vitro*.

Table 2 : Preliminary Screening of Aqueous Extract of Brassicaceous Crops against *Fusarium oxysporum* f. sp. *lycopersici*.

Treatments	Mycelial growth (mm)	% Inhibition over control
T ₁ : Mustard (<i>Brassica juncea</i> var. PM-26)	63.30 ^f	28.88
T ₂ : Mustard (<i>Brassica juncea</i> var. NRCHB101)	57.45 ^g	35.45
T ₃ : Mustard (<i>Brassica juncea</i> var. PM-25)	57.97 ^g	34.85
T ₄ : Mustard (<i>Brassica juncea</i> var. PM-27)	34.69	58.13 ^g
T ₅ : Mustard (<i>Brassica juncea</i> var. TM-2)	28.68	63.47 ^f
T ₆ : Mustard (<i>Brassica juncea</i> var. Binoy)	28.76	63.40 ^f
T ₇ : Rapeseed (<i>Brassica napus</i> var. TS-36)	55.47 ^h	37.67
T ₈ : Rapeseed (<i>Brassica napus</i> var. TS-38)	40.47 ⁱ	54.22
T ₉ : Rapeseed (<i>Brassica napus</i> var. TS-46)	55.30 ^h	37.86
T ₁₀ : Rapeseed (<i>Brassica napus</i> var. TS-67)	55.13 ^h	38.06
T ₁₁ : Rapeseed (<i>Brassica napus</i> var. M-27)	54.15 ^h	39.15
T ₁₂ : Green mustard (<i>Brassica rapa</i> subsp. <i>pekinensis</i>)	66.50 ^e	25.28
T ₁₃ : Radish (<i>Raphanus sativus</i>)	74.95 ^b	15.78
T ₁₄ : Turnip (<i>Brassica rapa</i> subsp. <i>rapa</i>)	68.45 ^{de}	23.08
T ₁₅ : Cabbage (<i>Brassica oleracea</i> var. <i>capitata</i>)	63.98 ^f	28.11
T ₁₆ : Cauliflower (<i>Brassica oleracea</i> var. <i>botrytis</i>)	66.53 ^e	25.25
T ₁₇ : Broccoli (<i>Brassica oleracea</i> var. <i>italica</i>)	69.23 ^d	22.21
T ₁₈ : Knol-khol (<i>Brassica oleracea</i> var. <i>gonylodes</i>)	72.50 ^c	18.53
T ₁₉ : Control	89.00 ^a	
SEd (±)	1.002	
CD(P=0.05)	2.012	

Data are the mean of five replications.

genera of Imperfect fungi” (Barnett and Hunter, 1972).

Pathogenicity test

The pathogenicity of the pathogen causing fungal wilt of tomato was confirmed by Koch's postulates. Garden soil was first sterilized in an autoclave at 15 lbs pressure, 121° C for two successive days. Earthen pots were filled up with five kgs of sterilized soil and inoculated by mixing the freshly prepared *Fusarium* inoculum (multiplied on sand maize medium) @ 50g/kg soil (Muthusamy, 1972). Tomato seedlings were planted in each pot and maintained properly by regular watering and constantly observed for the development of symptoms. The tomato plants exhibited typical fungal wilt symptoms and the fungus was again

isolated from the infected plant, purified, and maintained in PDA slants. The fungus reproduced a similar kind of cultural characteristic.

Preliminary Screening of aqueous extract of brassicaceous crops against the pathogen

In the *in vitro* experiment, aqueous extracts of eighteen selected brassicaceous crops (Table 1) were prepared, and preliminary screening was carried out at 25 percent concentration to test their biofficacy against *FOL*, causing fungal wilt of tomato by adopting the Poison Food Technique (Nene and Thapliyal, 2000).

The result presented in Table 2 revealed that all the brassicaceous crops showed inhibitory activity against *F. oxysporum* f.sp. *lycopersici* *in vitro* at a 25 percent concentration. Out of eighteen brassicaceous crops tested, Rapeseed (*Brassica napus* var. TS-38) recorded the highest inhibition (54.22%) of radial mycelial growth of the pathogen over control, followed by Rapeseed (*Brassica napus* var. M-27) and Rapeseed (*Brassica napus* var. TS-67) with 39.15 % and 38.06 % inhibition of radial mycelial growth of the pathogen, respectively. The Radish (*Raphanus sativus*) extract recorded the lowest inhibition (15.78%) of mycelial growth of the pathogen (Fig. 1).

Three promising brassicaceous crops, viz. *Brassica napus* var. TS-38, *Brassica napus* var. M-27 and *Brassica napus* var.

TS-67, selected based on their antifungal activity against *FOL* at 25 percent concentration during the course of preliminary screening, were further evaluated to test their efficacy against the tested pathogen *in vitro* at different concentrations (10%, 20%, 30% and 40%).

Effect of aqueous extract of brassicaceous crops at different concentrations against *Fusarium oxysporum* f.sp. *lycopersici* *in vitro*

The three promising brassicaceous crops selected through preliminary screening at 25 percent concentration were further evaluated to test their efficacy at different concentrations against *F. oxysporum* f.sp. *lycopersici* *in vitro*.

All the aqueous extracts significantly inhibited the

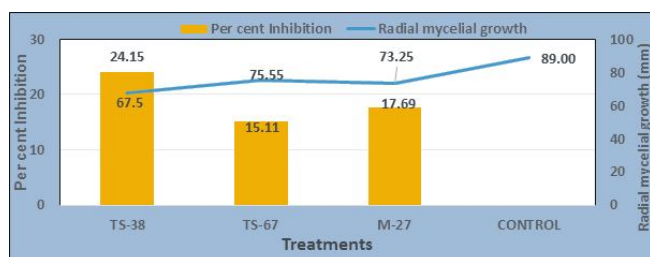


Fig. 2 : Effect of Aqueous extract of Brassicaceous crops at 10% concentration against *Fol* in vitro.

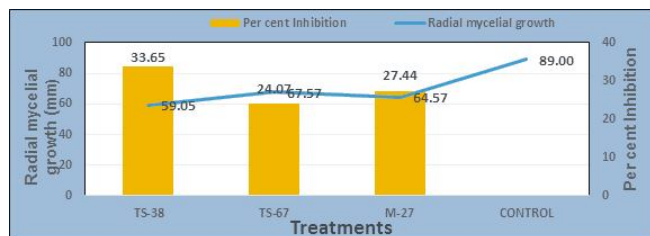


Fig. 3 : Effect of Aqueous extract of Brassicaceous crops at 20% concentration against *Fol* in vitro.

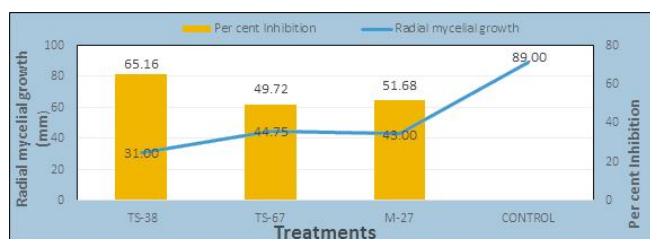


Fig. 4 : Effect of Aqueous extract of Brassicaceous crops at 30% concentration against *Fol* in vitro.

Table 3 : Effect of aqueous extract of brassicaceous crops at 10% concentration against *Fusarium oxysporum* f.sp. *lycopersici* in vitro.

Treatments	Mycelial growth (mm)	% Inhibition over control
T ₁ : Rapeseed (<i>Brassica napus</i> var. TS-38)	67.50 ^c	24.15
T ₂ : Rapeseed (<i>Brassica napus</i> var. TS-67)	75.55 ^b	15.12
T ₃ : Rapeseed (<i>Brassica napus</i> var. M-27)	73.25 ^b	17.69
T ₄ : Control	89.00 ^a	
SEd (±)	1.12	
CD(P=0.05)	2.47	

Data are the mean of five replications

radial mycelial growth of *F. oxysporum* f.sp. *lycopersici* at 10% concentration (Table 3). Among all, *Brassica napus* var. TS-38 (Colony diameter 67.50 mm), was found most effective against *FOL* recorded the highest inhibition (24.15%) of radial mycelial growth over the control followed by *Brassica napus* var. M-27 (Colony diameter

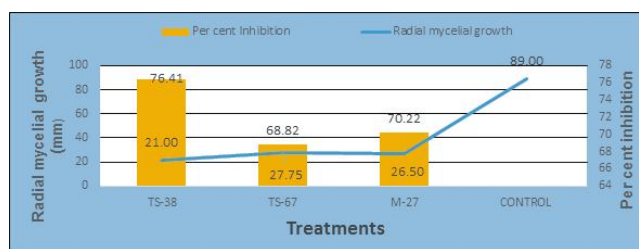


Fig. 5 : Effect of Aqueous extract of Brassicaceous crops at 40% concentration against *Fol* in vitro.

Table 4 : Effect of aqueous extract of brassicaceous crops at 20% concentration against *Fusarium oxysporum* f.sp. *lycopersici* in vitro.

Treatments	Mycelial growth (mm)	% Inhibition over control
T ₁ : Rapeseed (<i>Brassica napus</i> var. M-27)	64.57 ^c	27.44
T ₂ : Rapeseed (<i>Brassica napus</i> var. TS-38)	59.05 ^d	33.65
T ₃ : Rapeseed (<i>Brassica napus</i> var. TS-67)	67.57 ^b	24.07
T ₄ : Control	89.00 ^a	
SEd (±)	1.012	
CD(P=0.05)	2.42	

Data are the mean of five replications.

Table 5 : Effect of aqueous extract of brassicaceous crops at 30% concentration against *Fusarium oxysporum* f.sp. *lycopersici* in vitro.

Treatments	Mycelial growth (mm)	% Inhibition over control
T ₁ : Rapeseed (<i>Brassica napus</i> var. M-27)	43.00 ^c	51.68
T ₂ : Rapeseed (<i>Brassica napus</i> var. TS-38)	31.00 ^d	65.17
T ₃ : Rapeseed (<i>Brassica napus</i> var. TS-67)	44.75 ^b	49.71
T ₄ : Control	89.00 ^a	
SEd (±)	0.67	
CD(P=0.05)	1.47	

Data are the mean of four replications.

73.25 mm) and *Brassica napus* var. TS-67 (Colony diameter 75.55 mm) with 17.69%, and 15.12% inhibition of radial mycelial growth of the pathogen over control, respectively (Fig. 2).

Data presented in Table 4 showed that at 20% concentration, all the aqueous extracts significantly

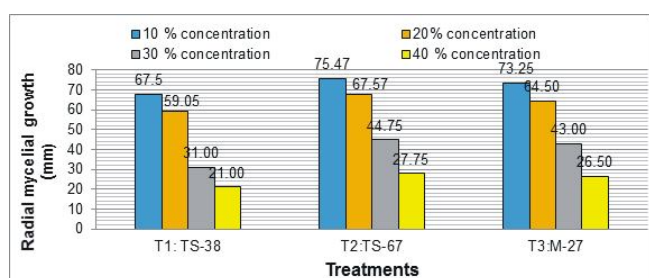


Fig. 6 : Effect of Aqueous extract of Brassicaceous crops (10, 20, 30 and 40%) on Radial Mycelial Growth of *Fol* in vitro.

Table 6 : Effect of aqueous extract of brassicaceous crops at 40% concentration against *Fusarium oxysporum* f.sp. *lycopersici* in vitro.

Treatments	Mycelial growth (mm)	% Inhibition over control
T ₁ : Rapeseed (<i>Brassica napus</i> var. M-27)	26.50 ^c	70.23
T ₂ : Rapeseed (<i>Brassica napus</i> var. TS-38)	21.00 ^d	76.41
T ₃ : Rapeseed (<i>Brassica napus</i> var. TS-67)	27.75 ^b	68.82
T ₄ : Control	89.00 ^a	
SEd (±)	0.57	
CD(P=0.05)	1.25	

Data are the mean of five replications.

Table 7 : Effect of aqueous extract of brassicaceous crops (10, 20, 30 and 40%) on radial mycelial growth of *Fusarium oxysporum* f.sp. *lycopersici* in vitro.

Treatments	Radial mycelial growth (mm)				Mean
	10 %	20 %	30 %	40 %	
T ₁ : <i>Brassica napus</i> var. M-27	73.25	64.05	43.00	26.50	51.81
T ₂ : <i>Brassica napus</i> var. TS-38	67.50	59.05	31.00	21.00	44.64
T ₃ : <i>Brassica napus</i> var. TS-67	75.47	67.57	44.75	27.75	53.89
	Plant Extracts (P)		Concentrations (C)		Interaction (P X C)
SEd (±)	0.51		0.59		1.02
CD (P=0.05)	1.03		1.19		2.07
CV(%)					2.90

inhibited the radial growth of *FOL*, the highest being the *Brassica napus* var. TS-38 (Colony diameter 59.05 mm), with 33.65% inhibition, followed by *Brassica napus* var. M-27 (Colony diameter 64.57 mm), and *Brassica napus* var. TS-67 (Colony diameter 67.57 mm) with 27.44% and 24.07% inhibition of radial mycelial growth of *FOL* over control, respectively (Fig. 3).

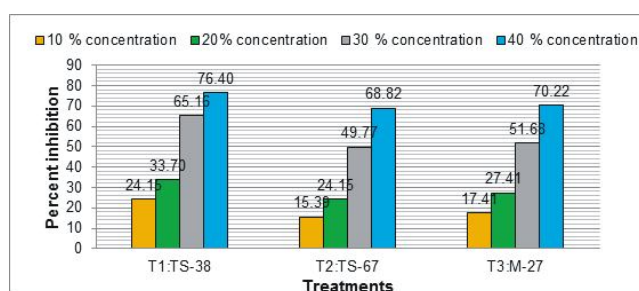


Fig. 7 : Effect of Aqueous extract of Brassicaceous crops (10, 20, 30 and 40%) on Inhibition of Mycelial growth of *Fol* in vitro.

With respect to percent inhibition at 30% concentration, the data presented in Table 5 indicated that the aqueous extract of *Brassica napus* var. TS-38 showed the highest inhibitory effect on the radial mycelial growth of *F. oxysporum* f.sp. *lycopersici* followed by var. M-27 and var. TS-67 as compared to the control. *Brassica napus* var. TS-38 recorded the highest inhibition (65.17%) of radial mycelial growth of *FOL* over the control. This was followed by *Brassica napus* var. M-27 and var. TS-67 with 51.68% and 49.71% inhibition, respectively (Fig. 4).

Data presented in Table 6 revealed that out of all three aqueous extracts, *Brassica napus* var. TS-38 was found most effective at 40% concentration, resulting in the highest inhibition (76.41%) over control, followed by var. M-27 and var. TS-67 with 70.23% and 68.82 %

inhibition of mycelial growth, respectively (Fig. 5).

The data presented in Table 7 showed that the aqueous extract of brassicaceous crops significantly influenced the radial mycelial growth of *Fusarium oxysporum* f.sp. *lycopersici* at all the concentrations (10%, 20%, 30% and 40%). Irrespective of the concentrations, all the aqueous extracts significantly

Table 8 : Effect of aqueous extract of brassicaceous crops (10, 20, 30 and 40%) on inhibition of mycelial growth of *FOL in vitro*.

Treatments	Inhibition of mycelial growth over control (%)				Mean
	10	20	30	40	
T ₁ : <i>Brassica napus</i> var. M-27	17.41(24.86)	27.41(32.14)	51.68(45.94)	70.22(56.90)	41.68(39.83)
T ₂ : <i>Brassica napus</i> var. TS-38	24.15(29.42)	33.70(35.44)	65.16(53.81)	76.40(60.92)	49.85(44.89)
T ₃ : <i>Brassica napus</i> var. TS-67	15.39(22.92)	24.15(29.34)	49.77(44.82)	68.82(56.03)	39.53(38.28)
	Plant Extracts (P)		Concentrations (C)		Interaction (P X C)
SEd(±)	0.39		0.46		0.79
CD (P=0.05)	0.81		0.94		1.62
CV(%)					2.85

*Data within the parentheses are arcsine-transformed data

Data are the mean of five replications.

reduced the radial mycelial growth of *Fusarium oxysporum* f.sp. *lycopersici* *in vitro*. Out of three crops evaluated, *Brassica napus* var. TS-38 recorded the lowest mean radial growth (44.64 mm) at all the concentrations, followed by *Brassica napus* var. M-27 and *Brassica napus* var. TS-67 with mean radial growth of 51.81 mm and 53.89 mm, respectively (Fig. 6).

With respect to per cent inhibition of mycelium, the data presented in Table 8 showed that the aqueous extract of *Brassica napus* var. TS-38 was found to be most effective, resulting in the highest mean inhibition (49.85%) of radial mycelial growth of *FOL* over control in all four concentrations (10%, 20%, 30%, 40%), which was followed by *Brassica napus* var. M-27 and *Brassica napus* var. TS-67 with a mean radial inhibition of mycelial growth of 41.68% and 39.53% respectively, over the control. However, the effect of aqueous extract increased with the increase of concentration, which was reflected at 40% concentrations, showing the highest inhibition (76.40%) of mycelial growth of *FOL* exhibited by the aqueous extract of *Brassica napus* var. TS-38 followed by *Brassica napus* var. M-27 and *Brassica napus* var. TS-67 with 70.22% and 68.82% inhibition, respectively, over control (Fig. 7).

Discussion

The fungal wilt of tomato caused by *Fusarium oxysporum* f.sp. *lycopersici* has been reported as one of the important factors for the reduction of the yield of tomatoes. The present study was conducted to evaluate the effect of brassicaceous crops against *FOL in vitro*.

Pathogenicity test

Identification of the pathogen

In the present investigation, a pathogenicity test was conducted based on Koch's postulates to confirm the

pathogen. A similar pathogenicity test was also carried out by Jasnic *et al.* (2005). Pathogenicity tests were also performed by other researchers (Abdulkadir *et al.*, 2023; Kumar *et al.*, 2013).

Effect of aqueous extract of brassicaceous plants against the pathogen causing fungal wilt of tomato *in vitro*

The preliminary screening results revealed that all eighteen brassicaceous plants were effective at 25 per cent concentration in reducing the radial mycelial growth of *Fusarium oxysporum* f.sp. *lycopersici* *in vitro*. Out of eighteen brassicaceous crops, three promising ones were further evaluated at different concentrations (10, 20, 30 and 40%). The aqueous extract of the brassicaceous plants significantly influenced the radial mycelial growth of *FOL* at all concentrations. Irrespective of concentrations, all the aqueous extracts significantly reduced the radial mycelial growth of *FOL in vitro*. Out of three plants evaluated, *Brassica napus* var. TS-38 recorded the lowest mean radial growth (44.64 mm) at all the concentrations (10%, 20%, 30%, 40%), followed by *Brassica napus* var. M-27 and *Brassica napus* var. TS-67 with radial growth of 51.81 mm and 53.89 mm, respectively.

With respect to per cent inhibition of mycelium, the data presented in Table 7 showed that the aqueous extract of *Brassica napus* var. TS-38 was found to be most effective, resulting highest mean inhibition (49.85%) of radial mycelial growth of *FOL* over control in all four concentrations (10%, 20%, 30% and 40%), which was followed by *Brassica napus* var. M-27 and *Brassica napus* var. TS-67 with a mean radial inhibition of mycelial growth of 41.68 % and 39.53 % respectively, over control. However, the effect of aqueous extract increased with

an increase in concentration, which was reflected at 40% concentrations, showing the highest inhibition (76.40%) of mycelial growth of *FOL* exhibited by aqueous extract of *Brassica napus* var. TS-38 followed by *Brassica napus* var. M-27 and *Brassica napus* var. TS-67 with 70.22% and 68.82% inhibition, respectively, over the control. The inhibition of the mycelial growth of the pathogen may be due to the toxic effect of the chemical (Isothiocyanate), produced during the enzymatic breakdown of glucosinolates, present in the *Brassica* crops (TS-38).

The results of the present investigation were in agreement with the findings of Rao and Viswanath (2023), who reported that the rapeseed-mustard leaf tissue showed the highest mycelial inhibition (77.65%), followed by broccoli leaf tissue (65.50%). However, radish leaf tissue showed the least inhibitory effect (11.77%), respectively.

A similar study was also conducted by Prasad and Kumar (2017), who reported that the isothiocyanates, a byproduct of the enzymatic breakdown of the glucosinolates from the Brassicaceae plant, were a promising substitute for conventional biofumigants. According to their findings, *Brassica alba* was the most toxic because it inhibited the pathogen's (*Fusarium oxysporum* f.sp. *ciceris*) mycelial growth to the highest extent possible, followed by the tissues of *B. nigra* L. and *B. juncea* L. Bharat and Sharma (2015) also reported that among different biofumigant crop residues (fresh as well as dry) tested *in vitro*, rapeseed variety taramira showed the highest inhibition in mycelial growth of *F. oxysporum* f. sp. *lycopersici* i.e. 62.59 and 60.74 per cent, respectively. Bhandari *et al.* (2015) reported the profiles and concentrations of glucosinolates (GSLs) in nine Brassica species (cauliflower, cabbage, broccoli, radish, baemuchae, pakchoi, Chinese cabbage, leaf mustard and kale). Broccoli showed the highest total GSL concentration in seeds ($110.76 \mu\text{molg}^{-1}$) and sprouts ($162.19 \mu\text{molg}^{-1}$) among the nine crops analyzed, while leaf mustard showed the highest total GSL concentration in shoots ($61.76 \mu\text{molg}^{-1}$) and roots ($73.61 \mu\text{molg}^{-1}$). The lowest GSL concentrations were observed in radish.

Conclusion

The fungal wilt disease of tomatoes is one of the limiting factors of production. The severity of this disease can be reduced significantly by using brassicaceous plants as a biofumigant. Among all the common brassicaceous crops evaluated, *Brassica napus* var. TS-38 can effectively be used as a biofumigant crop for eco-friendly and sustainable management of fungal wilt disease of

tomatoes.

References

- Abdulkadir, H.K., Ekefan E.J. and Gwa V.I. (2023). Pathogenicity of *Fusarium oxysporum* f.sp. *lycopersici* (Sacc.) Isolates in Causing Tomato Wilt Disease on Two Tomato (*Solanum lycopersicum* L.) Varieties. *Bio-Sci. Res. Bull.*, **39**(2), 60-68.
- Anonymous (2018). Horticultural Statistics at a glance, Ministry of Agriculture and Farmers Welfare, Govt. of India. <https://agricoop.nic.in/en/statistics/horticulture>. Retrieved on 13/07/22.
- Anonymous (2021a). Agricultural Production Statistics, Food and Agriculture Organisation of the United States. <https://www.fao.org/faostat/>. Retrieved on 13/06/23.
- Anonymous (2022a). Statista. <https://www.statista.com/statistics/1039712/india-production-volume-of-tomatoes/>. Retrieved on 13/06/23.
- Anonymous (2022b). APEDA, National horticulture board. <https://agriexchange.apeda.gov.in/India%20Production/IndiaProductions.aspx?cat=Vegetables&hscode=1088>. Retrieved on 20/06/23.
- Barnett, H.L. and Hunter B.B. (1972). *Illustrated Genera of Imperfect Fungi*. Burgess Publication Ltd. St. Paul, Minnesota, USA, pp. 241.
- Bhandari, S.R., Jo J.S. and Lee J.G. (2015). Comparison of glucosinolate profiles in different tissues of nine Brassica crops. *Mol.*, **20** (9), 15827-15841.
- Bharat, N.K. and Sharma J. (2015). Management of Fusarium wilt of tomato under protected conditions using Brassica crop residues, AM fungi and biocontrol agents. *Indian Phytopathol.*, **68** (3), 287-292.
- Devi, N., Venkataramana M., Srivastava R.K., Uppalapati S.R., Gupta V.K. and Yli-Mattila T. (2016). Molecular phylogeny, pathogenicity and toxigenicity of *Fusarium oxysporum* f.sp. *lycopersici*. *Sci. Rep.*, **6**, 21367.
- Gomez, K.A. and Gomez A.A. (1984). *A statistical procedure for agricultural Research*. John Wiley Sons, New York
- Jasnic, S., Vidic M., Bagi F. and Dor Devi C.V. (2005). Pathogenicity of *Fusarium* species in soybean, **109**, 113-115.
- Karavina, C. and Mandumbu R. (2012). Biofumigation for crop protection: potential for adoption in Zimbabwe. *J. A. P. Sci.*, **14**(3), 1996-2005.
- Kumar, D., Ratan V., Kumar A., Kumar S.J., Pal K. and Kumar M. (2013). Pathogenicity tests and antagonistic effect of bioagents on *Fusarium oxysporum* f. sp. *lycopersici* (*FOL*). *Pharma. Inno. J.*, **10**(11), 1887-1891.
- Muthusamy, M. (1972). Studies on damping-off of tomato incited by *P. aphanidermatum* (Edson.) Fitz., *M.Sc.*, (Ag.), Thesis. Tamil Nadu Agriculture University, Coimbatore, Tamil Nadu, p. 110.
- Nene, Y.L. and Thapliyal P.N. (2000). *Poisoned food technique. Fungicides in Plant Disease Control*. 3rd Edn, Oxford and IBH Publishing Company, New Delhi. pp. 531-533.

- Parmar, U., Tembhre D., Das M.P. and Pradhan J. (2019). Effect of integrated nutrient management on growth development and yield traits of tomato (*Solanum lycopersicon* L.). *Int. J. Pharmacogn.*, **8(3)**, 2764-2768.
- Prasad, P. and Kumar J. (2017). Management of Fusarium wilt of chickpea using brassicas as biofumigants. *Legume Res.*, **40(1)**, 178-182.
- Rao, V.G. and Viswanath H.S. (2023). Biocidal efficacy of plant volatile obtained from brassica species against Fusarium wilt of eggplant caused by *Fusarium oxysporum* f. sp. *melongenes*. *Biol. forum- Int. J.*, **15(4)**, 775-782.
- Ricker, A.J. and Ricker R.S. (1936). *Introduction of research on Plant Diseases*. John Swift Co., New York, p. 177.
- Rudolph, R.E., Sams C. and Steiner R. (2015). Biofumigation performance of four brassicacrops in a Green Chile pepper (*Capsicum annuum*) rotation system in Southern New Mexico, *Hort. Sci.*, **50 (2)**, 247 – 253.
- Shekhawat, P.S. and Prasad R. (1971). Antifungal properties of some plant extract and their inhibition of spore germination. *Indian Phytopathol.*, **24**, 800-802
- Sheoran, O.P. (2006). Online statistical tool (OPSTAT). [www.hau.emet.in/ about/ opstat.php](http://www.hau.emet.in/about/opstat.php). CCS HAU, Hissar.
- Tamura, K., Stecher G., Peterson D., Filipski A. and Kumar S. (2013). MEGA6: molecular evolution genetics analysis version 6.0. *Mol. Biol. Evol.*, **30**, 2725-2729.
- Vincent, J.M. (1947). Distortion of fungal hyphae in presence of certain inhibitors. *Nat.*, **159**, 850.