



Impact of the Interaction between *Meloidogyne javanica* and *Fusarium oxysporum* on the Okra Plantation

Manisha ^{a*}, S. P. Bishnoi ^a, Mamata ^b,
Sharad Kumar Meena ^c, Ramavath Abhi ^d,
Santosh Yadav ^e and Rahul Meena ^a

^a Division of Nematology, Rajasthan Agricultural Research Institute, Durgapura, Jaipur- SKN Agriculture University, Jobner 303329, India.

^b Department of Agronomy, MPUAT, Udaipur 333042, India.

^c Division of Entomology, Rajasthan Agricultural Research Institute, Durgapura, Jaipur- SKN Agriculture University, Jobner 303329, India.

^d Department of Nematology, MPUAT, Udaipur 333042, India.

^e Department of Soil Science & Agricultural Chemistry, MPUAT, Udaipur 333042, 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/v35i183470

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/103620>

Original Research Article

Received: 28/05/2023

Accepted: 31/07/2023

Published: 08/08/2023

ABSTRACT

The development of diseases in cultivated crops depends on the complex interrelationship between the host, pathogen, and prevailing environmental conditions. The interaction between the root knot nematode *Meloidogyne javanica* and the fungus *Fusarium oxysporum* was studied for the okra cultivar Pusa Sawani. Sequential and simultaneous inoculation of the nematode and fungus was performed to observe the disease severity in the plants due to the individual and interactive effects

*Corresponding author: E-mail: manishameena4361@gmail.com;

of the tested nematode and fungus. The results indicated that plant growth was adversely affected in all cases where the plant was inoculated with *M. javanica* and *F. oxysporum*, compared to the uninoculated control. The study clearly showed that nematodes act as predisposing factors in the spread of secondary fungal pathogens.

Keywords: Okra; wilt; fusarium; meloidogyne; interaction.

1. INTRODUCTION

Okra (*Abelmoschus esculentus* (L.) Moench) is a commercially grown vegetable crop commonly known as 'Bhindi'. It is one of the most important vegetable crops during the summer and rainy seasons and is cultivated in various states of India. The geographical origin of okra is disputed, with contributions from West Africa, South Asia, and Ethiopia. According to Khalid et al. [1] and Thompson & Kelly [2], the crop was first cultivated by Egyptians in the 12th century. Okra is typically grown in tropical, subtropical, and warm temperate regions worldwide. The okra crop is susceptible to various harmful microorganisms, including fungi, bacteria, viruses, nematodes, and insect pests. Among these microorganisms, the root-knot nematode *Meloidogyne javanica* is the most damaging, causing destructive diseases. Sikora [3] discovered that the okra crop is highly susceptible to the root-knot nematode *M. javanica*. Osman et al. [4] found that root-knot nematodes infest a wide range of important crop plants and pose a significant threat to vegetable crops in tropical and subtropical regions. Root-knot nematodes have also been found to be associated with bacterial and fungal pathogens. Both the nematodes and the associated pathogens can cause severe damage to the crop, leading to complex diseases and potentially complete crop destruction [5].

2. MATERIALS AND METHODS

An experiment was conducted at the Division of Nematology, Rajasthan Agricultural Research Institute, Durgapura, Jaipur, during 2019-2020 to study the Interaction between *Meloidogyne javanica* and *Fusarium oxysporum* on okra plant. The experiment utilized 20 cm-sized pots filled with sterilized soil. Soil samples of approximately 200 cc were randomly collected from the rhizosphere of okra plants at 4-5 different locations in each field. The samples were obtained at a depth of 15-20 cm, homogenized, and stored in labeled polythene bags. These bags were then tied and refrigerated at approximately 10°C. A composite sample of

200 cc, along with 5g of roots, was collected from a one-acre field.

The experiment included several treatments: nematode alone (N), fungus alone (F), nematode and fungus inoculated simultaneously (NF), nematode at the time of sowing and fungus after one week (N1F7), fungus at the time of sowing and nematode after one week (F1N7), and a control group without fungus and nematode.

A consistent number of 2000 second-stage juveniles of the nematode were inoculated per pot in the N, N+F, N1+F2, and F1+N2 treatments. Inoculation of the soil with the fungus, *F. oxysporum*, involved mixing 20g (mycelial weight of fungus) content from each flask containing sorghum grain inoculated with the fungus into each pot in the F, N+F, N1+F2, and F1+N2 treatments. For cases where the fungus and nematode were inoculated one week after nematode/fungus inoculation, three glass rods were placed equidistantly in a circle with a 2 cm diameter, and the holes were sealed with sterile soil after introducing the actively growing *F. oxysporum* derived from the sorghum grain. The nematodes were then pipetted around the seedlings. Precautions were taken to prevent contamination between pots. The pots were randomized according to a completely randomized design (C.R.D.) and watered as needed. The experiment was concluded 45 days after inoculation, and observations were made on plant growth characteristics and nematode reproduction.

2.1 Statistical Analysis

After completion of experiment, data was statically analyzed for interpretation of findings. The critical difference was calculated for comparison of treatment significant at 5% level of significance.

3. RESULTS

The interaction between *M. javanica* (root-knot nematode) and *F. oxysporum* (wilts disease-causing fungus) leads to the development of the

Root knot wilt disease complex, which is highly destructive to okra plants. In this complex, both the nematode and the fungus play a role in causing severe damage to the plants. All stages of *M. javanica*, including the eggs, juveniles, and adults, are harmful and result in the formation of root galls, which are visible symptoms on the roots. These root galls disrupt the normal functioning of the roots and negatively impact plant growth and productivity.

3.1 Shoot Length

The data obtained from the experiment showed that the treatment where the nematode (*M. javanica*) was inoculated first and the fungus (*F. oxysporum*) was introduced after seven days resulted in the minimum shoot length recorded, measuring 25.55 cm. This was followed by the treatment where the fungus was inoculated first and the nematode was introduced after seven days, with a shoot length of 29.07 cm. The simultaneous inoculation of both organisms resulted in a slightly higher shoot length of 30.05 cm. Additionally, both the nematode alone (43.90 cm) and fungus alone (43.25 cm) treatments caused a reduction in shoot length compared to the uninoculated control. The maximum shoot length observed was in the uninoculated control group, measuring 52.27 cm. These results suggest that the presence of either the nematode or the fungus, as well as their interaction, negatively impacted the shoot length of the okra plants. The uninoculated control plants exhibited the best growth in terms of shoot length (Table 1).

3.2 Root Length

Similarly, a similar trend was observed with the length of roots in the experiment. The treatment where the nematode (*M. javanica*) was inoculated first and the fungus (*F. oxysporum*) was introduced after seven days resulted in the minimum root length recorded, measuring 11.87 cm. This was followed by the treatment where the fungus was inoculated first and the nematode was introduced after seven days, with a root length of 13.27 cm. When both the nematode and fungus were inoculated simultaneously, a slightly higher root length of 14.05 cm was observed. Furthermore, both the nematode alone (16.50 cm) and fungus alone (18.50 cm) treatments resulted in a reduction in root length compared to the uninoculated control. The uninoculated control plants exhibited the maximum root length, measuring 20.72 cm.

These results indicate that the presence of the nematode or the fungus, as well as their interaction, negatively affected the length of roots in the okra plants. The uninoculated control plants exhibited the longest root length among all the treatments (Table 1).

3.3 Shoot Weight

The results of the experiment demonstrated that shoot weight was reduced in the treatment where the nematode (*M. javanica*) was inoculated first and the fungus (*F. oxysporum*) was introduced after seven days, with a shoot weight of 18.45 grams. This was followed by the treatment where the fungus was inoculated first and the nematode was introduced after seven days, resulting in a shoot weight of 21.70 grams. When both the nematode and fungus were inoculated simultaneously, a slightly higher shoot weight of 22.25 grams was observed. Similarly, when the nematode alone was inoculated, the shoot weight was 23.67 grams, and when the fungus alone was inoculated, the shoot weight was 27.37 grams. The maximum shoot weight was recorded in the uninoculated control group, measuring 30.25 grams. These findings suggest that the presence of either the nematode or the fungus, as well as their interaction, negatively impacted the shoot weight of the okra plants. The control group, without inoculation of the pathogens, exhibited the highest shoot weight among all the treatments (Table 1).

3.4 Root Weight

The results of the experiment indicated that the minimum root weight was observed in the treatment where the nematode (*M. javanica*) was inoculated first and the fungus (*F. oxysporum*) *F. oxysporum* was introduced after seven days, with a root weight of 6.30 grams. This was followed by the treatment where the fungus was inoculated first and the nematode was introduced after seven days, resulting in a root weight of 6.47 grams. When both the nematode and fungus were inoculated simultaneously, the root weight increased to 7.37 grams. Similarly, when the nematode alone was inoculated, the root weight was 9.4 grams, and when the fungus alone was inoculated, the root weight was 10.52 grams. The maximum root weight was observed in the uninoculated control group, measuring 12.42 grams. These findings indicate that the presence of either the nematode or the fungus, as well as their interaction, negatively affected the root weight of the okra plants. The control

group, without inoculation of the pathogens, exhibited the highest root weight among all the treatments (Table 1).

3.5 No. of Galls/Plant

The results of the experiment demonstrated that the maximum number of galls per plant was observed in the treatment where only the nematode (*M. javanica*) was inoculated, with an average of 43.00 galls per plant. This was followed by the treatment where the nematode was inoculated first and the fungus (*F. oxysporum*) was introduced after seven days, with an average of 38.25 galls per plant. When both the nematode and fungus were inoculated simultaneously, the number of galls per plant decreased to an average of 31.50. On the other hand, the minimum number of galls per plant was observed when the fungus was inoculated prior to the nematode, with an average of 28.50 galls per plant. No galling was observed in the control group or in the treatment where only the fungus was inoculated. These findings suggest that the presence of the nematode, either alone or in combination with the fungus, resulted in a higher number of galls on the okra plants. In contrast, inoculating the fungus before the nematode reduced the number of galls. The control group and the treatment with only the fungus did not exhibit any galling symptoms (Table 2).

3.6 No. of Egg Masses/Plant

The results of the experiment showed that the maximum number of egg masses per plant was observed when only the nematode (*M. javanica*) was inoculated, with an average of 32.67 egg masses per plant. This was followed by the treatment where the nematode was inoculated first and the fungus (*F. oxysporum*) was introduced after seven days, with an average of 28.32 egg masses per plant. When both the nematode and fungus were inoculated simultaneously, the number of egg masses per plant decreased to an average of 24.35. On the other hand, the minimum number of egg masses per plant was observed when the fungus was inoculated first and the nematode was introduced after seven days, with an average of 23.55 egg masses per plant. No egg masses were noticed in the control group or in the treatment where only the fungus was inoculated. These findings suggest that the presence of the nematode, either alone or in combination with the fungus, resulted in a higher number of egg masses on the okra plants. However, when the fungus was inoculated first and the nematode was introduced

later, the number of egg masses decreased. The control group and the treatment with only the fungus did not show any egg masses per plant (Table 2).

3.7 No. of Eggs/Egg Masses

The results of the experiment showed that the maximum number of eggs/egg masses was observed when only the nematode (*M. javanica*) was inoculated, with an average of 221.25 eggs/egg masses. This was followed by the treatment where the nematode was inoculated prior to the fungus (*F. oxysporum*), with an average of 206.00 eggs/egg masses. When both the nematode and fungus were inoculated simultaneously, the number of eggs/egg masses decreased to an average of 191.25.

On the other hand, the minimum number of eggs/egg masses was observed when the fungus was inoculated prior to the nematode, with an average of 188.75 eggs/egg masses. No eggs/egg masses were observed in the control group or in the treatment where only the fungus was inoculated. These findings suggest that the presence of the nematode, either alone or in combination with the fungus, resulted in a higher number of eggs/egg masses on the okra plants. However, when the fungus was inoculated first and the nematode was introduced later, the number of eggs/egg masses decreased. The control group and the treatment with only the fungus did not show any eggs/egg masses (Table 2).

3.8 Nematode Population/200 cc Soil

The results of the experiment revealed that the maximum nematode populations per 200cc soil were observed in the treatment where only the nematode (*M. javanica*) was inoculated, with an average of 1257.50 nematodes. This was followed by the treatment where the nematode was inoculated prior to the fungus (*F. oxysporum*), with an average nematode population of 1070.00. When both the nematode and fungus were inoculated simultaneously, the nematode population decreased to an average of 945.00. On the other hand, the minimum nematode population was observed when the fungus was inoculated prior to the nematode, with an average nematode population of 937.50. The number of the nematode population was observed in the control group or in the treatment where only the fungus was inoculated. These findings indicate that the presence of the nematode, either alone or in combination with the

fungus, led to a higher nematode population in the soil. However, when the fungus was inoculated prior to the nematode, the nematode population decreased. The control group and the treatment with only the fungus did not exhibit any nematode populations (Table 2).

3.9 Final / Total Nematode Population

The results of the experiment indicate that the maximum final nematode population was

observed in the treatment where only the nematode (*M. javanica*) was inoculated, with an average population of 14,773.50 nematodes. This was followed by the treatment where the nematode was inoculated prior to the fungus (*F. oxysporum*), with an average final nematode population of 12,251.38. Similarly, when both the nematode and fungus were inoculated simultaneously, the final nematode population was 10,323.25.

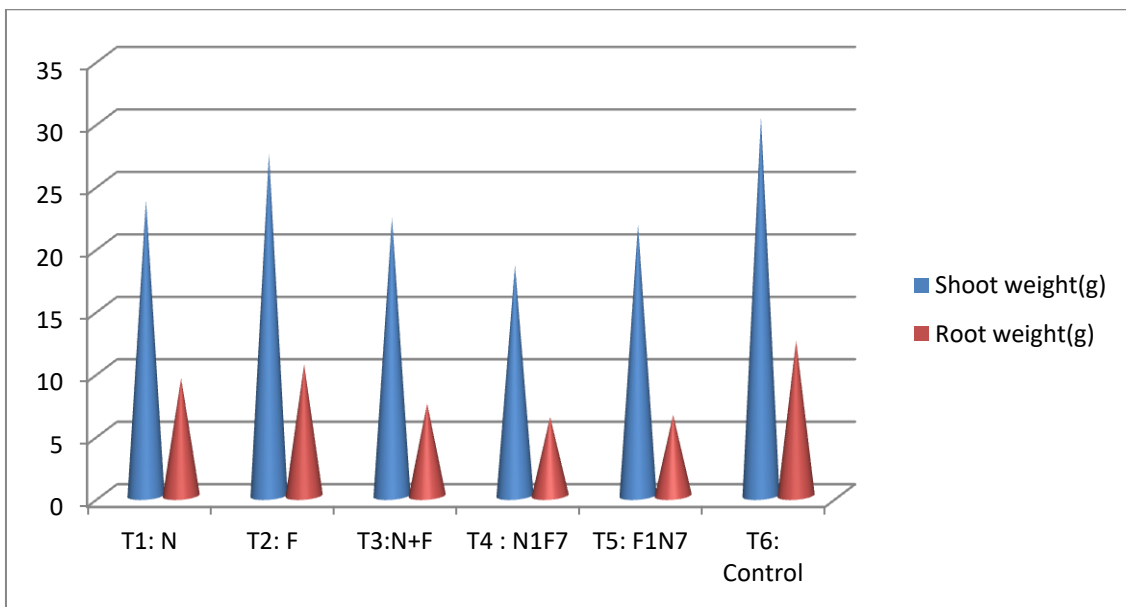


Fig. 1. Interaction between *Meloidogyne javanica* and *Fusarium oxysporum* on okra plant

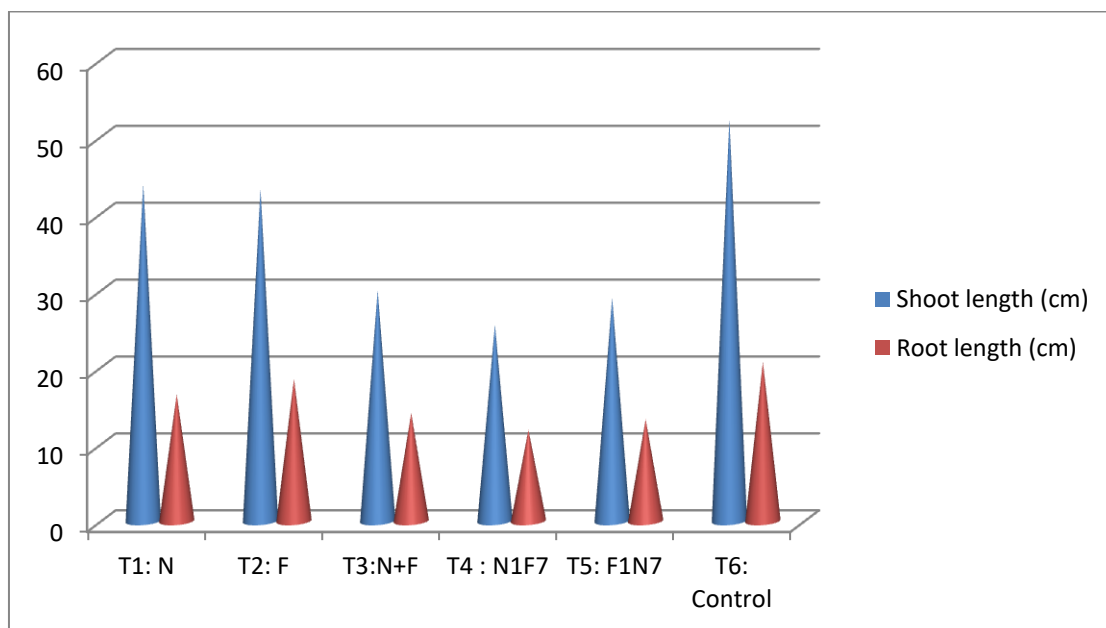


Fig. 2. Interaction between *Meloidogyne javanica* and *Fusarium oxysporum* on okra plant

Table 1. Interaction between *Meloidogyne javanica* and *Fusarium oxysporum* on okra plant

Treatments	Plant Growth Parameters			
	Shoot length (cm)	Root length (cm)	Shoot weight (g)	Root weight (g)
T1: Nematode alone(N)	43.90	16.50	23.67	9.40
T2: Fungus alone(F)	43.25	18.50	27.37	10.52
T3: Nematode and fungus inoculated at a time (N+F)	30.05	14.05	22.25	7.37
T4: Nematode at the time of sowing and fungus after one week (N ₁ F ₇)	25.55	11.87	18.45	6.30
T5: Fungus at the time of sowing and nematode after one week(F ₁ N ₇)	29.07	13.27	21.70	6.47
T6: Control (without fungus and nematode)	52.27	20.72	30.25	12.42
S.Em(±)	0.60	0.34	0.39	0.16
C.D.	1.79	1.04	1.17	0.48
C.V.	3.21	4.41	3.28	3.70

Table 2. Interaction between *Meloidogyne javanica* and *Fusarium oxysporum* on okra plant

Treatments	Plant Growth Parameter				
	No. of galls/ Plant	No. of egg masses/ plant	No. of eggs/egg masses	Nematode population/ 200cc soil/pot	Final/ Total nematode population/ pot
T1: Nematode alone(N)	43.00	32.67	221.25	1,257.50	14,773.50
T2: Fungus alone(F)	0.00	0.00	0.00	0.00	0.00
T3: Nematode and fungus inoculated at a time (N+F)	31.50	24.35	191.25	945.00	10,323.25
T4: Nematode at the time of sowing and fungus after one week (N ₁ F ₇)	38.25	28.32	206.00	1,070.00	12,251.38
T5: Fungus at the time of sowing and nematode after one week(F ₁ N ₇)	28.50	23.55	188.75	937.50	10,067.38
T6: Control (without fungus and nematod-e)	0.00	0.00	0.00	0.00	0.00
S.Em(±)	0.53	0.39	2.81	16.85	124.86
C.D.	1.60	1.18	8.42	50.46	373.88
C.V.	4.56	4.37	4.18	4.80	3.16

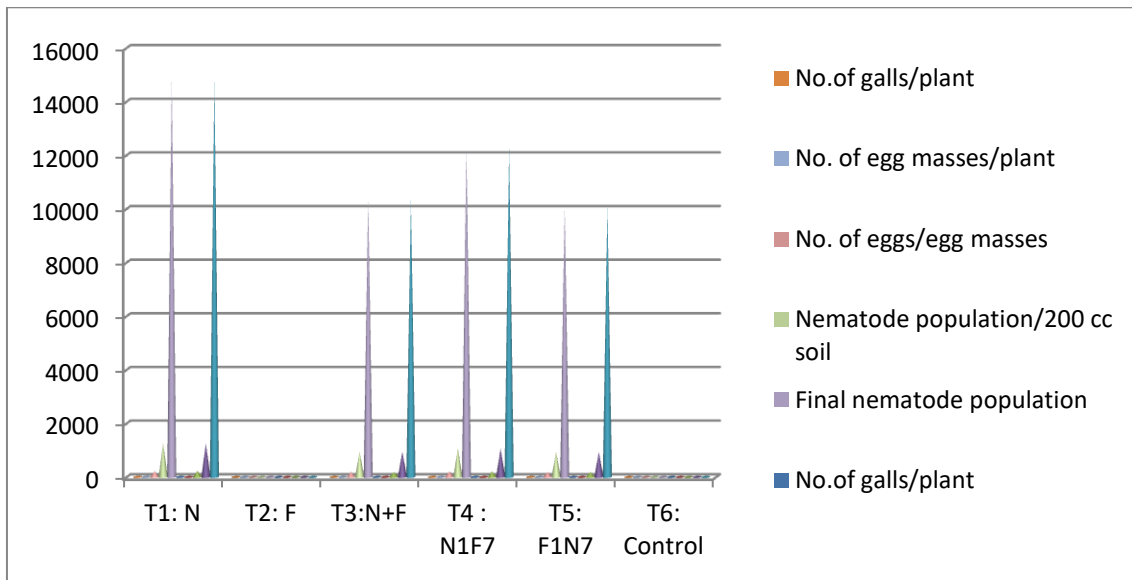


Fig. 3. Interaction between *Meloidogyne javanica* and *Fusarium oxysporum* on okra plant

On the other hand, the minimum final nematode population was observed when the fungus was inoculated prior to the nematode, with an average population of 10,067.38 nematodes. No final nematode population was observed in the control group or in the treatment where only the fungus was inoculated. These findings suggest that the presence of the nematode, either alone or in combination with the fungus, resulted in a higher final nematode population. However, when the fungus was inoculated prior to the nematode, the final nematode population decreased. The control group and the treatment with only the fungus did not exhibit any final nematode populations (Table 2).

4. DISCUSSION AND CONCLUSION

The results of the present investigation confirm that the growth of plants was negatively impacted when they were inoculated with both *M. javanica* (root knot nematode) and *F. oxysporum* (fungus), compared to the uninoculated control group. This is similar to the findings reported by Meena et al. [6] who observed that the severity of wilt disease was more pronounced when nematodes were sequentially inoculated before the fungus, compared to the fungus alone treatment. Kumar et al. [7] also reported similar findings, highlighting the synergistic effect of nematode and fungus inoculation in inhibiting plant growth. Furthermore, significant reductions in plant height were observed in pots inoculated with only the nematode or only the fungus, indicating the individual detrimental effects of these pathogens

on plant growth. Overall, these findings emphasize the negative impact of the interactive effect between *M. javanica* and *F. oxysporum* on plant growth and highlight the importance of considering the combined influence of multiple pathogens in disease severity and crop productivity. Meena et al. [8] evaluated the interaction between *Fusarium oxysporum* f. sp. *lycopersici* and *Meloidogyne incognita*. The results showed that inoculation of nematode at 7th day prior to inoculation of fungus caused maximum reduction in plant growth parameters and increase per cent disease incidence followed by simultaneous inoculation of nematode and fungus as compared to control. Kumawat et al. [9] studied the interactive effect of *Meloidogyne javanica* in enhancing the incidence of root rot by *Fusarium solani* in fennel (*Foeniculum vulgare* Mill.). The results of the experiments showed that inoculation of nematode at the time of sowing and fungus one week after caused maximum reduction in plant growth parameters and increased disease incidence followed by simultaneous inoculation of nematode and fungus in comparison to check. Whereas, maximum nematode reproduction was recorded in nematode alone followed by N+F. Conclusively, presence of root damaging nematodes in rhizosphere zone of plants plays an important role in enhancing incidence of root rot diseases.

Monika et al. [10] Disease development in crops depends on the complex interaction between host, pathogen and environmental conditions.

The significant role of nematode in the development of nematode-fungus interaction in demonstrated in many crops throughout the world. Therefore, the main objectives of this study were to evaluate the interaction between *Fusarium oxysporum* and *Meloidogyne* spp. in cucumber. The experiment was laid out in a completely randomized design with four replications in pot conditions. The results show that inoculation of nematode at 7th day prior to inoculation of fungus caused maximum reduction in plant growth parameters and increase per cent disease incidence followed by simultaneous inoculation of nematode and fungus as compared to control.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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