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# Cohabitation of *Fusarium oxysporum* f. sp. *pisi* and *Meloidogyne incognita* in pea (*Pisum sativum* L.) Var. Kashi Nandini and its Effect on Plant Growth

***Balkishan Chaudhary* and Jayant Bhatt***
*Department of Plant Pathology, College of Agriculture,*
*Jawaharlal Nehru Krishi Vishwa Vidyalaya, Jabalpur- 482004 (Madhya Pradesh), India.*

*(Corresponding author: Balkishan Chaudhary*)*
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**ABSTRACT: In glasshouse conditions, a disease complex involving *Meloidogyne incognita* and *Fusarium oxysporum* f. sp. *pisi* was investigated on pea. The pot experiments were carried out to decide the pathogenic effects of pathogens individually as well as collaboratively. *M. incognita* and *F. oxysporum* f. sp. *pisi*, individually caused a significant reduction in plant height, fresh and dry weight, in comparison of un-inoculated control. Various plant growth parameters were observed significant related with the reduction of plant growth when simultaneous and sequential inoculation of *M. incognita* and *F. oxysporum* f. sp. *pisi* was done. However, the reduction was more prominent when both the pathogens inoculated simultaneous. Nematode multiplication, galls formation in numbers, egg masses, eggs/egg masses and population of nematode were adversely exaggerated on simultaneous and sequential inoculation of both the pathogens in all the treatments. It was concluded that both the pathogens *viz.*, *M. incognita* and *F. oxysporum* f. sp. *pisi* are virulent against the pea *Var.* Kashi Nandini which defined the seriousness of disease and ultimately adaptation of management if such condition formed.**

**Keywords:** Cohabitation, *Fusarium oxysporum* f. sp. *pisi,* plant growth, pathogenic effects.

## INTRODUCTION

One of the most important pulse crops is Pea (*Pisum sativum*) in India as well as in the world. The crop having protein (22.5%), carbohydrate (56.5%), fat (1.1%), minerals (2.2%), fibre (4.5%) and important vitamins like vitamin B1 and B5 (Fageria *et al*., 2002). Pea is prone to various abiotic and biotic factors. The important biotic factors which showed the higher losses to the pea crop (Engqvist, 2001; Grunwald *et al*., 2004) is Root-knot nematode (*Meloidogyne* spp.). The *Meloidogyne incognita* (Kofoid & White) Chitwood and the wilt fungus, *Fusarium oxysporum* f. sp. *pisi* both infect the pea crop and forms disease complexes (Kumar and Kamalwanshi 2009). Combined infection of nematodes and fungus affects the crop in terms of quantitative and as well qualitative terms to the pea than their seperate infection. It is a well known fact that nematodes exposes to roots of the plants to the secondary invasion by the fungal pathogens and aggravate the disease severity (Mai and Abawi, 1987). Nematodes and fungal pathogens when forming disease complexes may cause significantly more crop losses than singularly (Hussey and McGuire, 1987). This complexion of disease i.e. the interaction between fungus which infects the plant roots and nematode shows the reduction in seed germination and enhance factors like more gall formation and fecundity in nematodes (Kassab and Ali 1996). The root knot nematode and wilt fungi found to be associated with the loss of growth and wilting in plants (Shawadfy and Mousa, 1997). Interactive effect of fungus and nematodes has been documented earlier in various crops like banana (Jonathan and Gajendran, 1998), chickpea (Maria *et al.,* 1998), crossandra (Mallaiah *et al.,* 2014) and gerbera (Meena *et al.,* 2015). Present investigation has been carried out to study the interactive effect of *Fusarium oxysporum* f. sp. *pisi* and *Meloidogyne incognita* in pea at different time of inoculation.

## METHOD AND MATERIALS

This experiment was performed in the pot house during *Rabi* season 2018 and 2019 for study the combined interaction of *Meloidogyne incognita* and *Fusarium oxysporum* f. sp. *pisi* on pea. The statistical Completely Randomized Block Design (CRBD) were utilized for data analysis. Variety Kashi Nandini (susceptible to nematode and moderately resistant to *F. oxysporium* f. sp. *pisi*) was used in the present investigation. The fungus was cultured on autoclaved corn meal medium. Ten cm earthen pots holding 500 $cm^3$ of soil were used for wilt fungi and root knot nematodes. A constant level of 1000 second stage juveniles of root knot nematode was inoculated per pot as per treatment. The nematodes were pipetted and poured around the pre germinated seeds, growing in sterile moist chambers prepared. During the dibbling of seeds the pre-germinated seeds have the radical length of 0.3 to 0.5 mm and were dibbled in 2 cm deep. In the precautionary measures, for avoid contamination from one pot to another. Every one week when the nematodes were to be inoculated, three glass rods were fixed 2 cm deep in a circle 2 cm in diameter. The glass rods were removed before to inoculation, and the nematode solution was equally poured into the holes before being filled with sterile soil. *Fusarium oxysporum* f. sp. *pisi* was used to inoculate the soil, and 50g of the fungus's inoculated corn meal medium was combined with each pot for fungus, fungus + nematode one week later, and simultaneous inoculation of fungus and nematode was done. The uninoculated corn meal medium was combined with the other pots that will get the nematode, nematode + fungus, fungus one week later, and control inoculations. The Grewal *et al.* (1974) method of placing three glass rods evenly spaced in a circle of two cm diameter and plugging the holes with sterile soil after introducing the actively growing *Fusarium oxysporum* f. sp. *pisi* was used when the fungus was to be inoculated one week after the nematode inoculation.

The treatments included (a) nematode alone (N), (b) fungus alone (F), (c) simultaneous inoculation of nematode and fungus (NF), (d) simultaneous inoculation of fungus and nematode one week later (N1F2), and (e) nematode at the time of sowing and fungus one week after (N1F2) (F1N2). After CRBD, 30 pots were therefore randomly placed over the glass house bench and watered as needed with an equal amount of sterilised distilled water. After 45 days following inoculation, the experiment came to an end. Throughout the experiment, the temperature in the glass house varied between 21 to 26°C.

## RESULT AND DISCUSSION

*A. Effect on plant growth parameters*

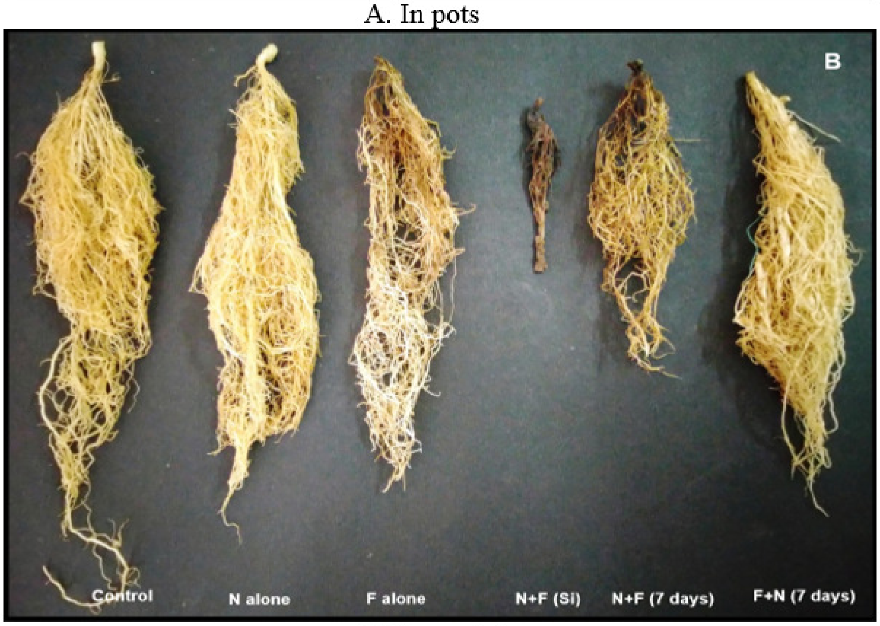
**a) Plant height and Root length:** Plant growth parameters *viz*., length of shoot and length of root were recorded maximum (46.32cm and 21.11 cm) in control while the treatment nematode and fungus inoculated simultaneously recorded minimum shoot and root length *i.e.* 16.25 & 9.60 cm respectively (Table 1 & plate 1). All the studies treatments were found differed significantly over the control. In contrast to the treatment in which the fungus was inoculated first and the nematode a week later, the treatment in which the fungus was inoculated first and the nematode a week later appeared to have a 36.48 cm shoot length and a 17.74 cm root length showed the second lowest shoot and root length (21.16 & 14.11cm). Significant reduction in shoot length and root length in pots with nematode alone (38.94cm & 18.58cm) and fungus alone (24.11cm & 16.64cm) were also observed. The similar results were also observed by Padilla *et al.* (1980) who investigated that decline in plant height and root length was significant where the treatment nematode and fungus was inoculated either alone or in different combinations at planting time and three weeks later. It was also seen that concomitant inoculation of the nematodes and the fungus at planting caused death of the plants within 45 days. In addition, Husain *et al.* (1985) revealed that simultaneous nematode and fungal inoculations on pea resulted in the greatest loss in plant growth features. It was discovered that the percent reduction in plant growth parameters was greater than the sum of the reductions caused by the two pathogens (*M. incognita* + *F. oxysporum* f. sp. *pisi*) when they were inoculated separately, suggesting that the two organisms had a synergistic effect when they shared a same host. The nematode-induced vulnerability of plants to fungal assault and subsequent increase in disease severity is the most likely cause of the synergistic impact. Haseeb *et al.* (2007) have also reported results of a similar kind. When compared to uninoculated plants, they looked into it and discovered a substantial decrease in all growth indices for *Pisum sativum*. The plant growth metrics were dramatically decreased when the wilt fungus and nematodes were both simultaneously inoculated. In their study, Kumar and Kamalwanshi (2009) noted that the presence of the root knot nematode (*M. incognita*), regardless of the presence of other organisms like *F. oxysporum* f. sp. *pisi*, caused a substantial reduction in shoot length. Nevertheless, when nematodes were infected one week before the fungus, the greatest loss in shoot length was seen. Kumar *et al.* (2017) observed that plant growth was adversely exaggerated in all the cases where the plant was inoculated with *M. inocgnita* and *F. oxysporum* when compared with uninoculated control. Siddiqui and Zehra (2012) observed that *M. incognita* and *F. oxysporum* f. sp. *pisi* alone caused a significant fall down in plant biomass (fresh & dry). Reduction in plant growth caused by *M. incognita* was statistically at par to that caused by *F. oxysporum* f. sp. *pisi*.

**Table 1: Effect of *M. incognita* and *F. oxysporum* f. sp. *pisi* on plant growth parameters of pea***.**

| Sr. No. | Treatments | Shoot length (cm)* | Root length (cm)* | Fresh shoot weight (g)* | Fresh root weight (g)* | Dry shoot weight (g)* | Dry root weight (g)* |
|---|---|---|---|---|---|---|---|
| 1. | Control | 46.32 | 21.11 | 7.37 | 6.41 | 1.98 | 0.78 |
| 2. | N alone | 38.94 | 18.58 | 5.84 | 5.34 | 1.43 | 0.47 |
| 3. | F alone | 24.11 | 16.64 | 3.77 | 2.66 | 0.84 | 0.29 |
| 4. | N + F (si) | 16.25 | 9.60 | 1.01 | 0.95 | 0.209 | 0.122 |
| 5. | N + F 7 day | 21.16 | 14.11 | 3.00 | 2.07 | 0.68 | 0.24 |
| 6. | F + N 7 day | 36.48 | 17.74 | 5.27 | 3.64 | 1.37 | 0.33 |
| **S. Em ±** | | **0.617** | **0.573** | **0.192** | **0.127** | **0.072** | **0.019** |
| **CD at 5 %** | | **1.812** | **1.682** | **0.563** | **0.372** | **0.211** | **0.056** |

**b) Fresh and dry shoot and root weight:** Minimum fresh shoot and root weights (1.01 & 0.95 g) was noted with *M. incognita* and *F. oxysporum* f. sp. *pisi* inoculated concurrently followed by nematode inoculated first and after seven days fungus (3.00 & 2.07 g) and fungus inoculated alone (3.77 & 2.66 g). The pots with the wilt fungus infected initially, nematode a week later, and nematode alone treated pots had respective shoot weights of 5.27 and 5.84 g and root weights of 3.64 and 5.34 g. Maximum shoot and root weights (7.37 & 6.41 g) were measured in control pots that had not been infected. The weights of the dried shoots and roots showed a similar tendency. When compared to control, all treatments considerably reduced the root and shoot weights (dry). Shoot and root weight were measured simultaneously inoculations on a dry weight basis at a minimum of (0.209 & 0.122 g), followed by plants injected with nematode first and fungus a week later (0.68 & 0.24 g), and wilt pathogen implanted alone (0.84 & 0.29 g). The pots with the fungus infected initially, nematode a week later, and nematode alone injected exhibited, respectively, dry shoot weights of 1.37 g and 1.43 g and root weights of 0.33 g and 0.47 g. (Table 1). The findings of the current study were consistent with those of Haseeb *et al.* (2007), who noted a substantial decrease in the fresh and dry weight of all the roots and shoots of *Pisum sativum* when compared to uninoculated plants. Prior to the fungus, simultaneous inoculation of pathogens and nematodes greatly increased. Siddiqui and Zehra (2012) concluded that *M. incognita* and *F. oxysporum* f. sp. *pisi* alone caused a significant reduction in plant biomass (fresh & dry) over un-inoculated control.


A. In pots




B. Up rooted

**Plate 1.** Interaction between *M. incognita* and *F. oxysporum* f. sp. *pisi* in pea var. Kashi Nandini.

When *M. incognita* and *F. oxysporum* f. sp. *pisi* were injected jointly, the amount of fresh and dried plant damage decreased more than when these pathogens were injected separately. According to Maqsood *et al.* (2020), both individual and combination inoculations of *M. incognita* and *Fusarium oxysporum* f. sp. *lycopersici* considerably increased the severity of the disease. The length of the shoots and roots, the total dry weight, and the total chlorophyll content in tomato all decreased as the disease intensity increased.

**Effect on nematode multiplication:** Maximum number of galls (58.80) and egg masses (6.32) were noted in the treatments with *Meloidogyne incognita* alone @ 1000 $J_2$/500g of soil inoculations while the minimum number of galls (18.10) and egg masses were seen when *Fusarium oxysporum* f. sp. *pisi* inoculated first and *Meloidogyne incognita* @ 1000 $J_2$/500g soil after one week and concomitant inoculations (Table 2). Statistics-wise, the treatments using *Fusarium oxysporum* f. sp. *pisi* alone were comparable to the control. 36.50 galls per plant and four 4.00 egg masses per gallon were seen, with nematodes infected initially and fungi a week later. When worm and fungus were inoculated simultaneously, there were 21.60 galls per plant and 2.22 egg masses per gallon. Nematode alone inoculations were followed by nematode inoculated first and fungus a week later, and the greatest (171.50) numbers of egg/egg masses and nematode population (6307.0) were noted (81.50 & 4186.40). Concurrent inoculations of *Fusarium oxysporum* f. sp. *pisi* and *Meloidogyne incognita* resulted in minimum (40.20) eggs and nematode population (2357.20). The findings corroborated Roy and Mukhopadhya's (2004) observations of the Brinjal crop. Haseeb *et al.* (2007) observed that the rate of reproduction of nematode and galling on roots decreased with pre-inoculation of fungus, while the infection of fungus increased in the presence of nematode. Siddiqui and Zehra (2012) observed highest number of galls and nematode multiplication in plants inoculated with *M. incognita* while inoculation of *F. oxysporum* with *M. incognita* resulted in less galling and nematode multiplication.

**Table 2: Effect of *M. incognita* and *F. oxysporum* f. sp. *pisi* on nematode multiplication*****.

| Sr. No. | Treatments | No. of galls/plant* | No. of egg masses/galls* | No. of eggs/egg mass* | Nematode population (root + soil)* |
|---|---|---|---|---|---|
| 1. | Control | 0.00 (1.00)** | 0.00 (1.00) | 0.00 (1.00) | 0.00 (1.00) |
| 2. | N alone | 58.80 (8.66) | 6.32 (3.51) | 171.50 (14.09) | 6307.00 (80.41) |
| 3. | F alone | 0.00 (1.00) | 0.00 (1.00) | 0.00 (1.00) | 0.00 (1.00) |
| 4. | N + F (si) | 21.60 (5.64) | 2.22 (2.48) | 40.20 (7.34) | 2357.20 (49.55) |
| 5. | N + F 7 day | 36.50 (7.04) | 4.00 (3.00) | 81.50 (10.20) | 4186.40 (65.70) |
| 6. | F + N 7 day | 18.10 (5.25) | 3.27 (2.80) | 58.90 (8.67) | 2714.90 (53.10) |
| **S. Em ±** | | **2.123** | **0.092** | **2.327** | **14.231** |
| **CD at 5 %** | | **6.233** | **0.269** | **6.834** | **41.785** |

* Mean of five replications, **Figure in Parentheses are √ n+1 transformed value, ***pooled data of two years

Ahmed *et al.* (2014) reported that maximum numbers of nematode population and number of galls/root system in the treatment similar in chilli. Kumar *et al.* (2017) investigated and recorded maximum number of root galling and nematode population in the presence of nematode alone followed by nematode seven days prior to fungus and fungus seven days prior to nematode with the presence of *Fusarium oxysporum* on black gram.

## CONCLUSION

The above exploration obviously clarify the relation between *M. incognita* and *Fusarium oxysporum* f. sp. *pisi* and their role in causing the disease in pea. The above study reported that the nematode, *M. incognita* acts as predisposing agent for the invasion of *F. oxysporum* f. sp. *pisi* and their combined infestation reduced the plant growth parameters and significantly changes modified the biochemical components in the plant. Cohabitation of nematode-fungus should be given emphasis while formulating the management strategies for the successful supervision of the both pathogens.

## FUTURE SCOPE

Genetic basis of mechanism of interaction, physiologically and histological changes in diseases complex have to be workout. Characterization of translocatable metabolites in nematode fungal interaction and determination of their biochemical nature and mode of action and specificity are the areas of further research.

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