



Reproductivity of *Meloidogyne incognita* on Fifteen Cucumber Cultivars

Muhammad Zameer Kayani^{1,*} and Tariq Mukhtar²

¹Green Belt Project, Department of Agriculture, Rawalpindi

²Department of Plant Pathology, Pir Mehr Ali Shah Arid Agriculture University, Rawalpindi

ABSTRACT

Root-knot nematodes are mainly controlled by chemicals and their use is often coupled with health hazards. The deleterious effects of pesticides can be averted by using non-chemical approaches, and resistant cultivars can prove a promising substitute. For their fitness as nematode-suppressive crops, the reproductive and developmental rates of the nematode must be assessed on these cultivars. As there is meager information on the reproductive potential of *Meloidogyne incognita* on different cucumber cultivars commonly cultivated in Pakistan, therefore, the objective of the present study was to evaluate the reproductivity of *M. incognita* on fifteen cucumber cultivars with varying levels of resistance and susceptibility. Significant differences were found among all the cucumber cultivars regarding formation of galls, egg masses, fecundity and reproductive factor. Maximum galls and egg masses were observed on highly susceptible cultivars followed by susceptible ones. On the other hand, minimum egg masses and galls were recorded on resistant and moderately resistant cultivars. The fecundity and reproductive factor of the nematode was found to be the minimum on resistant cultivar followed by moderately resistant ones. Contrarily, the highest fecundity and reproductive factor was observed on highly susceptible cultivars followed by susceptible ones. As the reproductive potential of *M. incognita* was found to be lowered on resistant (Long Green) and moderately resistant (Marketmore, Dynasty, Pioneer-II and Summer Green) cultivars and hence are recommended for cultivation in fields infested with *M. incognita*.

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Authors' Contribution

MZK and TM designed the study, executed experimental work, analyzed the data and prepared the manuscript. TM supervised the work.

Key words

Reproductive factor, Fecundity, Root-knot nematodes, Resistance.

INTRODUCTION

Many pests including plant pathogenic nematodes attack a large number of vegetables and are responsible for causing severe growth retardation (Ashfaq *et al.*, 2015, 2017; Riaz *et al.*, 2015; Fateh *et al.*, 2017; Javed *et al.*, 2017a, b; Kassi *et al.*, 2018; Mukhtar *et al.*, 2018; Nabeel *et al.*, 2018). However, one of the most important nematodes associated with low production of cucumber is the root-knot nematode (*Meloidogyne incognita*) (Kayani *et al.*, 2017, 2018; Tariq-Khan *et al.*, 2017). Root-knot nematodes are ranked at the top among the five major plant pathogens and the first among the ten most important genera of plant parasitic nematodes in the world (Mukhtar *et al.*, 2017a). They have wide geographic distribution, large host range and high destructive potential. They have been reported to be implicated with other plant pathogens and result in disease complexes and aggravation of wilt diseases (Shahbaz *et al.*, 2015; Aslam *et al.*, 2017a, b). In Pakistan *M. incognita* has been found one of the most dominant root-knot species and rampant in the cucumber-producing areas of Pakistan

and considerably reduces growth and yield (Kayani *et al.*, 2013). The worldwide distribution of this species is 47%. In Pakistan its overall occurrence is 52% and of all the root-knot nematode species associated with cucumber, *M. incognita* constituted 78.5% (Kayani *et al.*, 2013). Overall yield losses of 50 to 80% have been reported to be caused by root-knot nematodes in vegetables and about 33% yield losses due to root-knot nematodes have been estimated in cucumber (Sasser, 1979). Root-knot nematodes have become a serious threat to the profitable cultivation of cucumber in the country. The yield losses by root-knot nematodes are mainly caused due to buildup of inoculum of the nematode and repeated cultivation of same cultivars in the same land every year (Hussain *et al.*, 2016).

Root-knot nematodes are mainly controlled by the application of nematicides and resistant cultivars. Although nematicides can effectively manage nematodes but their use is often associated with hazards in underdeveloped countries like Pakistan and hence becoming unattractive for farmers. On the other hand, use of nematode resistant cultivars is considered to be innocuous and economically feasible (Mukhtar *et al.*, 2017b). These cultivars can also be integrated with other management practices in integrated nematode management (Shahzaman *et al.*, 2015; Khan *et al.*, 2017; Rahoo *et al.*, 2017, 2018a, b). Cultivars of various crops and vegetables are basically assessed for resistance

* Corresponding author: kianizmr@gmail.com

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to root-knot nematodes using root galling index as the only standard of damage to plants which is unreliable. This necessitates that other parameters like nematode reproduction on cultivars should also be considered while assessing resistance or susceptibility among crop cultivars to root-knot nematodes (Florini, 1997; Afolami, 2000). The key principles for accepting cultivars for their successful deployment in fields are their ability to suppress nematode populations and yield profitably in the presence of nematodes. For their fitness as nematode-suppressive crops, the reproductive and developmental potential and rates of *M. incognita* on these cultivars must be assessed. As there is meager information on the reproductive potential of *M. incognita* on different cucumber cultivars commonly cultivated in Pakistan, therefore, the objective of the present study was to evaluate the reproductivity of *M. incognita* on fifteen cucumber cultivars with varying levels of resistance and susceptibility.

MATERIALS AND METHODS

Nematode culture

A population of root-knot nematode (*Meloidogyne incognita*) initially isolated from cucumber roots, identified on the basis of perineal pattern and maintained on the highly susceptible cultivar of tomato (money maker) was used in the assessment. The nematode was mass produced on tomato cv. Money maker as described previously (Mukhtar *et al.*, 2013). Second stage juveniles (J2s) were extracted from the infected roots for inoculation of plants as described by Whitehead and Hemming (1965).

Assessment of cucumber cultivars for nematode reproductivity

Fifteen cucumber cultivars with different levels of resistance or susceptibility were assessed for reproductivity of *M. incognita*. These cultivars comprised of Long Green (resistant); Summer Green, Dynasty, Pioneer-II, Marketmore (moderately resistant); Poinsett, Cucumber Cetriolo, Green Wonder (moderately susceptible); Babylon, Cobra, Falcon-560 (susceptible); and Royal Sluis, Thamin-II, Mehran, Mirage (highly susceptible) (Mukhtar *et al.*, 2013). Three seeds of each cultivar were sown in plastic pots (20-cm-dia) containing 3 kg formalin sterilized soil (sand, 60%; silt, 20%; clay, 19%; organic matter, 1% and pH, 7.2). Ten days after emergence, one healthy seedling of each test cultivar was maintained in each pot. The plants of each cultivar were then inoculated with approximately 3000 freshly hatched J2s of *M. incognita* by making holes around the plants. The plants of each cultivar which were not inoculated with J2s served as control of that cultivar. Each cultivar was replicated

five times. The pots were maintained in a greenhouse in a completely randomized design at $25\pm 2^{\circ}\text{C}$ for sixty days. The plants were watered when needed.

Data collection

After stipulated period data regarding number of galls, egg masses, fecundity (eggs/egg mass) and reproductive factor were taken. Egg masses were counted after staining infected roots with Phloxin B (0.12 g Phloxin B dissolved in 1 L of water) for 20 minutes. The egg masses-stained roots were rinsed with tap water and counted under stereomicroscope at $25\times$. The final nematode population was computed by adding up the eggs extracted from the infected roots (Hussey and Barker, 1973) and nematodes extracted from the soil (Whitehead and Hemming, 1965). This final population was divided by the initial population to find out the reproductive factor.

Statistical analysis

All the data were subjected to Analysis of Variance (ANOVA) using GenStat package 2009 (12th edition) version 12.1.0.3278 (www.vsnl.co.uk). The means were compared by Fisher's Protected Least Significant Difference Test at 5%.

RESULTS

Significant differences were found among all the cucumber cultivars regarding formation of galls, egg masses, fecundity and reproductive factor. Maximum galls were observed on highly susceptible cultivars followed by susceptible ones. On the other hand, minimum galls were recorded on resistant and moderately resistant cultivars as shown in Figure 1A. Similarly, the nematode produced maximum egg masses on the highly susceptible cultivars followed by cultivars showing susceptible reactions. Contrarily, minimum egg masses were found on resistant and moderately resistant cultivars as shown in Figure 1B.

The cultivars showed significant variations regarding fecundity of *M. incognita* on fifteen cucumber cultivars. The fecundity of the nematode was found to be the maximum on highly susceptible cultivars followed by susceptible ones. The other way round, the nematode produced the minimum number of eggs per egg mass on resistant cultivar followed by moderately resistant cultivars (Fig. 1C). The reproductive factor of the nematode was also found to be the minimum on resistant cultivar followed by moderately resistant ones. Contrariwise, the highest reproductive factor was observed on the highly susceptible cultivars followed by susceptible ones. Significant variations in reproductive factor were also observed among cultivars showing different levels of susceptibility (Fig. 1D). The

reproductive factors of highly susceptible, susceptible, moderately susceptible, moderately resistant and resistant cultivars were found to be statistically different from each others' and were in the order: HS>S>MS>MR>R.

DISCUSSION

Vegetables are good hosts of root-knot nematodes and cucumber has been found an excellent host of *M. incognita* (Kayani *et al.*, 2013). The current study deals with the comparative reproductive potential of *M. incognita* on fifteen cucumber cultivars having varying degrees of

resistance or susceptibility. One of the most significant key factors for selecting cultivars for cultivation is their reproductive factors. Cultivars having lower reproductive factors will be appropriate for the management of root-knot nematodes. The host status of any crop is determined by the reproductive factor of the nematode which quantifies its reproductive potential on a specified crop plant (Windham and Williams, 1988). When the reproductive factor of a nematode on a selected host is less than one, it means the nematode is unable to reproduce on that host. On the other hand, if the reproductive factor exceeds one, the nematode can successfully multiply on that host (Pofu *et al.*, 2010).

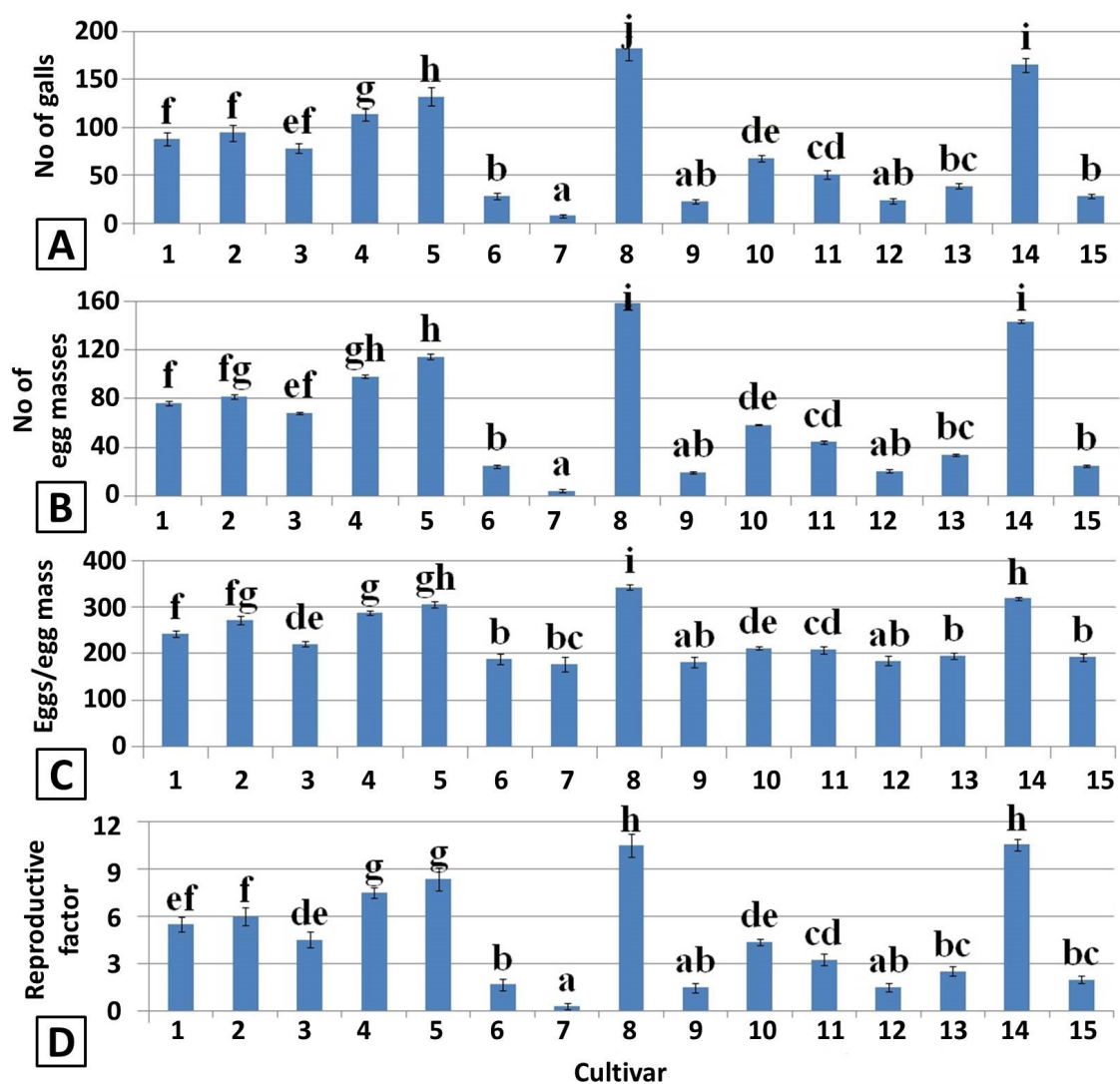


Fig. 1. Effect of cucumber cultivars on number of galls (A), number of egg masses (B), production of eggs/egg mass (C) and reproductive factor of *Meloidogyne incognita* (D). Means are average of five replications. Means followed by the same letters are not different according to Fisher's Least Significant Difference test ($P > 0.05$). 1, Babylon; 2, Cobra; 3, Falcon-560; 4, Mehran; 5, Mirage; 6, Marketmore; 7, Long Green; 8, Thamin-II; 9, Dynasty; 10, Green Wonder; 11, Cucumber; 12, Pioneer-II; 13, Poinsett; 14, Royal Sluis; 15, Summer Green.

The sensitivity of a host is assessed on the basis of host status and its responses to nematode infectivity (Seinhorst, 1967). When a host permits the nematode to reproduce on it and incurs yield losses, the host is referred to as susceptible, whereas if a host does not suffer yield losses, it is considered to be tolerant to the nematode. However, if the host does not allow the nematode to reproduce and resultantly there is no yield loss, the host will be a resistant one (Seinhorst, 1967). In the present study, cucumber cultivars showed highly significant differences regarding reproduction of *M. incognita* appraised on the basis of number of egg masses, fecundity and reproductive factor. Infection and production of egg masses on roots by the nematode were the principal determinants of variations among cucumber cultivars and these variations subsequently determined final nematode populations and reproductive factors (Fig. 1C, D). The variations in reproductive rates might partially be the result of genetic factors which impart resistance or susceptibility to the host or due to genetic variations in nematode populations (Griffin, 1982; Jacquet *et al.*, 2005; Castagnone-Sereno, 2006).

The differences in the host can affect different phases of the life cycle of the nematode. The resistant host does not allow the nematode to enter the roots or kill the nematode after it has penetrated the roots or the nematode is unable to develop or reproduce in the host. The variations in reproduction and multiplication of *M. incognita* on cucumber cultivars are owing to variations in their genetic makeup which can be explained in terms of number of egg masses. The production of maximum egg masses and eggs on the roots of highly susceptible and susceptible cultivars explains that maximum numbers of juveniles entered the roots and were successful in completing their life cycles in the host. The other way round, in case of resistant and moderately resistant cultivars only few juveniles made their way into the roots and got matured which is obvious by the number of egg masses and their reproductive factors. There are reports that resistant cultivars contain a limited number of developed nematodes as compared to susceptible cultivars (Dropkin and Nelson, 1960). Hindrances in invasion by second stage juveniles of the nematode have been ascribed to failure of maximum numbers of juveniles to develop in the infected roots and/or hypersensitive reactions in the host (Dropkin, 1969). In case of susceptible hosts, the juveniles had the maximum potential to fully develop as evident by their reproductive factors on the highly susceptible and susceptible cultivars in the present study (Fig. 1D). On the other hand, in resistant and moderately resistant cultivars the development of the juveniles was either curtailed or delayed (Nelson *et al.*, 1990).

CONCLUSION

The reproductive potential of *Meloidogyne incognita* was found to be significantly low on resistant (Long Green) and moderately resistant (Marketmore, Dynasty, Pioneer-II and Summer Green) cultivars. These cultivars are likely to suffer less damage by the nematode as compared to susceptible ones with highest rate of nematode multiplication and hence are recommended for cultivation in fields infested with *M. incognita*.

Statement of conflict of interest

Authors have declared no conflict of interest.

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