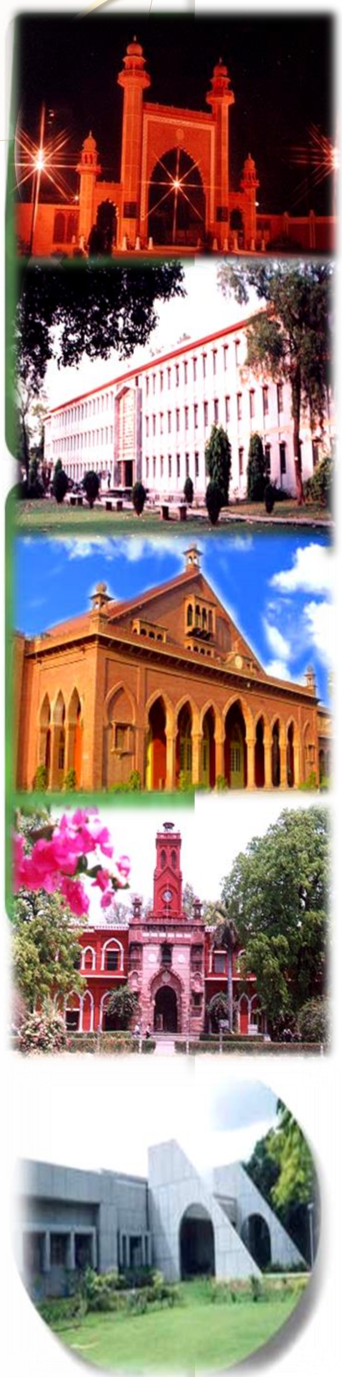
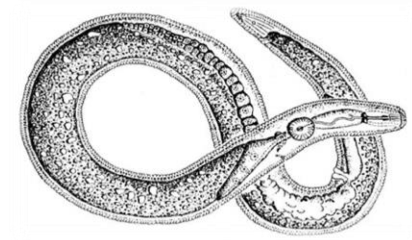
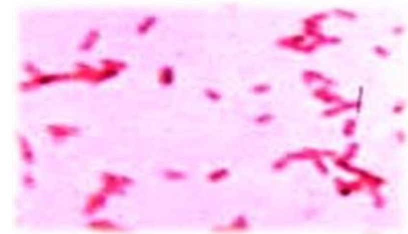






Interactions of Plant Pathogenic Bacteria and Plant Growth Promoting Rhizobacteria with Plant Parasitic Nematodes

Zaki A. Siddiqui
Plant Nematology and Pathology Section
Department of Botany
Aligarh Muslim University
Aligarh-202 002-India

NEMATODES

- Unsegmented worms
- Usually microscopic
- Phylum Nematoda

Nematodes are heterotrophic, multicellular, unsegmented, bilaterally symmetrical, pseudo-coelomate animals.

NEMATODES

Many different groups and habits

➢ Decomposers	19%
➢ Predators	
➢ Insect parasites	8%
➢ Animal parasites (often host-specific)	33%
➢ Plant parasites	7%
➢ Others (freshwater, marine, etc.)	33%

Plant Parasitic Nematodes – Habits and Habitat

- **Ectoparasites**

  many kinds inhabit soil around plant roots, feed on roots

- **Endoparasites**

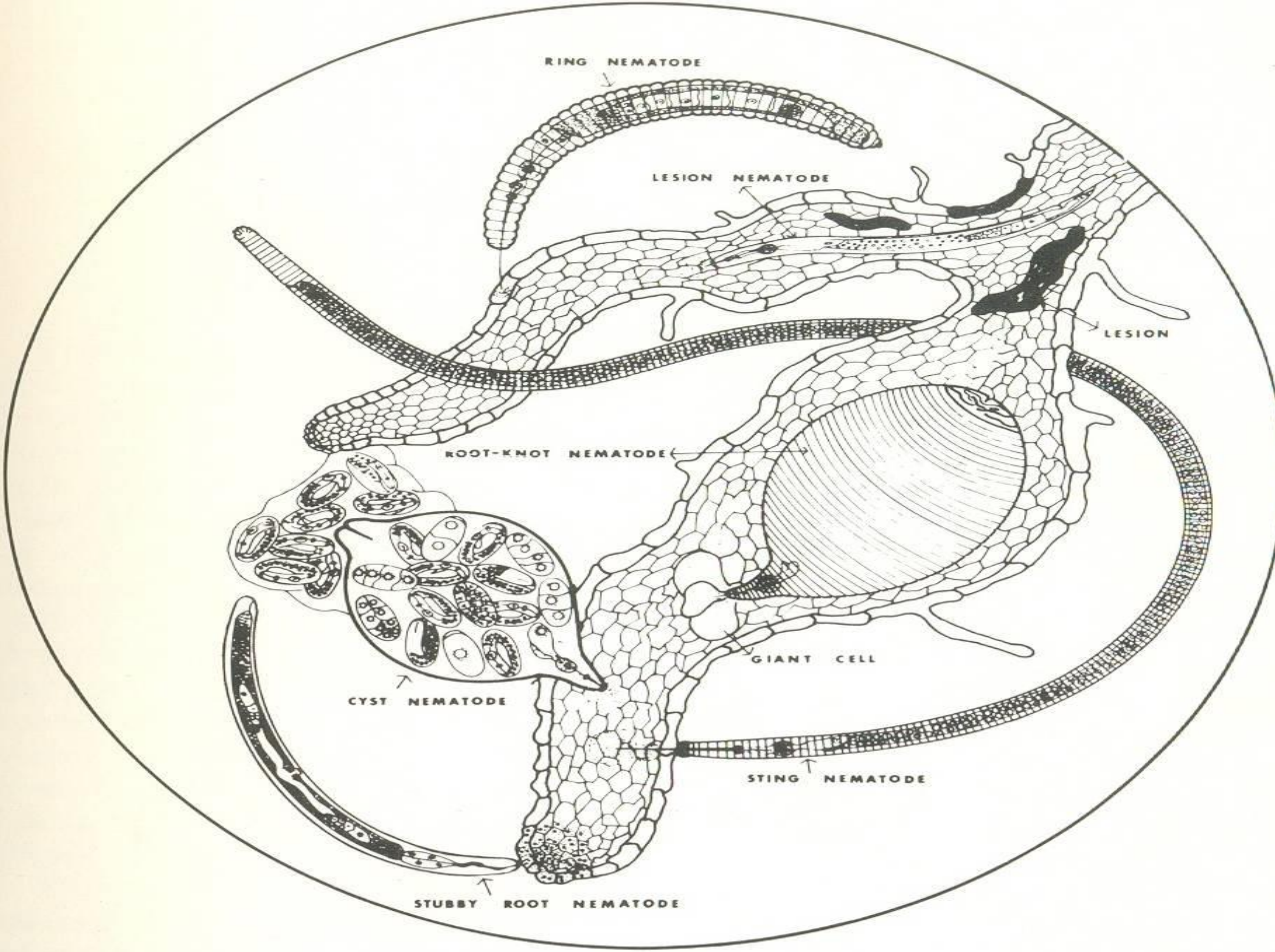
  some kinds enter roots (bulbs, and other below-ground plant parts) and feed internally

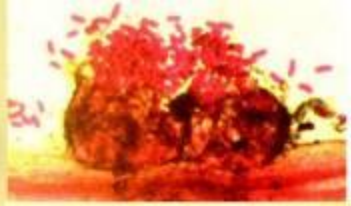
- **Semi-endoparasites**

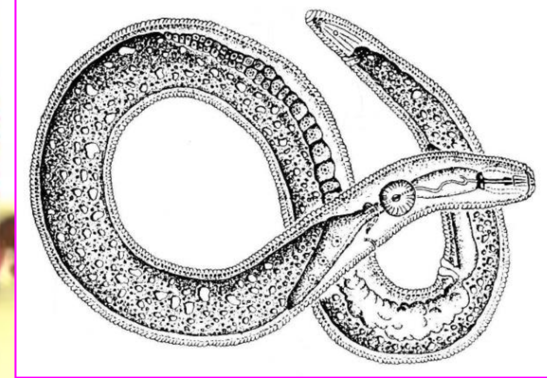
  partially enter into roots

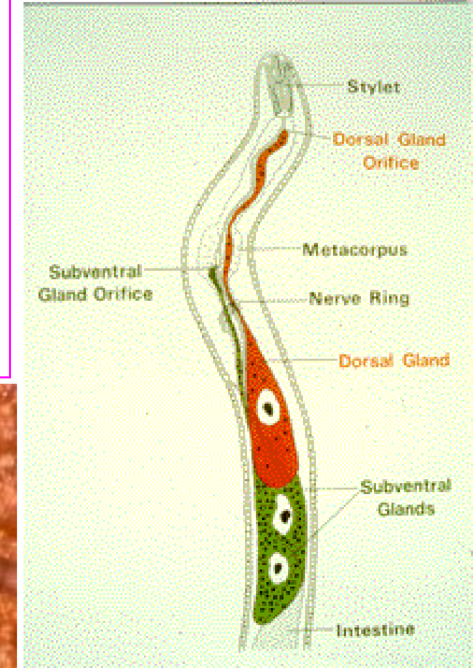
- **Foliar nematodes**

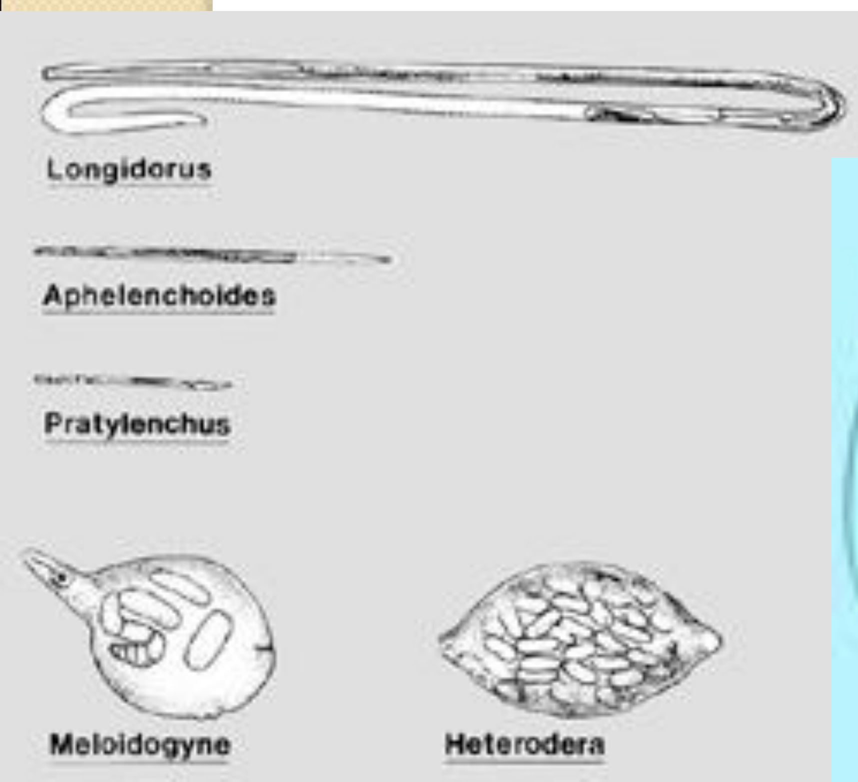
  a few kinds enter above-ground plant tissue (leaves, seeds, stems)

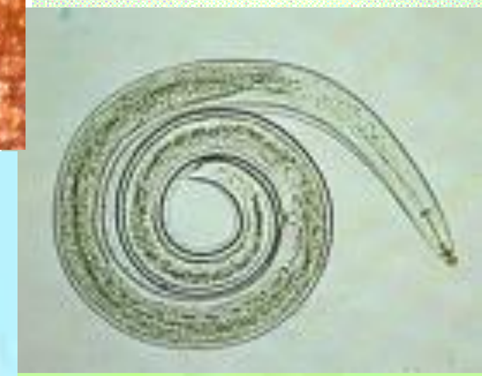

RING NEMATODE
LESION NEMATODE
LESION
ROOT-KNOT NEMATODE
GIANT CELL
CYST NEMATODE
STING NEMATODE
STUBBY ROOT NEMATODE






Stylet
Dorsal Gland Orifice
Metacorpus
Subventral Gland Orifice
Nerve Ring
Dorsal Gland
Subventral Glands
Intestine


Longidorus
Aphelenchoides
Pratylenchus
Meloidogyne
Heterodera



Summary of estimated annual yield losses due to damage by plant-parasitic nematodes worldwide*

Life-sustaining Crops**	Loss (%)	Economically important crops	Loss (%)
Banana	19.7	Cacao	10.5
Barley	6.3	Citrus	14.2
Cassava	8.4	Coffee	15.0
Chickpea	13.7	Cotton	10.7
Coconut	17.1	Cowpea	15.1
Corn	10.2	Eggplant	16.9
Field bean	10.9	Forages	8.2
Millet	11.8	Grape	12.5
Oat	4.2	Guava	10.8
Peanut	12.0	Melons	13.8
Pigeon pea	13.2	Misc. other***	17.3
Potato	12.2	Okra	20.4
Rice	10.0	Ornamentals	11.1
Rye	3.3	Papaya	15.1
Sorghum	6.9	Pepper	12.2
Soybean	10.6	Pineapple	14.9
Sugar beet	10.9	Tea	8.2
Sugar cane	15.3	Tobacco	14.7
Sweet potato	10.2	Tomato	20.6
Wheat	7.0	Yam	17.7
Average	10.7%	Average	14.0%
	Overall average 12.3%		

*Information based on worldwide survey of nematologists (371 resposes).
**Crops that stand between man and stravation, according to Dr. S. H. Wittwer to Michigan State University.
***Additional miscellaneous crops of economic importance, especially for food or exports.

First report on Interaction

- Atkinson (1892) was the first to report nematode fungal interaction on cotton. He reported that infection of root-knot nematode (*Meloidogyne* spp.) increased the severity of Fusarium wilt on cotton.
- Although large number of studies have been carried out on interactions involving different nematodes, fungi, bacteria and viruses but it appears impossible to estimate the number of such complexes that are likely to occur under natural condition.

Early observations on nematode- bacterial interactions

- **Hunger (1901) was first to observe nematode-bacterium interaction. He observed that tomato plants were readily infected with *Pseudomonas solanacearum* in nematode infested soil but remained healthy in nematode free soil.**
- **Johnson and Powell (1969) showed that root-knot nematodes acts as modifiers of infested tissue in such a way that the infested tissue and surrounding cells become more suitable for bacterial colonization.**
- **Field resistance in potato to *P. solanaceraum* was broken down when the plants were infected with *M. incognita acrita* (Jatala and Martin, 1977).**
- **Yellow ear-rot or 'tunda' disease of wheat is example of an obligatory relationships requiring both the nematode *Anguina tritici* and bacterium *Clivibacter tritici* (*Corynebacterium michiganense* pv. *tritici*)for expression of the complex syndrome (Gupta and Swarup, 1972).**

- Plant Parasitic nematodes have evolved clever strategies to live inside their hosts for a long time. Nematode secretions are in fact complex mixtures of hundreds of different proteins. Infection by nematode can alter host response to subsequent infection by other pathogens or other microorganisms. ***Interactions of nematode with pathogenic bacteria may generally lead to increase in disease severity while interactions of nematodes with plant growth promoting rhizobacteria reduce disease severity by reducing nematode population leading to disease control.***

Nematode-bacterial disease interactions

- Plant parasitic nematodes as primary pathogens favor the establishment of secondary pathogens like bacteria which otherwise can not infect the plant under normal conditions.
- Primary pathogens induce changes in a host, leading to synergistic association for disease development, whereas secondary pathogens participate actively and alter the course of pathogenesis.
- It is now established that two or possibly more pathogens rather than only one is required to cause some diseases.
- It is important to consider the role of primary participating pathogen, its interrelationship with secondary pathogen and their ultimate effects upon host plant.

General characteristics of vascular bacterial wilt diseases

- The typical wilt inducing bacterial diseases are restricted to plants where the functional conductive elements for transporting water are the vessels.
- These bacteria also have the ability to grow and reproduce in xylem vessels in the dilute sap of the transportation stream. Once inside the plant the bacteria live and reproduce in the intercellular spaces.
- The bacteria may also secrete enzymes that break down the middle lamella resulting in the maceration of tissues accompanied by cell death.
- When the bacteria invade vessels they reproduce, spread and carry on all their metabolic activities.
- The mode of infection of *Agrobacterium* however, differs from those of other pathogenic soil bacteria. Instead of causing growth reduction or destruction of cellular tissues, the bacteria stimulate cell division (hyperplasia) and cell enlargement (hypertrophy) in the affected tissues.

Physiological changes of plants infected with vascular wilt bacterial pathogens

- The vascular wilt bacteria produce extra-cellular enzymes during the infection process and these substances are transported apically through vessels and water.

- Plants infected with *Pseudomonas solanecearum* are able to produce higher molecular weight of polysaccharides (Husain and Kelman, 1958). These polysaccharides appear to be retained in xylem vessels and increase the resistance of the stem to flow of water. Endo-polygalacturonase activity and cellulose activity of tomato plants infected with *P. solanacearum* was also high (Kelman, 1953).

- Pegg and Sequeira (1968) reported on increase in phenylalanine, trytophan, tyrosine, dihydroxy-phenylalanine and aromatic acids in tobacco plants inoculated with *P. solanacearum*. Formation of tyloses reduces the flow of sap through the plants and ultimately reduces the water economy of plants and expressed as wilt disease.

Plant disease complexes involving nematodes can be grouped into

- Obligatory relationship
- Fortuitous relationship

- In obligatory relationship one member is completely dependent on another or directly influenced by it. Therefore, expression of plant disease symptoms occurs only when both nematode and bacterium are present together.

- In a fortuitous relationship each member acts independently and is not directly influenced by the other member. In this type of relationship the presence of nematode is not required for the expression of disease symptoms but they enhance the incidence and severity of the disease.

The possible roles played by the nematode in the interactions with bacterial pathogens are

- Nematodes act as predisposing agent by providing wounds for the entry of bacterial pathogens.
- Nematode increases susceptibility by modification of host tissue.
- Nematodes break resistance.
- Nematodes act as vector.
- Nematode root infection increases foliar bacterial diseases
- Nematodes infection changes the rhizosphere microflora

Nematode – *Agrobacterium* sp. interactions

Bacterium	Nematode	Host	Role of nematode in bacterial disease	Reference
Agrobacterium				
A. rhizogenes	***Pratylenchus vulnus***	**Rose**	**The bacteria may penetrate roots through injuries caused by *P. vulnus***	**Munnecke *et al.* (1963)**
A. tumefaciens	***Meloidogyne javanica***	**Peach**	**Nematodes increase crown gall incidence**	**Nigh (1966)**
A. tumefaciens	***M. hapla***	**Raspberry**	**Crown gall infections occurred only in presence of *M. hapla* in two of the three cultivars evaluated**	**Griffin *et al.* (1968)**
A. tumefaciens	***M. javanica***	**Almond**	**Increased crown gall incidence was observed in the presence of nematodes**	**Orion and Zutra (1971)**
A. tumefaciens	***Pratylenchus penetrans***	**Raspberry**	**Galled root supported high nematode population**	**McElroy (1977)**
A. tumefaciens	***Rotylenchulus reniformis***	**Grapevine**	**Speculated that nematode facilitate field infection by bacterium**	**Lele *et al.* (1978)**
A. radiobacter var. tumefaciens	***M. incognita***	**Cotton**	**Disease severity was increased by presence of nematodes**	**Zutra and Orion (1982)**
A. tumefaciens	***P. penetrans***	**Raspberry**	**Number of galls in both susceptible and resistant cultivar increased with the increase in nematode inoculum**	**Vrain and Copeman (1987)**
A. tumefaciens	***M. incognita***	**Tomato**	**Bacterium stimulated development and reproduction of *M. incognita* when applied to the opposite split root**	**El-Sherif and Elwakil (1991)**
A. vitis	***M. hapla***	**Grapevine**	**Combined inoculation of roots with *M. hapla* and *A. vitis* resulted in an increased level of root infestation**	**Sule and Lehoczky (1993)**
A. tumefaciens	***Meloidogyne* spp.**	***Prunus* spp.**	**Root-knot nematode and *A. tumefaciens* galls were numerous and homogenous under high inoculum pressure**	**Rubio-Cabetas *et al.* (2001)**

Nematode – *Clavibacter* sp. interactions

***Clavibacter* (*Corynebacterium*)0**				
C. michiganense* subsp. *insidiosum	***Ditylenchus dipsaci***	**Alfalfa**	**Bacterium was transmitted to alfalfa by nematodes.**	**Hawn (1963,1965)**
C. insidiosum	***M. hapla***	**Alfalfa**	**Observed relationship between incidence of bacterial wilt and *M. hapla*.**	**Hunt *et al.* (1971)**
C. tritici	***Anguina tritici***	**Wheat**	**Nematode is essential as vector of bacterium for yellow rot disease.**	**Gupta and Swarup (1972)**
C. michiganense	***M. incognita***	**Tomato**	***M. incognita* increased bacterial canker on tomato.**	**De Moura *et al.* (1975)**
C. rathayi	***Anguina sp.***	**Ryegrass**	**Toxin produced in the plant tissues in response to presence of bacterium.**	**Stynes *et al.* (1979)**
C. fascians	***Aphelenchoides fragariae***	**Strawberry**	**Demonstrated that nematode and *C. fascians* are necessary to produce “cauliflower” disease of straberry.**	**Crosse and Pitcher (1952); Pitcher and Crosse (1958)**
***Clavibacter* sp.**	***Anguina funesta and A. tritici***	**Wheat**	***Clavibacter* sp. adhered to both *Anguina funesta* and *A. tritici*, but differences in the nature of adhesion were noted**	**McClure and Spiegel (1991)**

Nematode- *Pseudomonas* / *Ralstonia* interactions

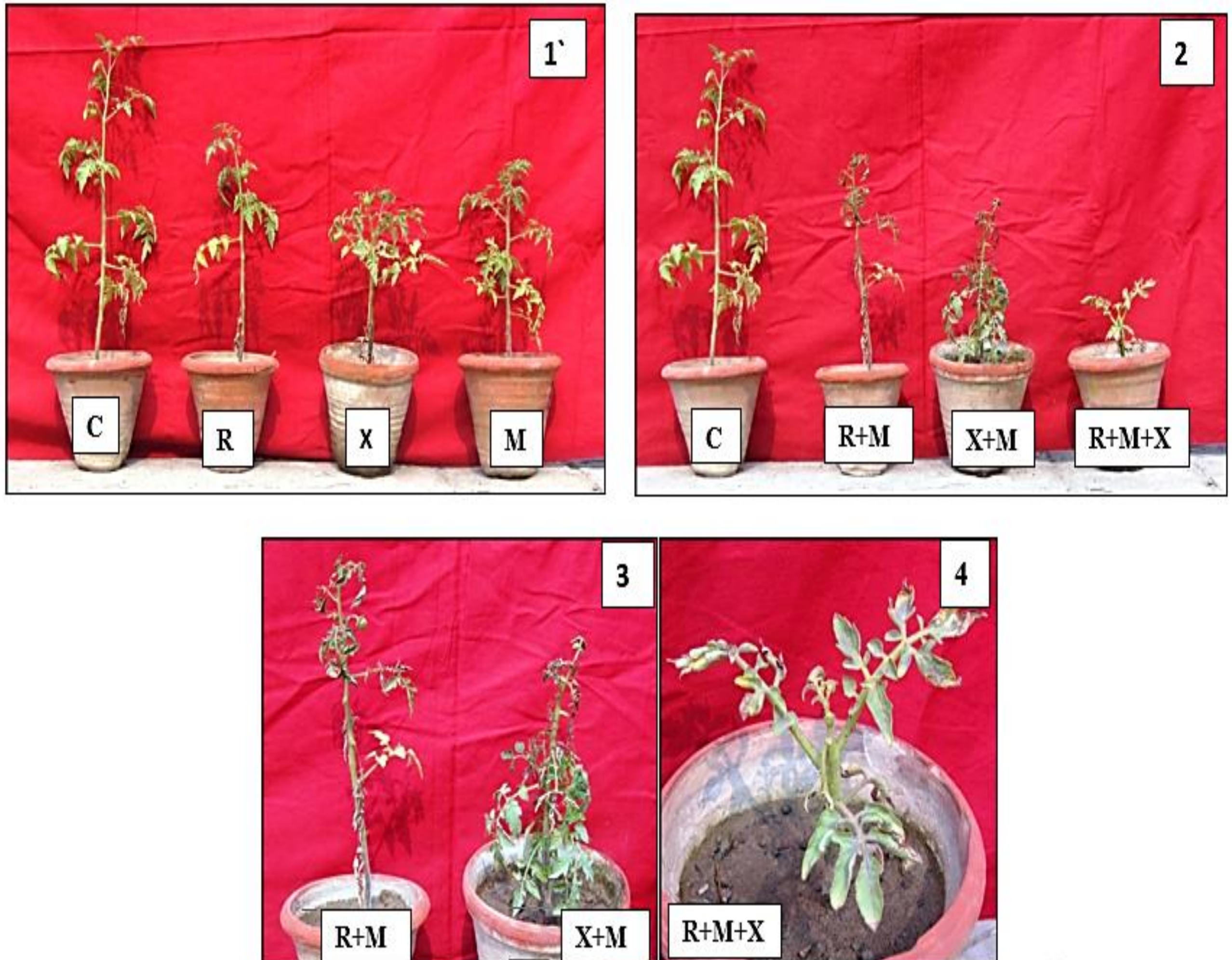
Pseudomonas				
P. solanacearum	***M. incognita acrita***	**Tobacco**	**Wounding of roots by nematodes larvae facilitate infection of bacterium.**	**Lucas *et al.* (1955)**
P. caryophylli	***Helicotylenchus nannus*** ***Meloidogyne* spp.**	**Carnation**	**Wounding the roots by nematodes facilitate entry of *Pseudomonas* into the roots.**	**Stewart and Schindler (1956)**
P. solanacearum	***H. nannus*** ***M. hapla***	**Tomato**	**Simulated by substituting mechanical injury for nematode feeding.**	**Libman *et al.* (1964)**
P. marginata	***M. javanica***	**Gladiolus**	**Nematodes increased severity of gladiolus scab.**	**El-Goorani *et al.* (1974)**
P. syringae	***Criconemoides xenoplax***	**Plum**	**More extensive canker developed on trees infected with nematodes.**	**Mojtahedi *et al.* (1975)**
P. solanacearum	***M. incognita acrita***	**Potato**	**Nematodes kill roots and promote emergence of pathogenic bacteria.**	**Jatala and Martin (1977a, b)**
P. solanacearum	***M. incognita***	**Eggplant**	**More number of plants wilted (40%) when nematodes and bacterium were inoculated simultaneously.**	**Reddy *et al.* (1979)**
P. solanacearum	***M. incognita***	**Tomato**	**Caused synergistic effect on wilt symptoms.**	**Napiere and Quinio (1980)**
P. marginalis ***P. viridiflava*** ***P. corrugata***	***Helicotyl. dihystera*** ***M. hapla*** ***P. penetrans*** ***D. dipsaci***	**Alfalfa**	**Nematodes interacted with three pseudomonads to produce greater growth reductions than obtained with single pathogen.**	**Bookbinder *et al.* (1982)**
P. fluorescens	***Ditylenchus dipsaci***	**Garlic**	**Mixed inoculation showed that *P. fluorescens* penetrated the plant tissues better when *D. dipsaci* was present.**	**Caubel and Smason (1984)**
***P. solanacearum* biotype 3**	***M. javanica***	**Eggplant**	**The combined effects of bacterium and nematode on eggplant were greater than independent effects of either.**	**Sitaramaiah and Sinha (1984a, b)**
P. solanacearum	***M. incognita* Race 1**	**Tomato**	**Nematode infection increased the severity of the vascular wilt disease.**	**Chindo *et al.* (1991)**

Nematode- *Pseudomonas* / *Ralstonia* interactions

P. mindocina	***M. incognita***	**Tomato**	**Bacterium inhibited nematode multiplication.**	**Siddiqui and Husain (1991)**
Ralstonia solanacearum	***M. incognita*** ***R. reniformis***	**Tomato**	**At high temperature *M. incognita* greatly increased wilt severity in susceptible and resistant cultivars but the *R. reniformis* had no such effect.**	**Deberdt *et al.* (1999)**
P. solanacearum	***M. incognita***	**Banana**	**Presence of nematode population 10 days prior to bacterium accelerated the wilt disease development to the maximum .**	**Pathak *et al.* (1999)**
R. solanacearum	**Nematodes**	**Potato**	**Disease severity increases if bacterium is found in association with root nematodes.**	**Stansbury *et al.* (2001)**
R. solanacearum	***Meloidogyne* spp.**	**Potato**	**Controlling *Meloidogyne* spp. did not decrease bacterial infection incidence .**	**Charchar *et al.* (2007)**
R. solanacearum	***M. hapla***	**Alfalfa**	**Bacterium and nematode together result in increased incidence and disease severity.**	**Partridge (2008)**
R. solanacearum	***M. incognita***	**Coleus and Withania**	**Simultaneous inoculation of both pathogens with *Fusarium* exhibited more early disease symptoms.**	**Mallesh *et al.* (2009)**
R. solanacearum	***M. incognita***	**Eggplant**	**In the presence of bacterium, nematode activities including population build up in soil and in roots were reduced.**	**Hussain and Bora (2009)**
R. solanacearum ***X. campestris***	***M. javanica***	**Tomato**	**Inoculation of *M. javanica* prior to *R. solanacearum* plus *X. campestris* caused the highest reduction in plant growth than other treatments except when all the three pathogens were inoculated together.**	**Singh and Siddiqui (2012)**
R. solanacearum	***M. incognita***	**Potato**	**Inoculation of *M. incognita* plus *R. solanacearum* caused a greater reduction in plant growth than the damage caused by either pathogen.**	**Siddiqui *et al.* (2014)**

Nematode - *Pactobacterium* and *Xanthomonas* interactions

Xanthomonas				
X. campestris	***M. incognita***	**Chickpea**	**Inoculation of both pathogens in combination caused a greater reduction in plant growth than the damage caused by each of them singly.**	**Alam and Siddiqui 2013**
Pectobacterium				
P. carotovorum	***M. incognita***	**Potato**	**Inoculation of *P. carotovorum* prior to *M. incognita* caused lesser reduction in plant growth than simultaneous inoculation of both pathogens.**	**Siddiqui *et al.* (2014)**
P. carotovorum ***X. campestris***	***M. javanica***	**Carrot**	**Inoculation of *M. javanica* 15 days prior to either bacterial pathogen caused a higher reduction in plant dry weight compared to inoculation of a bacterial pathogen prior to inoculation with nematodes.**	**Nesha and Siddiqui (2013)**



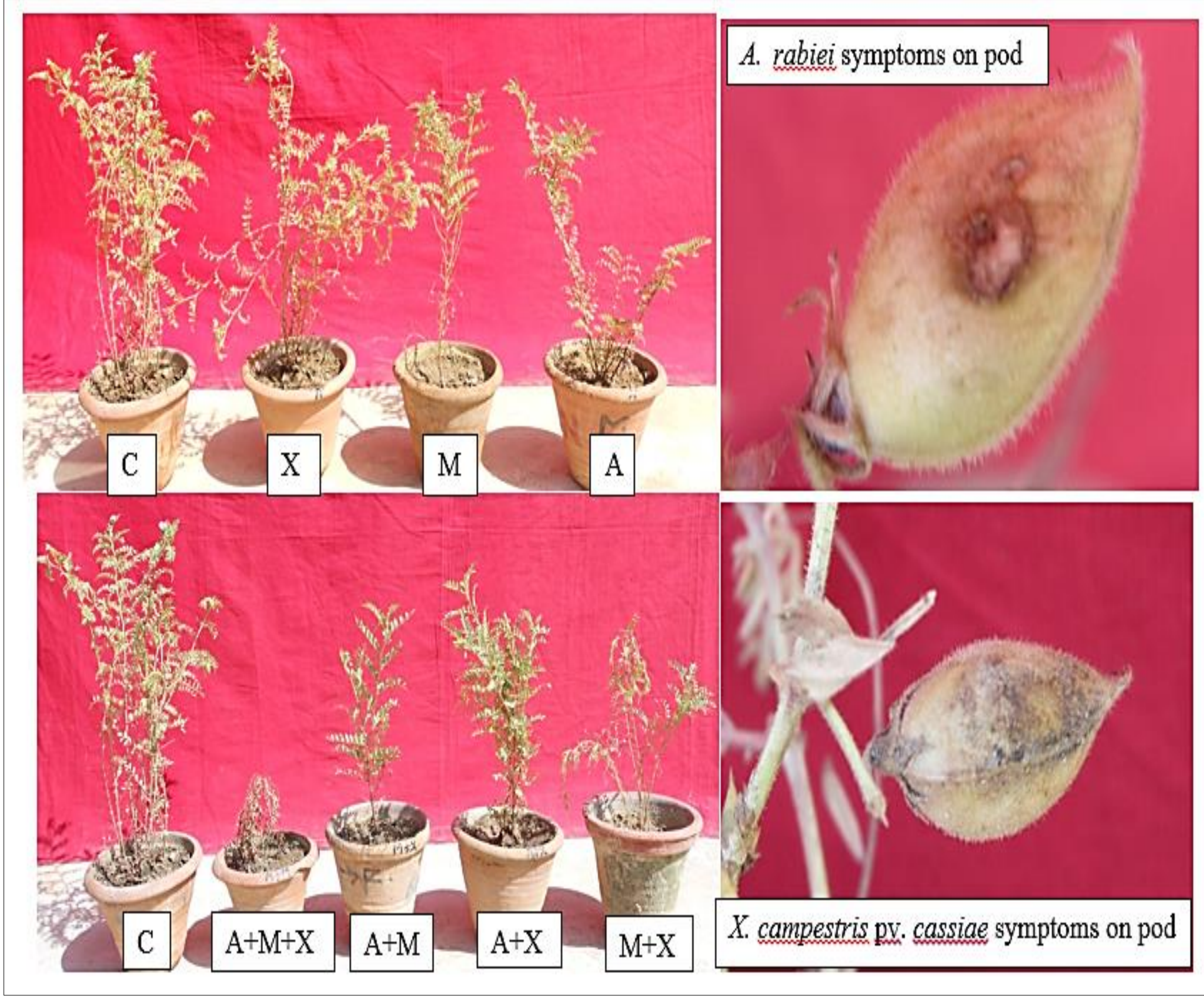
C=Control, R= *Ralstonia solnacearum*, X= *Xanthomonas campestris* pv. *vesicatoria*, M=*Meloidogyne javanica*

3. Symptoms on R+M and X+M inoculated plants 4. Symptoms on R+M+X inoculated plants

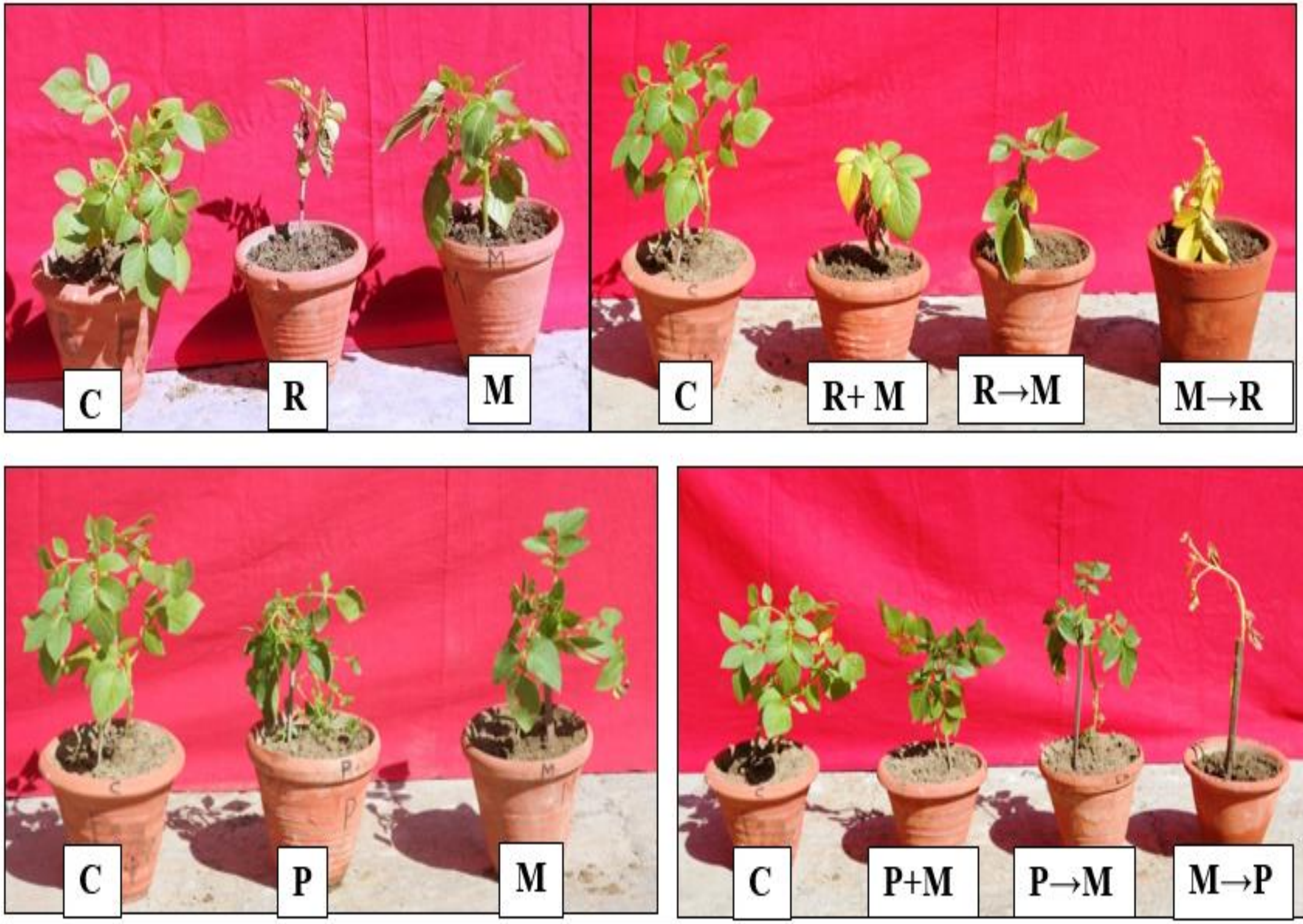
Singh N. and Siddiqui Z.A. (2012). Inoculation of Tomato with *Ralstonia solanacearum, Xanthomonas campestris*, and *Meloidogyne javanica. Intern. J. Veg. Sci.* 18: *78–86.*

Control (C); *Meloidogyne javanica* (M); *Pectobacterium carotovorum* pv. *carotovorum* (P); *Xanthomonas campestris* pv. *carotae* (X)

Nesha R. and Siddiqui Z.A. (2013). Interactions of *Pectobacterium carotovorum* pv. *carotovorum*, *Xanthomonas campestris* pv. *carotae* and *Meloidogyne javanica* on the Disease Complex of Carrot. *Intern. J. Veg. Sci.* 19(4):403-411.



Alam S. and **Siddiqui, Z.A.**,(2013). Interactions of *Meloidogyne incognita*, *Ascochyta rabiei*, *Xanthomonas campestris* pv. *cassiae* and *Rhizobium* sp. on the disease complex of chickpea. *Acta Phytopath. et Entom. Hung.*48:227-236.



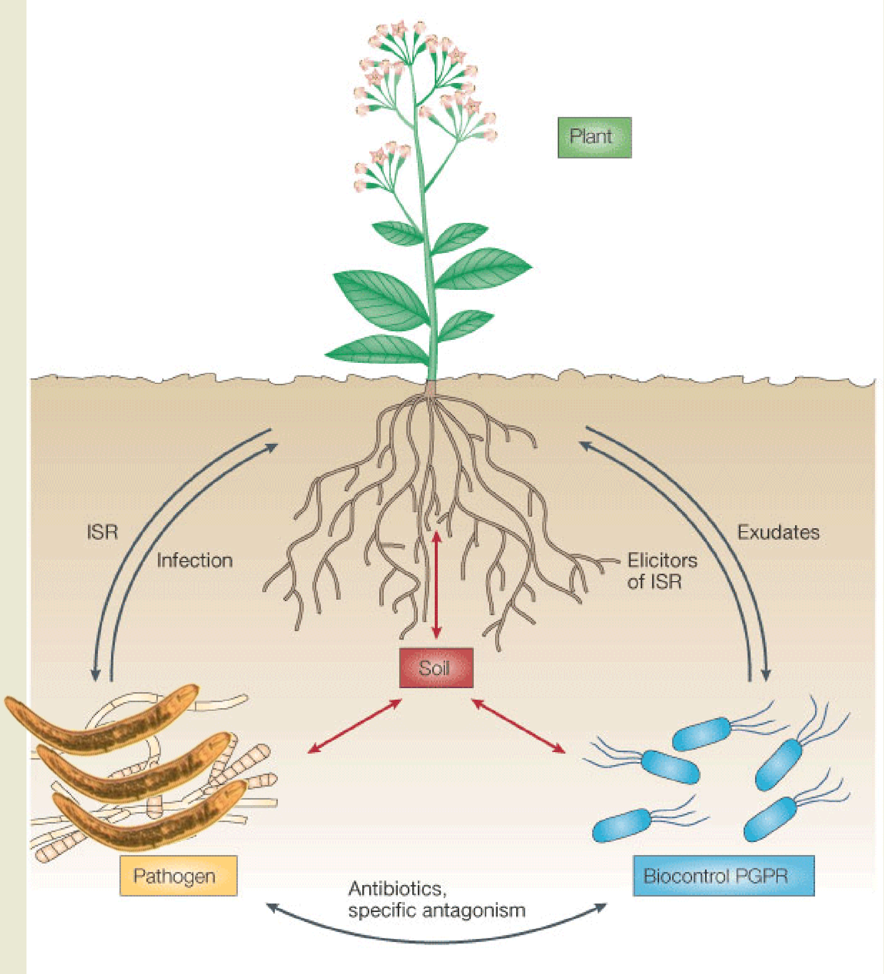
C=Control; M= *Meloidogyne incognita*; R=*Ralstonia solanacearum;* P=*Pectobacterium carotovorum*

+= Simultaneous inoculation; →=Second inoculation 15 days after first inoculation

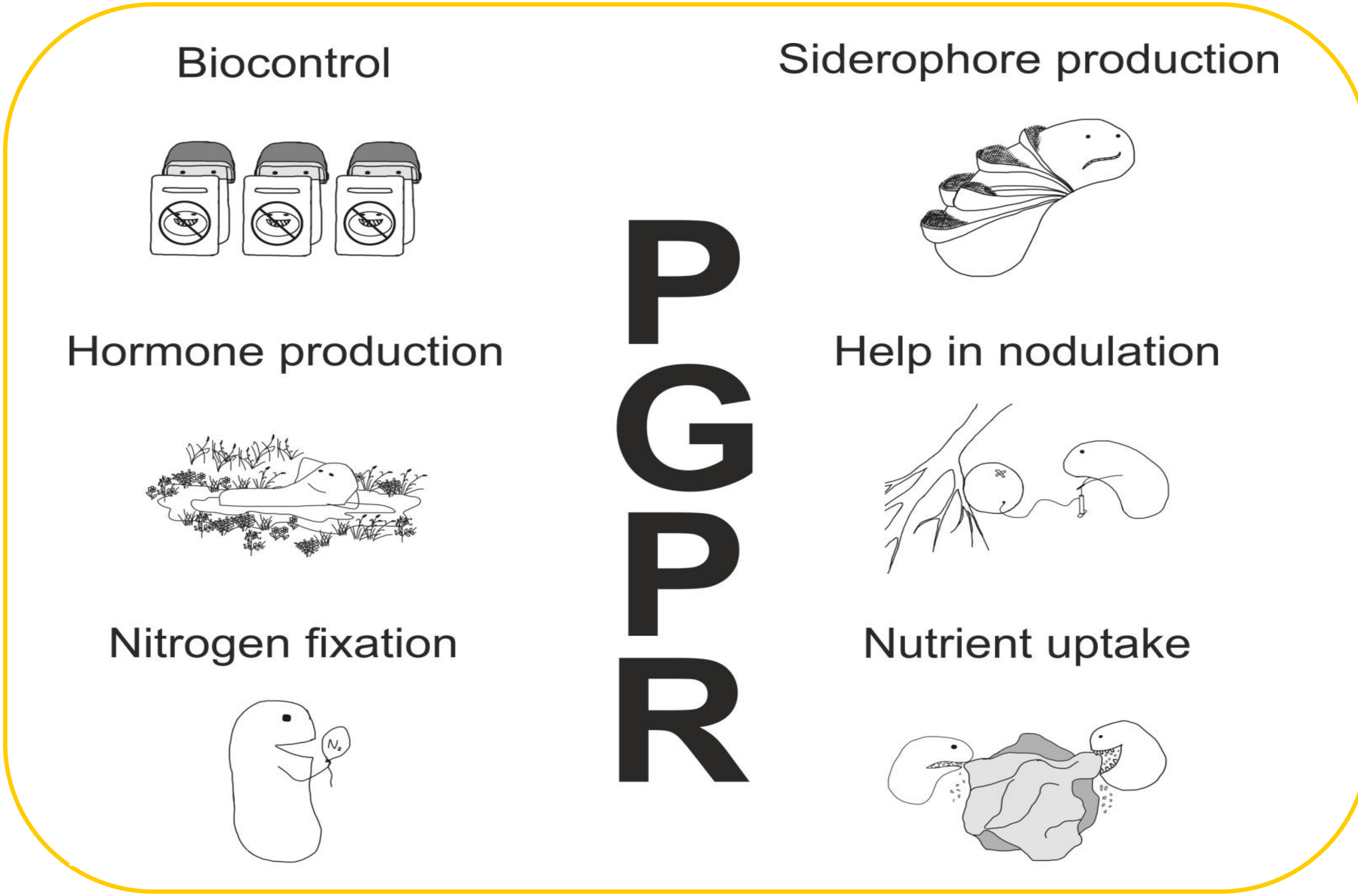
Siddiqui, Z.A., Shehzad M. and Alam S. (2014). Interactions of *Ralstonia solanacearum* and *Pectobacterium carotovorum* with *Meloidogyne incognita* on potato. *Arch. Phytopathol. Plant*

Plant Growth Promoting Rhizobacteria

Some bacteria are associated with roots of crop plants and exert beneficial effects on their hosts and are referred to as ***Plant Growth Promoting Rhizobacteria (PGPR)*** *(Kloepper and Schroth, 1978). The rhizosphere is subject to dramatic changes and the dynamic nature of the rhizosphere creates interactions other soil microorganisms.*



Interaction of PGPR-Nematode-Host Plant


Biocontrol
Siderophore production
Hormone production
Help in nodulation
Nitrogen fixation
Nutrient uptake
PGPR
N_2

Well known genera of PGPR

- *Alcaligenes*
- *Arthrobacter*
- *Azospirillum*
- *Azotobacter*
- *Bacillus*
- *Burkholderia*
- *Enterobacter*
- *Klebsiella*
- *Pseudomonas*
- *Serratia*

Plant Growth Promoting Rhizobacteria (PGPR)

- ❖Plant Growth Promoting Rhizobacteria are part of the natural microflora of healthy plants, they may be considered to be important contributors to plant health and general soil suppressiveness.
- ❖Approaches to use bacteria for control of plant diseases caused by nematodes generally involve either application of specific introduced bacterial strains as biological control agents to induce suppressiveness via alterations in the community of rhizosphere microorganisms.

Ninety four isolates of *Bacillus* and *Pseudomonas* spp. were collected from different localities and various crops. These isolates were tested in following studies on for their effects on plant growth and nematode reproduction. Fifteen isolates were found to have potential for the control of nematode diseases. These isolates have less antibiotic sensitivity and greater inhibitory colonization of roots and greater plant growth promoeffect on the hatching and penetration of nematodes, higher tion.

Siddiqui, S., **Siddiqui, Z. A**., and Ahmad, A. (2005) *World J. Micr. Biotechnol*. 21(5):729-732.

Siddiqui Z.A., Sharma B. and Siddiqui , S. (2007) *Acta Phytopath. et Entom. Hung*. 42:25-34.

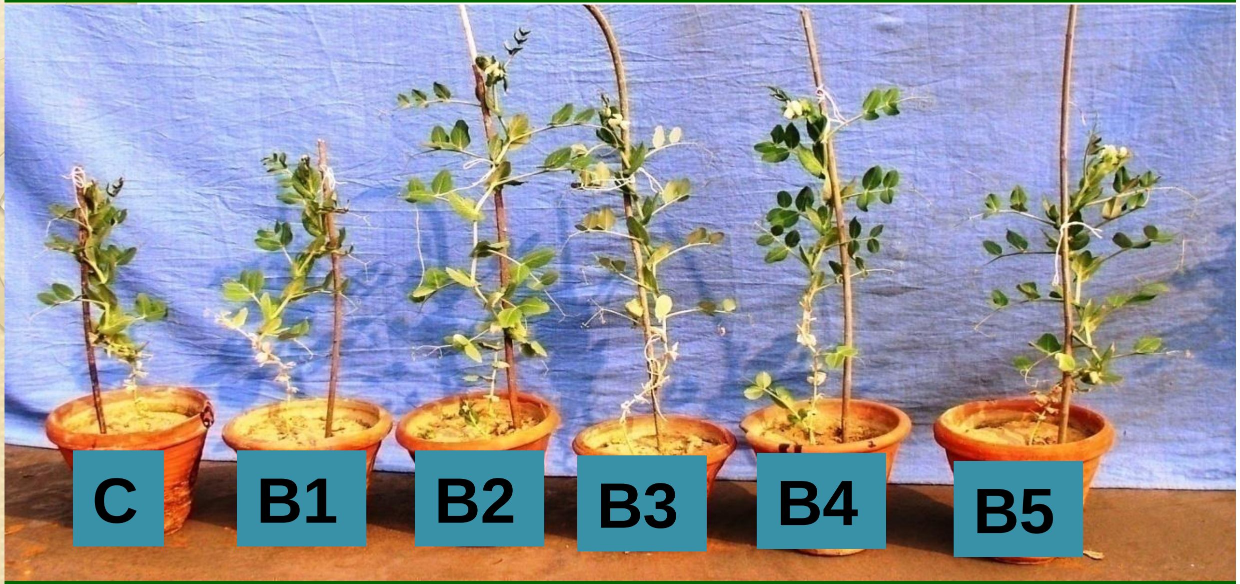
Siddiqui, Z.A. and Shakeel, U. (2007) *J. Plant Pathol*. 89:179-183

Siddiqui, Z.A., Shakeel, U., and Siddiqui, S. (2008) *Acta Phytopath. et Entom. Hung*.43:77-92.

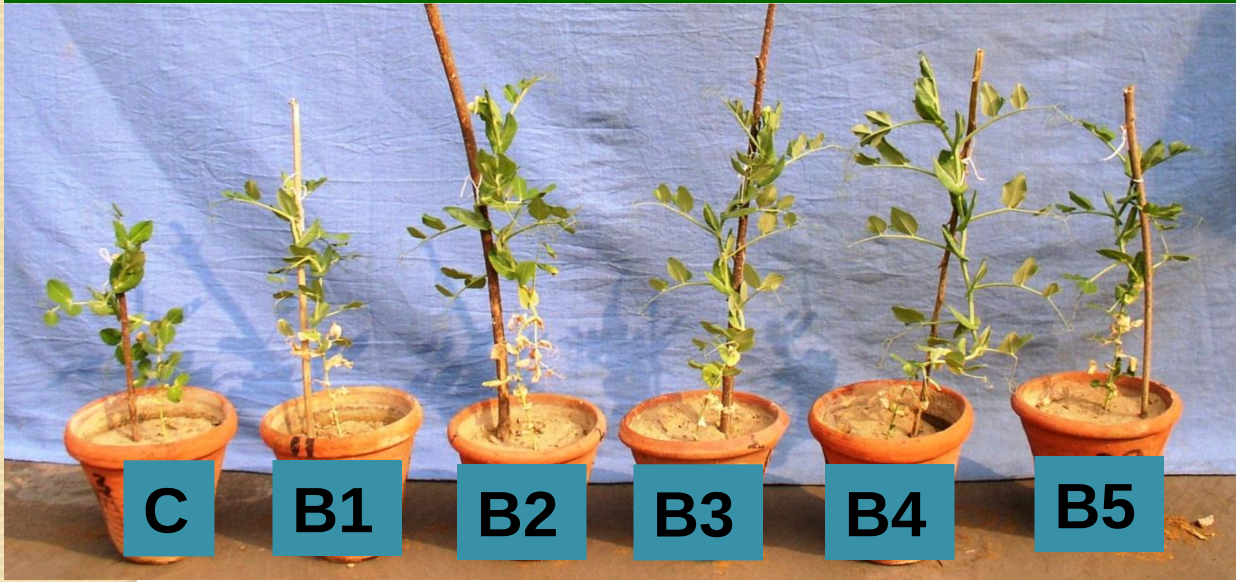
Singh P. and **Siddiqui, Z.A.,** (2010) *Arch. Phytopathol. Plant Protec*. 43:552-561

Singh, P. and **Siddiqui, Z.A.** (2010) *Arch. Phytopathol. Plant Protec*. 43:1423-1434

Effects of *Bacillus* isolates on pea (*Pisum sativum* L.)

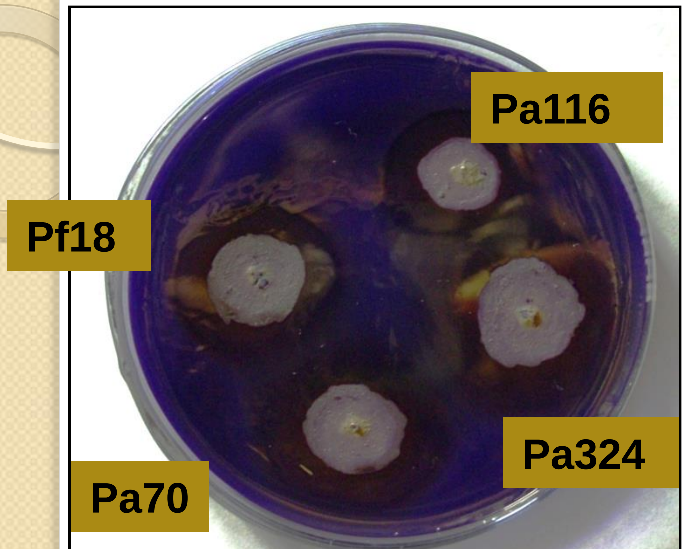


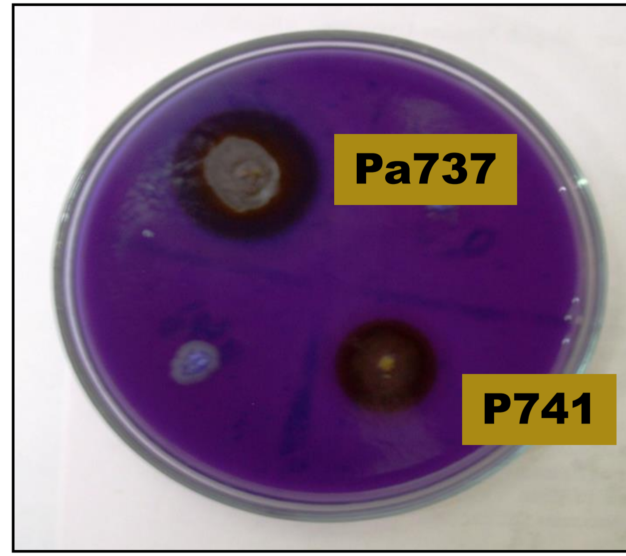
Without root-knot nematode *Meloidogyne incognita*

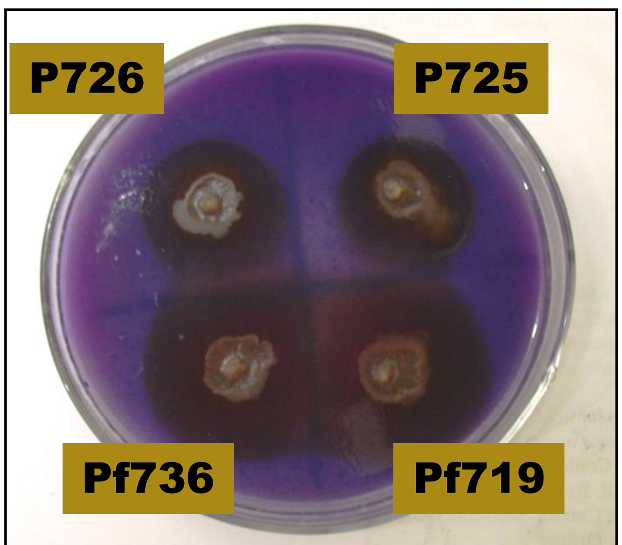


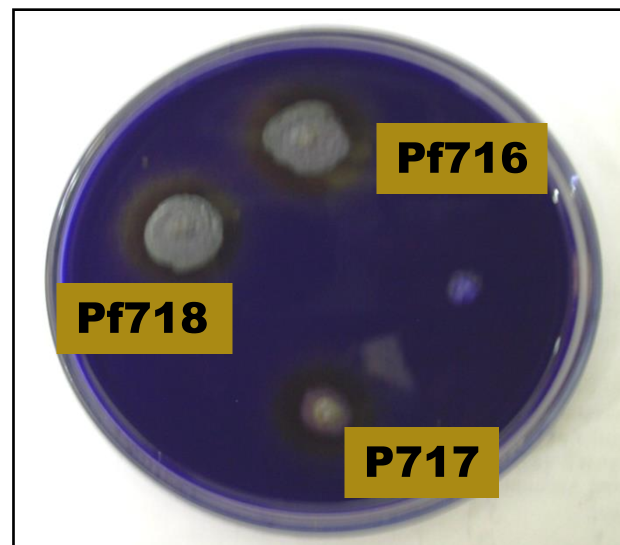
Inoculated with root-knot nematode *Meloidogyne incognita*

Siderophore production by *Pseudomonas* isolates









Use of Plant Growth Promoting Rhizobacteria with other microorganisms to achieve greater Bio-control of Plant Parasitic Nematodes

Combined inoculation of ***Paecilomyces lilacinus*** and ***Bacillus subtilis*** to chickpea plants with *Meloidogyne incognita* caused higher reduction in nematode multiplication and also improved plant growth significantly.

Siddiqui, Z.A. and Mahmood, I. (1993)

Fundam. appl. Nematol. 16: 215-18.

Increase in shoot dry weight and reduction in nematode multiplication and galling was greater when plants inoculated with *M. incognita* were treated with ***Bacillus subtilis*** plus filtrate of ***Aspergillus niger***.

Siddiqui, Z.A. and Mahmood, I. (1995)

Fundam. appl. Nematol. 18: 71-76.

Siddiqui, Z.A. and Mahmood, I. (1998) *Appl. Soil Ecol. 8:* 77 – 84.

The effects of *Pseudomonas fluorescens and Glomus mosseae* on the growth of tomato and on the reproduction of *Meloidogyne javanica* and morphometrics of nematode females were studied on tomato. Applications of *G. mosseae* or *P. fluorescens* caused reduction in galling, nematode reproduction and morphometric parameters of females. **Combined use of both organisms caused a higher reduction in galling, nematode reproduction and morphometrics than their individual application.**.

Siddiqui, Z.A. and Mahmood, I. (2001)*Bioresource Technol.* 79: 41-45

The effects of rhizobacteria, i.e. *Pseudomonas fluorescens*, *Azotobacter chroococcum* and *Azospirillum brasilense*, alone and in combination with *Rhizobium* sp. and *Glomus mosseae*, on the growth of chickpea, and reproduction of *Meloidogyne javanica* were studied. **Combined use of *P. fluorescens* with *G. mosseae* was better at improving plant growth and reducing galling and nematode multiplication than any other combined treatment**.

Siddiqui, Z.A., Iqbal A. and Mahmood I.(2001) Appl. Soil Ecol.16:179-185

Influence of two strains of *Pseudomonas fluorescens* (GRP3 and PRS9), organic manure, and inorganic fertilizers (urea, diammonium phosphate (DAP), muriate of potash and monocalcium phosphate) were assessed alone and in combination on the multiplication of *Meloidogyne incognita* and growth of tomato. ***P. fluorescens*** **GRP3 with organic manure was the best combination for the management of** ***M. incognita*** **on tomato.**

Siddiqui, Z. A. and Mahmood, I. (2003) *Boil. Agri. Horti*. 21:53-62

The influence of *Pseudomonas fluorescens*, *Azotobacter chroococcum*, and straws of *Triticum aestivum*, *Oryza sativa*, *Zea mays*, *Sorghum vulgare* and *Pennisetum typhoides* alone and in combination on the multiplication of *Meloidogyne incognita* and on the growth of tomato. *P. fluorecens* was better at improving tomato growth and reducing galling and nematode multiplication than was *A. chroococcum.* Among straws, *P. typhoides* was better than *Z. mays* followed by *S. vulgare and T. aestivum* in improving tomato growth and reducing galling and nematode multiplication. **Use of *P. fluorescens* with the straw *of P. typhoides* was the best combination for the management of *M. incognita* on tomato.**

Glasshouse experiments were conducted to assess the influence of *Pseudomonas fluorescens*, *Azotobacter chroococcum*, *Azospirillum brasilense* and composted organic fertilizers (cow dung, horse dung, goat dung and poultry manure) alone and in combination on the multiplication of *Meloidogyne incognita* and growth of tomato. **Poultry manure with *P. fluorescens* was the best combination for the management of *M. incognita* on tomato**.

Siddiqui, Z.A. (2004) *Bioresource Technol*. 95:223-227.

***Pseudomonas straita* and *Rhizobium* sp. improve plant growth and reduce galling and nematode reproduction and increase rate of transpiration of pea.** Use of both organisms together had greater adverse effect on galling and nematode multiplication than caused by either of them alone. Highest reduction in galling and nematode multiplication was observed when both organisms were used in 40 % fly ash amended soil.

Siddiqui, Z.A. and Singh, L.P. (2005) *J. Environ*. *Biol*.26:117-122.

Siddiqui, Z.A. and Akhtar M.S. (2007). *J. Plant Pathol*. 89:67-77

The effects on chickpea (*Cicer arietinum*) of the phosphate- solubilizing microorganisms ***Aspergillus awamori*****,** *Pseudomonas aeruginosa* (isolate Pa28) and ***Glomus intraradices*** in terms of growth, and content of chlorophyll, nitrogen, phosphorus and potassium and on the disease of chickpea caused by *Meloidogyne incognita*. *Pseudomonas aeruginosa* reduced galling and nematode multiplication the most followed by *A. awamori* and *G. intraradices*. **Combined inoculation of these microorganisms caused the greatest increase in plant growth and reduced the root-knot disease more than individual inoculations**.

Siddiqui, Z.A., Baghel,G., and Akhtar M.S.(2007) *World J. Micr. Biotechnol.* 23:435-441.

Pseudomonas putida caused greater inhibitory effect on the hatching and penetration of *M. javanica* followed by *P. alcaligenes*, *P. polymyxa* and *B. pumilus*. Inoculation of any PGPR species alone or together with *Rhizobium* increased plant growth both in *M. javanica*-inoculated and -uninoculated plants. Among PGPR, *P. putida* caused greater increase in plant growth and higher reduction in galling and nematode multiplication followed by *P. alcaligenes*, *P. polymyxa* and *B. pumilus*. Combined use of *Rhizobium* with any species of PGPR caused higher reduction in galling and nematode multiplication than their individual inoculation. **Use of *Rhizobium* plus *P. putida* caused maximum reduction in galling and nematode multiplication followed by *Rhizobium* plus *P. alcaligenes*.** *Pseudomonas putida* caused greater root colonization and siderophore production followed by *P. alcaligenes*, *P. polymyxa* and *B. pumilus*.

Akhtar, M.S. and **Siddiqui, Z.A.** (2007) *Austalasian Plant Pathol.* 36: 175-180.

Inoculation of plants with *G. intraradices, P. putida* and *P. polymyxa* alone and in combination significantly increased plant growth, pod number, chlorophyll, nitrogen, phosphorus and potassium contents and reduced galling, nematode multiplication. Inoculation of plants with *P. putida* most effectively reduced galling and nematode multiplication, followed by *G. intraradices* and *P. polymyxa*. **Combined inoculation of plants with *G. intraradices, P. putida* and *P. polymyxa* caused the greatest reduction in galling and nematode multiplication**.

Akhtar, M. S. and **Siddiqui, Z. A.** (2008) *Crop Protec.* 27: 410-417

Combined inoculation of *Glomus intraradices* with *Pseudomonas. straita* plus *Rhizobium* to *M. incognita* inoculated plants caused greater increase in plant growth, number of pods, chlorophyll, nitrogen, phosphorus and potassium contents than by inoculation of *G. intraradices* plus *Rhizobium* or *G. intraradices* plus *P. straita*. Inoculation of *Rhizobium* caused higher reduction in galling and nematode multiplication, followed by *P. straita* and *G. intraradices.* Maximum reduction in galling and nematode multiplication was observed when *G. intraradices* was inoculated with both bacteria. **Biocontrol of root-knot disease of chickpea may be achieved by the combined use of *Rhizobium*, *G. intraradices* and *P. straita* or use of *Rhizobium* plus *P. straita.***

Siddiqui, Z.A. and Akhtar, M. S. (2008) *J. Plant Interact*. 3: (3): 263-271

The effects of *Glomus intraradices* and *Pseudomonas putida* were observed alone and in combination with fertilizers (composted cow manure and urea) on the growth of tomato and on the reproduction of *Meloidogyne incognita*. Root colonization by *G. intraradices* and *P. putida* was increased with composted manure while urea had an adverse effect on root colonization. **The maximum reduction in galling and nematode multiplication was observed when *P. putida* was used with *G. intraradices* together with composted manure.**

Siddiqui, Z.A. and Akhtar, M. S. (2008) *Phytoparasitica* 36(5):460-471

Effects of organic wastes (biosolids, horse manure, sawdust and neem leaf litter [NLL]), *Glomus intraradices*, and *Pseudomonas putida*, were studied on the growth of tomato and on the reproduction of *Meloidogvne incognita.* When *P. putida* and *G. intraradices* were applied together, the increase in tomato growth was greater than when either agent was applied alone. Of the organic wastes, use of sawdust with *P. putida* caused a greater increase in root colonization by fluorescent pseudomonads followed by NLL, horse manure and biosolids. **Combined use of NLL with *P. putida* plus*G. intraradices* was better in reducing galling and nematode multiplication than any other treatment.**

Akhtar, M.S. and Siddiqui, Z.A. (2008) *J. Gen. Plant Pathol*. 74:53-60

The effects of *Glomus intraradices*, *Pseudomonas alcaligenes* and *Bacillus pumilus* on the root-knot disease chickpea caused by *Meloidogyne incognita* was assessed by quantifying differences in the shoot dry mass, pod number, nodulation, and shoot content of chlorophyll, nitrogen, phosphorus and potassium. *P. alcaligenes* caused a greater increase in shoot dry mass, pod number, chlorophyll, nitrogen, phosphorus and potassium in plants with pathogens than did *G. intraradices* or *B. pumilus*. **Combined application of *G. intraradices, P. alcaligenes* and *B. pumilus* to plants inoculated with pathogens caused a greater increase in shoot dry mass, pod number, nitrogen, phosphorus, and potassium than did an application of *P. alcaligenes* plus *B. pumilus* or of *G. intraradices* plus *B. pumilus*.**

Siddiqui, Z.A. and Akhtar, M. S. (2009) *J. Gen. Plant Pathol.* 75:144-153.

Effects of *Aspergillus niger* CA, *Penicillium chrysogenum* CA1,*Burkholderia cepacia* 4684, *Bacillus subtilis* 7612,*Glomus intraradices* KA and *Gigaspora margarita* AA were assessed alone and in combination on the growth of tomato and on the reproduction of *Meloidogyne incognita*. Application of *Bu. cepacia* 4684 caused a 36.1% increase in shoot dry mass of nematode-inoculated plants followed by *Ba. subtilis* 7612 (32.4%), *A. niger* CA (31.7%), *Gl. intraradices* KA (30.9%), *Gi. margarita* AA (29.9%) and *P. chrysogenum* CA1 (28.8%). Use of *Bu. cepacia* 4684 with *A. niger* CA caused a highest (65.7%) increase in shoot dry mass in nematode-inoculated plants followed by *Ba. subtilis* 7612 plus *A. niger* CA (60.9%). ***Burkholderia cepacia* 4684 greatly reduced (39%) galling and nematode multiplication, and the reduction was even greater (73%) when applied with *A. niger* CA**.

Siddiqui, Z.A., Qureshi A. and Akhtar M.S. (2009). *Arch. Phytopathol. Plant Protec.* 42:1154-1164

Biocontrol of *Meloidogyne incognita* was studied on *Pisum sativum* using isolates of fluorescent Pseudomonads (Pf1, Pa2, Pa3, Pa4 and Pf5) and *Bacillus* (B1, B2, B3, B4 and B5). *Pseudomonas* isolates caused greater inhibitory effect on hatching and penetration of *M. incognita* than caused by isolates of *Bacillus*. Among *Pseudomonas*, isolate **Pf1** caused greater inhibitory effect on the hatching and penetration of *M. incognita* followed by Pa3, Pa4, Pa2 and Pf5. Similarly, among ***Bacillus* isolates, B2 caused greater inhibitory effect on the hatching and penetration of *M. incognita* followed by B4, B3, B5 and B1**. **Isolates Pf1 showed greater production of siderophores followed by Pa3 and Pa4.** Similarly, isolate Pf1 produced greater amount of IAA followed by Pa3, Pa4, Pa2 and Pf5. On the other hand, Pf1 and Pa3 showed greater production of HCN followed by Pa4 and Pa2 and Pf5.

Siddiqui, Z.A. and Futai, K. (2009). *Pest Manag. Sci.* 65: 943-948

The effects of antagonistic fungi [*Aspergillus niger* v. Teigh., *Paecilomyces lilacinus* (Thom) Samson and *Penicillium chrysogenum* Thom] and plant-growth-promoting rhizobacteria (PGPR) [*Azotobacter chroococcum* Beijer., *Bacillus subtilis* (Ehrenberg) Cohn and *Pseudomonas putida* (Trev.) Mig.] were assessed with cattle manure on the growth of tomato and on the reproduction of *Meloidogyne incognita* (Kof. & White) Chitwood. The highest increase (79%) in the growth of nematode-inoculated plants was observed when *P. putida* was used with cattle manure. **Application of *P. lilacinus* or *P. putida* with cattle manure was useful for achieving greater biocontrol of *M. incognita* on tomato**.

Siddiqui, Z.A. and Akhtar, M. S. (2009).*Australasian Plant Pathol*. 38: 22-28.

The effects of ***Paecilomyces lilacinus, Pochonia chlamydosporia, Trichoderma harzianum, Bacillus subtilis, Paenibacillus polymyxa*** and ***Burkholderia cepacia***, were studied alone and in combination in glasshouse experiments on the growth of tomato and on the reproduction of *Meloidogyne incognita*. Application of antagonistic fungi and PGPR caused a significant ($P < 0.05$) increase in tomato growth (based on shoot dry weight) both with and without nematodes. *P. lilacinus* was more effective in reducing galling and improving the growth of nematode-inoculated plants than *T. harzianum*, while *P. polymyxa* was more effective than *B. subtilis*. **The greatest increase in growth of nematode-inoculated plants and reduction in nematode galling was observed when *P. polymyxa* was used with *P. lilacinus* or *P. chlamydosporia.***

Siddiqui, Z.A. and Akhtar, M. S. (2009). *Biocon. Sci. Technol*.19:511-521

Effects of ***Pseudomonas putida*** MTCC No. 3604 and ***Pseudomonas alcaligenes*** MTCC No. 493 and parasitic fungi (***Pochonia chlamydosporia*** KIA and ***Paecilomyces lilacinus*** KIA) were studied, alone and together with ***Rhizobium*** sp. on the growth of chickpea and multiplication of *Meloidogyne javanica.* Individually, *P. putida* 3604*, P. alcaligenes* 493 and *Rhizobium* caused a significant increase in the growth of chickpea in both nematode inoculated and uninoculated plants. **Inoculation of *Rhizobium* with a parasitic fungus or with plant growth promoting rhizobaterium caused a greater increase in the growth of plants inoculated with nematodes than caused by either of them singly.** *P. lilacinus* KIA caused a greater reduction in galling and nematode multiplication followed by *P. chlamydosporia* KIA*, P. putida* 3604 and *P. alcaligenes* 493*.* Combined use of *P. lilacinus* KIA with *Rhizobium* was better in reducing galling and nematode multiplication than any other treatment. *P. putida* 3604 caused a greater colonization of root than *P. alcaligenes* 493.

Akhtar, M. S. and **Siddiqui, Z.A.** (2009) *Australasian Plant Pathol.* 38: 44-50.

The effects of *Pseudomonas putida, Pseudomonas alcaligenes* and a *Pseudomonas* isolate (Ps28) on the hatching and penetration of *Meloidogyne incognita* in chickpea (*Cicer arietinum*) roots were studied. Root colonisation and the production of siderophores, hydrogen cyanide (HCN) and indole acetic acid (IAA) were also estimated for each bacterial isolate. ***P. putida*** **had the greatest inhibitory effect on hatching and root penetration of *M. incognita* followed by *P. alcaligenes* and Ps28, respectively.** Similarly, *P. putida* colonised roots more effectively than *P. alcaligenes* or Ps28. In addition, *P. putida* produced the greatest amounts of siderophores, IAA and HCN compared with *P. alcaligenes* and Ps28. *P. putida* caused the greatest reduction in galling and nematode multiplication followed by *P. alcaligenes* and Ps28, respectively. **The present study suggests that *P. putida* has potential for the biocontrol of root-knot disease of chickpea**.

Singh, N. and **Siddiqui, Z.A.** (2015). *Phytoparasitica.43:*61-75*.*

The effects of *Bacillus subtilis*, *Aspergillus awamori* and *Pseudomonas fluorescens* on the wilt–leaf spot disease complex of tomato caused by *Meloidogyne javanica, Ralstonia solanacearum* and *Xanthomonas campestris* pv. *vesicatoria* were observed. Inoculation of *P. fluorescens* caused a greater increase in plant growth and chlorophyll contents of pathogen-inoculated plants than that caused by *A. awamori*. Application of *P. fluorescens* with *B. subtilis* caused a greater increase in plant growth and chlorophyll contents of pathogen-inoculated plants, but the maximum increase was observed when all the three biocontrol agents were inoculated together. Inoculation of *P. fluorescens* caused a greater reduction in galling and nematode reproduction, followed by *B. subtilis* and *A. awamori*. **Maximum reduction in galling, nematode reproduction, wilt and leaf spot disease indices was observed when all three biocontrol agents were used together.**

Mixtures of biocontrol agents with different plant colonization patterns are generally useful for the biocontrol of different plant pathogens via different mechanisms of disease suppression. Moreover, mixtures of biocontrol agents with taxonomically different organisms that require different optimum temperature, pH, and moisture conditions may colonize roots more aggressively, improve plant growth and efficacy of biocontrol. Naturally occurring biocontrol results from mixtures of biocontrol agents rather than from high populations of a single organism. The greater suppression and enhanced consistency against multiple pathogens was observed using strain mixtures of PGPR or use of PGPR with other biocontrol agents.


Thank You!