National Symposium

Nematodes: A Challenge Under Changing Climate and Agricultural Practices

16th - 18th November, 2011

ABSTRACTS

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PREFACE

Plant-parasitic nematodes cause loss of about Rs. 210 crore to Indian agriculture annually. Their populations are more prevalent in the world's warmer regions, and that is what may make them more destructive to Indian agriculture in relation to global warming. Mechanization of agriculture, smaller land holdings and economic strength of Indian farmer have to be kept in mind for formulating strategies to manage the nematodes. The Nematological Society of India (NSI) aims to bring the nematologists and other research scientists on a common platform to deliberate and review new research developments as well as identify new research areas of national and regional interest at this National Nematology Symposium. We need to ascribe different goals to different institutions of the country for an effective nematode management strategy to develop, in view of the available human resource, expertise, geographical location, nematode problems, cropping patterns, etc. Premier research institutions need to concentrate on basic research in varied fields of nematology with greater collaboration with molecular biologists, agricultural chemists, entomologists and plant pathologists so as to understand the nematode-plant, nematode-fungus, nematode-bacteria, nematode-insect and nematode-virus interactions. Revelation of these interactions can provide us valuable leads for effective nematode management as chemical management strategies are expensive and environment damaging.

I, on behalf of the President, NSI and the Executive Committee of the NSI expresses deep sense of gratitude to the Vice Chancellor, KAU and Dr. M.S. Sheela along with her organising team for hosting the Symposium at such a beautiful venue. I personally thank all the participants of this Symposium for giving such a warm response. With respect, I acknowledge the guidance and other inputs received from Dr. H.S. Gaur, Jt. Director Edn. & Dean, PG School, IARI, New Delhi; Dr. R.K. Jain, Project Coordinator, AICRP Nematodes; Dr. A.K. Ganguly, Head, Division of Nematology and Dr. (Mrs.) Sudershan Ganguly, Professor, Division of Nematology for conducting this Symposium. I am extremely grateful to Dr. Anju Kamra, Dr. Pankaj and Dr. Sharad Mohan for their untiring efforts in publishing the, "Abstracts" of the Symposium and managing several other works associated with the Symposium. I am sure this compilation of lead talk and other abstracts will be of good use to the researchers in nematology and allied subjects.

In the end, warm greetings to the delegates on behalf of the NSI Executive, as well as the National and Local Organising Committee. We wish you a pleasant stay and fruitful deliberation at the Symposium.

Date: 7th November 2011
New Delhi

(Anil Sirohi)
General Secretary, NSI

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Lead Talks

Nematode Biodiversity- Future Implications

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Nematodes are amongst the most abundant and diverse group of invertebrate organism belong to animal kingdom. A high proportion of plant and soil nematodes species occur in the subtropical and tropical regions than in the temperate zone yet few large scale and long –term surveys on nematode biodiversity are available for the temperate region but different approaches such as species number and tropic groups, are used to assess diversity and their interactions with plants, plant pathogens, plant viruses and bacteria. As inhabitants of the soil rhizosphere nematodes are involved in energy flux, carbon, mineralization and other nutrient cycles, and as plant parasites, either alone or in synergism with other pathogens, are responsible for plant disease complexes and major crop losses. Nematodes have more generations per crop season at higher temperatures, thereby placing crops in the tropics under much greater pressure than those in temperate regions. This phenomenon is usually aggravated by simultaneous infection by more than one species of plant nematodes. A general description is provided of the morphology, tropic groups, parasitic habits, and interactions of nematodes within the soil and rhizosphere environment. Diversity in the tropics, host response to the various plant- parasitic, as well as relevance to some ecological processes where soil and plant nematodes participate. Any estimate of worldwide nematode species diversity is highly speculative since available figures are so variable. It is generally considered that about 20,000 species have been described till date although there may be as many as 500,000 species in total. A more integrative and complex approach refers to biodiversity as the variability found amongst nematodes from all sources including terrestrial, marine and other aquatic system which they are a part, including diversity within and between species ecosystems. High diversity is often attributed to an elevated level of niche specialization, food being an important driving factor, but functional redundancy does occur. In order to assess nematode diversity, several approaches have been used including identification and classification of individual belonging to a population, nematodes are arranged in community level according to feeding habits rather than by exclusive taxonomic traits, and genomic and Meta-genomic level where DNA samples are isolated from individuals and subjected to PCR, host tissue and soil samples are treated using Meta-genomic approaches. In both approaches DNA samples are sequenced, and than compare using BLAST search against Gene Bank Data to identify related taxa using statistical Algorithms based on sequence similarity.

Nematode Taxonomy: Where it Stands and How to Rejuvenate it?

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There has been tremendous spurge in interest in nematodes by zoologists, plant nematologists, entomologists, pathologists, geneticists, developmental and marine biologists in the recent years. However, the major lacuna has always been the correct identification of the concerned species without which every information may go waste. Classifying and identifying nematodes require vast experience and skills of very high order in addition to thorough knowledge of morphology, biology, host parasite relationships, etc. of the group.

Nematode taxonomy at infancy stage largely relied upon morphological characters as seen with a light microscope that were further supplemented by scanning electron microscopic observations. Later, classifications have been proposed using biological characteristics like infective and/ or resistant stage, number of generations in a year, host parasite relationships etc., particularly in case of heteroderids and anguinids. Use of such characters has increased the predictive value of classification. In 1990"s, two diverse classification schemes were proposed, one by Siddiqi and other by Maggenti *et al.* that showed healthy signs of development of nematode taxonomy. Cytogenetic, biochemical and molecular and several other approaches are also being applied to supplement result of conventional taxonomy and for solving subtle problems in identifying species, races and deducing phylogeny. Computer aided identification tools have been prepared for a number of genera.

Siddiqi, Jairajpuri, Das and Khan during Nineteen seventies laid the sound foundation of Nematode taxonomy in India. Their contributions brought Indian Nematology on the international map and led to all round growth and development of Plant Nematology in India. Indian taxonomists continued to produce not only high quality research papers but also came out with classical books on soil-inhabiting nematodes belonging to orders Tylenchida, Doylaimida and Mononchida which have become an essence in all the Nematology laboratories of the world toady. Recently, biochemical tools have been used to discriminate closely related species/ host races of *Meloidogyne* and *Heterodera*. Database have been prepared for species of *Aphelenchoides* and anguinids occurring in India. However, only around 850 species of plant parasitic nematodes, including very few of aerial plant parts and endo-, semiendo- parasites of roots, have been recorded till date from a vast country like India that occupies an area of over 3.28 million sq. km, and is diverse in relief, land forms, terrain, soil, range of daily and annual temperature, and varying amount of rain fall which in turn supports diverse flora of more than 45,000 plant species. Further, there has been some stagnation in taxonomic research in India in the recent years, so much so that dearth of taxonomists is now being felt. A major factor responsible for scenario is the distraction of plant nematode taxonomists to other groups like rhabditids, cephalobids and diplogasterids or to the field of applied Nematology. Other major factor of distraction is the difficulty being faced by taxonomist in publishing their findings in highly reputed journals due to lack SEM observations for which facilities should be developed in the ICAR institutes and SAUs.

To rejuvenate nematode taxonomy, it is fore most to produce taxonomists and to realize that in this era when emphasis is on non chemical methods of management like crop rotation, quarantine and regulatory measures, use of resistant/ tolerant varieties, biocontrol agents, etc. which are highly specific, taxonomic research is very critical. For this, SAUs and ICAR institutes should develop and strengthen their own taxonomic units and ensure that persons appointed for taxonomic work continue to serve in this field only and frequently interact with colleagues working on other aspects. These laboratories should be enriched with type material and identified slides besides regular exchange of material with other institutes. For catering to the needs of applied nematologists, National Identification Net work may be created which would facilitate identification of species and races, maintain pure culture of specified species/races of important pests *in vitro* and *in vivo,* supply them to other nematological centers, and organize refresher courses. It is heartening to see that National Core Group constituted by ICAR, New Delhi to revise curricula and syllabi of PG programme in disciplines of agriculture has recommended courses on Principles of Taxonomy (2+0) and Classification of Nematodes for students perusing M.Sc. degree programme in Nematology in SAUs. To over come teething problems in teaching these courses due to paucity of expertise available, expert taxonomists may be assigned the responsibility for preparing teaching material and to impart training to needy institutes. Conventional taxonomic methods are likely to continue as a major tool of identification but taxonomists should keep themselves updated with the non-traditional approaches e.g., biochemical, molecular, immunological, and use them wherever feasible. Frequent interactions with applied nematologists will further broaden their vision and make taxonomic studies more interesting and fruitful.

Nematode Barcoding and Computer Applications in Nematology

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There are millions of species on earth. We still do not know most of the species that exist on this planet. Only 5% of nematodes, the most abundant metazoans, have been described. The lack of knowledge on the biodiversity of nematodes on earth is a major constraint in knowing the contribution these little organisms make in different ecosystems and hence to our lives. The traditional system of nematode description and identification, based on morphological and morphometric information, is time consuming, tedious and inadequate to distinguish all the species or subspecific categories. With new DNA based tools it is possible to build a digital list of all the nematode species, that scientist can lay their hands-on, irrespective of life stage(s) encountered, in much less time. Concept of molecular barcode of organisms has come from the Universal Product Code used for commercial commodities. Barcoding is a technique for characterizing species of organisms using a short DNA sequence from a standard and agreed-upon position(s) in the genome. It is based on the idea that every species has its own 'diagnostic' sequences i.e. unique sets of base-pair mutations. These DNA barcodes are useful for identifying clades and evolutionary relationships amongst organisms. in nematode taxonomy most of the efforts are based on the study of rDNA, ITS or mitochondrial DNA. Like certain weaknesses of morphological nematode identification, validity and relevance of molecular approaches and the concepts on which these techniques are based have been subject to variety of criticism.

In the past decade lot of advancements have taken place in the field of Computers, digital communications and image analysis systems. The technology is used for storing, comparing and analyzing DNA sequences of unknown species and in evaluating the evolutionary relationships amongst different organisms. Comparisons of morphological characters are used in ascertaining identity of an individual. Software and hardware are available that identify humans by scanning the iris texture of an eye. Computer systems for fingerprints identification are commonly used in forensic sciences. Common commercial cameras have the feature to identify facial textures such as smile. These give us the opportunity to explore the technology for nematode diagnostics. Few attempts for nematode identification have been made wherein the data of an unknown species is compared with that of known species using certain algorithms.

All the techniques of nematode diagnostics are subject to failure in isolation. The talk will deal v

understanding

Free-Living to the Rescue of the Parasite: The Usefulness of *Caenorhabditis elegans* for Studies on Plant-Parasitic Nematodes

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Plant-parasitic nematodes (PPN) are a group of major agricultural pests. Due to their life underground, their impact on agricultural yield is often underestimated. However, recent estimates reveal a world-wide loss of over $150 billion annually to PPNs. Despite this importance, current PPN control strategies are either inefficient or unsafe. Therefore, there is an urgent need to develop new methods to tackle PPN. The primary bottleneck is that we don't know enough about them: our knowledge of their biology at the mechanistic level – what proteins enable their parasitic life style, what molecules help them identify the suitable host, etc – are poorly understood. Due to their parasitic nature, these worms have not been well suited for genetic analysis, the approach that has helped us understand a great deal about their free-living cousin *Caenorhabditis elegans*. Recent advances in genome sequencing and the application of the reverse genetic tools are set to change the scenario very rapidly in the near future. In this talk, I will review some of these developments and highlight how studies using *C. elegans* hold great promise in significantly enhancing our understanding of PPN biology.

Arabidopsis thaliana as a Model System to Study Plant-Nematode Interactions

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In order to understand a group of organism it is imperative that we study in detail all molecular, functional and developmental aspects of a representative/model organism of the group. The goal of a "Model organism" paradigm is to create a standard organism that can be used to elucidate themes fundamental to the growth and development of a group of organisms. The knowledge gained from a model organism can be used as a standard meter rod to compare and contrast the properties of other members of the group.

Some of the important features of *Arabidopsis* are its small size, short life cycle, very high fecundity and a very small genome size. The plant can be easily cultivated with minimal requirements in laboratories. Large number of plants can be grown in a small space. Under controlled environmental conditions a generation takes 6-8 weeks. The seeds are very hardy and can be stored for a long time without any significant loss in viability.

A combination of these attributes and the fact that (i) large number of mutant population was generated, identified and studied: (ii) a saturated genetic map was developed; (iii) several groups of dedicated workers extensively pursued work on *Arabidopsis*; (iv) availability of complete genomic sequence (v) an easy and reproducible *in planta* transformation system has made *Arabidopsis* a model plant for a variety of molecular studies.

Most genes found in Arabidopsis are common to all flowering plants. The availability of the entire *Arabidopsis* genome sequence and the tools available for functional genomics like molecular markers, ESTs, microarrays, insertional mutagenesis, protein expression and protein-protein interaction analysis will continue to provide useful information about entire complement of genes expressed by a typical flowering plant and will be helpful in thorough analysis of the function of these genes in all plant developmental processes including plant pathogen interaction.

Root-knot nematodes and cyst nematodes are obligate, biotrophic pathogens of several plant species. Nematodes cause dramatic changes in the morphology, physiology and biochemistry of their hosts. *Arabidopsis* has been shown to act as hosts for several nematodes. Arabidopsis-nematode interaction has been exploited to identify a large number of plant genes differentially expressed in giant cells and syncytia, and improved our understanding of genes involved in the control of cell cycles and cytoskeletal changes. The role of several new nematode effector molecules is being elucidated. Using the Arabidopsis as a host plant several hitherto unknown strategies adopted by nematodes to usurp plant development processes have been identified. Molecular characterization of several plant and nematode genes involved in the process has become possible because of the use of Arabidopsis as a model system. The information generated on nematode and Arabidopsis genomics and transcriptomics is likely to bring better insight into the plant nematode process and help us in identication of useful candidate genes to combat the menace of this important group of plant pathogen.

Is Plant Resistance Engineering the Only Answer for Nematode Management

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Agriculture is one of the strongholds of the Indian economy as it is the principal means of livelihood for more than 58% of the population, but because of rising population there is an ongoing need to increase food production. With land resources remaining the same, and weather vagaries beyond human control, one of the practical means of achieving greater yields is to minimize the pest associated losses to the crops. Plant parasitic nematodes, though inconspicuous in size are a major constraint to agriculture production. On an average we loose about 10.7% of the world production of the 20 life sustaining crops to nematodes. In countries like India the loss is predicted at about 14.6% and could as well go as high as 50-80% in some crops. The loss to Indian agriculture is estimated at about Rs. 210 crore annually

Over the years, nematologists have experimented and devised several measures to contain phytophagous nematodes, but the most effective way to contain them is by using high doses of toxic nematicides/insecticides which pose multiple dangers to the environment as well as the user. The use of nematode resistant crops, either alone or within integrated management programs, may be the most effective means by which the use of current pesticides can be reduced or even eliminated, but the constraint is of the availability of natural resistance in crops. Nematode resistant plants avoid the need for long crop rotations between host crops, don't require any new practices to be followed upon by the farmer and are appropriate for India's low cost technology agricultural systems. With greater understanding at molecular level of the plant - nematode interaction, parasitism, survival, genomic information and availability of DNA recombinant and RNAi technologies, we are in position to design resistance against specific or a group of plant parasitic nematodes.

Using molecular tools about nine R-genes against some plant parasitic nematodes have been identified and cloned from a number of crops. Cloned genes offer a prospective for transfer of resistance to other related crop plants which possibly will have suitable intracellular signaling pathways and therefore be competent to build up a resistance response. The potential disadvantages of transgenic R-genes include limitations on the spectrum of activity (a small subset of parasites are recognized) and the likelihood of rapid resistance breaking.

Novel strategies for designing resistance against PPN have been possible because of new knowledge generation in the area of nematode-plant interaction in the past decade. These include anti-invasion and migration strategies, feeding-cell attenuation, and anti-nematode feeding and development strategies. Such strategies can be nematode specific or against broader spectrum of nematodes and offer low or insignificant risk to man and environment besides being long lasting. The promising areas are engineering using genes expressing proteinase inhibitors (e.g. rice cysteine proteinase inhibitor (cystatin) Oc-IΔD86); lectins *(e.g. Galanthus nivalis, Pinellia ternata* and *Lycoris radiata*); toxins (e.g *Bt7*N, *Bt*Den, *Bt*18, *Bt*K73, *Bt*Soto and *Bt7*) and utilizing RNAi technology aimed at silencing of critical nematode processes using host delivery method. Our laboratory has successfully shown reduction in nematode infection to a mximum level of 65% by silencing esophageal gland proteins, though some other labs have attained even higher reduction in nematode infection using variety of genes. Bioengineering root-knot nematode resistant crops by expressing dsRNA that silence nematode parasitism genes has several advantages. In addition to achieving broad-spectrum resistance, this type of resistance has the potential to be more durable. The specificity of RNAi-mediated resistance is based on RNA hybridization rather than receptor-ligand binding interactions characteristic of traditional plant resistance genes, which should make the resistance highly durable. A key advantage of RNAi-mediated resistance is that dsRNA has no inherent translational ability to produce a functional protein, thus raking less biosafety issues.

The last decade has seen a boom in the understanding of nematode plant interactions using the tools of comparative and functional genomics. The molecular understanding of nematode biology and parasitism has opened new avenues for gene silencing and engineering resistance apart from using the natural R genes or toxin molecules. **An 80- fold increase in biotech crop hectares between 1996 and 2009, is an indication of acceptance of biotech or GM crops in the world agriculture.** In view of the said facts the day is not far when we may have a commercial release of genetically modified crop plants resistant to a single species/ genera or more of the plant parasitic nematodes. However, there still a need for increased understanding of the mechanism by which pathogen effectors are recognized and operate in the plant system. Also we need to develop effective nematode responsive promoters which will restrict the expression of resistance to only the feeding tissue or around.

Bacterial Nematode Biological Control Agents: Where we stand?

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Nematodes and bacteria are the two prominent biological components of any agro-ecosystem and have co-existed since ages. While bacteria are numerically most abundant organisms in agricultural soils, nematodes are considered most numerous among metazoans. Occupation of same ecological niches, consequently, has resulted in the development of varied relationships between these two types of organisms. Biological control of plant parasitic nematodes envisages interactions of the kind of parasitism and antibiosis. The parasitic relationship is exemplified by only one group, i. e., members of *Pasteuria*. Antibiosis includes various kinds of rhizobacteria, which are not parasitic to nematodes *per se*.

Pasteuria penetrans is a potent candidate for development as a component of INM despite perceived drawbacks of recognition, epidemiology, host specificity and production. Recognition and host specificity are bane or boon is debatable; however, in ecological perspective these should be construed as merits. *P. penetrans* has several merits of a highly successful bio-control agent. These include – demonstrable field efficacy, high level suppression of nematode populations, very high reproductive rates, long persistence in soil, and no special handling or storage conditions etc. On environmental front, the advantages of this bacterium are – controls nematodes without harming other organisms, natural microbe, no known human exposure risks, no contamination risks to soil or water table, no toxic soil or crop residues, and safe for birds and wildlife. Recent report on the successful *in vitro* cultivation of different strains of *Pasteuria* infecting cyst, lesion, ring and sting nematodes has raised the commercial prospects of this very potent bio-control agent of phytonematodes (www.pasteuriabio.com). *Pasteuria* Bioscience LLC – a Florida-based company has launched the first commercial product – ECONEM that is targeted against sting nematode in turf grass industry (www.pasteuriabio.com). Applications have been filed for patening materials and methods for controlling nematodes with *Pasteuria* spores in seed coating and slow release bio-degradable granules of *P. penetrans*. Nevertheless, till the commercial formulations based on *in vitro* system are available, alternate methods of *in vivo* mass multiplication relevant for semi-commercial scale can be adopted to enhance

Climate Change and Emerging Nematode Problems of Horticultural Crops

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Climate changes, of late, has been affecting the biological world in a significant way. General rise in temperatures has increased nematode problems of crops grown under poly-house or protected conditions. Root-knot nematode population densities in the rhizosphere of capsicum, okra, tomato, brinjal, cucumber, gerkhins, musk melons, gerbera and carnations have gone up from 455 – 724 nematodes/ 100 cc of soil. The average number of eggs per egg-mass of *Meloidogyne incognita* has gone up from 412 – 570.

Further, the frequency of occurrence of root-knot nematode (*M. incognita*) used to be very high 4 – 5 years back. Because of changes in the climate, the frequency of occurrence of *Rotylenchulus reniformis* and *Pratylenchus* sp. have been quite higher. These observations were made by us during the surveys conducted on various horticultural crops grown in the open field and protected conditions for the last 4 years in various agro-climatic regions in southern India.

Keeping in view these emerging problems due to climatic changes, efforts were made to develop integrated approaches for the management of nematodes using various bio-pesticides and botanicals. The results of these investigations will be discussed during the presentation.

its use in nursery bed soils and as seed treatment, and introduce it to the main-field. Cultivation of susceptible nematode host crops continuously should help build-up of *P. penetrans* populations to nematode suppressive levels in due course. Low populations of *P. penetrans* can similarly be amplified by following susceptible cropping sequences.

Plant growth promoting rhizobacteria are a contemporary subject of intensive research. These are double-edged weapons, and therefore, should be advocated with caution for nematode suppression. Proliferation of root system with the application of these bacteria may lead to enhanced nematode populations in the absence of nematoxicity in the bacterial strain. Several strains (*Pseudomomas fluorescens*, *P. putida*, *Azotobacter chroococcum*, *Gluconoacetobacter diazotrophicus* etc.) with nematoxic traits have been identified. PGPR have the advantage of easy mass multiplication and demonstrable results in terms of plant growth improvement in nematode-infested soils. Production of allelochemicals, alteration of root exudates, induced systemic resistance and increased plant tolerance are the major mechanisms of action for nematode suppression.

Fungal Nematode Biocontrol Agents: Where We Stand?

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Plant diseases including nematodes are among the major constraints in the production of food crops and inflict significant losses to global agriculture. Pesticides are widely used to control plant diseases but their application is costly and in some cases, may bring more disadvantages than benefits. Bioinoculants or biocontrol agents are the microorganisms that induce stimulatory effects on pests or pathogens through a variety of mechanisms when applied in an ecosystem. Use of bioinoculants to control plant diseases is an economically viable and ecologically sustainable method of disease management.

Kinds of Fungal Nematode Bioincoulants

A large number of bioinoculants is available among them fungal bioinoculant constitute majority and are widely used in different cropping systems. Important nematode fungal bioinoculants are classified in to four groups that directly attack/ influence the nematode developments include: i. nematode trapping fungi (*Arthrobotrys oligospora, Drechslerella dactyloides, Nematoctonus robustus*), ii. Endoparasitic fungi (*Drechmeria coniospora, Hirsutella rhossiliensis, Nematoctonus pachysporus*), iii. Eggs and female parasitic fungi (*Lecanicillium lecanii, Pochonia chlamydosporia, P. rubescens, Paecilomyces lilacinus*) and iv. Toxin producing fungi (*Coprinus* spp., *Pleurotus* spp). Other kinds of fungi which are not classified under the above scheme are plant growth promoting fungi which are active phosphate-solubilizing capacity which also suppress nematodes [*Aspergillus, Penecillium, Trichoderma, Fusarium* spp (Non-pathogenic endophytes)].

Lacunae in Fungal Nematode Bioinoculants

In India, most of the investigations on fungal nematode biocontrol agents are not robust today; since, the studies conducted are arbitrary, for instance, any projects/ investigations mainly highlighting the survey for isolation of biocontrol agents, testing their relative efficacy on nematodes either in pot culture/microplot studies. No doubt that, we have a large pool of diversity in fungal biocontrol agents too. Although, these fungi have been proposed as promising candidates for biological control of plant parasitic nematodes, experimental trials for this purpose have met little or no success (Stirling, 1991). This is partly due to our scarce understanding on the ecological performance of this group of fungi under non-controlled field conditions. The major lacuna in the successful implemention of the fungal nematode biocontrol agents are non-availability of efficient immobilizing systems for delivery and survival of bioinoculants and the other the ecological preference of the introduced fungus which needs to be studied in depth.

Mechanisms of action

Bioinoculants suppress plant pathogens by direct parasitism, lysis, competion for food, direct/indirect antibiosis through production of volatiles substances *viz.*, ethylene, hydrogen cyanide, alcohols, monoterpenes and aldehydes (Juan *et al.*, 2005). Activity of bioinoculants mainly depends on the physicochemical environmental condition to which they are subjected. These mechanisms are complex and what has been defined as biocontrol is the final result of varied mechanisms acting antagonistically to achieve disease control. These have been established for disease control. Very few studies established the effect of fungal bioinoculants on nematode at molecular and or genome level.

Future directions

a. Improvement in the already identified fungal bioinoculants can be made. Since, several isolates of *Pochonia chlamydosporia* are capable of producing enzymes (proteolytic, stilbenes) and toxins/metabolites in *Trichoderma* spp. (trichodermin), modulating culture condition or substituting the culture media with inducers of those compounds or genetic manipulation can be done for potential field efficacy.

b. Recently, endophytism has been found new aspects of the mode of action of fungal parasitism of invertebrates which has potential relevance in biocontrol performance. Fungal endophytes provide several benefits to plants *viz.*, production of secondary metabolites, protection against pests, herbivore resistance. Some endophytes may also improve the phosphorous uptake by the plant and its photosynthetic efficiency or increase the host tolerance to abiotic stress (drought, metal, salts and high temperature).

c. Many new technologies have begun to improve our understanding of the relation between fungal bioinoculants and nematodes. Eg. *Pochonia chlamydosporia* and its plant and nematode hosts and the trophic switch to nematode egg parasitism followed by fungal spore formation. This will enhance our ability in manipulating cultivation practices to favour egg parasitism and to optimize the performance of fungal inoculants.

d. In a recent *In vitro* study using *Trichoerma viride* against *Meloidogyne incognita* egg hatching and juvenile mortality were studied. The SEM photograph revealed fungal conidial attachment in 48 hrs of inoculation. The penetration of the hypha at 72 hrs inside the egg resulted in shrinkage of the egg shell (1200 x) which ultimately caused protoplasmic leakage of the eggs (Jonathan *et al.*, 2011). Hence, there is a huge potential to isolate effective native strains for the management of nematodes in crop plants.

e. Finally, tritrophic direct and indirect interaction should be further studied from ecology to molecular levels. The mechanisms of the biocontrol process might contribute to improve the implementation of the fungal bioinoculant as a successful commercial product for the control of nematodes.

New Molecules for Nematode Management

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Nematodes are a highly diverse group of organisms causing worldwide crop losses estimated at $157 billion per annum that is about 12.3% (overall) and, about 14% in the developing countries. In India, nematode is responsible for about Rs.21 billion every year. To avoid such huge loss nematode management is the major task in front of researchers. There are several methods used for nematode management among them chemical method is very effective and efficient. History of chemical control begins with the discovery of Carbon bisulphide (CS_2) and led to usage of Organophosphate (OP) & carbamates. But because of the environmental concern these fumigants, OP and carbamate compounds were banned from market. So the only way to overcome this problem is to develop a new generation nematicides which are safe to environment, required in lower dose, show lower residual activity and target specific in action.

Now this is the era of newer molecules which aims at safe crop protection. Recently the nematicidal activity of several newer molecules and their formulations were tested. Fluensulfone, a new nematicide of the fluoroalkenyl group, has proved to be very effective in controlling root-knot nematodes, *Meloidogyne* spp., by soil application. The systemic activity of this compound against *M. incognita* on peppers via soil drenching and foliar spray was evaluated. Among them the promising results were obtained from the plant products like saponins, prosaponin and sapogenin from *Medicago* spp, products like DIBOA and DIMBOA from Rye, flavone-C-Glycosides from *Arisaema erubescens*, flavanones from *Phyllanthus nirurii*, monoterpenoids like Borneol, carveol, citral, geraniol, menthol, terpinen-4-ol ,á-terpineol and neem products like raw neem seed powder, azadirachtin. Compounds isolated from fungus like 2,6-pyridinedicarboxylic acid Penipratynolene, *6-methoxycarbonylpicolinic acid (Penicillium bilaiae),* Paeciloxazin, Leucinostatins (*Paecilomyces* spp.), Fumiquinone A and B, Spinulosin, Pseurotin, LL-$S490_B$ (*Aspergillus fumigates)* also act as potential nematicides. Two new azaphilone metabolites, named pseudohalonectrin A (1) and B (2), were isolated from the culture of the aquatic fungus *Pseudohalonectria adversaria* YMF1.01019, originally separated from submerged wood in Yunnan Province, China. Pseudohalonectrin A and B were assessed for their nematicidal activity against the pine wood nematode *Bursaphelenchus xylophilus* and their structures were defined after spectral analysis. Nanotechnological approach has also been applied to control release formulations of Carbofuran showing better result than commercial formulations..

Integrated Pest Management in Green House Vegetables and Flowers: Status in India

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Protected cultivation in India has made rapid progress due to enormous advantage of protection against seasonal adversities, provision of homogeneous conditions for better growth and healthy produce, water conservation, extension of cropping period and off-season cultivation, besides protection against pests. However, prospects and constraints of the technology vary across vast area with diverse agro-climatic conditions. Many of these ideal conditions provided by protected cultivation also result in very high pest build ups, especially the soil-borne pathogens such as root rot, damping off fungi, powdery mildew, nematode parasites; besides other arthropod pests such as mites, aphids, whiteflies and thrips inflicting heavy damage to the plants, particularly in greenhouses that are more than a few years old. Extensive surveys in greenhouses have revealed serious build up of pests mainly on account of sucking pests and soil-borne pathogens. Due to rapid multiplication of these pests, growers often resort to repeated sprays of chemical pesticides leading to environmental degradation besides further aggravating the problems due to pest resurgence coupled due to mortality of natural enemies and non-target organisms.

A holistic integrated pest management (IPM) approach is quintessential to address such the whole gamut of wide ranging problems. IPM options initiate right from problem identification to agro-ecosystem analysis including the history of the cropping pattern etc. Baseline studies should be undertaken to collect the information on pest-scenario, popular plant-protection measures so that measures for pest avoidance such as exclusion of sick-fields, crop rotation, removal of weeds supporting the off-season pest propagules, level of the surviving pest etc. before taking control measures. Baseline studies should be followed with laboratory studies on the economic threshold levels for commensurate planning on pest management. Cultural practices including soil-solarization, mulching is planned much in advance of transplanting the vegetable seedlings. Preparation of nursery beds with proper enrichment with bioagent-colonized FYM including botanicals minimizes pest damage and provides concomitant nutrient for healthy seedling transplantation. Similarly, the sources of resistant germplasm provide eco-friendly management options. Old greenhouses which are heavily infested with soil-borne problems require special interventions like fumigation with chemicals such as metam sodium for disnfestation of planting beds.

Studies have been carried out on comprehensive pest management by comparing IPM and non IPM modules for the management of key pests on different greenhouse crops, development and validation of IPM strategies in vegetable and flower crops. Also, a thorough screening of 275 tomato germplasm lines was carried out and a rootstock was identified for the grafting of greenhouse tomato varieties over the identified rootstocks. As a challenge to counter soil-borne pathogens, exhaustive efforts ranging from soil fumigation to bioremediation involving combination of bioagents like *Pseudomonas flurorescens* and *Trichoderma harzianum* have given good results in tomato and cucumber. These results have been further improved in combination with organic amendments enriched with combination of bioagents gave significant increase in yield (33%) through reduction in wilting (16%) and root-knot nematode, *Meloidogyne incognita* population (18%). There was an overall reduction (19.81%) in soil-borne pathogens, using combined application of bioagents and organic amendments like enriched farm yard manure as compared to non-IPM module (37.56%). Similarly, sucking pests like two spotted spidermites were effectively controlled in greenhouse cucumber by combination of azadirachtin and agrospray® (0.5%).

Newer initiatives for the control of sucking pests of greenhouses using arthropod predators and parasitoids (*Phytoseiulus persimilis, Amblyseius swirskii, Diglyphus isea, Orius laevigatus* etc.) imported from Israel to suppress and manage spidermites, whiteflies and leaf miner in both vegetables and cut-flowers under protected cultivation are giving renewed and re-strengthened confidence in comprehensive management of greenhouse pests. Apart from reduced pest incidences, the economic analysis indicated that IPM approaches were economically favourable.

It is felt inspite of greenhouse industry multiplying rapidly; the growers are ironically getting disenchanted with overall production system in the wake of lack of technical support particularly due to pest problems. Therefore, concerted efforts on greenhouse pest management are required for proper support of growers, as the technology holds lot of promise, especially when the farm holdings are reducing very fast in a country like India and greenhouses definitely provide economical options on small land holdings.

Nematode Problems and their Management in Indian Pulses

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Pulses constitute an integral part of Indian diet. Pulse crops are grown in different agro ecological zones in around 22-23 million ha producing 14-15 million tons of grains till 2009-10 and 18.2 million tons during 2010-11. The average productivity of 600-625 kg/ha of these crops is far below the potential yield of over 2000 kg/ha. Several factors are responsible for poor and unstable yield of pulses, the most important being biotic and abiotic stresses. Around 8-10% of pulse crops are lost annually due to ravages of diseases. Among various disease causing organisms, plant parasitic nematodes are one of the major biotic constraints in production of pulse crops in India. Plant parasitic nematodes are causing yield losses ranging from 5-60% in different states of India. However, the quantum loss depends upon the nematode species and their population densities. Although a number of plant parasitic nematodes are infesting pulse crops but root-knot nematodes (*Meloidogyne incognita* and *M. javanica*), reniform nematode (*Rotylenchulus reniformis*), pigeonpea cyst nematode (*Heterodera cajani*) and root lesion nematode (*Pratylenchus thornei*) are considered to be the most potential nematodes of pulse crops. Many times, the infestation of nematodes is over looked and is confused with other soil problems. Once the nematode problem is identified, nematode population can be managed up to some extent by deep summer ploughing after irrigation. Suitable cropping sequences play important role in regulating the nematode populations. Pulse crops can be included in place of wheat, barley, oats, maize, and sorghum to control the cereal cyst or maize cyst or sorghum cyst nematodes. If the field is infested with root knot, reniform or cyst nematodes, pulses can be grown in rotation with cereals and millets. Growing of mustard with chickpea as intercrop has been found to reduce infestation of root knot nematodes in chickpea while growing of sorghum with pigeonpea as intercrop is highly effective in reducing the population of *Heterodera cajani*. Use of trap crop can also help in reducing the nematode damage to the main crop. Use of soil amendments such as non-edible deoiled seed cakes of neem (*Azadirachta indica*), *Melia azadirachta*, karanj (*Pongamia glabra*), mahua (*Madhuca indica*), castor (*Ricinus comunis*), *etc.*, improve soil fertility and organic matter status besides reducing nematode population density. Efforts have been made to identify resistant sources against key nematodes in different pulse crops. Biological control agents like *Pasteuria penetrans,* *Paecilomyces lilacinus, Verticillium chlamydosporium, Trichoderma viride, T. harzianum, Aspergillus niger, etc.,* can help in reducing the nematode populations. Chemical options are limited. Two chemicals phorate and carbofuran can be applied in soil as last option. Seed treatment with carbosulfan is also recommended to avoid initial nematode infection. Although, number of methods is available for the management of nematodes, yet, hardly any method of nematode control in pulses has become a consciously adopted practice by the common farmers. Interest has been generated recently with training of the farmers and other extension workers about the seriousness of the nematode problems and manage them to avoid losses in pulse crops.

Biofumigation for Management of Plant-Parasitic Nematodes: Prospects and Constraints

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In the present agricultural scenario, as nematologists have limited number of chemicals to recommend as nematicides and the concern for environment and soil health has increased considerably, the use of alternative methods of nematode management becomes imperative. Plants belonging to the families Brassicaeae, Capparidaceae and Caricaceae possess a class of organic compounds, called glucosinolates that contain sulfur and nitrogen and are derived from glucose and amino acid. These water soluble anions are present in cell vacuoles separated from the enzyme myrosinase in the cell cytoplasm. This enzyme hydrolyzes glucosinolates to release methyl isothiocyanate (MIC) which is nematicidal. If plant biomass from the above mentioned families is chopped and incorporated in the soil for degradation, MIC is liberated that inhibits activities of plant-parasitic nematodes in the soil. *Brassica juncea* leaves incorporated @10-30% w/w in root–knot infested soil ($30J_2$/cc soil) in polythene bags and use of chopped twigs @10tonnes/ha in root-knot infested polyhouse soil resulted in undetectable levels of nematode juveniles within a month (Kamra, unpublished). A significant increase in free-living nematodes was also observed in the treated soil, compared to untreated soil. in US, the greatest reduction in Columbia root-knot nematode, *M. chitwoodi* was attained by cropping rapeseed for 2 months and incorporating it in the soil as green manure. In a trial in Spain, incorporation of *B. juncea* and *Eruca sativa* alone or in combination with seed meal pellets and cadusaphos showed significant reduction of *M. incognita* infecting melon, watermelon and tomatoes. Biofumigation was found to be more effective than solarization for the management of *Xiphinema index* spreading grape fan leaf virus. in UK, ploughing of white mustard, *Sinapis alba* in potato field resulted in significant reduction of potato cyst nematode, *Globodera pallida*.

No management strategy is without pitfalls. In biofumigation too, care needs to be taken that the biomass to be incorporated is free of plant-pathogenic fungi like *Rhizoctonia, Pythium* or *Plasmodiophora*. Besides, the incorporation of the biofumigant biomass in chopped form with sufficient moisture facilitates degradation. Covering the biomass with soil or polythene mulch prevents escape of gases. The practice results in an increase in organic matter and water holding capacity and improves the nutrient status of the soil. Although it cannot be a replacement of soil disinfestation using chemicals, this practice alone or in combination with reduced dose of chemicals can help to manage plant-parasitic nematodes while maintaining soil microbial diversity in favour of plant growth.

Medicinal Plants - Nematode Problems and Problem to Nematodes

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Development of medicinal plant industries can be enhanced by ensuring that plants and planting materials are disease free. Among the various plant pathogens, plant parasitic nematodes are a serious threat to the future prospects of medicinal plants worldwide. It constitutes one of the most important groups of pathogenic organisms prevalent in and around the root, playing a significant role in the plant growth and yield reductions. Among the different plant parasitic nematodes, root-knot nematode (*Meloidogyne incognita & M. javanica*), root lesion nematode (Pratylenchus *thornei*) and stunt nematode (*Tylenchorhynchus vulgaris*) influence the yield of major medicinal plants. The economically important medicinal plants which suffer root-knot nematode infestation are: **Ashwagandha, Serpagandha, Brahmi, Menthol mint, Henbanes, Basil, Opium poppy, Coleus, Qinghao** and **Safed Musli**. It has become inevitable to manage nematode problems in medicinal plants through alternative methods because chemical nematicides have an adverse impact on human health and environment. Though, various nonchemical nematode management practices viz. using fallow land, alternation in sowing timings/planting materials, tillage practices, use of antagonistic crop, utilization of nematode free planting materials or seeds, organic amendments and microbial management are available, due to economics (cost-benefit ratio, more pressure on limited piece of land) only selected methods are applied in field to tackle the nematode problems in medicinal plants. This notorious nematode problem in medicinal plants may also be tackled with plant based chemical/products for enhancing the crop production. Large number of plants is known to possess nematicidal activities in their different parts like roots, shoots, leaves, flowers, seeds etc. Therefore utilization of plant based formulations could be more useful to manage nematode problems in medicinal plants.

Nematode Management: Success and Failures in India

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Agriculture has been accorded high priority in our national economy as it accounts for 17 per cent of GDP, contributes for 10.23 per cent of total exports, supplies raw materials to industries and directly supports livelihood of about 50 per cent of our populations. The country has made substantial progress in terms of food grain production since its independence when the food grain production in India was around 50.8 million tonnes in 1950-51 which rose to nearly 240.0 million tonnes during 2010-11. However, in terms of productivity India still lags behind many developing and developed countries. In view of the globalization, liberalization and intense pressure on land as a result of increasing human populations, diversification in agriculture in India has become need of the day. In the present scenario of agricultural production system, vagaries of pests and diseases have increased considerably due to intensification in agriculture. In order to further enhance the crop production for meeting the food requirements of increasing human populations, the crop protection sciences must receive equal attention of policy planners, agricultural scientists and above all the farming community as crop production sciences. Among the various biotic stresses, plant parasitic nematoades constitute an important group of pests and and diseases and has assumed a significant proportion due to the insidious disease symptoms/syndromes in different crops in the country. The Indian Nematology has made significant progress since first report of root-knot nematode in India by Barber in 1901. In view of the damage potential of these tiny hidden enemies of crop plants, All India Coordinated Research Project on Nematode Pests of Crops and their Control started functioning under the aegis of ICAR, New Delhi as a Plan project w.e.f. April 1, 1979 to undertake studies on nematode distribution, assessment of crop losses and for developing effective nematode management technologies. The studies carried out by fifteeen cooperating centres of AICRP (Nematodes) has generated useful information on the effect of these noxious nematode pestss on crop yield from the identified hot spot areas. Based upon the surveys conducted over the years, nematode distribution in the form of **"Nematode Distribution Atlas"** has been prepared and published by the ICAR, New Delhi during 2010. The national loss due to economically important plant parasitic nematodes in 24 different crops in monetary terms has been estimated to the extent of Rs.21,068.73 millions which needs due attention for working out the cost effective nematode management technologies for reducing the yield losses caused by them.

Under this project, a few successful nematode management technologies, which are low cost eco-friendly and practically feasible have been demonstrated at the farmer's field. A number of such technologies have been included in the package of practices of different State Agricultural Universities for adoption by the farmers in the nemtode affected areas. The rice root nematode, *Meloidogyne graminicola* and rice root nemeaotde (*Hirschmanniella* spp.) can be managed by treatment of nursery beds with carbofuran @ 0.3 g a.i./m^2 and field application of carbofuran @ 1 kg a.i./ha 40 days after transplanting. Soil solarization of nursery beds area using a thin transparent polyethylene sheets (50-100 um thick) for 2-4 weeks in summer and application of of carbofuran @ 0.3 g a.i./m^2 before sowing could provide nematode free seedlings of transplanted solanaceous vegetable crops, fruits, ornamental etc. which could perform better in field and reduce the spread of nematodes. Deep summer ploughings of nematode infested fields in States having hot and dry summers have helped in reducing the infestation of root-knot nematodes, cereal cyst nematodes and other plant parasitic nematodes. Management of cereal cyst nematode (*Heterodera avenae*) infecting wheat and barley by practising crop rotations with mustard, coriander, garlic etc. led to reductions in its populations. Methods have been developed for reducing the cost and dose of nematicides. Nursery bed treatment with carbofuran@ 0.3 g a.i./m^2 + bare root dip treatment of seedlings at transplanting with carbosulfan (25 EC) @ 500 ppm, protected transplanted vegetables like tomato, brinjal, chilli and pointedgourd etc. against root-knot nematodes (*Meloidogyne* spp.). Seed dressing of direct seeded crops with carbosulfan (25 DS) @ 3 per cent a.i. w/w in mungbean, cowpea, blackgram, okra and cucurbits etc. reduced the attack of root knot, reniform (*Rotylenchulus reniformis*) and lesion nematodes (*Pratylenchus thornei*). Use of organic amendments including neem and castor cakes @ 1 t/ha and their combinations with seed treatment with carbosulfan (25 DS) @ 3 per cent w/w has been found to reduce root-knot nematode damange in vegetables and groundnut. Paring and hot water treatment of banana suckers at 55^0 C for 20 minutes combined with aplilcation of neem cake @ one kg/ plant and carbofuran @ 16.6 g/plant in the pit before planting was effective against root-knot and burrowing nematodes. Among various bio-agents tested, seed treatment with *Pseudomonas fluorescens* + *Trichoderma viride* each @ 5 g/kg seed was found most effective for the management of root-knot nematode and reniform nematode, *Rotylenchulus reniformis* infecting various pulse crops. In future emphasis will be laid on to further develop cost-effective integrated nematode management technologies for different cropping systems under different agro-climataic zones of the country and also for organic farming systems.

However, despite untiring efforts on the part of Nematologists, there are some failures for achieving successful and desired management of nematodes at farmers level. These failures may be attributed to some of the following reasons :-

- Microscopic size of phyto-parasitic nematodes and their soil borne character and hidden mode of life.
- No clear cut above ground symptoms
- Uneven distribution of nematode populations in the field.
- Border subject and less importance among plant protection disciplines.
- Lack of trained manpower.
- Lack of recognition as limiting factor
- Non availability and high cost of nematicides.
- Limited options other than use of chemical nematicides
- Breeders accord low priority for programme on host plant resistance against nematode pests.
- More reliance on the part of farmers on pesticides dealers for nematode management solutions rather than the Nematologists.

Roadmap Ahead

- New nematode problems are likely to crop up in near future due diversification in agriculture. Hence, there is a need to maintain close vigil on these problems and studies on nematode biodiversity, host parasite relationship and crop loss estimation against such nematode pests of economic importance has to be taken up vigorously.
- In the present regime of WTO and IPR, there is a need to generate epidemiological data on important nematode pests so as to fix their tolerance limit for developing and identifying pest risk analysis and pest free paradigms of such; nematode pestss. Hence, the first and foremost option for nematode management is the prevention of introduction of nematode problems in areas where they do not exist.

- There is a need for active collaboration of all the plant protection scientists for working out pest management strategies in a cropping based system rather thán the individual crop in a holistic manner.
- Use of ITK (Indigenous technical know-how) particularly using cultural methods of nematode management needs field validation.
- Capacity enhancement in nematology
- Creasing awareness regarding nematode problems and their management through multi media gadgets.

Entomopathogenic Nematodes – Bacteria Interactions with Insect Immunity

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Successful infection by entomopathogenic nematodes (EPNs) initially involves host location and penetration into the hemocoel. However, entering an insect host is not always easy for the nematode as it faces a series of resistance from the host. Insects avoid or repel the nematode actively which is termed as behavioural resistance. The insects can detect nematodes on its cuticle and initiate evasive and aggressive behaviors which are effective in removing and even killing nematodes. Mechanical barrier on the insect body mediated via sieve plates, mandibles, rasters etc prevents the entry of nematodes in many soil dwelling insects. Scarab larvae also have morphological defenses against nematode entry due to thick peritrophic membrane that serve as barrier of nematode infection via the midgut. High gut pH or presence of protease can be detrimental to the infective juveniles of *EPNs.* Physiological resistance in the form of **'behavioural fever'** response is seen in grasshoppers/locusts which elevate their body temperatures higher than the ambient called **'basking'.** Once the EPNs reach the haemocoel they along with the regurgitated symbiotic bacteria encounter the host immune response of two types- i). *Cellular* - the haemocytic mechanism comprising of phogocytosis, nodule formation and cellular encapsulation, and ii). *Humoral*- includes melanotic encapsulation and production of antimicrobial peptides. Both, the nematodes and the bacteria utilize a variety of pathogenic strategies to overcome the cellular and humoral defense reactions of the hosts, and the response varies with the insect species, their physiological state and the virulence potential of the bacteria. Nematode encapsulation is reported in the orders Orthoptera, Coleoptera, Diptera and Lepidoptera. However, there are species within these orders that do not encapsulate invading nematodes. A counter mechanism for EPNs to cope with the host immune system is also available in the form of i). *Evasion* (avoid from being recognized), ii). *Tolerance* (tolerate the encapsulation response, and iii). *Suppression* (suppress the encapsulation response. Symbiotic bacteria produce toxins that inhibit encapsulation, and melanization. Within minutes of their release in the haemolymph the bacteria are recognized by the insect's haemocytes and get collected into a mass or nodules known as 'haemocyte-bacteria complex'. The bacteria can multiply within the nodules and re-emerge to destroy the haemocytes in case of Lepidoptera and Orthoptera species and go on to cause disintegration of insect fat body which is the main source of antimicrobial peptides.

The tripartite interaction between nematode-bacteria and insect host with respect to the immune response from the insect side is still being explored and their might be fascinating insights into this interaction in the future especially with sequencing of the genomes of *H. bacteriophora* and *P. luminescens.*

IPR and Commercialization of Nematode Bioagents & Entomopathogenic Nematodes

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Biodiversity and soil biodiversity The Convention on Biological Diversity (CBD) defines biodiversity as "the variability among living organisms from all sources, including terrestrial, marine and other aquatic ecosystems and the ecological complexes of which they are part; this includes diversity within species, between species and ecosystems".

Nematodes (Phylum: Nematoda) are one of the most diverse groups of organisms on the planet and occur worldwide in virtually every environment ranging from e.g., deep sea sediments to arid deserts, or from interstitial bacterivores to obligate parasites with multiple intermediate hosts. In the animal kingdom, nematodes are second only to insects in the number of species described, but are the most abundant animals on earth. While only a few nematode species are parasitic on humans, domesticated animals and plants, the majority of species are beneficial to agriculture and the environment.

One group, the entomopathogenic (or insect-parasitic) nematodes have emerged as excellent biological control agents of important insect pests. Since the late 1970s, there has been a tremendous research and commercial interest in entomopathogenic nematodes, Heterorhabditidae and Steinernematidae, and their associated bacteria in their gut. Accordingly, there is an intense interest to isolate these nematodes from different regions of the world that are climatically adapted and have the potential for biological control of pests in that area. In addition, many countries are concerned about the introduction of exotic entomopathogenic nematodes because they may have negative impact on non-target organisms. More than 30 nematode families are known to host taxa that parasitize or are associated with insects. However, because of their biocontrol potential, research has concentrated on seven families: Mermithidae, Allantonematidae, Neotylenchidae, Sphaerularidae, Rhabditidae, Steinernematidae and Heterorhabditidae, the latter two currently receiving the most attention as control agents of soil insect pests. Besides insect hosts, the *Phasmarhabditis hermaphrodita* (Schneider), a member of the family Rhabditidae, is reported to suppress several slug species, and has been developed as a biological molluscicide. Moreover, several predatory mononchids, dorylaimids, nygolaimids, diplogasterids and the fungal-feeding nematode (*Aphelenchus avenae* Bastian) studied as potential biocontrol agents of plant-parasitic nematodes and plant pathogens. However, India is yet to utilize the diversity of these nematodes and realize the potential of the novel gene pool which has a high biological control potential in combination with the gene pool of their naturally symbiotic bacteria.

Explosion in knowledge and scientific explorations in the world for the past 2-3 decades has lead to the fact that the genetic resources of biota *in toto*, be it microbial organisms, plants, animals, fish, arthropods, nematodes etc., are invaluable gene pools that have most varied applications in agriculture, health, environment, etc. The conservation, maintenance, preservation and judicious utilization of the gene pool of respective biota *in toto* has become the mandate of most of the developed countries. The best examples of the outcome of such global mandate culminated in to the deciphering of several hundred GENOMES, which significantly includes *E. coli, Caenorhabditis elegans, Drosophila melanogaster*, Honey bee, mouse, *Oryza sativa, Homosapiens sapiens* and, several other microbials and metazoans. Ironically, *Caenorhabditis elegans, A NEMATODE,* has the distinction of being the first multicellular organism to have its entire genome (genetic code) sequenced. More recently, with the advent of biotechnology, a third discipline has developed. These are the "*C. elegans* ".

With the advent of diversification of research interests for the general good, most research findings in each field are observed to be directly or indirectly finding a link to development of technologies and commercial value. Therefore, there is a growing awareness of intellectual rights protection and benefit sharing of the novelty in research findings. Globally, the information on novel technologies displayed in public domain either with IP protection/copy rights/trade marks, or priced. Under this back drop, the science of Nematology has several proven findings that are patentable or IP protectable. The pioneering biological Model used for most metabolic pathways for diseases and drug discoveries is *C. elegans*. Development of several mutants and populations of *C. elegans* is now instrumental in deciphering several biological and metabolic pathways for the benefit of life on earth. These research areas have opened up new vistas in applicability of science of nematodes for commercial use and thus scope for patent protection.

IPR in biological control of plant parasitic nematodes has been more or less in the lines of IPR of biological control of plant pathogens in terms of evolving the knowledge on mass production, formulations and application methods of antagonistic fungi. The first success has been the commercialization of *Paecilomyces lilacinus* strain 251 for biological control of root-knot nematodes in vegetables and *Arthrobotrys oligospora* for the control of mycelophagous nematodes in mushroom cultivation in Europe. Further, the success of commercial scale production technologies related to *Pasteuria penetrans* in fermentors is far more significant keeping in view the necessity to evolve a system for overcoming the obligate in vivo reproduction of the bacterium. In India, the first such technology applied for IPR protection was from Khan et al., (2005), AMU, Aligarh on Novel process for commercial production of biopesticides, which was granted US patent number.

This was followed by Dr. M. S. Rao, IIHR, Bangalore (A PROCESS FOR PRODUCING A BIO-PESTICIDE COMPOSITION CONTAINING *Trichoderma harzianum* AND *Pseudomonas fluorescens* 05/03/2010), which has been granted the US patent number. Nagesh et al., (2006) have evolved A SIMPLE AND NOVEL DESIGN FOR SMALL-SCALE SOLID STATE MASS PRODUCTION UNIT FOR ANTAGONISTIC FUNGI and applied for an Indian Patent. Subsequently This was commercialized to two firms.

Further, Nagesh et al., (2010) have applied for Indian patent on technologies for developing WP formulation of *Pochonia chlamydosporia* var. *Chlamydosporia* as bio-nematicide and scale-up production and down-stream processing for commercial use.

Table 1. World patents on biological control agents for nematode control

Inventor	Title
Antunes Franco, Carlos Manuel	Liquid media for chlamydospore production of *P. chlamydosporia*.
Antunes Franco, Carlos Manuel	*P. chlamydosporia* strain pcmr and method to use it in biological control of the root-knot-nematode.
Kerry *et* De Leij	nematicidal composition of *V. chlamydosporium* strain AC

Table 2. World patents related to bioefficacy and biological control applications

Title	Country
Entomopathogenic nematodes and methods of their use	USA
Isolated species of *Steinernematid* nematode and methods of white grub control therewith	USA
Heterorhabditis sp. Gsn2 and method for biological control of harmful insects using the same	Korea
Application of entomopathogenic nematode-infected cadavars from hard-bodied arthropods for insect suppression	USA
Entomopathogenic nematode for control of insect pests	USA
New entomopathogenic nematodes active at low temperature	Italy
Application of partially-desiccated EPN for biological pest control	USA
Insecticidal bacteria	Italy
Heterorhabditis bacteriophora insecticide compositions and related processes	USA
Steinernema sp. nematode for suppression of *Helicoverpa zea* and *Spodoptera frugiperda*	USA
Biological pesticide based on chitosan and EPN	Spain
Biological control of orthoptera pest insects	USA
Biological control of mushroom flies by use of *Steinernema bibionis*	NZ
Biological fight against the flies of mushrooms	AU
Biological fight against the harmful orthopteres	USA
H. bacteriophora insecticide compositions & related processes	USA

Table 3. World patents related to EPN formulations and storage

Title	Country
A novel bio-pesticidal formulation with improved shelf-life and the method for its preparation	India
Biological pesticide based on chitosan and entomopathogenic nematodes	Spain
Method and apparatus for the storage of entomopathogenic nematodes	Australia
Process for producing entomopathogenic nematode preparation and method of storing the same	USA

Culture medium for proliferation of nematode parasitic on insect	Japan
Process of storage of nematodes entomopathogenic	AU
Granular formulation of nematodes with improved storage stability	USA
Method for the storage of entomopathogenic nematodes	Australia
Storage of entomopathogenic nematodes	USA
Commercial storage and shipment of entomogenous nematodes	USA
Nematode storage and transport	AU
Production of hydrogel encapsulated nematodes	USA
Hydrogel encapsulated nematodes	USA

There are a few applications for IP protection for technologies on EPN from India. They are

Dr. Sharad Srivastava et al., (2009), IARI, New Delhi, on DEVELOPMENT OF ENTOMOPATHOGENIC NEMATODE - BASED TERMITE BAIT AND A TECHNIQUE TO DISSEMINATE THE BAIT FOR ATTRACTING AND KILLING SUBTERRANEAN TERMITES.

Dr. S. Ganguly et al., (2006), *IARI, New Delhi, on A novel bio-pesticidal formulation with improved shelf-life and the method for its preparation.* This technology was commercialized through transfer of technology to one firm for commercial use.

Dr. M. Nagesh et al., (2010), NBAII, Bangalore on DEVELOPMENT OF NOVEL INSECTICIDAL FORMULATIONS OF HETERORHABDITIS INDICA STRAIN NBAII HI1 AND HETERORHABDITIS BACTERIOPHORA STRAIN NBAII HB5 FOR THE BIOLOGICAL CONTROL OF INSECT PESTS, AND THE METHODS THEREOF FOR THEIR PREPARATION AND USE. This technology was commercialized through transfer of technology for commercial use.

So is the case in the field of agriculture, where technologies related to the control of plant parasitic nematodes through biological control agents, novel genes similar to Bt, plant varieties/hybrids with nematode resistance, RNAi constructs and transformants etc.; or entomopathogenic nematodes and their symbiotic bacteria in terms of their improved tolerance to stresses, virulence, production, formulations, application techniques etc.

The first miRNAs were discovered by forward genetics approaches in the nematode *C. elegans*. Since then, the availability of a reference genome in combination with experimental and computational approaches has resulted in the description of 155 miRNAs in this species. Since recently, the *C. briggsae* and *C. remanei* genomes are available as well, allowing for similar approaches. Within this group, *C. briggsae* and *C. remanei* are sister species and *C. elegans* represents an outgroup. The evolutionary distance between *C. elegans* and *C. briggsae* and *C. remanei* is estimated to be about 100 million years (Myr). While *C. remanei* reproduces gonochoristically, both *C. briggsae* and *C. elegans* reproduce in a hermaphroditic fashion, which likely evolved independently in both species (Kiontke et al. 2004). The genome size of *C. elegans* and *C. briggsae* is ~100 Mb, whereas the genome of *C. remanei* is ~140 Mb. The natural habitat for these species is compost or garden soil, where they feed on microorganisms (Kiontke and Sudhaus 2006). A fourth species for which genomic sequence information is available is *Pristionchus pacificus*, a necromenic nematode, which had a last common ancestor with the *Caenorhabditis* species 280–430 Myr ago (Dieterich *et al.* 2008). Although genomic contigs are not completely assembled yet, its genome size is considerably larger than that of *C. elegans*. Already, research groups at IARI, New Delhi are active on evolving miRNA technologies for disrupting nematode invasion/damage to plant systems and impart heritable nematode resistance in plant hosts, which not only have direct benefit to agriculturists but also generate IPR related competence in the contemporary sciences and researchers.

It is summarized that there is a vast scope for the science of nematodes to have a commercial value and application for the benefit of agriculture and general good.

The Implications of Global Climate Change on Plant-Parasitic Nematodes and Nematology

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The plant parasitic nematodes, as primary consumers, play an important role in food-web. At certain population densities one or more species of plant parasitic nematodes attain the pest status for one or more crops in a particular ecological niche. Abiotic alterations, changes in the physiology of the plant and diversity and activities of the macro- and microfauna and flora also influence the distribution and abundance of nematodes. Climate change usually expressed as elevated temperature and CO_2 level, altered seasonal and rain patterns and extreme events influences agricultural productivity in many direct and indirect ways. The niche characteristics are altered with climate change and so the occurrence and dominance of particular nematode species, their competitors, antagonists and associates in a community. There are a few reports of changes in geographical distribution of species, and possibilities of establishment of present subtropical and tropical species in regions that are subtemperate or temperate today. Increased temperatures will amount to greater cumulative degree hours that would allow shorter life-cycles and more generations of certain species of nematodes. The increased frequency of extreme low or high temperatures will have sub-lethal or lethal effects that may bring down population densities.

Elevated CO_2 levels are expected to enhance photosynthesis and probably increase availability carbohydrate rich food for the nematodes that may, however, be low in protein due to altered C:N ratio. At the same time host plant tolerance may be altered. The increased infestation and crop damage commonly observed in polyhouses forecast the situation that may occur in open fields in future. The changes in duration of summer and winter months will alter the population dynamics of nematodes, requiring readjustments of cropping schedules to escape crop damage due to nematodes and other pests. The altered ecosystem function, especially decomposition processes, will modify microbial community structure that will have positive or negative impact on nematodes. Nematodes will use survival adaptations to face extreme events including heat and desiccation. The existing nematodes are likely to gradually adapt over generations to the rate of climate change and thus persist. Emergence of heat and desiccation tolerant races, invasive alien species and increased susceptibility of crops to nematodes under stress will be new challenges.

The coinhabant microflora and fauna that has various kinds of interactions with nematodes will be simultaneously affected by climate change. Of special interest to Nematologists will be the disease complexes and nematode antagonistic microorganisms that may be coinhabitants or used as bio-control agents. Expertise in morphological and molecular taxonomy will be essential to understand changes in community structure.

Developing resistant crop varieties and newer methods of nematode management will be new opportunities for both fundamental and applied nematologists. Climate induced alteration in expression of resistance or susceptibility related genes will also need to be considered. Collaboration between nematologists and agronomists will be required since some of the agronomic practices designed for time and energy saving and soil & moisture conservation, including zero tillage, SRI, raised-bed etc. have been found to aggravate certain nematode problems. For adopting such modified agronomy, management of plant parasitic nematodes will be even more important.

Many species of nematodes also naturally regulate or serve as biocontrol agents for insect, mollusc and other pests of importance in agriculture, veterinary and human health. Climate change will also influence the activity, distribution and abundance of such species. Investigations on the adaptations, selection and effective use of such useful nematodes will also be required.

Nematodes can also serve as sources of stress tolerance genes that may be useful for crop improvement to meet the impending climatic challenges. Nematodes may serve as natural and inexpensive indicators for forecasting and monitoring climate change. Use of nematodes as model organisms and bioindicators, together with GIS and simulation models can help in climate change research for adaptation and mitigation. Thus, the natural and anthropogenic climate change at the micro and macro levels will have significant impacts on plant parasitic nematodes that will throw more challenges and provide greater opportunities for Nematologists and their collaboration with other disciplines of fundamental and applied environment and agricultural sciences. Broader horizons and increasing role of fundamental and applied nematology can be visualized in the emerging scenario.

Ecological Manipulations for Nematode Management

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Almost all plant parasitic nematode lead part of their lives in the soil. Many of them live freely in the soil feeding on root and underground stem and cause injury to plants Even in the most highly specialized sedentary parasites the eggs , the pre-parasitic larval stages and the males are found in the soil for all or part of their lives. Soil temperature, structure, moisture and aeration affect the survival and movement of nematodes in the soil . Most of the nematodes occur in great density and abundance at a depth of 0-30 cm in the soil.

Preventing the nematode problem is far better than trying to manage them after it is established. The main goal in managing the nematodes is to keep the population below the injury level or the population density as low as possible, since they can not be eliminated completely from soil and our crops have to live with them . Many pesticides have been tried over the years but only a few nematicides are presently being used . In this context ecological manipulation is the best alternative to minimize the population of nematodes in soil. .Many of these are oldest practices of its own included as cultural or physical methods of management but now in the context of organic cultivation gaining importance it can be grouped as the ecological manipulation techniques . The practices include fallowing , deep summer ploughing , addition of FYM, crop rotation , trap cropping , addition of organic amendments, organic wastes , flooding , green mauring , mulching , bio fumigation etc . These practices were done mainly to reduce the nematode population in the soil but it also improve the nutritional status of the soil , moisture conservation , management of weeds and other pests etc. Indirectly application of biological control agents in the soil and soil solarization also help in changing the ecology . Even adjusting the sowing and planting time, spacing and fertilizer application also got an impact on the ecology. By manipulating the biotic and some abiotic factors in green and glass house condition these tiny organisms can be reduced to a manageable level . The interventions in the above traditional methods can be effectively used for the management of nematode without polluting the environment, the need of the hour.

Session I : Nematode Biodiversity and Biosystematics

Two New Species of the Genus *Lenonchium* from the Malnad Tracts of Karnataka

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The Malnad Tracts of Karnataka, a major part of the Western Ghats receives regular rainfall during most of the months in a year. Moisture maintained almost throughout the year makes a very conducive environment to soil microfauna including nematodes. Soil samples collected from this area yielded two new species of the genus *Lenonchium* Siddiqi (1965). This paper deals with the description of the species, *L. anamicus* sp.n. and *L. singulatus* sp.n. with illustrations. *L. anamicus* sp.n. is characteristic in having a large sized body, about 3 mm long, slightly ventrally arcuate upon fixation, tapering towards extremities; a,b,c values 56-63.5 (60.4 ± 3.7); b = 6.1-7.0 (6.6 ± 0.4); c = 11.3-14.2 (13 ± 1.3) respectively; female with amphidelphic gonads and a long, filiform, 9-11 anal body widths long tail, tip always with four finger like projections, caudal pores two on each side; distinct sphincter present at oviduct-uterus junction. Males with large spicules, arcuate, with well developed, lateral guiding pieces which are bifurcated at the tip. Ventromedian supplements 19-25, and an adanal pair, mammiform, arranged serially. *L. singulatus* sp.n. is characterized by having L = 4.2 mm; a = 65.6; b = 8.1; c = 15.4; c' = 6.7; V = 46.3; G1 = 21.3; G2 = 23; odontostyle 28 μ; odontophore 30 μ; oeosphagus 525 μ; expanded part of oesophagus 334 μ; prerectum 478 μ; rectum 41 μ; tail 275 μ; ABD 41 μ; female with amphidelphic gonads with a long filiform tail about 6.7 anal body widths long, tip with 4 finger like projections, caudal papillae two on each side.

Survey for Nematode Infestations Under Protected Cultivation in Haryana

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This study was carried out to conduct survey for nematode infestations in vegetable, ornamental and forest crops, grown under polyhouse conditions in Haryana. For this purpose, thirty two soil samples, collected from different polyhouses of Hisar, Sonepat, Karnal, Kurukshetra and Ambala districts of Haryana were analyzed for the occurrence of different nematode fauna. The results revealed that in bitter gourd crop, root-knot nematode was most predominant with 50% frequency of occurrence having low population. In capsicum, root-knot nematode was the most predominant (80%) followed by *Tylenchorhynchus* (20%), *Aphelenchoides* (10%), *Aphelenchus* (10%), *Hoplolaimus* (10%), *Paratrichodorus* (10%) and *Rhabditids* (10%). In cucumber, *Tylenchorhynchus* was the most predominant nematode (80%), followed by root-knot nematode (40%), *Hoplolaimus* (40%) and *Dorylaimids* (20%). Tomato crop harboured root-knot nematode (28.5%), *Tylenchorhynchus* (72.5%), *Aphelenchus* (14.5%) and *Longidorus* (14.5%). In Gerbera (flowering crop), *Hoplolaimus* and *Tylenchorhynchus* were the predominant nematode species with low population. The polyhouses at Govt. Forest nursery at village Seonthi (Kurukshetra), showed the presence of *Tylenchorhynchus (100%), Ditylenchus* (50%) and *Longidorus* (50%) in Eucalyptus crop.

Rice Root-Knot Nematode, *Meloidogyne graminicola* – Emerging as Serious Pest of Rice – Wheat Cropping Pattern of Ghagar River Belt of Hanumangarh District in Rajasthan

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Hanumangarh District of Rajasthan is known as food basket, where farmers following Rice – wheat cropping system since last half decade in Ghagar river belt. Yellowing and stunting of rice were reported by state extensive agencies particularly in nursery of rice in some village of Hanumangarh District. To find out reason soil and plant samples were collected from Shere-Khan village of Hanumangarh District. Roots of weak plant were stained and examined in laboratory. Mature females of root knot nematode were teased out from root to study the perineal pattern under stereoscopic binocular microscope. Perineal pattern were prepared and mounted on glass slides in a drop of anhydrous glycerine and taxonomic measurements of the perineal pattern were taken under compound microscope. Presence of *M. graminicola* was confirmed and it reported first time from state. IInd and IIIrd stage juveniles were found near root tip. Some of female also observed in cortex area of root. In a preliminary survey, incidence was also reported from Pilibanga Tehsil of Hanumangarh District.

Biodiversity and Biosystematics of Plant Parasitic Nematodes Associated with Banana in India

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Banana and plantain have been the most accepted food-fruit across the globe. India has emerged as the largest producer of banana in the world with total production of 26.2 million tonnes per annum from 6.5 lakhs hectares having share of 37 per cent of the total fruit production. Crop losses caused by nematodes to bananas are very high, with an average annual yield loss estimated at about 20 per cent worldwide. Wide diversification has been noticed in the nematode community under varied ecosystems. Over the years, banana cultivation is gradually making forays in the eastern part and northwest Tarai region of Uttar Pradesh. More than 50 species of plant parasitic nematodes have been reported in association with banana root systems and are mainly responsible for limiting banana production to a greater extent. But no systematic survey has been conducted on banana except few reports on the occurrence and distribution of plant parasitic nematodes associated with banana in India. National Research Centre for Banana conducted an extensive survey to examine the biodiversity of plant parasitic nematodes in major banana growing areas of the country covering different agro climatic zones and different soil types. The results of the study revealed that the root-lesion nematode, *Pratylenchus coffeae*, was the most predominant species and ranked first in prominence and importance value. This was followed by root-knot nematode, *Meloidogyne incognita*, spiral nematode, *Helicotylenchus multicinctus*, burrowing nematode, *Radopholus similis* and cyst nematode, *Heterodera oryzicola*. Analysis of the soil samples collected from banana revealed the presence of 50 species belonging to 28 genera of plant parasitic nematodes. Among them, *Rotylenchulus reniformis, Helicotylenchus multicinctus, H.dihystera, H. punicae, H. africanus, Meloidogyne incognita, M.. arenaria, M.hapla, M.javanica, Hoplolaimus indicus, H. seinhorsti, Pratylenchus coffeae, P. minutus, P. brachyurus, P. goodeyi, P. penetrans, P. scribneri, P. thornei, Paratrichodorus renifer, Scutellonema branchyurum, S. mangifera, Macroposthonia ornata, Hemicrionemoides cocophilus, Tylenchorhynchus brevidens, Trichodorus porosus, Xiphinema americanum, X. ensiculiferum, X. basiri and X. insigne. Criconemoides* spp. and *Tylenchus* spp.

Meloidogyne spp., *Hoplolaimus* spp., *P. coffeae, Tylenchorhynchus* spp., were the most predominant species associated with banana. The community analysis of plant parasitic nematodes such as frequency, density and prominence value were also calculated.

Occurrence of Lesion Nematode, *Pratylenchus thornei* in Aerobic Rice Under Field Conditions

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In Asia, 75% of rice is produced in irrigated lowland fields with high irrigation requirements to sustain a layer of ponded water for most of the growing season. However, irrigation water is becoming increasingly scarce, and estimated that about 2 million ha of Asia's dry-season irrigated rice area will suffer water shortages by 2025. One of the adaptation strategies for water shortage is the system of aerobic rice, in which especially developed varieties that are high-yielding, input-responsive, and adapted to mild or medium water stress are grown under aerobic, non-saturated soil conditions. Especially on lighter soils with high seepage and percolation rates, aerobic rice has an advantage over systems that include saturated or partially flooded soil conditions.

Root lesion nematodes are migratory endoparasites. Two species-*Pratylenchus indicus* have been reported in India and Pakistan, causing yield losses in upland rice. The infestation of *P. thornei* was observed for the first time in the aerobic rice in field at the Institute. The host range of *P. thornei* includes weeds and food crops such as maize, groundnut, wheat, oat, and sorghum.

The aboveground symptoms were similar to general symptoms of plant root stress. Low to moderate populations of lesion nematodes caused visible aboveground symptoms. The plants turned yellowish and the growth was retarded. The nematodes produced characteristic necrotic lesions (darkened areas of dead tissue) on the surface and throughout the cortex of infected roots. The lesions turned from reddish-brown to black and are spotty along the root surface. As the nematodes continue to migrate and feed within the roots, the lesions can coalesce to become large necrotic areas of tissue that may eventually girdle the root. Severe damage from high populations of lesion nematodes can result in a stunted and necrotic plant root system.

Aerobic rice is a water-saving rice production system for water-short environments with favorable soils and adapted, potentially high-yielding varieties that are direct dry seeded. Soils remain aerobic but supplementary irrigation is applied as necessary. Under such conditions we have to develop management options other than crop rotations.

Nematode Diversity in Forest Ecosystem of Gujarat, India

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Nematodes are small worm-like organisms present in forest ecosystem. They interact directly and indirectly with plants and other microfauna, regulating decomposition and release of nutrients to the plants. Gujarat is one of the state having three types of forest viz., Open, Dense and Mangrove forests because the state has wide variations in climate, topography and soil composition. The study of nematode fauna from 2007-2009 in different localities of Gujarat State covering all type of forest. Sixty seven species of nematode belonging to 50 genera, 22 families under the following 7 orders: Tylenchida, Aphelenchida, Aphelenchoida, Dorylaimida, Mononchida, Rhabditida and Enoplida have been identified. Thirteen species of nematode have been recorded for first time from India. In open forest the free living nematode population have been found dominantly 42% followed by predaceous (22%), phytophagous (19%), bacteriovorus (10%) and fungivorus (7%). The abundance of the phytophagous nematode have been found 38% followed by freeliving 21%, predaceous 20%, fungivorus 16% and bacteriovorus 5% in undisturbed dense forest. Seventeen species of nematodes have been recorded in mangrove forest, of which 40% of bacteriovorus nematode followed by free living (30%), fungivorus (12%), predaceous (10%) and 8% of phtophagous nematodes.

Taxonomic Studies on Rice Root-Knot Nematode Species of Haryana

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Rice root knot nematodes is a matter of major concern to the farmers of Haryana state who have, in recent years, started taking paddy crop in non-traditional rice areas having coarse-texured soil. J2, females and males isolated from most of the infected root and soil samples were similar to *Meloidogyne graminicola* Golden & Birchfield, 1965. However, soil samples from Hansi (Hisar), Rohtak and Kunjpura (Karnal) harboured two type of J2 distinguishable on the basis of total body and tail lengths- i. Short forms (similar to *M. graminicola*) measuring 420-452 µm in length with 62-71 µm long tail, and ii. Long forms (similar to *M. oryzae* Maas *et al.*, 1978) 527-567 µm in length and having 86-93 µm long tail. Detailed studies were therefore, undertaken to establish the identity of these two forms of J2 by conducting a series of experiments and examinations.

Both forms of J2 were present in galled paddy roots that were collected from Hansi (Hisar), Rohtak and Kunjpura (Karnal), soil samples of which harboured both the forms of J2. Even a single egg mass could contain both forms of J2. ii. When inoculated separately @ 100 J2/ seedling raised in 1% water agar in Petri plates, both forms of J2 developed to females in 9-10 days. iii. In case of simultaneous inoculations (50 J2 of each form) both forms of J2 could be recovered from the same gall formed near the root tip, three days after inoculation. iv. When inoculated separately on rice seedlings grown in 100 cc plastic cups containing steam sterilized soil, nearly 10% of J2 recovered from soil 40 days after inoculation belonged to other form. v. No morphological differences were discernible in females (stylet, position of excretory pore, perineal pattern), and males (stylet, number of lateral lines) raised separately from different forms of juveniles.

Thus, there is no difference in the biology of two forms of J2 and progeny of one form contains phena of other type also. Therefore, both forms of J2 belong to same species. Incidentally, *M. oryzae* that has been reported from infecting rice in Surinam differs from *M. graminicola* chiefly on the basis of differences in body length and tail length only.

Lesion Nematode, *Pratylenchus* spp., the Major Nematode Problem of Chickpea in Bundelkhand

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Chickpea is the major crop grown in Budelkhand region during *rabi* season. It is grown over an area of about 0.5 million ha. To find out the status of nematode problems in chickpea in Bundelkhand region, survey was under taken during 2008-09, 2009-10 and 2010-11. Seventy seven soil and root samples were collected from chickpea fields covering areas of Hamirpur, Mahoba, Jhansi, Jaluan, Kanpur, Fatehpur, Tikamgarh and Banda districts. Soil samples were brought to laboratory and processed for extraction of nematodes. Plant parasitic nematodes present in soil were counted and their absolute and relative frequencies, absolute and relative densities and prominence values were calculated for each plant parasitic nematode present in chickpea fields. Roots were stained in boiling lactophenol acid fuschin and observed for nematode infections. Out of fourteen nematode genera observed in soil samples, *Pratylenchus* sp. was most prominent with its prominence value 424.4, density 574.7 nematodes/100 cc soil and frequency 54.5%. Other nematode genera commonly observed were *Tylenchorhynchus, Hoplolaimus, Tylenchus* and *Helicotylenchus*. *Pratylenchus* sp. was most prominent in Hamirpur district with prominence value 805.5, density 1046 nematodes/100 cc soil and frequency 59% followed by Banda, Jhansi, Tikamgarh, Mahoba, Jalaun districts. It was least prominent in Kanpur and Fatehpur districts. *Pratylenchus* sp. was also observed in roots. The sparse and reduced growth of chickpea plants with drying of the lower leaves was observed in *Pratylenchus* sp. infested fields. Lesions were observed on the roots and root mass was reduced due to nematode infection. Root-knot and reniform nematodes were not observed in chickpea in Bundelkhand region. *Pratylenchus* sp. was the major nematode infesting chickpea in this region and needs to be managed to avoid losses due to this nematode.

Occurence and Distribution of *Meloidogyne incognita* in Ornamental Plants of Jorhat District

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In the present investigation an effort was made to study the occurrence and distribution of root-knot nematode (*Meloidogyne incognita*) in important ornamentals of Jorhat districts. In a random roving survey programme during 2009-10, 174 soil and root samples were collected from different nurseries, homested gardens, orchards etc. The soil and root samples were collected from the rhizosphere of different ornamental crops. The study revealed that root-knot nematode is abundantly present in most of the surveyed ornamental crops. The maximum frequency of occurrence (76.92 percent) was recorded from Chipahikhola block and minimun (48.38 percent) from Titabor. The nematode is more or less uniformly distributed through out the surveyed area. Out of all ornamentals gladiolus (*Gladiolus hortulanus*) was recorded to be the most susceptible ornamental crop.

Management of *Meloidogyne incognita* and *Rhizoctonia solani* Disease Complex with Organic Amendments in Tea Seedling

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An experiment was conducted during 2009-10 under pot condition to evaluate certain organic amendments *viz.*, neemcake, mustard cake, jatropha and vermicompost each at the rate of 15 g and 30 g per kg of soil in comparison with carbofuran @ 3 g per kg of soil as soil treatment for the management of *Meloidogyne incognita* and *Rhizoctonia solani* disease complex in tea seedling. All the four organic amendments at all the doses and the treatment with carbofuran @ 3 g per kg of soil were found to be effective in increasing the plant growth characters as well as reducing the number of galls, egg masses, collar rot incidence and final nematode population as compared to that of control. However, carbofuran @ 3 g per kg of soil was recorded to be most effective treatment followed by neem cake and mustard cake at 30 g per kg of soil. Among the two doses of all the four organic amendments tested, the higher dose i.e. 30 g per kg of soil was found to be effective as compared to the lower dose.

Host Status of Various Forest Seedlings to Root–Knot Nematode (*Meloidogyne spp.*)

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The present study was carried out to the see the host response of various forest seedlings to root- knot nematode (*Meloidogyne* spp.).For this purpose, three seedlings, each of 11 plant species obtained from various forest nurseries of Hisar and Bhiwani districts of Haryana, were transplanted in 30 cm sized pots filled with root- knot nematode infested soil having a mixed population of *Meloidogyne incognita* and *M. javanica* (INP 320 J2/ 200CC soil) during August, 2010.Two months after planting, the seedlings were uprooted, washed and observed for root–knot index (1 to 5 scale) and final nematode population for assaying their host status.

The results revealed that seedlings of Tamarind (*Tamarindus indica*), Bakain (*Melia azadirach*), Ailanthus (*Ailanthus* sp) and Amaltash (*Cassia fistula*) showed non-host response without any galls and egg masses. Neem (*Azadirachta indica*) and Jamun (*Syzygium cumini*) showed poor host response with a root-knot index less than 2.0. Desi kiker (*Acacia nilotica*) and Shisham (*Dalbergia sissoo*) showed moderate host response with a root-knot index 2.1 to 4.0. Mulberry (*Morus alba*), Papaya (*Carica papaya*) and Sukhchain (*Pongamia pinn*ata) showed good host response with a root-knot index of 4.0 and above.

Occurrence of Root-Knot Nematode, *Meloidogyne incognita* in West Indian Cherry, (*Malpghia emarginata*) Orchard

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Samples were collected in root zone of west Indian cherry, (*Malpghia emarginata*) orchard at Anbil Dharmalingam Agricultural College and Research Institute, Navalure kuttapattu, Where the trees exhibits unthrifty growth characters. Both soil and root samples were collected at a depth of 15-25 cm during the month of May 2010 when the soil temperature was 34.11°C and the soil moisture was in 4.31 %. Each sample contains 100 gram of soil and 10 gram roots. The soil samples were processed for nematode extraction by Cobb's wet sieving method, immediately after collection, All the nematode juveniles were in tightly coiled and in non motile position and they were caught only in the 100 mesh sieve none of them were caught in 350 mesh sieve. The non motile coiled nematodes in the nematode suspension become active after 20 minutes. Each sample contains an average of 953 root-knot juveniles per 100 gram of soil. The root samples contains multiple galls with root pitting symptoms. The roots were graded for the root-knot index and it was comes under scale 5. Species identification was done based on perennial cuticular pattern and it was confirmed that it was Southern root-knot nematode species, *Meloidogyne incognita*.

Plant-Parasitic Nematodes Associated with Small Onion (*Allium cepa* var. *aggregatum*)

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A random survey was conducted to determine the occurrence of plant parasitic nematodes associated with small onion, *Allium cepa* var. *aggregatum* in Alathur block of Perambalur district in Tamil Nadu. A total of 51 soil and root sample were collected from onion fields. Nematodes were extracted from 100g of soil per sample using a modified Bearmann extraction tray technique. The endo-parasitic nematodes were extracted by using modified maceration and filtration technique. A total of five genera of plant-parasitic nematodes *viz., Hoplolaimus* sp., *Helicotylenchus* sp., *Tylenchus* sp., *Criconema* sp. and *Xhiphinema* sp. belonging to three families of the order *Tylenchida* and one family of *Dorylaimidae* were identified from the soil samples. The population of various plant-parasitic nematodes differed significantly in different samples. The lance nematode, *Hoplolaimus* sp. was detected in 75 per cent of the samples collected with the density of 54.40/100 g of soil, followed by the spiral nematode, *Helicotylenchus* sp. as 65 per cent frequency with the density of 52.60. The ring nematode, *Criconema* sp. occured with relatively lower frequency (30 %) and density (18.00) level. The study concluded that small onion is a potential host to plant-parasitic nematodes and particularly for *Hoplolaimus* sp. and its use as a rotation crop should be evaluated. Endo-parasitic nematodes were not detected in this survey.

Session II : Molecular and Physiological Nematology

New Gene Targets of Root-Knot Nematode, *Meloidogyne incognita* for RNAi Engineered Resistance in Tomato

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Complete genome sequence of the root-knot nematode, *Meloidogyne incognita* has paved the way for selecting potential gene targets for functional validation through RNA interference. *In silico* search was conducted to identify three genes having specific role in the development process in *M. incognita*, namely, Cathepsin-L-Cysteine Proteinase (*Mi-Cpl-1*), Collagenase (*Mi-Col-1, Mi-Col-2*) and Acetylcholine Esterase (*Mi-ace-1*). Genes were aligned with the homologous gene sequences of other nematode species using ClustalW programme after BLAST search. A unique 366 bp sequence had been identified as conserved region of *Mi-cpl-1* spanning between 787 to 1152 bp using BioEdit programme. To amplify the gene, total RNA was isolated from J2 of *M. incognita* followed by conversion of cDNA through RT-PCR. Specific restriction sites were incorporated into the primer sequences for cloning the gene in sense and antisense orientation into the RNAi vector. The gene construct has a potential to provide resistance against root-knot nematode, *M. incognita*.

Identification of Antigens of Root-knot Nematodes, *Meloidogyne graminicola* and *M. incognita* Involved in Differential Host Preference by Proteomics Approach

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Meloidogyne graminicola normally infect rice, wheat and several other graminaceous plants but can infect several dicotyledonous plants. *M. incognita* is a serious pest of dicotyledonous crops and occasionally infects cereals. Evolutionary adaptation of these root-knot nematodes to their preferred hosts might have led to variability in their gene/protein profile which could contribute to their differential behaviour outside and inside of the different host crops. Polyclonal antibodies raised against live pre-parasitic second-stage juveniles (J2) of *Heterodera avenae* as well as against surface coat extract and Secreted-Excreted (SE) products of *M. arenaria* showed cross-reactivity to antigens with different molecular weights present in the whole body homogenate of *M. incognita* and *M. graminicola* J2. Monoclonal antibodies raised against whole J2 and SE products of *H. avenae* showed very high levels of cross reactivity with a couple of specific antigens of *M. incognita* J2 homogenate but did not show any reaction with the J2 homogenates of *M. graminicola*. This variability in antigenicity may correspond to specific functions of these molecules in *M. incognita* and *M. graminicola*. Using proteomics approach possible amino acid sequence of those antigens was elucidated through MALDI-TOF and Q-TOF, showed sequence similarity with several proteins like signal recognition particle protein, heat shock protein, zinc finger domain etc. from the genomic database of several nematode species. To investigate the function of the identified nematode genes, RNA interference, could be used to reduce the expression of these selected genes and determine their importance for nematode development, survival, or parasitism.

Sustainable Management of Root-Knot Nematode by RNAi Mediated Silencing of Parasitism Genes of *Meloidogyne incognita*

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Plant parasitic nematodes, especially root-knot nematodes, are the hidden enemy of crops and *Meloidogyne incognita* is the most pathogenic species with yield losses in the range of 10-27%. A recent survey in cotton growing areas of north India has revealed widespread infestation of root-knot nematode in Bt cotton. The root-knot nematode-fusarium wilt disease complex is becoming prevalent now even in *hirsutum* cotton. Lack of effective nematicides along with concomitant dangers of environmental pollution limits use of chemicals for nematode control. Resistant varieties are not available and crop rotations designed to reduce nematode density are not economically practicable. This necessitates the need for exploration of novel methods for management of root-knot nematode and RNA interference (RNAi) mediated crop protection fits the bill. Using RNA interference, the nematode's biology can be turned against itself resulting in endogenous nematode control.

At CICR Nagpur, work has been initiated on RNAi-mediated protection of cotton against root-knot nematode. Primers complementary to the conserved regions of 10 key parasitism genes were synthesized and used for amplification of specific sized amplicons. Evaluation of dsRNA for ten parasitism genes viz.. including

MjTis11, ADF 1, ADF 2, ADF 3, ADF 4 for penetration and reproduction of root-knot and reniform nematode was carried out. Out of these MjTis11 reduced penetration of rootknot nematode by 58% while the effect on reniform nematode penetration was significantly less at 28%. More parasitism genes need to be screened to identify parasitism gene/s most critical for nematode penetration and reproduction for effective RNAi mediated nematode management.

Engineering Resistance in Tomato against Root-Knot Nematode, *Meloidogyne incognita*, by RNAi Silencing of Esophageal Gland Gene

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Root-knot nematode (*Meloidogyne incognita*) is highly evolved obligate sedentary plant endoparasites with a wide host range that infect many plant species causing significant economic losses worldwide. The parasitism genes expressed in the esophageal gland cells of root-knot nematodes encode proteins that are secreted into host root cells to induce the formation of the nematode feeding site and its maintenance during the parasitic process. These parasitism proteins offer putative targets for engineering resistance in otherwise susceptible crops using biotechnological tools. With the advent of RNA interference (RNAi) technology and the demonstration of host induced gene silencing in parasites, a new strategy to control pests and pathogens has become available, particularly for root-knot nematodes. The tomato, *Solanumesculentum,* (cv. Pusa Ruby) was transformed using *Agrobacterium tumefaciens*mediated method to express dsRNA of the subventralesophageal gland parasitism gene AF531170. The plants at T_0 and T_1 stage were evaluated for nematode infection by inoculating J2 into pots. The host-induced RNAi of the root knot nematode parasitism gene led to a significant reduction of 59.89% and 54.79% in the development of adult females over control in T_0 and T_1 plants respectively. There was reduction in the galls to tune of 61.10% and 47.05% in T_0 and T_1 plants expressing dsRNA of the parasitism gene respectively. The reduction in eggs per egg mass was 33.98% and 34.23% in T_0 and T_1 plants respectively. These observations demonstrate the relevance of the targeted parasitism gene during the nematode life cycle and more importantly, suggest potentiality of obtaining viable level of resistance in crop plants in the future using RNAi technology against root knot nematode.

Transformed Tomato Plants with Ability to Silence Esophageal Parasitism Gene Grow Better in Root-Knot Nematode *Meloidogyne incognita*, infested Soil

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Nematodes of the genus *Meloidogyne* spp. are major pests on tomatoes (*Lycopersiconlycopersicum*Karst) where they cause considerable losses in yields ranging from 28 to 68 per cent. Nematicides have been used to control these pests but, they bring about the problems of high costs and environment pollution. Engineering resistance usingRNAi strategy is one of a safer strategy to counter the nematode menace. In our lab we have generated plants expressing dsRNA construct to silence a subventralesophageal gene, AF531170. In this study we compared the general health of the transformed tomato plants (T_0 and T_1) with the untransformed control plants under pot condition, soil infested with root-knot nematode juveniles @ 2J2/g of soil. Plant growth parameters, root and shoot weight, root and shoot length were observed after 60 days of nematode inoculation in T0 and T1 generation plants. All the treatments were significant over inoculated control. The average per cent increase in shoot weight, root weight, shoot length and root length is 34.41, 39.86, 19.46 and 35.34 in T_0 plants and 38.26, 44.20, 14.30 and 31.44 in T_1 plants, respectively. These observations demonstrate that the targeted parasitism gene doesn't have any negative effect on plant growth parameters of the crop plant.

Session III : Nematode Ecology

Functional Ecology of Wetland Nematode of Genus *Plectus* (Plectidae: Plectida) from Wet Land Shekha Jheel, Uttar Pradesh

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The paper deals with the taxonomy, biology and ecology of genus *Plectus* in the wetland Shekha Jheel. *Plectus* was found to be the most frequent taxon in the samples collected from different biotopes of the wetland Shekha Jheel. The genus is important as it is one of the most widely distributed and largely continental taxa with species reported from freshwater, semi-aquatic and terrestrial habitats. Its prevalence in the samples reveals the quality, nature and status of the environment. The individuals were bacterivores and regarded as colonizers (r-strategists) with a value of 2 at *c-p* (colonizer-persister) scale. Three species of *Plectus* frequently occurring in samples,were identified and studied in detail using LM and SEM tools. The males could not be found and the populations comprised of females with parthenogenesis as the only mode of reproduction. An inference was drawn on the occurrence, distribution of these species of *Plectus* and their interaction with other cohabiting species to assess the environmental conditions at the wetland. The morphological adaptations were evaluated to assess their role in proper establishment of these species in variable conditions. The inference was also drawn about their niches that enabled them to survive without severe competition. Though some species of Plectus have the potential to form dauer larvae but in the present study dauer larvae could not be found.

A Statistical Model for Estimation of *Pasteuria penetrans* Spore Levels in Naturally Infested Soils

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Pasteuria penetrans is an obligate parasite of root-knot nematodes (*Meloidogyne* spp.). The spores of this bacterium remain in soil and are resistant to high temperatures and desiccation in edaphic ecosystems. There are reports of *P. penetrans* reaching suppressive levels to keep the populations of phytonematodes

below damaging levels. A statistical model was developed for the estimation of *P. penetrans* spore densities in naturally infested field soils, to assess the extent of natural suppression of nematode populations by this bacterium.

Field soil (sand = 90.4%, silt = 2.0%, clay = 7.6%, pH = 7, organic carbon = 0.77%) naturally infested with *Meloidogyne javanica* or *M. incognita* and *P. penetrans* was steam-sterilized at 140° C for 2 h to eliminate both the nematode and the bacterium. In a lab experiment, the steam-sterilized field soil was filled in 5 cm Petri-plates @ 20 g per plate. Required quantities of *P. penetrans* spores were added to the soil in 2 ml of water suspension to create spore levels of 1×10^3, 5×10^3, 1×10^4, 5×10^4, 1×10^5, 5×10^5, 1×10^6 and 5×10^6 per g soil. Second stage juveniles of *M. javanica* or *M. incognita* were added to each plate @ 1000 per plate in 2 ml water suspension. The plates were stored in a BOD incubator at 27° C. The J2 were extracted from each plate after 48 h by sugar centrifugal technique. Twenty J2 were examined microscopically at 400x to count the number of endospores on each juvenile.

A significant positive correlation was established between the number of endospores observed per J2 and the initial spore densities. The data were computed to draw regression equations. In case of *M. incognita,* it was $y = 9.127x - 23.348$ with $R^2 = 0.8467$, and for *M. javanica* it was $y = 8.557x - 22.047$ with $R^2 = 0.8505$. Thus, on the basis of field observations on the spore encumbrance on J2 of *M. incognita* or *M. javanica,* these statistical tools can be helpful to estimate the spore levels in the field soils under a given set of conditions.

Community Analysis of Nematodes Associated with Castor grown in and around Hyderabad

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Castor is a non edible oil seed crop grown in many parts of India. Andhra Pradesh is a leading producer of castor in the country. Seed oil is being used widely for various purposes. Pest and disease were found to be a major constraint for the production of castor. Among them infection due to plant parasitic nematodes were of major importance.

Soil samples were collected from rhizosphere of castor from DOR, Rajendra Nagar and were analyzed for different nematode communities. Samples were taken at the depth of 10-15 cm. Nematodes were extracted from composite soil samples.

The estimation of nematode population was done by multi-chambered counting dish under stereoscopic binocular microscope. Data obtained from soil samples were subjected to the nematode community analysis.

The findings of the investigation undertaken on the study of nematode communities associated with castor reveled that nematodes fall under four trophic groups namely plant parasites, bacterial feeders, omnivorous and Predators. The plant parasitic nematodes recorded were *Helicotylenchus* sp, *Rotylenchulus reniformis, Meloidogyne* sp, *Hoplolaimus* sp and *Aphelenchids*. Bacterial feeders recorded were Rhabditids and cephalobids, ominivores like Dorylamids, and predatory nematodes like Mononchids were also recorded.

Rotylenchulus reniformis was found to be the most abundant and frequent nematode associated with castor grown in DOR farm Rajendra Nagar. Rhabditids and Dorylamids were also found to be equally frequent. *R. reniformis* and dorylamids were found to be the most frequent nematode associated with castor grown in deep black soils of Narkhoda farm of DOR. *R. reniformis* was the most abundant nematode. On comparison of absolute densities of nematodes in both black and red soils of the farm revealed that there is not much difference in the densities of three nematode groups. However, the density of *R.reniformis* is slightly higher in black soil than in red soil and in contrast absolute density of dorylamids were slightly higher in red soil.

Life Cycle of Foliar Nematode, *Aphelenchoides besseyi* Infecting Tuberose, *Polianthes tuberosa*

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Foliar nematode , *Aphelenchoides besseyi,* has been reported to cause appreciable yield loss in tuberose. Since no systematic study about the life cycle of *A. besseyi* has been carried out in tuberose crop, same was investigated in vitro in laboratory condition with ambient temperature of 28^0 C to 35 ^{0}C .Five to six cm long leaf bits of tuberose cv. Calcutta single with spores of *Alternaria solani* were surface sterilized in 1:1000 $HgCl_2$ solution and incubated in a moist chamber for 36-48 hrs. for quick development of fungi. Such leaves were transferred to small ice cream cups with a thin film of moisture. Pre-adult stages of *A.besseyi* isolated from tuberose flowers were inoculated to leaf bits in the ice cream cups and kept inside moist chamber. Development of different stages of nematodes was studied daily by observing under a microscope. New leaf bits were transferred to ice cream cups to meet the feeding requirement of *A. besseyi* as and when required. Pre-adult nematodes fed the *A. solani* grown on leaf bits up to 3 days after which they molted to give rise to adults. Two days after feeding adult females started laying eggs singly on leaf bits. Embryonic development and hatching of second stage juveniles took place immediately within one day. Second stage juveniles took about four days for molting to third stage larvae. They took two days more to become pre-adult stages. The life cycle was completed within a period of 12 days for females and 10 days for males. The nematode reproduced amphimictically where male and females were inoculated to the leaf bits. Parthenogenetic reproduction was observed when only females were inoculated to the leaf bits. The ratio of male to female in amphimictic reproduction was 1:1. In parthenogenetic reproduction the population of females was more as compared to amphimictic reproduction. The population of males was very high in both the type of reproduction when the leaf tips were kept continuously for long time in the mist chamber.

Community Analysis of Plant Parasitic Nematodes Associated with Cashew Nut in and around Bhubaneswar

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Cashew nut as a perennial plantation crop is grown in vast rainfed and dryland areas in the state of Odisha. An attempt was made to investigate the association of plant parasitic nematodes in the rhizosphere of cashew nut plants in five locations in and around Bhubaneswar. Soil samples drawn from 67 cashew nut plants were processed through cobb's sieving and decanting technique followed by modified Baermann technique. Examination of nematode suspensions revealed that eight plant parasitic nematode species associated with cashew nut plants in descending order of their frequencies were *Aphelenchoides* sp. (82.08), *Tylenchus* sp. (53.73), *Rotylenchulus reniformis* (43.28), *Xiphinema insigne* (38.8), *Helicotylenchus dihystera* (17.91), *Hemicriconemoides cocophilus* (17.9), *H. mangiferae* (13.43) and *Hoplolaimus indicus* (7.46). But average density of nematode population in most of the samples was very low, ranging from 1-18.31/200cc soil. Among the eight species, *Aphelenchoides* sp. was found to be the most prominent having

prominence value (pv) = 16.58 followed by *Rotylenchulus reniformis* (7.11), the lowest being recorded for *Hemicriconemoides mangiferae* (0.17).

ECONEMA: Software for Ecological Analysis of Nematode Distribution

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Computation and analysis of the distribution of any organism is vital for studying their life functioning process. In Nematology, frequently surveys for the distribution of nematodes are conducted in various crops and their community structure with several ecological parameters are manually analyzing mainly based on Nortons formulae.

Software 'ECONEMA' has been developed by incorporating the Nortons formulae and other ecological parameters relevant to Nematology by the Scientists of All India Co-ordinated Research Project on Nematodes at the Tamil Nadu Agricultural University (TNAU) Centre. The ECONEMA software code has been written with Visual Basic and dot NET framework, which can be run on commonly used computer operating systems (OS) such as Windows XP and Windows 7. The following community analysis parameters can easily be obtained *viz.*, absolute frequency and density; relative frequency and density; prominence value; importance value; relative biomass and total biomass. In addition, similarity indices between habitats, ratio between fungal feeders and bacterial feeders, omnivours percentage, Simpsons and Shannon-Weiner indices etc. can also be computed easily. The output data is provided with 'PRINT' option for easy compilation of reports.

Effect of Organic Amendments and Nematicide and on Nematode Saprozoic Index in Soil

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A replicated trial was carried out in microplots of size 1.5 m X 3 m in the field to study the influence of three applications of neem seed cake@ 42Kg/ha and FYM

@10tons/ha and carbofuran@ 2Kg ai/ha, at an interval of 2 months, from October to February, to study the influence of agricultural inputs (nematicide and organic amendments) on population densities of free-living and plant-parasitic nematodes infecting Pusa Ruby. The plant-parasitic nematodes encountered were *Rotylenchulus reniformis, Tylenchorhynchus, Pratylenchus sp., Helicotylenchus* sp. and criconematids, with rhabditids, tylenchids and dorylaimids as saprophytic, non-target species. A high SNI (Saprozoic Nematode Index) of 0.68 in neem cake treated plots was indicative of a production system, rich in organic matter, with a greater abundance of free-living saprophytic nematodes. These nematodes play an important role in soil mineralization on interaction with soil bacteria and fungi. In FYM treated plots, a high PPN/FLN ratio of 2.5 was observed, indicating an increase in plant-parasitic nematodes, alongwith free-living species, with the highest average yield among the treatments given. The application of carbofuran reduced the beneficial nematodes and the SNI and gave the lowest yields.

Biology and Life Cycle of Rice Root-Knot Nematode, *Meloidogyne graminicola*

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Successful cultivation of rice largely depends on the careful management of pests and diseases. The rice root knot nematode, *Meloidogyne graminicola* assumes a status of an important nematode problem in rice growing regions of the world. In India, major rice cultivating states are severely affected by *M. graminicola* with considerable yield loss especially the loss is in increasing trend under raised bed/ aerobic system of rice cultivation. Several management options have been employed but with reduced control of nematodes, because of lack of thorough biological background and life cycle pattern. Since, *M. graminicola* is exhibits difference in life cycle duration in different regions of India. A study was conducted to study the biology and life duration of *M. graminicola* occurring in Tamil Nadu. The study revealed the following parameters on the development of the nematode under field conditions: J2-J3 - 1-6 days; J3-J4 - 7-11 days; J4-Adult - 12-18 days and egg mass production- 19-22 days. Life table studies of *M. graminicola* in rice are discussed.

Distribution of Root-Knot Nematode (*Meloidogyne incognita*) in Tuberose (*Polianthes tuberosa* L.) Bulbs and Comparisons of Extraction Methods

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Root-knot nematode, *Meloidogyne incognita* invades, infects and survives in tuberose (*Polianthes tuberosa* L.) bulbs. Detection and estimation of nematode infection in tuberose bulbs is vital to nematode management. Comparison of maceration (either in water or $MgSO_4.7H_2O$ solution), with centrifugal floatation, sieving or flocculation floatation showed better recovery of nematodes When maceration was done with $MgSO_4.7H_2O$ solution. Maximum numbers of eggs, J_2, J_3, J_4, males and females were recovered with maceration centrifugal floatation technique where maceration was done in solution of specific gravity 1.2. Most of the nematodes (58.35%) were present up to a depth of 5.2 mm, in tuberose bulbs. No nematodes were observed at a depth of > 13.0 mm.

Nematode Fauna Associated with Wheat in District Aligarh of Uttar Pradesh

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An extensive survey was carried out on nematode fauna associated with wheat grown in district Aligarh (UP) belt covering villages *viz.*, Jattari, Mohr, Gharbara, Takipur Fatehpur and Rajpur Cereal cyst nematode, the most important nematode pest was encountered in wheat crop with a high population of 25 cyst/250g soil and soil population was above 2 J2/g soil and with absolute frequency of 75% from Rajpur and 55% in Fatehpur. Absolute density of cereal cyst nematode juveniles was reported as one juvenile/g soil.

Further, it was observed that *Tylenchorhynchus* sp. had 60% frequency of occurrence followed by *Hoplolaimus* (50%) root-knot nematode (5%). Besides, population of reniform nematode was also encountered with absolute frequency of 5%. Members of *Tylenchidae* were also promiscuous to wheat crop. Presence of root-knot nematode, *M graminicola* and rice root nematode in wheat crop was perhaps due to rice-wheat crop rotation prevalent in the Aligarh region. It is important to keep track of population build up of cereal cyst nematode in these areas to assess the nematode related losses and damage to wheat crop. The cereal cyst nematode was not reported from these places earlier in wheat.

Current Status of Potato Cyst Nematodes in the Nilgiris of Tamil Nadu

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The potato cyst nematodes (PCN) *viz., Globodera rostochiensis* and *G. pallida,* also popularly called the golden nematodes, are one of the destructive pests affecting the potato production in the Nilgiris district of Tamil Nadu. In order to know the current status of PCN in the Nilgiris and its population dynamics in various cropping systems, a survey was conducted in June, 2011 and all the major potato growing Taluks *viz.*, Ooty, Coonoor, Kotagiri and Kundah were surveyed. Forty six villages were covered and soil samples were collected from the potato fields.

In Ooty taluk, very high intensity of nematodes (> 101 cysts/ 100 ml soil) was recorded in most of the villages surveyed. In Kundah taluk, Emarald, Lawrence and Gandhikandi villages recorded very high intensities while the other villages recorded medium to high intensity of PCN. Low intensity of PCN was recorded from Kheti, Ellanalli, Devarsholai and Gandhipettai villages of Coonoor taluk. In Kotagiri taluk, the intensity varied from low to very high according to the prevailing temperature and cropping pattern in the region.

The nematode population was found higher in fields wherein potato was grown for 3 seasons in a year without any rotation with other crops. The mean number of cysts in potato-potato-potato cropping sequence was 489 per 100 ml soil. Leaving the land fallow for one season and growing of potato for one season in rotation with beet root/cabbage/beans for the other season recorded the minimum population of PCN as 50 cysts per 100 ml soil.

The trend in PCN population was studied in the four taluks of the Nilgiris district based on the current and previous surveys conducted in 1982, 1987 and 2006. An increasing trend in the PCN population was observed in the past three decades. Between 2006 and 2011, the percentage increase in PCN population varied from 7 to 26 per cent in the Nilgiri hills. The increasing trend in PCN population is because of the continuous cultivation of potato and inadequate control measures followed by most of the farmers. This is an alarming sign and unless the farmers are made well aware of the impact of this PCN, in the long run, it becomes impossible to sustain the potato cultivation in this part of the country.

Session IV : Nematode Management

Herbal Control of *Meloidogyne incognita* in Iran

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Water extracts and powders of six medicinal plants (*Citrullus colocynthis* (L.) Schrad., *Tanacetum polycephalum* Schultz Bip., *Teucrium polium* L., *Artemisia sieberi* besser, *Achillea wilhelmsii* C. Koch and *Melia azaderach* L. were studied to find their nematicidal effects in laboratory as well as greenhouse conditions on *Meloidogyne incognita*. Maximum effects on nematode mortality and activity were seen by *M. azaderach* followed by *T. polycephalum A. wilhelmsii A. sieberi, T. polium* and *C. colocynthis*. Concentrations of 2% of water extract and 0.04 of powders were bests on reduction of nematode mortality and activities in laboratory and greenhouse conditions respectively. In comparison to control (tomato cv. Orobata), all plants except (*C. colocynthis*) significantly ($P<0.01$) reduced number of galls, number of egg masses, number of eggs, number of larvae in soil, number of larvae in tomato root and final nematode population. Treatments of 0.04 w/w of seed kernel of *M. azaderach* completely controlled nematode in tomato plant.

Weeds as Host and Non-hosts of *Meloidogyne incognita*

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Twenty species of weeds were examined to found their susceptibility to *Meloidogyne incognita* as a field experiment. Results showed that *Solanum nigrum, Chenopodium album* and *Verbena officinalis* were highly infected by the nematode. High nematode reproduction factors (more than 0.50) were also recorded on *Solanum nigrum* (0.783), *Chenopodium album* (0.567) and *Verbena officinalis* (0.539) in comparison to control which was 1.00. Moderate nematode reproduction factors (0.10 to 0.50) were seen on *Atriplex persicum* (0.206) and *Atriplex tartaric* (0.163). *Amaranthus*

chlrostachys and *Amaranthus reteroflexus* were poor host of the nematode by nematode reproduction factors of 0.055, 0.027 respectively. These two plants can be good in management of *M. incognita* as trap plants. Other examined weeds (*Atriplex lasiantha, Chenopodium foliusom, Digitaria nodosa, Erigeron candensis, Inula britanica, Lactuca serriola, Malva nicaensis, Menta langifolia, Omi majus, Polygonum aviculare, Polypogon monspeliansis* and *Sonchus asper*) were non host to the nematode.

Bio-Management *Meloidogyne incognita* using Fungi and Rhizobacteria

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An experiment was conducted to control root knot nematode by using antagonistic fungi and rhizobacteria. Out of eleven fungi and sixteen bacteria isolates and on the basis of antagonistic effect, only four fungi and six rhizobacteria isolates were selected for their antagonistic effect on *Meloidogyne incognita*. They were studied for suppression of *M. incognita* and root infecting fungi *Fusarium oxysporum* in laboratory and in pot experiment using *Lycopersicon esculentum*. All the selected isolates of fungi and bacteria they not only inhibited egg hatching of root knot nematode and the radial growth of root infecting fungi *in vitro* but also exhibited strong nematicidal activity by killing the second stage larvae of *M. incognita* to varying degree in pot experiment. All the antagonists were cultured in liquid protein supplemented broth medium and culture filtrates were prepared. The exposure of *M. incognita* eggs and juveniles to the culture filtrates showed strong nematicidal activity than the control ($P > 0.05$). The antifungal activity was determined by dual plate culture test between antagonists and root infecting fungi *in vitro*. The toxic principle of antagonistic fungal and bacterial isolates was partially characterized by quantitative determination and SDS- PAGE. All the antagonists showed reduction in root knot and root rot disease and promote plants growth as the length and weight of root and shoot in pot trials on tomato plants.

Chemical Control of *Meloidogyne graminicola* on Rice in Sandy Soils of Haryana under Rice-Wheat System

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To find out the possibility of chemical control of this nematode in rice, experiments were conducted in field and screen house during 2010-11. Rice variety Pusa 1121 was used in the experiments. A field naturally infested with *M. graminicola* was selected in Fatehabad district. Carbofuran @ 1.5 kg a.i./ha was applied 1 and 4 weeks after transplanting (WAT) in half of the field and half was left untreated. In pot experiment, soil collected from the same field in June 2010 was filled in 20 earthen pots (4 kg capacity). Rice was grown for two months for multiplying nematode. Soil of the pots was mixed well and refilled in same size pots (population = 8060J_2/ kg soil). Forty-day-old three healthy seedlings of rice were planted in each pot. The Treatments were: T_1= Carbofuran. @ 1.5 kg a.i./ ha1 WAT, T_2= Carbofuran. @ 1.5 kg a.i./ha 1 and 4 WAT, T_3= Cartap hydrochloride @ 1.5 kg a.i./ha 1 WAT, T_4= Cartap hydrochloride @ 1.5 kg a.i./ha 1 and 4 WAT, T_5= Phorate @ 1.5 kg/ha 1 WAT, T_6= Phorate @ 1.5 kg a.i./ha1 and 4 WAT, T_7= Control (No chemical),T_8= Carbofuran @ 1.5 kg a.i/ha 4 WAT, T_9= Root dip in carbosulfan 1000 ppm for 2 h. In field experiment, observations on root-knot index (RKI) and yield of rice at harvest and, RKI and yield of proceeding wheat crop were recorded. In pots, observations on phytotoxicity (1 WAT), plant growth and RKI (75 days after transplanting) were recorded.

There was no improvement in crop yield after application of carbofuran; and RKI in chemical treated and untreated fields was 5 on 1-5 scale. Yield of wheat (PBW343) remained unaffected with little nematode infection. In pot experiment, root dip treatment with carbosulfan (25 EC) 1000 ppm showed phytotoxicity. No effect of any chemical was observed on RKI and plant growth as compared to untreated control. These results suggest that even the higher doses of chemicals are not effective in controlling *M. graminicola* in sandy soil. Leaching of chemicals as well as nutrients and high nematode population build up in such soils are attributed to be the major factors for ineffectiveness of chemicals and poor yield of rice.

Integrated Pest Management in Vegetables

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Field experiments were conducted twice on tomato, brinjal, okra and cauliflower at Coimbatore, Tamil Nadu in the host country of India under the Integrated Pest Management Collaborative Research Support Programme (IPM-CRSP) funded by USAID, Virginia Tech, USA. A set of packages as Integrated Pest Management for the management of pest and disease including nematodes were experimented for its effectiveness in the above crops and compared with farmer's practice. The results of the experiments indicated that IPM components meant for root knot nematode, *Meloidogyne incognita* management *viz.,* (i) soil application of *Pseudomonas fluorescens* @ 2.5 kg/ha (ii) soil application of neem cake @ 0.25 t/ ha and growing *Tagetes* sp / *Brassica* sp as border crop is effective to suppress the root knot nematode incidence in terms of root gall index to the extent of 60, 60, 60 and 80 per cent and to increase the fruit/ curd yield by 16.66, 18.18, 20.00 and 20.83 per cent compared to farmer's practice in tomato, brinjal, okra and cauliflower respectively.

Management of Root-Knot Nematode, *Meloidogyne* spp. in Tomato Through Bio-Agents Under Protected Cultivation

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A trial was conducted against root-knot nematode, *Meloidogyne* spp. on tomato in the polyhouse at CCS HAU, Hisar during 2010-11 by using various bio-agents (*Paecilomyces lilacinus, Pseudomonas fluorescens* and *Trichoderma viride*) at two doses (1.8 and 2.0 g/m^2). Carbofuran (@ 7 g/m^2) and untreated check were also kept simultaneously to compare the efficacy of bio-agents. All the treatments were replicated thrice with randomized block design. At the time of maturity of the crop, the observations were recorded on plant height, root-knot index and final nematode population. The results revealed that *T. viride* @ 2.0 g/m^2 was found to be most effective in reducing root-knot index (2.5) and final nematode population in soil (163 J2/200 cc soil) over untreated check ((RKI = 4.7 and FNP= 353 J2/200 cc soil) followed by *P. lilacinus* check ((RKI = 2.8 and FNP= 181.8 j_2/200 cc soil).Plant height (262 cm) at harvest was also found to be maximum in the beds treated with *T. viride* followed by carbofuran (241 cm) and was minimum in untreated check (231 cm).

Management of *Heterodera cajani* by Seed Treatment in Pigeon Pea

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The experiment was conducted in the sick plot, Department of Nematology, CCS HAU, Hisar during July, 2009-10 and 2010-11 to find out the most effective non-chemical seed treatment for the management of *Heterodera cajani* infesting pigeon pea. The experiment consisted of seed treatments with neem seed kernel powder (NSKP) @ 10g/kg seed, *Trichoderma viride* @ 10g/kg seed (spore count= 10^8 spores/g talc powder), seed treatment with half dose of NSKP and *T. viride* (each @ 5g/kg seed). Besides these, seed treatment with carbosulfan @ 3.0% (w/w) and untreated check were kept as controls to compare the efficacy of various treatments. Among non-chemical treatments, *T. viride* @ 5 g/kg seed + NSKP @ 5 g/kg seed as seed treatment was found most effective in enhancing pigeon pea yield by 10.71% over untreated check and generated additional income of Rs. 3750/ha. Reduction in cyst and larval population /200cc soil was recorded to be 27.2% and 15.15%, respectively over check. The ICBR of this treatment was 1:20. *T. viride* @ 10 g/kg seed alone was the next effective treatment increasing pigeon pea yield by 8.85% and suppressing the cyst and larval population /200cc soil by 18.8% and 17.29%, respectively over untreated check.

Diversification in Existing Wheat Based Systems for Management of Cereal Cyst Nematode, *Heterodera avenae*

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Wheat is an extensively grown staple crop of India and occupy an unique position as a human diet. Cereal cyst nematode, *Heterodera avenae* is considered to be most important nematode infecting wheat on worldwide basis. In India, it causes a serious disease, locally known as 'Molya' of wheat and barley, was first reported in Sikar district of Rajasthan. Out of the 32 districts in the state, presence of CCN was reported from 18 districts (Ajmer, Aiwar, Bharatpur, Bhiwara, Bikaner, Dausa, Hanumangarh, Jaipur, Jhunjhunu, Karoli, Nagaor Pali, Rajasamund, Sawaimadhopur, Sikar, Sirohi, Tonk and Udaipur). Therefore, present investigation was undertaken to evaluate the diversification of existing wheat crop for management of cereal cyst nematode. An experiment was conducted at Agricultural Research Station, Durgapura, Jaipur in naturally infested soil. Inoculum level was 13.0 larvae/g soil of cereal cyst nematode. The experiment consisted of seven treatments *viz* Mustard, Pea, Gram, Fenugreek, Raj MR 1 (Resistant variety of wheat) along with treated check (Carbofuran @ 1.5 kg ai/ha) and untreated check (Susceptible variety-Raj 1482) in a completely randomized block design. Soil samples were taken from each treatment before the sowing and recorded population of cyst. Each treatment was replicated thrice. The yield was also taken at the time of harvesting of the crop in each treatment separately. Soil samples were taken from each treatment and recorded the number of cyst after the harvest. The results revealed that all the treatments gave significantly reduced the cyst in the soil as compared the control (Higher cyst). Carbofuran @ 1.5 kg ai/ha reduces the nematode population followed by mustard, fenugreek and resistant variety. Pea and gram also reduces the cyst as compared to the control. Diversification, the most effective method to reduce nematode population that affect the two crop season.

Development of *Pasteuria penetrans,* the Bacterial Parasite of Root-Knot Nematode, in *Planta* during *rabi* Season

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In a screen-house experiment the development of *Pasteuria penetrans* was studied on *Meloidogyne javanica* during *rabi* season. Chickpea plants (cv. HC 1) were inoculated with *P. penetrans* encumbered J2 of *M. javanica* on five different dates i.e., 25^{th} Oct., 10^{th} Nov., 25^{th} Nov., 10^{th} Dec. and 25^{th} Dec. The plants were uprooted at weekly intervals to examine the development of root-knot nematode as well as the bacterium. It was observed that the life cycle of host nematode, *M. javanica* was prolonged; although eggs were formed in the genital tracts of females but no egg masses were formed till the end of March. The sporogenesis phase of *P. penetrans* did not occur in any of the five sets. *P. penetrans* development was

restricted to vegetative phase only (microcolonies and thallus stages). The nematode development continued *albeit* slowly; while that of bacterium failed to synchronize with that of the nematode. It is inferred that the minimum temperature thresholds for the nematode and the bacterium are at variance.

Impact of Nutrient Supply on Plant Parasitic Nematodes in Cereal Based Cropping System

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Use of modern agricultural technologies to increase yield has become an effective substrate for land expansion. Moreover, better plant vigour can avoid the parasitic burdon of plant parasitic nematodes imposed on plants. Keeping this in view the present investigation have been under taken with an objective to develop soil ecosystem management strategies to protect against damage caused by plant parasitic nematodes ultimately leading to improvement in plant health.

An experiment to determine the impact of nutrient on population of plant parasitic nematodes, was conducted under field conditions employing 12 treatments of NPK and its combinations with organic fertilizers. Each treatment was replicated five times with fixed plots in both *kharif* and *rabi* seasons following rice–wheat crop sequence.

Soil samples from around the root zones of rice and wheat at a depth of 15 to 20 cm comprising of 10-12 cores /plot. The cores were mixed and homogenized to draw a sub sample (250 cm^3). Samples were analyzed by Cobb's sieving and decanting method followed by Baermann's Funnel Technique. Nematode extraction was carried out for 48 h at room temperature (30±3^{0}C), subsequently counted under stereo-binocular in a counting dish.

The data recorded from the paddy field indicated that the population of *Hirschmanniella oryzae* declined if NPK was incorporated with organic matter *viz.,* FYM, crop residue or green manure. Significant reduction was recorded with NPK 50% + FYM 50%, and NPK 50% + crop residue, NPK50% alone favoured population of *H. oryzae.* Whereas all the treatments favoured population of spiral nematode, *Helicotylenchus dihystera.* Maximum population was recorded with NPK 50% + FYM 50%. However, severe fluctuation in *Tylenchorhynchus* sp. in paddy was noticed during the course of investigations but decline was recorded in all the doses of NPK and NPK 50 % + FYM 50% . Treatments did not have any significant effect on population reduction of *Helicotylenchus* sp. and *Tylenchorhynchus* sp. in paddy

When the field was cropped with wheat during rabi, all the treatments reduced *H. oryzae* in soil. Maximum reduction was observed with NPK organic matter combination. Significantly reduced population was observed in NPK 50% and green manure 50% in the form of *Sesbania*. All the treatments, except no fertilizer, no organic matter, NPK 50% and NPK 75% reduced population of *Tylenchorhynchus* significantly in wheat. Lowest population was recorded with full dose of NPK + compost followed by NPK + green manure 25 per cent.

Pathogenicity of *Meloidogyne incognita* and *M. javanica* on Bottle Gourd

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Bottle gourd [*Lagenaria siceraria* (Mol.) Standl.], a paratropical species of cucurbitaceous family has good source of nutritional components and income to the farmers. India has 1,16,939 hectare area with 1.4 lakh tonne production and 12.21 t/ha productivity of bottle gourd. There are reports of a few diseases, among them, the root- knot nematodes (*Meloidogyne* spp.) adversely affect both yield as well as quality of bottle gourd in central Gujarat. Therefore, present investigation was carried out to determine the threshold level of *M. incognita, and M. javanica* pathotype- 1 and 2 on bottle gourd. Earthen pots of 15 cm diameter were washed and disinfected using 4% formaldehyde (Formalin 40 EC) solution. After drying, pots were filled with steam sterilized soil (1.0 kg/pot). Three seeds of ABG 1 (Anand Bottle Gourd 1), a susceptible bottle gourd variety were sown in each pot. On germination, plants were thinned down to one plant/pot. Second stage juveniles (J2) were extracted from the respective egg masses of *M.incognita, and M. javanica* pt 1 and 2 and inoculated in the rhizosphere of each plant. The inoculum levels tested were 0, 10, 100, 1,000 and 10,000 J2 of both the nematode species. The uninoculated plants served as control. Each treatment was repeated four times in Completely Randomized Design. Plants were uprooted carefully 45 days after nematode inoculation. The results showed that an inoculum level of 100 juveniles (J2)/plant/pot of *M. incognita* and 1,000 J2/plant/ pot of *M. javanica* pt 1 and 2 was detrimental to the growth and development of bottle gourd indicating more pathogenic effect of *M. incognita*. Total nematode population of nematode/plant increased progressively with an increase in inoculum levels from 10 to 10,000 J2/plant. Nematode reproduction rate decreased with an increase in inoculum levels and it was maximum in the level of 10 J2/plant and minimum in 10, 000 J2/plant for all the three populations of root-knot nematode.

Management of Root-Knot Nematode, *Meloidogyne javanica* by integration of Summer Ploughing and Seed Treatment in Okra

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Root-knot nematode, *Meloidogyne javanica* (Treub) Chitwood is a major limiting factor in the production of vegetable crops in general and okra (*Abelmoschus esculentus*) in particular. Considering its importance, among various management strategies, cultural practices and judicious use of nematicides are worth mentioning since these are feasible, effective and economical. So a cultural practice in the form of summer ploughing and carbosulfan seed treatment @ 3.0 per cent a.i. (w/w) were integrated for the management of *M. javanica* in okra at root-knot nematode infested field of Department of Vegetable Sciences, CCS HAU, Hisar. There were two treatments *viz.* untreated okra seeds sown in non-ploughed field (check) and carbosulfan treated seeds sown in summer ploughed field. The initial nematode population was recorded before and after the summer ploughing. There was 34.5 per cent increase in okra yield over check in summer ploughed field sown with okra seeds treated with carbosulfan @ 3.0 per cent a.i. (w/w). The final population of *M. javanica* decreased to the extent of 40.4 per cent over check by summer ploughing alone while at the time of harvest, the final population further reduced to the tune of 27.8 per cent over check by seed treatment with carbosulfan. Similar, trend was observed in respect of final root-knot index thus inferring that combination of summer ploughing during the month of June and seed treatment with carbosulfan @ 3.0 per cent a.i. (w/w) proved very effective for the management of root-knot nematode in okra.

Management of Root-Knot Nematode and Root-Rot Fungus Complex in Cotton Through Bioagents

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Since cotton is attacked by a number of insect pests including root-knot nematode and pathogenic fungi such as wilt and root-rot, the interaction among themselves is bound to happen under natural conditions, thus forming disease complexes in which losses are manifold and hence their management becomes inevitable. In the present study, a complex involving root-knot nematode, *Meloidogyne incognita* and root-rot fungus, *Rhizoctonia bataticola* was managed by use of certain bio-control agents applied as seed and soil treatment in 50 kg pots having root-knot nematode infested soil. Three days after mixing the pathogenic dose of *R. bataticola* in pots, the bio-agents were mixed in the soil and the pots were sown with bioagents treated/untreated seeds of American cotton var., H-1098. The results indicated highest (37.1) per cent increase in seed cotton yield over check by soil treatment of *Trichoderma viride* @ 2.5 kg/ha. The cotton yield, final nematode population in soil and root-knot index showed significant differences among various treatments including carbofuran @ 1.0 kg a.i/ha and untreated check. The final nematode population in soil as well as in roots and root-knot index was minimum and significantly lowest by *T. viride* having root-rot index as nil compared to 13.0 per cent in check. The incremental cost benefit ratio (ICBR) was highest (1:17.5) in seed treatment with *Pseudomonas fluorescens* @ 20 g/kg seed followed by 1:15.3 in *T. viride* indicating thereby that soil application with *T. viride* @ 2.5 kg/ha to be the most promising bio-control agent for the management of root-knot nematode and root-rot complex in cotton.

Endospore Adherence and Infection of Bacterial Parasite, *Pasteuria penetrans* on the Four Common Species of Root-Knot Nematodes, *Meloidogyne* spp. in Haryana (India)

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Four species of root-knot nematode, namely *Meloidogyne javanica, M. incognita, M. arenaria* and *M. graminicola* are considered economically important. The relative

efficacy of a local isolate of bacterial parasite, *Pasteuria penetrans* (isolate KKV) against these four species of *Meloidogyne* was tested. The J2s of four species were exposed to endospore suspension of *P. penetrans*. The endospores readily attached to J2s of *M. graminicola* (16.6 spores per J2), *M. javanica* (14.5 spores per J2) and *M. incognita* (16.7 spores per J2), but the encumbrance rates were nearly half on the J2 of *M. arenaria* (9.5 spores per J2) after 24 h of exposure of J2 in a water suspension of *P. penetrans* endospores. Several endospores attached to J2 of *M. graminicola* were inverted.

The spore-encumbered J2 were inoculated on brinjal plants for *M. incognita*, *M. javanica* and *M. arenaria*, while in case of *M. graminicola* these were inoculated on rice plants in a screen-house. After 40 days of inoculation, the *Meloidogyne* females were dissected out from the roots, crushed and examined microscopically for infection with *P. penetrans*. Spore-filled females were recorded in case of *M. arenaria* (100%), *M. incognita* (80%) and *M. javanica* (70%); while in case of *M. graminicola*, no infection of *P. penetrans* was recorded even in a single female. Thus, *M. graminicola* is a non-host of *P. penetrans* isolate used in this study.

Screening of Urd Bean (*Vigna mungo*) Germplasm for Resistance to Root-Knot Nematode, *Meloidogyne incognita*

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A total of 200 accessions of urd beans [*V. mungo* (L.) Hepper] were screened in potted soil for their host suitability to the root-knot nematode, *Meloidogyne incognita*. Pots were filled with naturally infested soil, containing 4 second stage juveniles (J2)/cc soil. Host status was assessed after 60-days of sowing on the basis of number of root galls induced by nematode. Five accessions *viz.*, IC-8262, IC-251913, IC-485566, IC-485651 and IC-506652 were found resistant with 1-10 root galls/ plant. Rests of the accessions were found susceptible to *M. incognita*. Further, a highly susceptible accession i.e. IC-472000 and five above stated resistant accessions were tested to observe nematode penetration rate (NPR) into the roots and egg mass formation. A significant difference was observed in the NPR between susceptible and resistance accessions. The NPR increased substantially in susceptible accession with the passage of time after inoculation. As NPR was 19.5% and 23.3% of the inoculums in susceptible accession at 5 and 10 days after inoculation, respectively. However, less than 5% NPR was recorded in roots of resistant accessions at 5 and 10 days after inoculation. The number of egg masses observed at 45 days after inoculation comprised 32.3% of the nematode inoculum on susceptible accession, while none or very few egg masses were found on resistant accessions.

Behaviour of International Wheat Nurseries against Indian Population of Cereal Cyst Nematode, *Heterodera avenae* and *Heterodera filipjevi*

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Cereals constitute the world's foremost source of food and feed. The global demand for wheat is expected to increase from 621 million tonnes in 2004-05 to 760 million metric tonnes (mt) in 2020, around 813 mt in 2030 and >900 million tonnes in 2050; this implies growth rates of 1.6% during 2005-20, 1.2% during 2005-30, and 0.9% over 2005-50. Furthermore, the effects of global climate change, namely, the increase in frequencies of drought and heat stress, rise in temperature, coastal inundations, aridity, high/low rainfall, humidity, salinity/alkalinity, lowering /rise of soil water tables, changes in atmosphere gaseous compositions, diseases/pests resurgence etc will ultimately impact on food and livelihood securities. To ensure food security, efforts are made globally so that food production is increased and sustained. There are various biotic and abiotic factors which decide the fate of crop and quantity of yield. For tackling attacks of diseases, insects and nematodes, the most successful method to date against nematode has been the use of genetic host resistance and non host rotational crops which maintain nematode populations below economic threshold for damage. Pursuing this, evaluation against cereal cyst nematode was made in India on wheat. Besides domestic sources, international sources were also utilized and on that line Soil Borne Pathogen Spring Wheat International and Soil Borne Pathogen Winter Wheat International Nurseries were evaluated at 5 locations in India during 2009-2011 against the population of *Heterodera avenae* and *Heterodera filipjevi*. These entries were evaluated under natural as well as artificially inoculated controlled conditions at the locations. The entries have behaved differently across the locations owing to presence of different biotypes of *H. avenae* and *H. filipjevi*. The entries which showed resistant reaction against *H. avenae* failed to express same results against *H. filipjevi*. Even entries failed to yield same reactions against *H. avenae* at different locations. Molecularly these entries, upon analyses, showed presence of *Cre 1, Cre 3, Cre 5* and *Crecon* CCN resistant genes among them and in some cases either in isolation or combination of genes but upon phenotyping they failed to express resistant reactions against Indian population. There are possibilities that the resistance to Indian population may be due to presence of some other minor genes. The analyses and interpretation of the findings shall be discussed at length during the actual presentation.

Plant Parasitic Nematodes Associated with Bt Cotton

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Presently, Karnataka occupies about 5% Bt cotton's acreage in our country. Present investigations on the plant nematodes associated with Bt cotton crop growth in northern Karnataka, India encompassed several aspects like a nematode random survey in Bt cotton growing areas of the state, nematode community analysis, pathogenecity & biology of a dominant nematode and reaction of Bt cotton cultivars and hybrids to that nematode. A random survey undertaken in the Bt cotton growing districts of northern Karnataka (which is a major cotton belt of the state) for the associated nematodes in cotton rhizosphere soil and root samples, showed the presence of plant pathogenic nematodes mainly, reniform nematode (*Rotylenchulus reniformis*), lesion nematode (*Pratylenchus* sp.) and some dorylaimid pathogens. Other plant pathogenic species were present in less numbers. Community analysis showed that the forementioned nematodes were important in the cotton growing areas surveyed. Pathogenic nature of reniform nematode on a Bt cotton cultivar was demonstrated in greenhouse studies, a first such effort in India, in respect of this important nematode pathogen: The said nematode required 27-29 days for the completion of one generation under Dharwad conditions. Many inter- and intra-specific *hirsutum Bt* hybrids were not found susceptible to the reniform nematode infection, but MRC-7918 and Tulsi 117 were found to be moderately susceptible.

Management of Nematodes Infecting Cardamom (*Eletteria cardamomum* Maton.) using Biodynamic Preparation

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Cardamom is cultivated as an under storey crop in the tropical evergreen forests of western ghats of South India. India is the second largest producer of cardamom in the world. Kerala is the leading producer in the country producing contributing 60 per cent of production. Plant parasitic nematodes were reported to be an important constraint in successful cultivation of cardamom. In this context, the present study gains importance.

Plant growth parameters of Cardamom were evaluated by the application of the following treatments. T1 – Recommended dose of FYM (30 t/ha), T2 – Recommended dose of Coir pith compost (15 t/ha), T3- Jeevamruta, T4- Recommended dose of FYM (30t/ha) + *Azospirillum* (10g. / clump)+ 10g. PSB/ clump + *Trichoderma* 10 g. / clump, T5- Jeevamruta+(10g. / clump)+ 10g. PSB/ clump + *Trichoderma* 10 g. / clump. T5 was found to be significantly better treatment in increasing the plant height in comparison to other treatments. Treatments T1 to T4 were found to be on par in their effect. Number of leaves were found higher in treatment T5 followed by T3 and T4. However, all these treatments were found to be on par. The treatments were found significant among themselves in the case of No. of panicles. T3 recorded highest panicle length followed by T5 in the case of panicle length. However, these two treatments were found on par. T3 recorded highest number of racemes and lowest was recorded in T1. In general, treatment T5 (Jeevemrutha+ Azospirillum 10g/ clump+PSB 10 g/ clump+ *Trichoderma* 10 g/ clump) was found to be the best treatment followed by T3 (Jeevamrutha alone). The increased plant growth and yield parameters as a result of application of organic inputs is due to increased activity of soil microflora like bacteria, fungi and actinomycets and soil fauna like nematodes and protozoa. Similar results were recorded by Kumar *et .al.,* 2009 on cardamom. The results of their experiment revealed that application of 100% organic manure in the form of FYM enhanced capsule yield by 34.1 per cent over control.

Integrated Management of Foliar Nematode *Aphelenchoides besseyi* Infecting Standing Tuberose Plantations

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Tuberose, *Polianthes tuberosa,* is one of the most important flowers cultivated throughout the country having high commercial value. Floral malady and floret necrosis of tuberose caused by foliar nematode *Aphelenchoides besseyi* has become a major concern for the tuberose growers of Odisha. This nematode affects tuberose plants at various stages causing reduction in growth, yield, quality and marketability of flowers. Farmers usually use infected planting materials and incur heavy loss subsequently being failed to manage this important nematode parasite in standing crop with various pesticides indiscriminately or replacing the plantation frequently by new crop. Therefore, a field experiment was conducted during 2008-09 to determine an ecofriendly and economic management schedule for this nematode in standing tuberose crop. The trial was carried out with 10 treatments replicated four times in a two year old nematode infested tuberose plantation (cv. Calcutta Single). Treatment combinations of field sanitation by removal of dried/ fallen leaves and old spikes with three sprayings of either Sesamum oil @ 0.2% along with 1 ml of sticker (Teepol) per liter of spray solution or Imidacloprid @ 0.025% at monthly interval as well as soil amendment by Sesamum oil cake @ 1.5 t/ha followed by three sprayings of either Quinalphos @ 0.2% or Imidacloprid @ 0.025% at monthly interval during peak flowering period managed the floral malady and floret necrosis problems of tuberose effectively with increased plant growth parameters and flower yield significantly reducing the nematode population both in soil as well as in plant parts.

Reaction of Coleus Cultivars against Root-Knot Nematode, *Meloidogyne incognita* and its Impact on Quality

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Roots and tuber crops are among the world's most important food crops, with a great potential to improve food security and alleviate poverty in resource poor countries. Koorka or coleus, *Solenostemon rotundifolius* is a short duration tuber vegetable, mainly cultivated in Kerala. One of the major constraints in production of tubers is the losses sustained due to attack by nematodes. The over dependence on pesticides can be avoided by identifying a resistant variety against major nematode pests In this context, the present study was undertaken with an objective to screen varieties /lines/accessions against *M. incognita* to select a suitable resistant one. Since the wild related forms with poor quality were coupled with resistance reaction, hence the impact on the quality of tubers was also assessed.

Two improved varieties, five lines, two accessions and one local variety were screened for comparing the relative tolerance to *M. incognita*. The cuttings collected from CTCRI, Vellanikkara, Pattambi and Palappoor were selected for screening. The soil was inoculated with *M.incognita* at an inoculum level of one juvenile per g of soil 15 days after planting. One litre denematized soil per pot was applied forty five days after planting to promote tuberisation.

The biometric characters, yield and nematode population characteristics were compared with the susceptible check, Palapoor local. The variety Sree Dhara ranked first in biometric characters and yield closely followed by the variety Nidhi. The variety Sree Dhara showed statistically significant variation from the rest of varieties/ lines accessions in reducing the nematode population characteristics. Lowest number of larvae, females, eggmasses in root and eggs per eggmass were recorded in Sree Dhara. This variety also recorded minimum root-knot index of 1.00. The level of total sugar and starch content decreased with increase in infestation. The susceptible check, Palappoor local recorded lowest content of total sugar, starch and crude fibre, while least reduction of the above content was observed in moderately resistant variety Sree Dhara. Hence variety Sree Dhara was identified as a resistant variety against root- knot nematode infestation in *S. rotundifolius.*

Host Status of Various Forest Seedlings to Root–Knot Nematode (*Meloidogyne* spp.)

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The present study was carried out to the see the host response of various forest seedlings to root- knot nematode (*Meloidogyne* spp.).For this purpose, three seedlings, each of 11 plant species obtained from various forest nurseries of Hisar and Bhiwani districts of Haryana, were transplanted in 30 cm sized pots filled with root- knot nematode infested soil having a mixed population of *Meloidogyne incognita* and *M. javanica* (INP 320 J2/ 200CC soil) during August, 2010.Two months after planting, the seedlings were uprooted, washed and observed for root–knot index (1 to 5 scale) and final nematode population for assaying their host status.

The results revealed that seedlings of Tamarind (*Tamarindus indica*), Bakain (*Melia azadirach*), Ailanthus (*Ailanthus* sp) and Amaltash (*Cassia fistula*) showed non-host response without any galls and egg masses. Neem (*Azadirachta indica*) and Jamun (*Syzygium cumini*) showed poor host response with a root-knot index less than 2.0. Desi kiker (*Acacia nilotica*) and Shisham (*Dalbergia sissoo*) showed moderate host response with a root-knot index 2.1 to 4.0. Mulberry (*Morus alba*), Papaya (*Carica papaya*) and Sukhchain (*Pongamia pinnata*) showed good host response with a root-knot index of 4.0 and above.

Evaluation of Antagonistic Potential of Rhizobacteria against Root-Knot Nematode (*Meloidogyne* spp.) in Vegetable Crops

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Six rhizobacterial strains (RKB-68 and RKB-91 of *Bacillus* spp.; AVK 41, AVK 51 and HT 54 of *Azotobacter chroococcum* and 35-47 strain of *Gluconacetobater diazotrophicus)* were evaluated against root-knot nematode in smooth gourd, okra and tomato in root-knot nematode infested field of the Department of Nematology, CCSHAU-Hisar, having a mixed population of *Meloidogyne incognita* and *M. javaica*

(INP: 220 J2/200 CC soil). Results revealed that in smooth gourd, AVK-51 strain showed maximum increase in yield (10.8%) followed by *G. diazotrophicus* 35-47 (9.5%) and strain HT 54 (8.1%) in comparison to untreated check. Root-knot index in various rhizobacterial treatments ranged between 3.2 to 3.8 in comparison to 4.2 in untreated check. In okra, maximum increase in yield was observed in HT 54 strain (10.8%) followed by *G. diazotrophicus* 35-47 (9.5%) and RKB-68(8.2%) in comparison to untreated check. At harvest, root-knot index was found minimum (3.3) in *G. diazotrophicus* 35-47 and RKB68 followed by 3.5 in HT 54 and AVK 51 in comparison to 4.3 in untreated check. In tomato nursery, no significant difference in plant height and weight was observed between treated and untreated check. However minimum number of galls per plant was observed in RKB-68(7.0) followed by HT 54 (7.6) in comparison to 13.3 in untreated check. At harvest, tomato yield/ plant in various treatments ranged between 154.2 g to 159.7 g/m^2 in comparison to 148.4g in untreated check which were non significant. Root-knot index at harvest in various bacterial treatments (3.5-4.0) were statistically at par with untreated check (4.3).

Avoidable Losses Due to Root-Knot Nematode (*Meloidogyne incognita*) in Fenugreek

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Fenugreek (*Trigonella foenum-graecum*), an important seed spice crop of Haryana, is highly vulnerable to root knot nematode(*Meloidogyne* spp). Keeping in view the common occurrence of this nematode on Fenugreek, a systematic experiment was planned at farmer's field infested with root-knot nematode (*M. incognita* having INP: 280J2/200CC soil) in Dharnia village of Fatehabad district of Haryana during Dec., 2010. The experiment consisted of two treatments (T1= Soil treatment with carbofuran @ 2.0 kg a.i. /ha, T2= Untreated check) with ten replications. At the time of maturity of the crop, Observations were recorded on yield, root-knot Index and final nematode population. The results revealed that avoidable loss in fenugreek due to *M. incognita* was found to be10.2% resulting in19.5% reduction in root-knot index and 25.0% reduction in final nematode population in comparison to untreated check.

In-vitro Evaluation of Native *Pseudomonas* spp. Isolates against Root-Knot Nematode, *Meloidogyne incognita*

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Pseudomonas spp. an nematicidal bacterium is widely used as a bio-agent for a number of soil borne diseases and root-knot nematode, *Meloidogyne* spp. During a Survey, 12 isolates of *Pseudomonas* spp. were isolated from rhizosphere of Cucurbits, Tomato, Brinjal, Bangalore blue grapes, Citrus, Guva, Cabbage and Groundnut crops cultivated in Bangalore (Rajankuntae, Singnayakanhali) and Pavgad region of Kolar district of Karnataka. These isolates were evaluated for their nematicidal potential under laboratory experiments. In laboratory studies eggs and juveniles were exposed to each isolates of *Pseudomonas* by diluting the standard filtrate to 50% and undiluted filtrate constituted 100% for different time intervals the cell free culture filtrate not only inhibited the hatching of eggs but also showed the mortality of second stage juveniles of root knot nematode at different time intervals. 4 isolates of culture filtrate inhibited hatching and induced significant levels of mortality of *M. incognita* compared to sterile water *in vitro*. The level of mortality increased with an increase in the culture filtrate concentration from 50 to 100% and with the length of the exposure period from 12 to 48hrs. The significant suppression of egg hatching and mortality of second stage juveniles of *M. incognita* increased with increase in culture filtrate concentration and exposure period recorded in 4 native *Pseudomonas* spp. isolates. The better suppression of egg hatching is recorded in isolate three (7.33 to 38.66%) followed by mortality of second stage juveniles (14 to 56%) at 50% concentration of culture filtrate exposure period up to 48 h compared to non treated control.

Comparative Virulence Studies of Sorghum Cyst Nematode, *Heterodera sorghi* Populations of India

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The sorghum cyst nematode, *H. Sorghi* is widely distributed in India and is presently growing in importance due to rapid increase in growing area under maize and sorghum crop in India. Originally first time described from Ghaziabad district of Uttar Pradesh later reported from Maharashtra, Himachal Pradesh, Jammu Kashmir, Andhra Pradesh and Haryana. Keeping in view the widespread distribution of *H. Sorghi* as a potential nematode pest on sorghum and maize crop 5 populations of *H. Sorghi* from Himachal Pradesh (Kulu, Chamba, Tihri), Uttar Pradesh (Ghaziabad) and Jammu & Kashmir (Kud) are selected for conducting comparative virulence studies in glass house condition on maize (cv Prabhat). The maize seeds were planted singly in 15cm earthen pots. Six days old seedlings were inoculated with three levels of inoculums namely, 2, 4 and 6 J2/cc soil. Besides, uninoculated seedlings without nematode served as check. The trial comprised of 4 treatments including checks and each treatment was replicated five times for all the populations. Observations on different plant growth parameters and nematode multiplication were recorded after 60 days of nematode inoculation. Decrease in shoot lengths were found to be non significant. However, reduction in fresh weights of shoot and root was found to be significant at initial inoculums level of 4 J2/cc soil and above. The maximum per cent reduction of fresh shoot weight (g) of 35.47, 35.27, 22.02, 27.16, 24.50 and in fresh root weight reduction of 43.94, 39.23, 19.79, 26.56, 34.85 in Ghaziabad, Kud, Kulu, Tihiri and Chamba respectively. While in dry shoot weight maximum reduction of 49.33, 68.84, 43.06, 63.10, and 47.75 and similarly in dry root weight maximum reduction of 53.71, 43.87, 62.23, 43.28 and 44.81 in Ghaziabad, Kud, Kulu, Tihiri and Chamba respectively.

Wider Area Validation of Adaptable Integrated Management Technology against Root-Knot Nematode, *Meloidogyne graminicola* in Farmers' Participatory Approach

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Wider area validation and economic analysis of adaptable Integrated Root-Knot Nematode (Meloidogyne graminicola) Management technology in rice in farmers' participatory approach was carried out in adopted village Akkihebbalu, K.R.Pet, Mandya district, Karnataka during 2008 (20 ha) , 2009 (20 ha) and 2010 in the 437 acre area covering 167 farmers' families. The Integrated Nematode Management technology (INMT) for rice comprised raising rice nursery in cabrofuran (0.3g a.i/sq.m) treated beds followed by its field application @ 1 kg a.i/ha 40 DAT(T1) or applying *Pseudomonas fluorescence* @ 20 g/sq.m in soil nursery (T2) or seed treatment of rice with *Trichoderma viride* @ 4 g/kg seed in nursery (T3) . The adoption of INMT resulted in reducing the nematode population from 320 J2 /200 c.c soil as initial nematode population to 135(T1) ,165 (T2) and 192 (T3) per 200 cc of soil in the respective treatments thereby leading to increase yield 47.25q/ha, 46.79q/ha and 42.99q/ha in T1,T2 and T3 respectively in comparison to 38.10q/ha in untreated control (T4).

Interactive Effect of Herbicide and Nematicides on Cereal Cyst Nematode, *Heterodera avenae* Infecting Wheat Under Field Conditions

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Weeds are alternative hosts for plant-parasitic nematodes and have long been recognized for their ability to maintain nematode populations targeted for suppression by various management strategies. Shrinking nematicide options and increasing environmental concerns are making integrated pest management (IPM) a necessity for nematode management in many crops. A prominent similarity between most major weeds and plant-parasitic nematodes is that both are place-bound organisms that are passively dispersed. Weed-nematode interactions in agricultural production systems may be more intricate and complex than the simple function of weeds as alternative hosts. Their relationship may represent a normal adaptation resulting from the limited mobility of both groups of organisms and the obligate parasitism of phytophagous nematodes. Thus a field trial was conducted at IARI, New Delhi to study the effect of post-emergence herbicide, Pinoxaden and other chemicals (carbofuran and phorate) on the cereal cyst nematode, *Heterodera avenae* population (initial population of 40 – 92 cysts per 200 cc soil) infecting wheat. Carbofuran and phorate @ 1 and 2 kg a.i./ha were applied in the field at the time of sowing followed by foliar application of Pinoxaden @ 40, 50 and 60g/ha 30 days after sowing (DAS) in the plots of size of 20 m^2. All the treatments were replicated thrice alongwith the control. It was observed that application of carbofuran @ 1 and 2 kg a.i./ha was effective in reducing the cyst population (16-20 cysts/ 200 cc soil), followed by the spray application of Pinoxaden at higher dosage (50 and 60 g/ha). However, combined application of carbofuran and phorate both @ 1kg a.i./ha followed by spray with Pinoxaden @ 60g/ha showed similar reduction in the cyst population.

The challenge that faces weed scientists and nematologists is to identify effective, compatible IPM strategies that address weed and nematode management collectively. The interactive study could be the viable option to manage weeds as well as cyst nematodes in wheat under field conditions.

Screening of Wheat Varieties for Resistance against *Pratylenchus thornei* and Effect of Temperature on its Reproduction

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Root lesion nematode, *Pratylenchus thornei* is an important nematode parasite of wheat in many parts of world. Twenty varieties/lines of wheat, *Triticum aestivum*, three varieties of *T. durum* and three known sources of resistance to *Heterodera avenae* were evaluated against *P. thornei*. Effect of sowing time variation (Oct.30, Nov.15 and Dec.5) using PBW 343 variety on nematode infection was also studied. Nematode population in soil plus root was estimated three months after inoculation. On the basis of reproduction factor (Rf) varieties / lines were categorised as resistant (Rf <1) or susceptible (Rf e"1). Of the 20 varieties/ lines, nine namely, AUS 15854, PBW 343, PBW 550, Raj MR 1, Raj 3765, CIMMYT line (CROC_1/AE. SQUARROSA (224)//OPATA), WH 542, WH 896 and WHD 943 were rated as resistant where as 11 varieties *viz.*, C 306, DBW 16, DBW 17, PBW 373, UP 2425, WH 147, WH 711, WH 912, WH 1021, WH 1025 and WH 1080 were rated as susceptible. All the three known sources of resistance against *H. avenae* (AUS 15854, CIMMYT line and Raj MR 1) were also found resistant to *P. thornei*. Final population and reproduction factor of *P. thornei* increased with the delay in sowing time. No lesions or root browning was discernible on inoculated plants.

Eco-Friendly Management of Potato Cyst Nematodes (*Globodera* sp.) Under Field Conditions

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The potato cyst nematode (PCN), *Globodera spp.* is responsible for large yield losses in potato crops in infested areas. The eggs present in the hard coated cysts hatch out only in the presence of root exudates of host plants i.e., susceptible varieties of potatoes. Non-availability of host plant along with the application of synthetic chemicals is expected to control this soil pest without much negative impact on the environment. Thus, it is desirable to find environmentally friendly alternative measures to control this nematode species. In this study we evaluated the effects of seven different crop rotations of susceptible and resistant varieties of potato along with three non-host crops of PCN on its population in a two year long rotation. The results were expressed as number of cysts per 100 ml of soil after harvest of each crop, Rf (Reproduction factor) value of PCN after two years of cropping, yield of each crop in terms of potato equivalent yield (PEY) and economics in terms of net returns and benefit: cost ratio. The lowest numbers of cysts were recorded (Rf value : 0.67) from the treatment in which cabbage along with a resistant crop of potato was grown in between two susceptible potato crops, along with the application of nematicide carbofuran @ 2.0 kg a.i./ha for each potato crop. However, higher economic returns (BC ratio : 3.40) were recorded in the treatment potato – cabbage – carrot – potato, along with carbofuran application to potato.

Assessment of Avoidable Loss Due to Stunt Nematodes in Bidi Tobacco Nursery

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Bidi tobacco (*Nicotiana tabacum* L.) grown in central Gujarat is found to be attacked by root-knot (*Meloidogyne incognita* and *M. javanica*), reniform (*Rotylenchulus reniformis*) and stunt (*Tylenchorhynchus vulgaris*) nematodes in nursery as well as field crop. Estimation of losses in production of transplants and cured leaf yield of bidi tobacco in Gujarat due to root-knot disease revealed 51 and 31-50 % loss, respectively. Infection of reniform nematode delayed the growth and development of bidi tobacco seedlings by 38 days causing 22.1% avoidable loss at I[st] pulling (38 days) and over all loss to the tune of 5.51% in production of transplants. However, no quantified information about avoidable loss is available with respect to stunt nematodes in bidi tobacco nursery. Hence, the present investigation was carried out for three consecutive years (2007-08 to 2009-10) to assess the loss due to the stunt nematodes in bidi tobacco nursery. Twenty eight beds, each of 1.2x1.2m size, were prepared in bidi tobacco nursery infested with stunt nematodes. Two treatments i,e. drenching of carbosulfan (Electra 25 EC)@ 2.5 l/ ha (CAR), using 2 litre solution/sq. m, a day prior to seeding and 25 days after seeding (25 DAS)and a control-only water (CON)were applied in paired design keeping fourteen replications. Each bed was seeded with bidi tobacco cv. Anand 119 @ 5 kg/ha. Soil samples from each bed were procured before applications of the treatments, at seeding and at the end of the experiment and analyzed for nematode assay. The results revealed that though the differences were not significant, infection of stunt nematodes caused 14.5% avoidable loss at I pulling and over all loss to the tune of 14.1% in production of transplants in bidi tobacco nursery. This ultimately caused loss of 70,000 transplants/ha. ICBR worked out for carbosulfan @ 2.5 l/ha comes to 1: 4.60 and net realization of Rs. 55,000/ha.

Integrated Disease Management in Bidi Tobacco Nursery

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Study on Integrated Disease Management in Bidi Tobacco Nursery was carried out at Bidi Tobacco Research Station, AAU, Anand in Randomized Block Design with bed size of 1.2 x 1.2 sq. m for each treatments replicating three times. All the effective and economical components *i.e.* soil solarization, rabbing, carbosulfan, metalaxyl MZ and carbendazim in management of either root-knot, damping-off or frog-eye spot disease were tried along with vermi compost and castor cake making six IDM schedules i.e., 1. Vermi compost @ 4 t/ha + soil solarization for 15 days during April-May using clear LLDPE film of 25 um + carbosulfan @ 2.5 l/ha drenching using 2 l solution/ sq. m 15 days after seeding (DAS)+metalaxyl MZ @ 2.16 kg/ha drenching as and when required+carbendazim @ 0.025% spraying at the initiation of frog-eyes pot disease, 2. Castor cake @ 4 t/ha + as in Treat.1 except vermi compost, 3. as in Treat.1 except vermi compost, 4. Rabbing with bajra husk @ 7 kg/ sq. m in the month of June + as in Treat.1 except vermi compost, 5. Soil soiarization + carbosulfan a day prior to seeding and 25 DAS +metalaxyl MZ + carbendazim, 6. As in Treat. 5 except soil soiarization. Proper control was also kept for comparison. Bidi tobacco Anand 119 was seeded @ 5 kg/ha and watered regularly. The results revealed that all the individual components *i.e.* soil solarization, rabbing, carbosulfan, metalaxyl MZ and carbendazim found effective in management of either root-knot, damping-off or frog-eye spot disease can be used as a IDM schedules for effective and economical management of damping-off, frog-eye spot, root-knot diseases and in increasing number of transplantable and total surviving seedlings in bidi tobacco nursery. Number of weeds was also reduced by all schedules compared to control barring the schedules of vermi compost and carbosulfan.

Effect of Biofertilizers on Root-Knot Nematode *(Meloidogyne incognita)* infecting Tomato

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An investigation was carried out to study the effect of some biofertilizers on root knot nematode (*Meloidogyne incognita*) infecting tomato var. Pusa Ruby in pot culture condition, The experiment comprising of 8 treatments each replicated thrice in complete randomized design, in order of complete biofertilizer *(Azospirillum + Azotobacter +* Phosphorus solubulising bacteria) @ 6kg/ha, VAM *(Glomus fasciculatum)* @ 100kg/ha, recommended dose of NPK@ 125 kg N- 50kg P-100kg K/ha, complete biofertilizer @ 6kg/ha + VAM@ 100kg/ha, VAM@ 100 kg/ha + NPK@ 125 kg N-50 kg P- 100kg K/ha, complete biofertilizer @ 6kg/ha + NPK@ 125 kg N-50 kg P- 100kg K/ha, Carbofuran @ 1 kg ai/ha as standard check and an inoculated check were treated in the respective replicated pots containing 1kg steam sterilized soil each. Fifteen days old tomato seedlings were transplanted one in each pot after treatment. Seven days after treatments, 1000 J_2 of *Meloidogyne incognita* were inoculated in each replicated pot. The experiment was terminated 45 days after inoculation. From the experimental findings it was evident that barring the inoculated check, all other seven treatments significantly increased the plant growth parameters and reduced the infection parameters and nematode multiplication. However, application of VAM *(Glomus fasciculatum)* @ 100 kg/ha + recommended dose of NPK @ 125 kg N-50 kg P-100 kg K/ha performed the best by increasing in shoot and root length, fresh shoot & root weight, increased biomass production

with the corresponding decrease in root galls (74.47%), egg masses (83.28%), root knot nematode population in soil (73.69%) and in root (64.87%) over inoculated check followed by VAM alone.

Bioefficacy Evaluation of Controlled Release Nano-Formulations of Azadirachtin-A against *Meloidogyne incognita* in Tomato

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Pesticides containing nano-scale active ingredients are already on the market, and many of the world's leading agrochemical firms are conducting R&D on the development of new nano-scale formulations of pesticides. In the present study, controlled release formulations of azadirachtin-A, a bioactive constituent derived from the seed of *Azadirachta indica* A. Juss (Meliaceae), have been prepared using commercially available polyvinyl chloride, polyethylene glycol and laboratory synthesized poly(ethylene glycol) based amphiphilic copolymers. Copolymers of polyethylene glycol and various dimethyl esters, which self assemble into nano micellar aggregates in aqueous media, have been synthesized. Kinetics of azadirachtin-A, release in water from the different formulations was studied. Release from the commercial PEG formulation was faster than the other CR formulations. The rate of release of encapsulated azadirachtin-A from nano micellar aggregates is reduced by increasing the molecular weight of PEG . The diffusion exponent (n value) of azadirachtin-A, in water ranged from 0.47 to 1.18 in the tested formulations. The release was diffusion controlled with a half release time ($t_{1/2}$) of 3.05 to 42.80 days in water from different matrices. The results suggest that depending upon the polymer matrix used, the application rate of azadirachtin-A, can be optimized to achieve insect control at the desired level and period.

The developed CR formulations were evaluated against *Meloidogyne incognita*. A seedling dip experiment was conducted in 15 cm earthen pots containing 500cc autoclaved soil sand mixture. A month old seedlings of tomato cv. Pusa Ruby was dipped in the solutions of Aza @ 5, 10, 20 and 40 ppm with polymer (AP0, AP1 and AP2). Each seedling was transplanted and freshly hatched juveniles (J2s) of root-knot nematode, *Meloidogyne incognita* were collected from pure culture, were used for inoculation @ 2J2s/cc soil. The observations on penetration and development of J2s were taken for 7, 21, 28 days post inoculation (Dpi). In general the penetration of second stage juveniles decreased with the increase in the concentration of Aza from 5 to 40 ppm. AP2 @ 10ppm improved the plant growth in terms of shoot length followed by @ 5 ppm. The effectiveness of polymers were as follows P2>P1 compared to without polymer. The Aza-A-PEG 1500 (AP2) which showed the lowest population of nematodes provided significantly superior control amongst all.

Influence of Fly Ash on *Meloidogyne incognita*

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Influence of fly ash of pH 8.1 and EC 0.15 on egg hatching, *in vivo* juveniles (J_2) and root penetration by root knot nematode *Meloidogyne incognita* in tomato under glasshouse conditions (± 28°C) was studied. The experimental results revealed that the fly ash extract inhibited egg hatching and caused mortality of juveniles of *M. incognita* and its effect was directly related to the concentration (0.5 to 0.1 per cent). However the per cent root penetration by *M. incognita* in tomato is inversely correlated to the concentration of fly ash extract. The highest per cent inhibition of egg hatch (29.50), *in vivo* mortality of juveniles (28.40) and lowest per cent root penetration (22.0) of *M. incognita* was observed with 0.5 per cent fly ash extract.

Screening of Banana Cultivars for Dual Resistance to *Pratylenchus coffeae* and *Meloidogyne incognita*

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The root-lesion nematode, *Pratylenchus coffeae* and root-knot nematode, *Meloidogyne incognita* are economically important nematode pests of banana and are widely distributed in all banana growing regions of the country. Crop losses caused due to *P.coffeae* in cv.Nendran and *M.incognita* in cv. Poovan are 44.4% and 30.4 respectively. Breeding for host plant resistance is a promising strategy for controlling nematodes. Hence, study was undertaken to evaluate 68 core

collection of *Musa* germplasms belonging to 24 diploids (AB-6; AA-4 &BB-14), 41 Triploids(AAA-6; ABB-15; AAB-20), Tetraploids(ABBB-2) and one hybrid (H-201) against root-lesion nematode, *P. coffeae* and root-knot nematode, *M. incognita* separately in pots under shade net condition for locating the resistant/tolerant reaction to both nematodes. The results of the study indicated that 5 diploids (Kunnan, Borkal baista, Gragric sarpara, Elavazhai and *Musa* ac.ssp.burmanica) and 9 triploids (Dasaman, Chirapunji, Kalibow, Amrithapani, Terabun, Thenkadali, Ladan, Padathi and Ennabenian) were found resistant to both nematodes. Individual nematode performance revealed that 3 diploids (Agniswar, Narmine and Andaman balbisiana,); 3 triploids (Bungan, Chinia and H-3) and one hybrid (H-201) were found resistant to root- lesion nematode, *P.coffeae* whereas 5 diploids and 15 triploids were found resistant to root-knot nematode, *M.incognita.*

Field Evaluation of Controlled Release Nano-Formulations of Carbofuran against *Meloidogyne incognita* infecting Tomato

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The root-knot nematode, *Meloidogyne* spp. is one of the most widespread nematode pests, limiting the world agriculture productivity. Out of four important *Meloidogyne* spp., most of the plants are susceptible to the infection of *M. incognita*. Carbofuran [2, 3–dihydro-2,2-dimethylbenzofuran-7-yl methylcarbamate], is a systemic insecticide-nematicide for soil and foliar treatments. Being a carbamate it inhibits the cholinesterase enzyme and inactivates the nervous system. The effective duration of carbofuran from its controlled release (CR) formulation is higher than its commercial formulation for managing *M. incognita*. Thus, a CR formulation could be one of the options to manage the important nematodes in vegetable crops like tomatoes.

In the present investigation, bioefficacy of developed carbofuran formulations, with PEG-600 (CP1) & PEG-900 (CP2) @ 5, 10 and 20 ppm, alongwith commercial formulation of carbofuran 3G (CP0) were evaluated against the root-knot nematode, *Meloidogyne incognita* infecting tomato (cv. Pusa Ruby) in pot and field conditions. The bioefficacy data indicated that the formulations developed by utilizing polymers having PEG – 900 as hydrophilic segment were effective even at 14 days post inoculation (dpi) as evident from shoot and root length. In pot experiment, it was observed that the developed CR formulation at 5 and 10 ppm significantly increased the plant growth characters such as shoot and root length of tomato compared to commercial formulations (CP0) at all the time intervals 7, 14 and 21 dpi. Maximum shoot and root length was recorded with CP2 at 10 ppm concentration. Also, the reduction in penetration was found to be maximum with CP2 (3.6 – 4.6 J2s) at all concentrations compared to CP1 (6.6 – 16.4 J2s) and CP0 (29.3 – 32.6 J2s). Overall, CP2 was more effective in reducing the number of nematodes upto 14 days, compared to CP1 and CP0. Both the CR formulations (CP1 and CP2) in general significantly reduced the number of galls, when compared to CP0. The number of galls at 28 dpi decreased with the increase in the concentration with CP1and CP2. CP2 at 10 and 20 ppm was significantly more effective under pot conditions. However, under field conditions, lower concentrations (5 and 10 ppm) of CP2, were less effective in controlling the gall formation whereas, CP2 at 20 ppm, was most effective than other treatments. The study revealed that the developed CR formulations of carbofuran have the potential for effective management of *M. incognita* in tomato under field conditions.

Biology and Life Cycle of Rice Root-Knot Nematode, *Meloidogyne graminicola*

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Successful cultivation of rice largely depends on the careful management of pests and diseases. The rice root knot nematode, *Meloidogyne graminicola* assumes a status of an important nematode problem in rice growing regions of the world. In India, major rice cultivating states are severely affected by *M. graminicola* with considerable yield loss especially the loss is in increasing trend under raised bed/ aerobic system of rice cultivation. Several management options have been employed but with reduced control of nematodes, because of lack of thorough biological background and life cycle pattern. Since, *M. graminicola* is exhibits difference in life cycle duration in different regions of India. A study was conducted to study the biology and life duration of *M. graminicola* occurring in Tamil Nadu. The study revealed the following parameters on the development of the nematode under field conditions: J2-J3 - 1-6 days; J3-J4 - 7-11 days; J4-Adult - 12-18 days and egg mass production- 19-22 days. Life table studies of *M. graminicola* in rice are discussed.

Effect of Certain Biocontrol Fungi and Bacteria on Root-Knot of Rice caused by *Meloidogyne graminicola*

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Investigations were carried out to study the biocontrol potential of *Trichoderma harzianum, Aspergillus niger, Pochonia chlamydosporia, Bacillus subtilis* and *Pseudomonas fluorescens* against *Meloidogyne graminicola* infesting rice cv. Sugandh-5. Pure cultures of the biocontrol agents were applied to seedlings as root dip and soil application (2 ml/pot) in both nematode infested and non-infested (autoclaved) soil with five modes of application viz., root-dip, single soil application, root-dip + one soil application (15 days), two soil applications (15 and 30 days) and root-dip + two soil applications (15 and 30 days). Plants grown in non-infested soil and applied with biocontrol agents showed better growth in respect to improved fresh and dry weight of root and shoot. Maximum growth promoting effect was recorded with *P. fluorescens* applied by root dip + 1 or 2 soil applications (*P d"* 0.01). In the nematode infested soil, terminal and spiral galls developed on the roots and plants suffered 20-31% decrease in the plant growth parameters. Application of *P. chlamydosporia* or *A. niger* as root dip + one soil application was found highly effective and suppressed the gall formation (22-26%), egg mass production (25-27%) and soil population (16-60%) of *M. graminicola*, and subsequently increased the plant growth variables by 17-22%. Root-dip treatment with *P. chlamydosporia* or *A. niger* also significantly suppressed the nematode population build up (7.4-19.5%) and improved the plant growth (7.3-10%).

Effect of Different Modes of Application Carbofuran, Carbosulfan, Phorate and Chlorpyriphos on *Meloidogyne graminicola* Infesting Rice

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An experiment was conducted to determine damage potential of *Meloidogyne graminicola* on commonly grown rice cv. Sugandh-5 and to assess the effectiveness

of four nematicides against the nematode. The nematicides were applied through root-dip (200 ppm solution) and soil application of 2kg/ha phorate 10G (25 mg a.i/pot), carbofuran 3G (83.3 mg a.i/pot and 1L/ha), carbosulfan 20EC (5µl/pot) and chlorpyriphos 20 EC (6.25 µl /pot) in both nematode infested and non-infested soil with five modes of application viz., root-dip, single soil application (15 days after transplanting), root-dip + one soil application, two soil applications, and root-dip + two soil applications (15 and 30 days). The nematicides did not cause toxicity to rice plants. The plants grown in nematode infested soil showed terminal and spiral galls on the roots, and exhibited 19-29% decrease in the plant growth parameters. Among different treatments, root-dip followed by single soil application of carbofuran and phorate suppressed galling (16-20%), egg mass production (18-22%) and soil population (27.5-58.2%) of *M. graminicola,* and increased the plant growth of rice by 9-19%. Root-dip treatment alone with carbosulfan also significantly suppressed the galling (10-12%) and improved the dry weight of root and shoot (7-10%). Root-dip + two soil applications induced greater plant growth promotion and nematode suppression but the treatment was statistically equal to root dip + one soil application.

Evaluation for Resistance in Indigenous Germplasm of Rice against *Meloidogyne graminicola* and Its Management

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The study was undertaken to screen commonly grown cultivars of rice against *Meloidogyne graminicola* and to evaluate effectiveness of carbofuran through soil application against the nematode. Eleven rice cultivars viz. Anjali, Abhishek, R-Dhan, Hazari, Sadabhar, Samba Mehsuri, Sharbati, Sugandh, Swarna, Vandana and Virendra were grown in rice root knot nematode infested soil filled in 15 cm pots (1000J_2/kg soil) to evaluate host reaction of the germplasm. The screening of the cultivars was done based on the galling, egg mass production, soil population of *M. graminicola* and plant growth parameters. All the cultivars except Abhishek was found susceptible to *M. graminicola* and developed characteristic terminal galls. The cvs Sugandh and R-Dhan were found highly susceptible and developed 50-57 galls/root system and exhibited 11-37 % decrease in the plant growth variables. The cvs. Anjali, Swarna, Vandana and Virendra showed 20-30 galls/root system. These cultivars also supported reproduction of *M. graminicola* which produced 20-30 egg masses leading to 60-90% increase in the soil population of the nematode.

The cultivar Abhishek expressed tolerance against the nematode, and developed 10-12 galls/root system without exhibiting significant decline in the plant growth variables. Soil application of carbofuran at 15 or 30 days after transplanting suppressed the galling, egg mass production and soil population of *M. graminicola* and improved the plant growth of rice cultivars. The study has revealed that one soil application at 15 days after transplanting with carbofuran can significantly control the root-knot disease and improve the plant growth.

Root-Knot Nematode, *Meloidogyne incognita* Management in Mulberry (*Morus alba* L.)

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Mulberry (*Morus alba* L.), the sole food plant of silkworm (*Bombyx mori* L.), is cultivated both in tropical and temperate countries of the world. India is the second largest country in the world having 3.42 lakh hectares under mulberry cultivation. Root knot nematode, *Meloidogyne incognita* plays an important role in reducing herbage yield and quality of leaves besides the life span of mulberry plants. The disease is manifested by the formation of galls in the root accompanied by stunted growth, chlorosis and loss of vigour of the plant. Pot culture experiments were conducted for the evaluation of efficacy of biocontrol agents *viz.*, *Pseudomonas fluorescens* and *Trichoderma viride*, against *M. incognita* on mulberry variety S36 in comparison with standard chemical check of carbofuran 3G and an untreated control. The experimental results revealed that use of combination of *P. fluorescens* (@ 10 g/plant) + *T. viride* (@ 10 g/plant) as soil application was effective to check the root knot nematode disease and to improve growth of mulberry with increased leaf yield, moisture, protein, nitrogen, chlorophyll content and reduce the nematode population. *P. fluorescens* (@ 5 g/plant) + *T. viride* (@ 5 g/plant) soil application also recorded increased plant growth, moisture, protein, nitrogen, chlorophyll content and reduced nematode population compared to control followed by *P. fluorescens* @ 10 g/plant + *T. viride* @ 10 g/plant.

Screening of Banana Hybrids (Phase II Hybrids) for Resistance to *Meloidogyne incognita*

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Banana and plantains (*Musa* spp.), the second largest fruit crop in the world, are important staple foods in tropical America, Asia and Pacific. It is the most widely consumed, and exported fruit in the world. Though it is cultivated in tropics, the demand exists all over the world for its unique flavour and taste. Plant parasitic nematodes are one of the major biotic stresses affecting banana production. The reactions of eighteen new synthetic banana phase II hybrids to *Meloidogyne incognita* was studied under field as well as artificially inoculated pot conditions. Hybrid H 531 was found to be resistant and eight hybrids, H-02-34, H-03-05, H-03-11, H-03-13, H-03-17, H-04-12, H-04-24 and NPH-02-01 were found to be toterant to the root-knot nematode, *Meloidogyne incognita* and the remaining were rated as susceptible reaction. Biochemical studies like total phenols and enzymatic activity like peroxidase, polyphenol oxidase and phenylalanine ammonia lyase of hybrids in defense mechanism in response to nematode invasion indicated higher activities in resistant genotypes *viz-a-vis* susceptible ones. Hybrid H531 had the maximum biochemical content and enzyme activity among the hybrids taken for this study. The resistant hybrids had enhanced contents of total phenol, peroxidase, polyphenol oxidase and phenylalanine ammonia lyase.

Assessment of Different Genotype of Fieldpea for Resistance against Root-Knot Nematode, *Meloidogyne javanica*

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Fieldpea (*Pisum sativum* L.) is one of the important *rabi* pulse crops grown in India. It is grown over 0.7 million hectares yielding about 0.6 million tons. Root knot nematodes (*Meloidogyne* sp.) are serious nematode pests of fieldpea. It causes enormous yield loss in all fieldpea growing areas. Present study was carried out to assess the level of resistance against root-knot nematode, *M. javanica* on the basis of Root galls formation. Six genotypes of fieldpea i.e. HUDP-15, IPFD-06-5, IPFD-10, IPFD -99-13. HFP-4 and IPFD 99-25 were selected. Seeds were sown in pots filled with steam sterilized soil. After germination seedlings were inoculated with 500 second stage juveniles of *M. javanica*. After 40 days plants were uprooted and numbers of galls / plants were counted under stereoscopic microscope. Genotype HFP-4 was found resistant to *M. javanica*. Genotype HUDP-15 was found moderately resistant. IPFD-06-5, IPFD-10, IPFD-99-13 and IPFD_99-25 were found susceptible to *M. javanica*. Genotype HFP-4 can be used in breeding programme for developing nematode resistant varieties. We are attempting genetic transformation of the fieldpea cultivar HUDP-15 with RNAi constructs of salivary gland chromosomes to prevent the loss caused by the plant parasitic nematode. Detail characterization of the lines are in progress.

Effect of Chemical Application on Tomato Against Stunt Nematode, *Tylenchorhynchus vulgaris* and Reniform Nematode, *Rotylenchulus reniformis*

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An experiment was conducted to find the effect of chemical application such as root dip in triazophos (200 ppm for 20 min.), carbofuran 1 kg a.i./ ha, chlorpyrifos 2l

a.i/ ha, *Trichoderm harzianum* 10 kg/ha, root dip treatment and carbofuran or chlorpyrifos or *T. harzianum* with untreated control on tomato against stunt nematode, *Tylenchorhynchus vulgaris* and reniform nematode *Rotylenchulis reniformis*. Experiment was conducted in 16m^2 plot with three replication under completely ranodomise design. Initial and final nematode population was recoded. Fruit picking was done six times. Observations revealed that reniform nematodes declined in all the treatments except in *T. harzianum* wherein population was increased in relation to initial population. Maximum reduction (70%) in population of reniform was found in carbofuran and chlorpyrifos in relation to control final population. *T. harzianum* treated plants had increase in population of reniform nematode by 20% in relation to initial population however in control untreated had increase of 20%. Stunt nematode population increased in all the treatments in relation to initial population however it was lower than the final control population. Average yield per plot was highest in combined application of root dip and chlorpyrifos (19 kg/16m^2) while in other treatments it was in the range of 13-18 kg/16m^2. The lowest yield was recorded with root dip and carbofuran or *T. harzianum* (13 kg/16m^2) in comparison to control (12 kg/16m^2).

Population Behavior of Stunt and Reniform Nematode in Raised Bed Cultivation of Tomato Treated with Different Chemicals

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An investigation was carried out on the management of reniform and stunt nematode with root dip in triazophos (200ppm for 20min), carbofuran 1 kg a.i/ha, chlorpyrifos 2l a.i/ha, *Trichoderma harzianum* 10 kg/ha, root dip treatment and carbofuran or chlropyrifos or *T. harzianum* with untreated control. Experiment was done in 4m2 plot in completely randomize design. It has been observed that population of stunt nematode was increased by 50% in *T. harzianum* applications with respect to initial population. However based on final population in relation to control it was found to be higher in all the treatments excepting in chlorpyriphos and *T. harzianum*. In untreated control the population of stunt nematode increases by 20%. Population of reniform nematode was reduced in all the treatments (20-50%) except *T. harzianum* which showed increase of population by 60% with respect to initial population. While in relation to control final population reduction in reniform nematode was around 70%. Thus, stunt and reniform nematode respond similarly to the chemical application. Fruit yield was observed to be improved in all the treatments. In general yield was improved by 66%. While in combined application of root dip and chlorpyriphos yield increase was maximum (31 kg/16m^2) in comparison to control (19kg/16m^2).

Effect of Chemical Treatment on the Preceding Crop of Gram on the Population of Nematode in the Succeeding Crop of Mungbean

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Effect of chemical treatment of gram with carbofuran 1 and 2 kg a.i/ha, *Trichoderma harzianum* 10 and 15 kg/ha and seed soaking in triazophos 125 and 250ppm for 20 min with untreated control as preceding crop on the nemic population in succeeding crop of mungbean was investigated. Experiment was conducted in 16m^2 plot in completely randomize design. Observations were recorded on the initial and final population of two main nematodes *viz. Tylenchorhynchus vulgaris* and *Rotylenchulus reniformis*. Pod yield was also recorded. Data revealed that there has been significant increase in population of reniformis nematode. The population was as high as 17 pre adult of reniformis nematode in carbofuran 2 kg a.i/ha which was much more than carbofuran 1 kg a.i./ha (8 preadult /g soil). It means that increase of doses of carbofuran has also enhanced the population of reniformis nematode. With *T. harzianum* increase was observed with 7 preadult /g soil. While minimum increase was found with triazophos 250 ppm treatment (4 pre adult/g soil.) in comparison to control 11 preadult/g soil. While scenario would be different if the relationship is drawn with initial vs. final population basis. Here average population of 1 preadult lead to increase by 7 preadult/g soil in treated plant while in control increase happened from 3 preadult to 20 preadult/g soil of reniform nematodes. Thus treatment in general had beneficial effect in term of rise in population of reniform nematode. By and large population of stunt nematode was low in all the treatments in relation to control with respect to final population. However, carbofuran 1 kg a.i./ha and triazophos 250 ppm has stunt nematode population equivalent to control. As regards to pod yield it has been observed that pod yield was better in treated plot than the untreated control. While maximum pod yield was recorded in triazophos (0.5 kg/16m2) followed by *Trichoderma* sp. (0.45 kg/16m^2) Yield was by and large better by 18-45% by various treatments.

In vitro Screening of Microbial Antagonists Against Nematode Disease Complex in Tomato

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In vitro assay of native isolates of fungal and bacterial antagonists viz., *Pseudomonas fluorescens, Trichoderma viride, Bacillus subtilis* and *Paecilomyces lilacinus* was assessed *Fusarium oxysporum* f.sp.*lycopersici* by conducting hatching test and by dual culture technique. The native antagonists were tested for their ovicidal effect against *M. incognita* eggs. The culture filtrates of bacterial cell and fungal spore concentrations were maintained at 100, 75, 50 and 25 per cent concentration and distilled water served as control. Three replications were maintained for each treatment. The number of hatched juveniles was counted after 36, 48 and 60 hours of incubation. The results revealed that the lowest egg hatching was observed in *B.subtilis* followed by *P. lilacinus* and *T. viride* and.34.0 per cent respectitively at 100 per cent concentration of culture filtrate 36 h exposure period. All these treatments were on par with each other. Similar trend was observed at 75 and 50 per cent concentrations. The highest egg hatching was found in 25 per cent concentration at 60 h after exposure period with value of 65.0, 60.3, 58.2 and 57.3 per cent eggs hatched in *P. fluorescens, T. viride, P. lilacinus* and *B.subtilis* respectively compared with 50 per cent concentration. The highest egg hatching was recorded in control, Distilled water with 60.4% at 25 per cent concentration after 36 h of exposure period and 75.3 per cent after 60 h of exposure period. Among the antagonist, *P. fluorescens* recorded the lowest mycelial growth of 2.9 cm accounting for the highest growth inhibition of 68.5 per cent reduction followed by *T. viride, P. lilacinus* and *B. subtilis* recorded 3.7, 4.6 and 5.4 cm respectively in pathogen growth as against control which recorded 9.2 cm. The per cent inhibition of growth was 54.8, 50.0 and 41.3 respectively over control.

Evaluation of Biocontrol Potential of Native Antagonists for the Management of Nematode Disease Complex in Tomato

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Field trials were carried out to study the biocontrol potential of native antagonists viz., *Pseudomonas fluorescens, Trichoderma viride, Bacillus subtilis* and *Paecilomyces lilacinus* against root-knot nematode, *Meloidogyne incognita* and *Fusarium oxysporum* f.sp.*lycopersici* in tomato. The result revealed that, application of biocontrol agents enhanced the growth of tomato plants compared to carbofuran application. Among the biocontrol agents tested, *P. fluorescens* was fond effective in increasing the shoot length, shoot weight, root length, root weight and yield of tomato plants with 70.4%, 84.0%, 95.2%, 85.0% and 62.2% respectively over control. The effect of this treatment was followed by *T. viride* which was significantly different from *P. fluorescens*, whereas *P. lilacinus* and *B. subtilis* were on par with each other. The effect of carbofuran was on par with *P. lilacinus* and *B. subtilis*. Application of carbofuran was found effective in reducing the nematode population, but the effect was on par with *P. fluorescens*, which recoréd 61%, 51%, 42.2% and 44.0% reduction in number of females; number of egg masses soil and root population. Next to *P. fluorescens, T. viride* was found effective which recorded 38.6% and 38.0% reduction in soil and root population where as *P. lilacinus* and *B. subtilis* were on par with each other in reducing soil and root population. The per cent decrease was 31.4, 30.0, 31.2 and 25.0 respectively over control.

Biofumigation for Management of Root-Knot Nematode in the Soil

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Biofumigation is an agronomic practice that makes use of plant metabolites called glucosinolates that are present in cell vacuoles of plants belonging to family Brassicaceae, Capparidaceae and Moringdaceae. Upon degradation the enzyme myrosinase present in cytoplasm is released that converts glucosinolates to methyl isothiocyanates that cause nematode mortality in the soil. A laboratory and field trial were conducted to observe the effect of *Brassica* leaves and twigs on juveniles of root-knot nematode, Meloidogyne incognita. Fresh leaves of *Brassica juncea*(cv MC27 and Pusa Jagannath) were incorporated in root-knot infested soil (30J2/cc soil) @10%, 20% and 30% w/w and kept in a BOD incubator for 30 days. No root-knot J2 could be observed in the suspension on nematode extraction using Cobb's method. The buioassay of the soil using moong seedlings did not show the development of galls.In the field, chopped twigs of *B. juncea* were incorporated in root-knot infested soil in a polyhouse. Soil samples collected from treated rows showed an increase in free living nematodes with undetectable levels of root-knot juveniles. Biofumigation is a safe and ecocompatible method of nematode management.

Attempt to Delay the Emergence of Infective Juveniles of *H. indica* from *G. mellonella* Cadavers by Inducing Anhydrobiosis

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Multi-locational field trials conducted during 2009-2011 in the Ghaziabad district of western UP indicated the application of *H. indica* infected *G. mellonella* cadavers gave more effective results *vis-a-vis* inundation of infective juveniles (IJs) via irrigation for managing white grub infestation in sugarcane crop. However, this technology is constrained by the need to apply the cadavers before the emergence of the IJs. We addressed this challenge by attempting to delay the emergence of the IJs of *H. indica* through inducing partial anhydrobiosis while they were undergoing development inside the *Galleria* cadavers. Six-day old cadavers were coated with fine clay dough and incubated in 90, 93 and 97% relative humidity chambers for inducing anhydrobiosis. Non-coated cadavers and coated cadavers at 100%RH served as control. At 90 and 93% RH the cadavers dried within 48 hrs (8^{th} day from mortality) as compared to 72 hrs (9^{th} day from mortality) at 97% RH, which coincided with the emergence of the cadavers in the two control treatments. Emergence of IJs was delayed in all the three treatments by two days at 90% RH and three days at 93 and 97% RH, respectively. The emergence was negligible at 90 and 93% RH (500 IJs/cadavers) due to high incidence of mortality. However, at 97%RH the emergence started on the 12^{th} day ($1.2 x 10^5$ IJs/ cadaver) and continued till the 16^{th} day as compared to non-coated cadavers where emergence started on the 9^{th} day ($1.8 x 10^5$ IJs/ cadaver) and stopped by the 12^{th} day.

Absorption of Carbosulfan by *Polianthes tuberosa* L. bulbs and its Effect on Management of Root Knot Nematode, *Meloidogyne incognita*

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Carbosulfan 25emulcified concentration was evaluated as seed soaking for management of root-knot nematode, *Meloidogyne incognita* infecting bulbs of tuberose. Soaking of tuberose bulbs was done in 500, 1000 and 2000 µg/ml of carbosulfan 25EC for 2 and 4 h of exposure. Absorption of carbosulfan by tuberose bulbs evaluated by high pressure liquid chromatography (HPLC) ranged from 5.25 pg/ml to 35.44 µg/ml. With the increase in the time of exposure or the concentration of nematicide there was corresponding increase in incorporation of nematicide in the bulb tissues. Hatching from root-knot nematode, *M. incognita* eggs was reduced significantly by carbosulfan treatment at all the tested concentration (5-35 ug/ml). Penetration of juveniles, treated with 5-35 ug/ml concentration of carbosulfan, reduced by 41.44 to 85.51%. Reduction of 75.93 to 81.04% in the total number of nematodes in tuberose bulbs was recorded with soaking in 2000 µg/ml concentration of carbosulfan.

Biomanagement of Root-Knot Nematode (Meloidogyne incognita) infecting Okra in West Bengal

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Field experiments were conducted with bioformulations (talc based) at Central Research Farm, Bidhan Chandra Krishi Viswavidyalaya, Nadia, West Bengal with the objective to manage root knot nematode (*Meloidogyne incognita*) on okra. During *rabi* seasons of 2008 and 2009, the experiment was laid out with five treatments viz. T1- *Paecilomyces lilacinus* (cfu $2x10^6$) at 2.5kg in 2.5t FYM (Farm Yard Manure); T2-*Trichoderma harzianum* (cfu $2x10^6$) at 2.5kg in 2.5t FYM; T3- *Pochonia chlamydosporia* (cfu $2x10^6$) at 2.5kg in 2.5t FYM; T4- *Pseudomonas fluorescens*

a (cfu 2x10^8) at 2.5kg in 2.5t FYM and T5-untreated control. Each treatment replicated four times and the okra cv. Saguna/Arka Anamika was grown in the nematode infested plot of 2.0m x 2.5m size. The effect of biopesticides on the reduction (19% to 60%) of soil nematode population (J_2 = second stage juvenile) was recorded over the control plots. Soil application of biopesticides had significant effect on nematode population in soil, root galling and fruit yield of okra. The highest yield with greater suppression of soil nematode population was obtained in plots treated with *P. lilacinus* at 2.5kg in 2.5t FYM (T1). Further, it was found that combined application of *P. lilacinus* and *T. harzianum* as seed treatment and soil application with FYM was not significantly different from their individual application. Further, the bioformulations were also tested in a separate trial for consecutive two seasons of 2010 and 2011 with the nine treatments viz. T1- *P. lilacinus* (cfu 2x10^6) as seed treatment 20g/kg of seed; T2- *T. harzianum* (cfu 2x10^6) as seed treatment 20g/kg of seed; T3- *T. harzianum* (cfu 2x10^6) at 2.5kg along with 2.5t FYM/ha; T4- *P. lilacinus* (cfu 2x10^6) at 2.5kg along with 2.5t FYM/ha; T5- T1+ T4 ; T6- T2+T3 ; T7- 2.5 tons of FYM/ha ; T8- carbofuran 1.0 kg a.i./ha and T9-untreated control. Results showed that adoption of treatment enhanced the total yield of the okra over untreated plots. The effect of biopesticides on soil nematode suppression (J_2) was observed to the extent of 73% over the control plot. Besides, the adoption of *P. lilacinus,* either as seed treatment or soil application with FYM had significant effect on soil nematode population, root galling and yield of okra. The highest yield with relatively greater suppression of soil nematode population was obtained in plots treated with *P. lilacinus*. The application of *T. harzianum* as seed treatment at 20g/kg and soil application at 2.5kg along with 2.5t FYM/ha also proved effective to reduce soil nematode population and enhancing (14 to 34%) yield of okra. The carbofuran treated plots showed relatively low root galling but soil nematode was not suppressed to a considerable extent. Further, it was also found that application of *T. harzianum* as seed treatment or soil application with FYM, and FYM alone was not found to be effective for managing root knot nematode on okra.

Neem Seed Kernel Water Extract as Prophylactic Measure for Nematode Infestation in Button Mushroom

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During initial studies promising results were obtained by using 4% Neem Seed Kernel Water Extract (NSKWE) for managing nematodes infestation in white button mushroom (*Agaricus bisporus*). Further large scale studies were, therefore, undertaken to assess its efficacy as prophylactic and control measure.

For this purpose 4% NSKWE solution was prepared by dissolving dried neem seed kernels powder in water for 24 h. Solution was then filtered through a four-ply muslin cloth and was mixed with compost @ 7 L/ q compost at the time of filling the compost in mushroom beds. To assess its effect for managing 4000 *Aphelenchoides swarupi* were added 10 days after spawning in to mushroom beds, each containing 70 kg compost and another inoculation of 6,000 nemas, twenty days after casing in mushroom laboratory of department of Nematology, CCS HAU, Hisar. Appropriate checks were maintained to compare the effect of treatment. Observations were recorded on mycelial growth of *A. bisporus* 10 days after spawning, number of flushes, total production and nematode population at the end of crop season. Incidence of diseases, if any, was also recorded. In NSKWE treated and nematode inoculated beds, mycelial growth of button mushroom was better and yield was 8.2% higher (13.5 kg/ q compost) than nematode inoculated, untreated beds (12.4 kg/q compost). Treated beds harbourbed 96.6% less nematodes than untreated beds.

Similar experiments were conducted at farmers' mushroom houses at Siswala (Hisar) and Assan (Rohtak) villages to see of 4% NSKWE use as prophylactic measure. However, no nematode was inoculated and each bed contained 5 q compost. *A. swarupi* infestation was recorded only during 2007-08. NSKWE treated beds yielded 96% more and contained 51.9% less population of *A. swarupi.* During 2008-09, no myceliophagous nematode infestation was recorded in either of mushroom houses. However, bacteriophagous nematode population was very low in treated beds than untreated ones. NSKWE treated beds yielded 22.0 kg/ q as compared to 15.5 kg/q compost in untreated beds. Incidence of dry bubble (at Siswala village during 2008-09) was also significantly less in NSKWE treated beds.

New Strategies for the Integrated Management of Rice Root-Knot Nematode under Changing Climate in Karnataka

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Incidence of root-knot nematode *Meloidogyne graminicola* (Golden and Birchfield 1965) was observed for the first time during kharif 2001 in Bhadravathi taluk of Shimoga district (Krishnappa *et al.,* 2001). Rice seedlings at the young stage of

10-15 days old in the nursery have been found heavily infested with root-knot galls with stunted growth and yellowing symptoms. Subsequently, the same was reported from K.R. Pet taluk of Mandya district where it was also noticed in the nursery stage on the wild rice (*Echinocloa crusgalli*) in paddy field. Now, during this year, there is sudden outbreak of this nematode in all the rice growing areas of Karnataka state and especially heavy infestation of this disease is reported from Davanagere, Bellary, Uttara Kannada and Dhakshina Kannada districts. The nematode is spreading through irrigation water. The death of the seedlings will be more if this nematode is coupled with the fungus *Sclerotium* spp. The State Department of Agriculture, Government of Karnataka, UAS, Bangalore and Directorate of Rice research, Hyderabad had jointly taken necessary immediate steps to manage the disease. A technology was released to the farming community through package of practices of UAS, Bangalore (Ravindra *et al.,* 2006). However, though some control measures are available to the farming community, this nematode disease is increasing year after year and spreading to new areas also especially in monocropping of rice season after seasons and in canal irrigated areas. The infestations are relatively more frequent in fields under aerobic cultivation. It appears that the nematode is adopting to the submerged and floded conditions also. Since the nematode is devastating at the nursery stage itself detailed strategies are being taken up to develop a new schedule of management of the nematode for recommending to the farmers. Recommendations on integrated schedule for preplant, nursery stage and main field will be developed by employing soil solarization, organics like rice hull ash and poultry manure, bioagents like *Trichoderma viride* and *Pseudomonas flourescens,* nematicides like carbofuran etc.

Field Experiment on Efficacy of *Pseudomonas fluorescens* (Pfbv 22) and *Bacillus subtilis* (Bbv57) on *Meloidogyne incognita* and *Radopholus similis* in Black Pepper

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An on farm trial was conducted at Horticultural Research Station, Thadiyankudisai to evaluate the PGPR consortium for nematode management in black pepper. The commercial formulation of PGPR consortium is the combination of two bacterial strain *Pseudomonas fluorescens* (Pfbv 22) and *Bacillus subtilis* (Bbv57) developed by Jonathan (2008) Department of Nematology, Directorate of Plant Protection

Studies, Tamil Nadu Agricultural University, Coimbatore was used in this experiment. The field trail was conducted during the month of August 2010 to March 2011. Soil application of two bacterial formulation *of Pseudomonas fluorescens* (Pfbv 22) + *Bacillus subtilis* (Bbv57) each @ 5g per vine along with standard treatment of *Pseudomonas fluorescens* (Pf1) @ 10g / vine, carbofuran (1kg a.i./ha) + carbendazim (1g/ lit) and an untreated control were imposed in randomized block design with five replication. The trial was concluded after green pepper harvest. At the time of final harvest the observation such as soil nematode population (250g), female population in root (1g), number of egg mass in 1g of root and gall index of root knot nematode *Meloidogyne incognita.* Root and soil nematode population of *Radopholus similis* also recorded respectively at the time of harvest. Yield of green pepper per vine was weighed in every harvest and recorded. After data analysis it is found significant differences were found among the all treatments in the field experiment. Soil application of *P. fluorescens* (Pfbv 22) + *B. subtilis* (Bbv57) each @ 5g/vine recorded the lowest soil and root population of *M. incognita, R. similis* with 58.3%, 53.5% and 41.7%, 44.1% respectively compared to untreated control. Number of egg mass and adult female per g of root is also low when compared to other treatments. This result shows significant difference within each treatment. Soil application of *P. fluorescens* (Pfbv22) + *B. subtilis* (Bbv57) each @ 5g/vine had not only decrease the nematode population and also enhance the yield of green pepper (140%) compared with untreated control. The PGPR consortium also performed significantly different from recommended soil application of Pf1 and carbofuran+carbendazim. Regarding benefit cost ratio it was observed that soil application of Pfbv 22 + Bbv 57 each @5g/vine recorded 3.38 compared to the untreated control (2.44).

Study on Isolation and Effect of Glomalin on Bio-Ecological Variation of Root-Knot Nematode *Meloidogyne incogntia*

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Glomalin were extracted from *G. fasiculatum, G. messeae* and *G. intraradices* growing media under glass house condition. Glomalin were isolated from the soil by cooker method using sodium citrate as buffer and quantified by modified bradford method. From this studies *G. fasiculatum* recorded the highest level of glomalin as 1.32 mg/

g of soil followed by *G. masseae* (1.1 mg/g of soil) and *G. intraradices* (0.92 mg/g of soil). Glomalin isolated from the *G. fasiculatum* used as stock solution for further studies. The different concentrations of glomalin *viz.*, 5%, 6%, 7%, 8%, 9%, 10%, 11%, 12%, 13%, 14%, and 15% was prepared and used to study the effect on egg hatching, juvenile mortality, penetration and attraction of root knot nematode, *Meloidogyne incognita* towards tomato root.

The results revealed that all the treatments inhibited the egg hatching and increased the juvenile mortality. Among the treatments glomalin at 7%, 8%, 9% and 10% were on par with each other in inhibiting the egg hatching. Regarding juvenile mortality 6%, 7%, 10% and 11% were on par. The other treatments were significantly different from each other.

Glomalin treated tomato seedlings were uprooted every 24 hours interval after nematode inoculation for the period of seven days. The roots were stained with acid fuchsin lactophenol and destained with plain lactophenol and observed under stereo zoom microscope. All the treatments influenced the nematode penetration significantly. Among the treatments 10 per cent concentration of glomalin solution recorded the lowest number of nematode penetration. The movement of nematodes towards tomato roots was observed under stereo zoom microscope, 48 hr after inoculation. Among the treatments 10 per cent concentration of glomalin solution recorded the lowest number of nematodes attracted towards root.

Management of Root-Knot Nematode, *Meloidogyne* spp. in Tomato Through Bio-Agents under Protected Cultivation

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This trial was conducted against root-knot nematode, *Meloidogyne* spp. on tomato in the polyhouse at CCS HAU, Hisar during 2010-11 by using various bio-agents (*Paecilomyces lilacinus, Pseudomonas fluorescens* and *Trichoderma viride*) at two doses (1.8 and 2.0 g/m^2). Carbofuran (@ 7 g/m2)and untreated check were also kept simultaneously to compare the efficacy of bio-agents. All the treatments were replicated thrice with randomized block design. At the time of maturity of the crop, the observations were recorded on plant height, root-knot index and final nematode population. The results revealed that *Trichoderma viride* @ 2.0 g/m^2 was found to be most effective in reducing root-knot index (2.5) and final nematode population in soil (163 j_2/200 cc soil) over untreated check ((RKI = 4.7 and FNP= 353 j_2/200 cc soil) followed by *Paecilomyces lilacinus* check ((RKI = 2.8 and FNP= 181.8 j_2/200 cc soil).Plant height (262 cm) at harvest was also found to be maximum in the beds treated with *T. viride* followed by carbofuran (241 cm) and was minimum in untreated check (231 cm).

Synthesis and Bio-Efficacy Evaluation of Cyclohexenones and Indazoles against *Meloidogyne graminicola*

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With growing world population, there is an urgent need for the discovery and development of new classes of pest control agents with even higher activity against the target organisms. There is a demand for agrochemicals having higher efficiency, efficacy, economy, selectivity and safety with reduced risk for non-target organisms and the environment. The process of discovery, design and development of agrochemicals is an ongoing exercise and challenge against the phenomenon of 3 R's (resistance, resurgence and residues). Ecologically sound synthetic/semi-synthetic compounds with minimum hazards to the non-target organisms and higher level of safety to the environment will find acceptance by farmers and end users. Mining for new bioactive molecules from synthetic and natural sources and improvement of processes incorporating the principles of green chemistry are the essential ingredients of agrochemical development. Besides it is necessary to combat the continuing problem of insect resistance and to remove the toxic hazards presented by some of the existing classes of pesticides.

Over the years various innovative methods have been devised to speed up the chemical reactions. In these environmentally conscious days the development of technology is directed towards environmentally sound and eco-friendly methods. Microwave assisted organic reaction enhancement (MORE) is now a days a well established technique for synthesis of various heterocycles. All thermally driven reaction can be accelerated by microwave. The spectacular results obtained *viz.* shorter reaction time, experimental simplicity, selectivity of products and easy work up etc gave clear indication on the potentialities of this technique over conventional heating. Furthermore the reactions can be carried out under solvent free conditions making it an environmental friendly process. Thus the use of microwave energy for the synthesis of organic compounds forms a part of green chemistry.

Our group has been engaged in the area of design and development of bioactive heterocyclics and related compounds with pest control properties as well as design and study of nano-polymers as carriers of pest control agents. The present talk will deal in details the synthesis and bioefficacy evaluation of cyclohexenones and indazole derivatives against *Meloidogyne graminicola* in rice under pot conditions. The results indicate that both cyclohexenones and indazoles show promise as nematicide under pot conditions, the details of which will be presented in the conference.

Cross Generic Infection of *Pasteuria* Isolated from *H. cajani* to *Globodera* spp.

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The potato cyst nematode *Globodera* spp is major threat to potato cultivation in the Nilgiri Hills of Tamil Nadu and is restricted under State quarantine. The *Pasteuria* group of bacteria is Gram positive, endospore-forming nematode hyperparasites currently being developed into biological control agents. Cross-generic infection of *Pasteuria* is rarely seen as the bacterium is highly host specific. This is the first report from India where a population of *Pasteuria* originally isolated from the pigeon pea cyst nematode *Heterodera cajani* in Delhi adhered equally well to the cuticle of *Globodera* spp. Both conventional and altered spore attachment were observed and the infected nematode juveniles showed typical 'swarming' behavior characteristic to *Pasteuria* infection. After attaining the attachment the juveniles of *Glododera* spp encumbered with *Pasteuria* spores were inoculated to 10 days old potato seedlings (cv. Kufri Jyoti) raised in sterilized soil. Observation on the completion of the life cycle of the nematode was taken after a period of 10 weeks. The soil from the pots was processed to isolate the nematode cysts. Individual cysts were handpicked and observed for the presence of any *Pasteuria* infection. In total 97 cysts were recovered, out of which 54 were brown, 4 were yellow and the remaining 39 were creamy white. None of the brown and yellow females showed any infection. Out of the 39 white females, 20 were found to be infected with *Pasteuria* developmental stages. There was an asynchronous development of *Pasteuria* observed in the infected females which showed overlapping presence of the rods and sporangial microcolonies represented by diards, tetrads and the presence of mature endospores. None of the infected females contained any eggs. These observations suggest that *Pasteuria* originally isolated from the pigeon pea cyst nematode *H. cajani* shows cross-generic attachment to the potato cyst nematode *Globodera* spp. and further proceed to successfully complete its life cycle. However, as the population of *Globodera* in the Nilgiri Hills is a mixture of both the species viz. *G. rostochiensis* and *G. pallida*, therefore, we cannot scientifically conclude the infection of *Pasteuria* to a particular species at this stage.

Pathogenesis of Greasy Leaf Streak Disease in Tuberose by Foliar Nematode, *Aphelenchoides besseyi*

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Aphelenchoides besseyi, Christie1942 is one of the very few above ground plant parasitic nematodes inciting severe yield losses in many field and ornamental crops in India. In recent years , serious yield losses in tuberose resulting decline in quality and marketability of spikes have been recorded by this nematode species in the state of Odisha. Therefore, it was felt imperative to investigate into details of epidemiology and pathogenesis of this important disease in tuberose, through inoculated pot culture studies as well as field observations. Also the important weather parameters during the period under investigation were correlated with the disease development. Temperature and relative humidity played an important role in the development of disease symptoms. During this period relative humidity was more than 90% and average temperature ranged between 25.6-26.0.Initially the disease appeared as small water soaked spots in the form of narrow streaks/ stripes on the upper side of mid rib in the month of July and August during which the temperature and relative humidity were very high. Gradually these spots extended along the mid rib in both the direction in the longitudinal axis. However, the enlargement of these spots along lateral axis were very often limited by the parallel veins. In early stages of formation, these streaks were orange coloured. Later on , these spots turned to dark black in colour with a greasy appearance. Completely developed black greasy leaf symptoms on the leaf lamina measured up to twelve inches. These spots have a tendency to brittle and bend down. Rotting of leaves was frequently encountered during wet weather conditions. Large number

of nematodes were recovered from the margin of the leaf streaks. Rolling of the leaf as well as drying of the leaf tips were recorded during dry spells in the season. Healthy spikelets bore blemish free florets. Infected flower stalks were shortened ,twisted and distorted to different degrees. Very often spikelets failed to open. Petals exhibited red to orange brown streaks at the bases thereby resulting in the detoriation of flower quality.

Efficacy of Few Bioagents as Seed Treatment for Management of Root-Knot Nematode (*Meloidogyne incognita)* infecting Okra (*Abelmoschus esculentus*)

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Okra, *Abeimoschus esculentus* is an important crop of Malvaceae family and is grown extensively under various agro climatic zone of India. Among various biotic stresses, root-knot nematode, *Meloidogyne incognita* constitute one of the major constraint in successful production of okra and has been reported to account for 90% avoidable losses under field condition depending upon initial nematode population in the field. Use of bioagents offers an economic and eco-friendly management option which can be included as an important component of integrated nematode management of root-knot nematode.Hence, the present study were conducted under screen house conditions for evaluating the bioefficacy of some of the bioagents against *Meloidogyne incognita* infecting okra. Five bioagents viz. *Trichoderma harzianum, Trichoderma viride, Pochonia chlamydosporia, Paecilomyces lilacinus* and *Pseudomonas fluorescens* each having spore load of 2 x 10^6 cfu/g and *Pseudomonas fluorescens* with spore load 1 x 10^8 (cfu/g) were included for their comparative efficacy. In addition, seed treatment with carbosulfan (25 DS) @ 3% a.i. w/w as treated check alongwith untreated check and healthy (non-infested) check were also included. The seeds of okra (Cv. A-4) treated with the respective bioagents and chemical nematicides were sown in earthen pots (20 cm. diameter) containing 2 kg. soil having initial nematode population of 2 J2/g of soil. All these pots were arranged on the bench of screen house in completely randomized design (CRD) with four replications for each treatment.

The observations recorded 60 days after sowing revealed non-significant differences among all treatments in respect of root length and root weight. However, shoot length and shoot weight were significantly higher than untreated check in seeds treated with *Trichoderma viride* and *Paecilomyces lilacinus.* The number of galls/ root system and egg masses/root system were significantly less in all the treatments where bioagents have been used compared to untreated check. Further, the number of eggs/eggmass (274.55) was maximum in untreated control while it was minimum (231.58) in seeds treated with carbosulfan (25 DS) @ 3% a.i. w/w. The number of galls and eggmasses/root system were minimum in treated check i.e. carbosulfan 3% a.i. w/w. It has also been observed that all the treatments were significantly effective in reducing the final nematode populations per 200 g soil compared to untreated check. However, seed dressing with *Trichoderma viride* and *Paecilomyces lilacinus* were statistically at par in reducing the final nematode populations. Hence, use of bioagents as seed dressing treatment can be suitably included as a component of integrated nematode management for the management of root-knot nematode infecting okra.

Efficacy of Foliar Spray of *Heterorhabditis indica* and *Steinernema astiaticum* against *Pieris brassicae* on Cabbage in Field

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In two separate replicated field trials, indigenous entomopathogenic strains of *Heterorhabditis indica* and *Steinernema asiaticum* were sprayed in aqueous suspension to control a natural population of cabbage butterfly, *Pieris brassicae* on cabbage. In trial I, *H. indica* IJs were sprayed @ 50,000 in 50 ml water per plant in the evening hours. Malathion 50EC @ 0.05% and water alone served as checks. In trial II, in addition to above treatments, half dose of IJs (25,0000) was combined with Malathion @ 0.025% (half the recommended dose).

H. indica gave 71% control of *P. brassicae* compared to 50% in Malathion after 24 h; which increased to 81% after 48 h. The chemical insecticide killed lady bird beetles; however these were unharmed in treatments involving EPNs. In trial II, *S. asiaticum* @ 50,000 IJs dose gave 60.9% control compared to 53.7% in insecticide @ 0.05% dose. However, combining EPNs with insecticide at half of the recommended dose resulted in higher mortality (69.9%) compared to either *S. asiaticum* (35.3%) or insecticide alone (53.7%). Lady bird beetles were not killed by *S. asiaticum* too.

Session V : Entomopathogenic Nematodes

Influence of Ultraviolet Radiation on the Efficacy of Entomopathogenic Nematode, *Steinernema tami*

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Investigations were carried out to study the influence of ultraviolet radiation on the entomopathogenic nematode, *Steinernema tami.* Ultraviolet light emitted from a UV lamp of 15W caused 100 per cent mortality of *S. tami* after 180 min. of exposure. At the first observation interval of 30 minutes exposure to UV radiation, 6.3 per cent mortality of *S. tami* was observed. The highest mortality of 37.1 per cent of IJs was occurred between 60 to 90 min. followed be 17.2 per cent at 90 to 120 min. The nematodes exposed to UV light were failed to infect *Corcyra cephalonica*. UV rays from sunlight are determintal to entomopathogenic nematodes. The results of the present study showed that exposure of entomopathogenic nematodes to UV radiation not only caused mortality of IJs but also affect the infectivity of IJs on the host.

Attraction and Preferential Behaviour of Entomopathogenic Nematodes Towards Insect Pests and Various Factors Influence on Their Attraction

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Attraction and preferential behaviour of infective juveniles (IJs) of entomopathogenic nematodes (EPN), *Steinernema masoodi, S. mushtaqi* and *S. carpocapsae* towards gram pod borer, *Helicoverpa armigera,* tobacco caterpillar, *Spodoptera litura* and blue butterfly, *Lampides boeticus* larva were tested in petri-dish. Effect of various factors such as different temperatures (20, 25, 28, 30 and 35 °C), periods of insect incubation (30, 60, 90 and 120 min.), agar concentrations (1, 1.5, 2, 2.5 and 3 per cent) and agar thicknesses (2, 4 and 6 mm) on the attraction of *S. masoodi, S. mushtaqi* and *S. carpocapsae* were also tested towards *H. armigera.* All tested EPN species were found to have positive attraction responses towards tested insects but showed variation in their individual behaviour. Among these species, *S. mushtaqi* was found more responsive towards *H. armigera, L. boeticus* and *S. iitura* than

S. masoodi and *S. carpocapsae*. The differential responses of EPN towards different insect were attributed to the inert behaviour of infective juveniles, their preference for a particular insect, chemical composition, concentration, quality and quantity of attractant released by insects. *S. mushtaqi* was more attracted towards *H. armigera* (64 per cent), followed by *L. boeticus* (43 per cent) and *S. litura* (38 per cent). However, attraction responses of *S. masoodi* and *S. carpocapsae* were recorded more towards *S. litura* (52 and 39 per cent, respectively), followed by *H. armigera* (48 and 42 per cent, respectively) and *L. boeticus* (29 and 34 per cent, respectively). Results show that, various factors such as temperatures, periods of insect incubation, agar concentrations and agar thicknesses affect the attraction responses. Maximum attraction was found when insect incubated for 60 min, agar plates containing 2 mm thick layer of 1.5 per cent water-agar at 30°C. This opens a new opportunity in its utilization of EPN as biopesticides against insect pests management programme in future.

Effect of Temperature on Pathogenicity of *Heterorhabditis* and *Steinernema* spp. against White Grub *Holotrichia serrata* F. (Coleoptera: Scarabaeidae)

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The effect of temperature on pathogenicity of entomopathogenic nematode (EPN) against first instar white grub, *Hoiotrichia serrata* was tested under laboratory condition at two temperature levels 35°C and 25°C. Three isolates of *Heterorhabditis* spp. and six isolates of *Steinernema* spp. at different dosages of 0, 5, 10, 50, 100, 500, 1000, 2000 and 4000 IJs/grub was tested. In general, there was an increased mortality of white grub recorded as the dosage increased. All the EPN caused mortality of grubs at all the dosages tested except *Steinernema* sp. (324) which required higher dose to cause mortality of grubs. At 35°C, three steinernematids viz *S. glaseri*, *Steinernema* sp. (Puli 2) and *S. riobrave* recorded 100 per cent mortality. The *S. giaseri* (LN1) was found to be superior and it recorded 100 per cent mortality at all dosages at both temperatures 35°C and 25°C tested. The mortality of grubs due to Heterorhabditis spp. at 35°C was ranged between 40 to 86 per cent. At 25°C, six EPN recorded 100 per cent mortality and among these S. glaseri (LN1) and H. indica (LN2) recorded 100 per cent mortality at all the dosages tested. LD50 values were calculated for both temperatures and it was observed

that 25°C recorded low LD 50 value (ranged between 7 to 517 IJs/grub) than 35°C (ranged between 8 to 1251 IJs/grub) except *Steinernema* (Puli2) and *S. riobrave* which recorded low LD_{50} values at 35°C than 25°C.

Insecticidal Activity of *Photorhabdus luminescens* against *Galleria mellonella* L. and *Chilo sacchariphagus indicus* Kapur

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Insecticidal activity of *Photorhabdus luminescens* isolated from *Heterorhabditis indica* was studied against full grown larvae of greater wax moth, *Galleria mellonella* and sugarcane internode borer, *Chilo sacchariphagus indicus.* The last instar larvae of *G. mellonella* and sugarcane internode borer *Chilo sacchariphagus indicus* were injected under aseptic conditions with 10µl of bacterial cells in phosphate buffered saline (PBS). A 10 µl syringe was used to inject various dosages *viz.*, 0, 50, 100, 200, 500, 1000, 2000, 5000 and 10000 bacterial cells/larva. The larvae were incubated in Petri dishes at room temperature. Each treatment involved 5 larvae and was done at three replicates. The larvae were checked for mortality every day for four days. Mortality of *G. mellonella* was observed in 24 to 72 hours when treatments with different dosages of bacterial cells when injected into the host. 100 per cent mortality of larvae reported with in 24h in 5000 and 10000 cells/larva. In general increased rate mortality of G. mellonella observed due to increase in the cell concentration with more exposure time. There was only 16.67 per cent mortality of larvae recorded with dosages of 100, 200 and 500 cells/larva and there was no mortality of *G. mellonella* recorded with 50 cells/larva. The mortality of sugarcane internode borer *C. sacchariphagus indicus* was observed due to Photorhabdus bacteria. 100 per cent mortality of the internode borer larvae recorded with 2000, 5000 and 10000 cells /larva.

Biological Management of Cardamom Root Grub, *Basilepta fulvicorne* Jacoby with Entomopathogenic Nematode–A Successful Event

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Root grub, *Basilepta fulvicorne* Jacoby (Eumolpinae:Coleoptera), a major pest on roots of cardamom (*Elletaria cardamomum* Maton), causes about 30-60% crop loss at various levels of pest infestation. The management of the pest with chemical insecticides, though effective, is costly, laborious and polluting the environment. Search for alternate method resulted in unraveling of biological control agents viz. entomopathogenic fungi (EPF) (*Beauveria bassiana* and *Metarhizium anisopliae*, infesting on adult beetle and grub respectively) and entomopathogenic nematodes(EPN) (*Steinernema* spp. and *Heterorhabditis* spp. infesting on grub). Studies on comparative efficacy of EPF and EPN indicated the potential of EPN in effectively causing mortality on root grub. Search for local isolates of EPN in cardamom growing areas in the Western Ghats of Kerala and Tamilnadu yielded several isolates of EPN belonging to the genera of *Steinernema* and *Heterorhabditis*. Laboratory evaluation of these EPN local isolates and also with exotic EPN species on cardamom root grub in soil column bioassay indicated that local isolates cause higher mortality. Between the two genera of local isolates of EPN, *Heterorhabditis* strains caused higher mortality on root grub. Among the local isolates of *Heterorhabditis,* strain ICRI EPN 18, *Heterorhabditis indica* was highly virulent on root grub in terms of very low Lc 50 as well as in producing higher number of infective juveniles (ijs) per infected root grub.

Pot culture experiments with various local and exotic EPN strains of *Steinernema* and *Heterorhabditis* also brought to light the viruliferous strain ICRI EPN 18, *H.indica* in causing mortality on root grub. Field experiments were conducted in ICRI farm with various strains of *Heterrhabditis* and the grub reduction percentage ranged between 59 – 96% after 30 days of EPN application. Dose response study indicated that one lakh ijs per plant caused significant reduction of root grub in field condition. Comparative evaluation of various formulations of EPN *viz.* EPN infected *Galleria* cadaver, talc and liquid formulations indicated that cadaver formulation caused significantly higher reduction of root grub within short exposure period. Demonstration trials in farmers' plots at various agro climatic conditions with local isolate ICRI

EPN 18 (*H.indica*) in cadaver formulation have not only created awareness among farmers on biocontrol of root grub with EPN but also yielded information on method of delivery of EPN in the soil; the cadaver formulation is to be applied at a depth of 5 cm in the soil, at about 10 cm from plant base on four sides of the plant; the soil in plant base is ensured with adequate moisture with mulch cover and irrigation if the soil is dry.

Out reaching farmers with biocontrol technology of cardamom root grub was carried out with training on mass production of EPN through *Galleria mellonela* larvae as well as with demonstration trials. Comparative demonstration trials with chemical insecticide and EPN in farmers plot have shown the effectiveness of biocontrol of root grub with EPN. Several farmer groups are given hands on training on EPN mass production and a few of them have started producing EPN infected *Galleria* cadaver and using them in the field for management of cardamom root grub.

Evaluation of Entomopathogenic Nematode, *Steinernema asiaticum* Against Diamondback Moth, *Plutella xylostella* Under Screen-House and Field Conditions

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In a screen-house trial, ten larvae (3rd instar) of *Plutella xylostella* were released in the whorl of two-month old cabbage plants. Infective juveniles (IJs) of *S. asiaticum* were released either in single spray (30,000 IJs), or in two sprays (15,000 + 15,000 IJs) at four days interval in 50 ml sterile water containing Glycerine (1%), Triton x-100 (1%) and Congo red (0.05%). Observations recorded after 4 days of first spray revealed maximum insect mortality (36%) at 30,000 iJ dose. Split application (15,000 + 15,000) of *S. asiaticum* proved better and resulted in 48.3% mortality, and this was significantly higher than single application. The recovery of IJs from dead cadavers of *P. xylostella* after first/second spray in different treatments ranged between 95-439.

In field investigation, cabbage plants infested with larvae of *P. xylostella* were sprayed with – i). *S. asiaticum* @ 50,000 IJs alone (T1), ii). Malathion 50EC @ 0.05% alone (T2), iii). *S. asiaticum* @ 25,000 IJs + Malathion 50EC @ 0.025% (T3), iv). *S. asiaticum* @ 25,000 IJs alone (T4); besides untreated control. All EPN treatments were given in the evening on randomly selected plants in 50 ml sterile water per plant containing the three adjuvants used in screen-house experiment. Observations were recorded on dead larvae after 24 and 48 h of spray. Irrespective of time, maximum insect mortality (37.5%) was recorded in T3 (combined application); while EPN alone at 50,000 IJs (T1) caused 28.8% insect mortality. Malathion alone at 0.05% (T2) and *S. asiaticum* alone at 25,000 IJs, (T4) resulted in 18 and 11.3% mortality, respectively, and these two were at par. The effect of time and it is interaction with treatments was non-significant.

Compatibility of *Steinernema asiaticum* and *Heterorhabditis bacteriophora* with Chemical Insecticides and Bt Registered for the Control of *Plutella xylostella* (L.) Under Laboratory Conditions

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Two indigenous strains of EPNs (*Steinernema asiaticum* and *Heterorhabditis bacteriophora*) and three insecticides (Endosulfan 35EC, Malathion 50EC and Bt) were tested at recommended doses (to control *Plutella xylostella* in cabbage) for their compatibility in a laboratory test. The infective juveniles (IJs) of nematodes were immersion (water screening) in the solution of insecticidal formulations (endosulfan 35 EC @ 0.035% and 0.07%; malathion 50EC at 0.025% and 0.05%; and Bt formulation at 0.1 and 0.2 %). Observations were recorded on per cent mortality of nematodes after 24 and 48 h. Minimum nematode mortality 0.87 and 1.59% was recorded in Bt (@ 0.1%) in *S. asiaticum* and *H. bacteriophora*, respectively. Nematode mortality increased significantly at higher concentration. Maximum IJ mortality of 6.03 and 12.43% was recorded in endosulfan (@ 0.07%) in *S. asiaticum* and *H. bacteriophora*, respectively. The mortality of IJs did not increase significantly with time in case of chemical insecticides. Overall, negligible IJ mortality at recommended doses of endosulfan, malathion and Bt augur well for the incorporation of these EPN strains in the IPM programme for the management of *P. xylostella* on cabbage.

Effect of *Steinernema abbasi*, and Its Bacterial Symbiont, *Xenorhabdus* sp. on Root-Knot Nematode, *Meloidogyne incognita* on Tomato

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Treatment with 1000, 5000 and 10000 IJs of *Steinernema abbasi* caused 68-96% inhibition in egg hatching and induced mortality to *Meloidogyne incognita* juveniles in cavity blocks. The treatment with 75 and 100% cell free culture filtrate (CFCF) and 10^{10} CFUs of *Xenorhabdus* sp. inhibited the egg hatching by 99% and induced 100% mortality to juveniles of *M. incognita*. Application of 1000, 5000 and 10000 IJs *S. abbasi* caused 35, 43 and 54% decrease in the root penetration of *M. incognita* studied in coffee cups. Treatment with CFCF (50-100%) and CFUs (10^7-10^{10}) suppressed the penetration by 20-43 and 12-33%, respectively. In a pot trial inoculation with 2000 J_2 of root-knot nematode, *M. incognita* caused 16, 24 and 19% decline in the dry weight of root, shoot and yield of tomato. Application of 10000 IJs of *S. abbasi*/pot significantly decreased the number of galls (13%), egg masses (16%) and soil population of *M. incognita* (37%) and increased the dry matter (10 and 12%) and yield (17%) of tomato. The 100% CFCF reduced the galling and egg mass production by 11 and 8% and improved tomato yield by 15%. Fecundity of root-knot nematode was not influenced with any of the treatments. Overall inhibitory effect of the treatments on the root-knot nematode was greatest with IJs followed by CFCF and CFUs.

Suitability of Gel Formulation for Entomopathogenic Nematode, *Heterorhabditis indica* (Strain ICRI EPN 18) for The Management of Cardamom Root Grub, *Basilepta fulvicorne*

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Entomopathogenic nematodes (EPN) are being used as effective biocontrol agents on soil inhabiting/root damaging pests. A local isolate of EPN, *Heterorhabditis indica*

(strain ICRI EPN 18) is found to be an effective biocontrol agent on cardamom root grub, *Basilepta fulvicorne* Jacoby, a major pest on small cardamom, *Elettaria cardamomum* Maton. Evaluation of various formulations of *H.indica viz.* EPN infected *Galleria* cadaver, talc and liquid formulation indicated that cadaver formulation could significantly reduce root grub under field condition within short time and with higher percentage. Gel formulation of EPN is also reported to be an effective formulation with longer shelf life than cadaver formulation. Commercial gel formulation of *H.indica* and *Steinernema thermophilum* were evaluated under field condition in comparison with cadaver formulation of *H. indica*. The field experiment, laid out in Exploded Block Design with about 100 plants per treatment was conducted at research farm in Myladumpara. The result indicated that even though gel formulation of *S. thermophilum* (57.9 %) and *H. indica* (43.2%) could reduce root grub considerably as compared with control treatment, cadaver formulation of *H. indica* could result in statistically significant reduction of root grub (63.2%) than gel formulation on 30 days of EPN application. The paper discusses various factors on the differential efficacy of gel and cadaver formulations in reducing root grub under field condition.

Compatibility of Entomopathogenic Nematode *Steinernema thermophilum* (Nematoda: Rhabditida) with Pesticidal Formulations

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The compatibility of entomopathogenic nematode, *Steinernema thermophilum* with four different dozes of the twenty six pesticidal formulations, namely Tagride, Talstar, Marshal, Superkiller, Decis, Confidor, Karate, Tracer, Hostathion, Tagtara, Dursban, Thiodan (Insecticides), Stomp, Atrataf, Isogourd, Fernoxone, Clincher, Validamycin (Herbicides), Zinthane, Contaf, Bavistine (Fungicides), Baba, Metarhizium, Mycomite, Niyantran, Sparsha, Nisarga and Varsha (Formulations of bio-agents) was tested. More than 98 % survival of the infective juveniles of *S. thermophilum* was recorded even after 7 days of treatment in all the tested doses of formulations except Hostathion, Dursban, Thiodan and double the highest recommended dose of Fernoxone. The virulence of the infective juveniles exposed to the highest recommended doses of various formulations was evaluated in laboratory filter paper bioassay by treating them on the 4th instar larvae of *Galleria mellonella* after an exlposure period of 48 h. The bioassay study confirmed the retention of virulence by the infective juveniles after their exposure to the above-mentioned formulations except Hostathion, Dursban and Thiodan. Our study confirms that these formulations except Hostathion, Dursban and Thiodan in their recommended doses are absolutely compatible with *S. thermophilum* and could be tank mixed in spray mixtures of respective pesticidal formulations.

In vitro Studies on the Attraction of Entomopathogenic Nematodes Towards Insect Pests and Various Factors Influencing their Attraction

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Attraction of infective juveniles (IJs) of entomopathogenic nematodes (EPN), *Steinernema masoodi* (Ali *et al.*, 2005), *S. mushtaqi* (Pervez *et al.*, 2009) and *S. carpocapsae* (Weiser, 1955) Wouts, Mracek, Gerdin and Bedding, 1982) towards 3rd instar larva of gram pod borer, *Helicoverpa armigera,* tobacco caterpillar, *Spodoptera litura* and blue butterfly, *Lampides boeticus* were tested in petri-dish (Bilgrami *et al.*, 2000). Effect of various factors such as different temperature, periods of insect incubation, agar concentrations and agar thicknesses on the attraction were also tested. All tested EPN species were found to have positive attraction responses towards insects but showed variation in their individual behaviour. Among these species, *S. mushtaqi* was found more responsive towards *H. armigera* (64 per cent), followed by *L. boeticus* (43 per cent) and *S. litura* (38 per cent). However, attraction responses of *S. masoodi* and *S. carpocapsae* were recorded more towards *S. litura* (52 and 40 per cent, respectively), followed by *H. armigera* (48 and 42 per cent, respectively) and *L. boeticus* (29 and 34 per cent, respectively). Results show that, various factors such as temperatures, periods of insect incubation, agar concentrations and agar thicknesses affect on the attraction responses. Maximum attraction was found when insect incubated for 60 min, agar plates containing 2 mm thick layer of 1.5 per cent water-agar at 30°C. This opens a new opportunity in its utilization of EPN as biopesticides against insect pests management programme in future.

Attempt to Delay the Emergence of Infective Juveniles of *H. indica* from *G. mellonella* Cadavers by Inducing Anhydrobiosis

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Multi-locational field trials conducted during 2009-2011 in the Ghaziabad district of western UP indicated the application of *H. indica* infected *G. mellonella* cadavers gave more effective results *vis-a-vis* inundation of infective juveniles (IJs) via irrigation for managing white grub infestation in sugarcane crop. However, this technology is constrained by the need to apply the cadavers before the emergence of the IJs. We addressed this challenge by attempting to delay the emergence of the IJs of *H. indica* through inducing partial anhydrobiosis while they were undergoing development inside the *Galleria* cadavers. Six-day old cadavers were coated with fine clay dough and incubated in 90, 93 and 97% relative humidity chambers for inducing anhydrobiosis. Non-coated cadavers and coated cadavers at 100%RH served as control. At 90 and 93% RH the cadavers dried within 48 hrs (8th day from mortality) as compared to 72 hrs (9th day from mortality) at 97% RH, which coincided with the emergence of the cadavers in the two control treatments. Emergence of IJs was delayed in all the three treatments by two days at 90% RH and three days at 93 and 97% RH, respectively. The emergence was negligible at 90 and 93% RH (500 IJs/cadavers) due to high incidence of mortality. However, at 97%RH the emergence started on the 12th day (1.2×10^5 IJs/ cadaver) and continued till the 16th day as compared to non-coated cadavers where emergence started on the 9th day (1.8×10^5 IJs/ cadaver) and stopped by the 12th day.

DJ Raski
Award

Effect of Root-Knot Nematode, *Meloidogyne graminicola* on the Nutrient Uptake and Physiological Characteristics of Rice Plant and Quality and Vigour of Seed

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Root-knot nematodes cause hypertrophy and hyperplasia of the vascular and cortical root tissues that reduce the absorption and transport of nutrients and water from soil. Aboveground symptoms are often mistaken for nutrient deficiencies. Farmers tend to apply higher doses of fertilizers than strictly necessary for a nematode-free crop, which not only causes economic loss, but also avoidable soil and groundwater pollution. Investigations were carried out to know the effect of different population densities of *Meloidogyne graminicola* on nutrient uptake, physiological parameters, quality of grain and vigour of seed produced from a non-basmati Pusa-44 and basmati Pusa Sugandh-5 varieties of rice grown in soil infested with varying levels of *M. graminicola* under pot and field conditions. The concentrations of N, P, K, Fe and Zn in rice leaves were significantly reduced in both rice varieties and were negatively correlated to the nematode infestation level under both field and pot conditions. The reductions did not significantly differ between the two varieties. The plant height and shoot dry weight were also negatively correlated to the infestation level of *M. graminicola* infestation. The residual contents of N, P, K, Fe and Zn in soil were greater at higher levels of nematode infestation than in nematode-free or lower infestation levels, with a strong positive correlation. The macro- and micronutrients remained unutilized in the soil while the plant suffered severe nutrient deficiencies and growth reduction in the presence of nematode infestation. The photosynthetic rate, photosynthetic water use efficiency, rate of transpiration and chlorophyll content were significantly reduced in both the rice varieties and were negatively correlated to the infestation levels of *M. graminicola* under both field and pot conditions. The degree of reduction did not vary significantly between the two varieties. Respiration rate significantly increased in both the rice varieties and was positively correlated to the infestation level of *M. graminicola* under both field and pot conditions. The reduced anabolism and enhanced catabolic activities would result in decreased biomass production leading to poor plant growth and yield. The results also proved that rice grains produced on plants infected by the nematode, *M. graminicola* were lighter in weight and had poorer nutrient qualities such as amylose and protein contents that would affect consumer interests. Further, if these

grains were used as seed, the germination percentage was lower and the seed vigour was poor compared to the seed obtained from plants grown in *M. graminicola* free soil, harming crop performance and farmers' economic interests. Investigations on physiological parameters of plants and performance of varieties would be vitiated if the roots are infected by nematodes. Similar effects can be expected in case of other crops and root parasitic nematodes.

Steinernema harryi n.sp., A New Entomopathogenic Nematode from Southern India, along with its Characterization for Managing Homopteran Insect Pests

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A new species of entomopathogenic nematode, *Steinernema harryi* n. sp. was recovered from a soil sample collected from Ambattur region of Chennai, Tamil Nadu, India in March 2009. A combination of morphological characteristics and morphometrical measurements of all stages distinguishes *S. harryi* n. sp. from rest of the members of *Steinernema*. On the basis of body length 486 (435-508 µm) of infective juveniles, this new species belongs to the "*carpocapsae* group" of small infectives. The infective juveniles are characterised by the much posterior position of excretory pore 38 (35-41µm), relatively longer oesophagus 105 (97-110 µm), D % 36 (35-39), E % 82 (73-89) and the lateral field with 6 or 7 incisures. First generation males are differentiated from other species of the *carpocapsae* group by having the largest GS ratio 0.79 (0.71-0.85) and also differ in the number and general disposition of genital papillae. Furthermore, there are clear-cut morphological differences between the two generation of males in the shape and size of spicules and gubernaculums. Females of this species are recognised by the presence of well developed, double flapped epiptygmata and protruding vulval lips. The first and second generation females lack a prominent post-anal swelling and mucron, respectively. Additionally, the molecular characterization of ITS region of rDNA also confirmed *S. harryi* n. sp. as a new and distinct species.

Bioefficacy of four Indian strains of entomopathogenic nematodes *S. thermophilum* (New Delhi strain), *S. carpocapsae* (Meghalaya strain), *S. riobrave* (Gujarat strain), *S. harryi* n. sp. (Tamil Nadu strain) were evaluated against mealybugs, aphids and whiteflies in two doses (50 and 500 IJs/ml) under laboratory conditions. *S.*

thermophilum caused significant mortality of mealybugs, 83% within 72 h after inoculation at 50 IJ/ml and 100% within 48 h at 500 IJs/ml. *S. riobrave* and *S. harryi* n. sp. produced only 66% mortality within 60 h at 500 IJs/ml. Against aphid, *S. thermophilum* caused 66% and 83% mortality at 50 and 500 IJs/ml, respectively within 3 days post inoculation. None of the *Steinernema* spp. caused significant mortality of whitefly at 50 IJs/ml; only the higher concentration produced mortality. At 500 IJs/ml *S. riobrave* caused maximum mortality of about 66% within 72 h after inoculation. *S. carpocapsae* was the least effective strain as it caused only 33 % mortality of all tested insects at both low and high doses. Emergence was observed only in 16.6% of the mealybug cadavers infected with *S. thermophilum* and *S. harryi* n. sp, while no emergence was observed in whitefly and aphid cadavers. Among the evaluated indigenous strains, *S. thermophilum* proved to be effective against homopteran insect pests as it caused significant mortality of mealybugs and considerably good mortality of aphids and whiteflies.

The most effective strain from the previous laboratory experiment, *S. thermophilum* was tested for its efficacy against *Phenacoccus solenopsis* infecting brinjal under field conditions. Mealybugs were artificially inoculated on plants, allowed for establishment and sprayed with nematodes in three different concentrations (1000, 2000 and 3000 IJs/ml) along with the adjuvant 0.033% APSA80. For comparison, one insecticide Chlorpyriphos 20% EC and a control without nematodes were used. The results revealed that the mortality percentage increased with increasing concentration. At 1000 IJs/ml, 25% mortality was obtained vis-á-vis 29% in 2000 IJs/ml and 32% in insecticide treatment, while the highest mortality of 34% was achieved with *S. thermophilum* applied at 3000 IJs/ml. From our study, it is concluded that *S. thermophilum* is an effective strain for the management of mealybugs as its ability to search and kill the insects seemed promising as bioinsecticide.

Development of a Molecular Marker for the Genes Responsible for Nematicidal Activity in Strains of *Trichoderma harzianum* (*Hypocrea lixii*) isolated in India and Field Evaluation of Bio-Efficacy

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Trichoderma harzianum (Rifai), a versatile bio-control agent has been extensively used for the management of pathogenic fungi infecting various crops. In our preliminary studies a few strains of this bio-agent were found to be infecting root knot nematodes (*Meloidogyne incognita* Chitwood) besides other pathogenic fungi. However a few other strains were only infecting pathogenic fungi. As there are variations in the efficacy of the strains, it was thought to carryout research on the molecular characterization of the strains of *T. harzianum* which are infecting the nematode as well as pathogenic fungi so as to control nematode induced disease complexes. These studies resulted into a molecular marker for the nemticidal activity of the 48 strains of *T. harzianum* collected from different agr-climatic regions of southern India. Though a lot research has been made in the field of applied aspects of *T. harzianum* for controlling nematodes, not much work has been done to understand basic aspects of its nematicidal activity at molecular level. In this study the work was highly concentrated on the molecular identification of the efficient strains that control both the nematodes and pathogénic fungi using *in silico* and molecular methods. The strains used in this work has been initially identified by phenotypical, biochemical and physiological tests. It was not sufficient for the confirmation of the traits of the closest related species leaving them un differentiable even after they were subjected to PCR for confirmation of ITS regions.

It was thought to develop molecular markers for these traits, as it was known that each trait is associated with some gene. A novel gene *PRA1* was identified in the investigations leading to the nematicidal activity during the egg stage in the life cycle of nematodes which was confirmed by mode of action experiments in the culture filtrates. With this potential information the research was further carried out to find out efficient strains antifungal activity along with nemticidal activity. As it was well known that *â-tubulin* gene was responsible for the antagonistic properties of certain bio-agents against pathogenic fungi. Molecular markers were developed for

PRA1 along with *â-tubulin* and all the strains identified as *T. harzianum* were investigated for their presence. A very few strains of *T. harzianum* have shown the presence of both the genes. Where as many of the strains have only *â-tubulin* gene and few again with *PRA1* alone. The work was further carried to confirm of the traits *in vitro* and *in vivo* by field evaluation which proved that the strains exhibiting the presence of both the genes were potential in controlling the disease complexes induced by nematodes than the strains with either of the genes alone were poor in controlling the disease complexes in crops.

Hence the markers developed in this investigation helps in identifying the best strain of *T. harzianum* which can also control the disease complexes induced by root knot nematodes. It is also useful in identifying the strains with the nematicidal activity. The gene sequences identified were submitted in the gene bank NCBI.

Production of Nematode Free Bio-Agent Colonized Seedlings of Cabbage

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Cabbage (*Brsassica oleraceae* L. var *capitata*) is a popular leafy green vegetable. It is a major source of food and has been used in medicinal applications. Our surveys in Karnataka indicated the infestation of the seedlings of cabbage by pathogenic fungus (*Fusarium oxysporum*) and nematodes (*Meloidogyne incognita*). However the seedling producers are not aware of this. Even farmers without knowing anything transplant these seedlings in the main field. In this manner pathogenic fungi and nematodes are carried along with the seedlings to the farmer's fields. Later on, farmers have to spend lot of amount for the management of these pathogens in their fields. *Meloidogyne incognita* (Chitwood) and *Fusarium oxysporum* together cause disease complex on cabbage. It is important that the seedlings should be free from the infestation of pathogenic fungi and nematodes. Hence it was thought to standardize a holistic approach to manage these pathogens in cabbage by the use of combinations of potential bio-agents *Trichoderma harzianum, Pseudomonas fluorescens* and *Paecilomyces lilacinus.* The experiments were also conducted to find the most effective strains of *T. harzianum, P. fluorescens* and *P. lilacinus* for the combined application by evaluation of their compatibility and bio-efficacy against *Meloidogyne incognita* and *Fusarium oxysporum in vitro.* Experiments were conducted using various IIHR- formulations of *T. harzianum* ($2x10^6$CFU/g), *P. fluorescens* ($2x10^8$CFU/g) and *Paecilomyces lilacinus* ($2x10^6$CFU/g) along with neem cake. One hundred kg of neem cake was enriched for 15 days mixing 1kg of each *T. harzianum* ($2x10^6$ cfu/g), *P. fluorescens* ($2x10^8$cfu/g) and *Paecilomyces lilacinus* ($2x10^6$CFU/g). Optimum moisture was maintained in neem cake for better enrichment. Bio-pesticide enriched neem cake was mixed at the rate of 50-60g/kg of cocopeat. One kg cocopeat is used for filling in one protray. By this method we got pathogen free seedlings of cabbage. The seedlings were vigorous and healthy and were colonized by these useful bio-agents.

Engineering Resistance against Root-Knot Nematode, *Meloidogyne incognita*inTomato by RNAi

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India is the world's third largest tomato (*Lycopersicumesculentum*Mill.)producer, producing about 124.30 lakh MT annually. The crop productivity is constrained because of several biotic stresses namely, insects, fungi, bacteria, viruses and nematodes.Root-knot nematodes (*Meloidogyne* species), reniform nematodes (*Rotylenchulus reniformis*), cyst nematode (*Globoderarostochiensis*) and several ecto-parasitic nematodes parasitize the crop worldwide. World overthe root-knot nematodes (*Meloidogyne* spp.) are most damaging and cause an average loss of about 27% to the crop. As of date we don't have environmentally acceptable and effective nematode control methods. The management strategies mainly rely on use of crop rotation and hazardous chemicals. Crop resistance is the most preferred and cost effective strategy to combat the nematode damage. But as resistance sources in nature are few, we need to design or engineer resistance in crops, so as to offer environmentally amenable and economical management options to the Indian farmer.

The identification of nematode parasitism genes, genome sequenced data of nematodes, molecular understanding of nematode host interactions by using tools of comparative and functional genomics has providednew targets for engineering resistance using RNAi technology. RNAi is genetic technique which permits ablation of mRNA induced by double stranded RNA (dsRNA), aimed to disrupt flow of genetic information. The most interesting aspects of RNAi is that, it is highly precise, remarkably powerful and the interfering activity can cause interference in cells and tissues even far from the site of introduction which offers a promise of as cheap and environmentally safe way of managing plant parasitic nematodes.

In this study we transformed tomato (cv. Pusa Ruby)using *Agrobacterium tumefaciens* method so as to express*inplanta*dsRNAaimed at knocking down AY134442, a parasitism gene of the subventral esophageal gland of *M. incognita*. The transformed plants were confirmed for presence and expression of the gene using PCR and RT PCR techniques.The 1kb band amplicon obtained on using AY134442gene specific primers confirmed the presence and expression of the gene in T_0 and T_1 generations tomato plants.Molecularly confirmed tomato transgenic lines (T_0 and T_1) were used for root-knot nematode, *M. incognita race 1* infection analysis.The nematode penetration studies conducted in T_0 and T_1 generation plants showed significant per cent decrease over control in both the generations. The maximum overall reduction in penetration of juveniles over control was 85%, 61.94% and 63.4% in T_0 while it was 78.72%, 76.43% and72.72% in T_1 generation after 1, 3, and 7 days of inoculation respectively. The nematode development and reproductionstudies were conducted after 35 days of inoculation. The maximum per cent reductionin the development of adult females over control in T_0 and T_1 was 62.64% and 60.56% respectively.There was significant reduction in the number of root galls in the transgenic lines as compared to control. The maximum reduction in number of galls per plant in T_0 and T_1 generation was 62.78% and 60.56% respectively.All the transgenic events had significantly reduced number of eggs per egg mass over control.The overall per cent reductionin eggs per egg mass over control in T_0 and T_1ranged between 40% and 57% respectively.Transformed tomato plants showed better growth parameters as compared with nematode inoculated untransformed tomato plants.

It can be summarized that, *in planta*dsRNA production strategy for silencing the parasitism gene of the root-knot nematode considerablyprotects tomato plant from nematode damage, and could be utilized for engineering resistance. Other important/crucial genes related to important pathways, body systems, or genes involved in development of plant parasitic nematodes could be good targets for silencing and designing RNAi based resistance against specific nematodes. Future research can address tissue specific gene silencing, with the use of nematode responsive promoters.However, there is still need of increased understanding of mechanism by which pathogen effectors are operated. The future promises to hold for molecular management strategies that are target oriented and environment decontaminating.

Molecular Characterization of *Pochonla chlamydosporia*, Zare and Bio-Management of Disease Complex in Okra (*Abelmoschus esculentus* L. Moench)

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Okra (*Abelmoschus esculentus* L. Moench) is one of the important vegetable crops and is produced in most parts of the country. The productivity of this vegetable crop is seriously hampered by various diseases and pests. It is highly susceptible to root-knot nematode *Meloidogyne incognita* (Kofoid & White) Chitwood and *Fusarium oxysporum* f.sp.*vasinfectum* . Both these organisms cause diseases complex leading to reduction in its production to the tune of 35 to 50%.

Pochonia chlamydosporia, Zare is a nematophagous fungus has proved to be promising bio-agent with tremendous potential in its application in a field conditions. These bio-agent strains show a lot of variations in their bio-efficacy. Hence it was thought to undertake research on molecular characterization on this potential bio agent and also develop a molecular profile of efficient strains of *P. chlamydosporia.*

Successful isolation of this fungus was done on semi selective medium. Two different strains were isolated and number PC-3 and PC-N. Fungus produced verticillate nature of mycelium, which bears conidia and chlamydospores.

Chlamydospore production by the different isolates of *P. chlamydosporia* confirmed that the isolated filamentous fungus was *P. chlamydosporia.*

The universal primers ITS1 and ITS4 were used for PCR analysis for species level identification of newly isolated strains of *P. chlamydosporia* . ITS primers generated a single band of 600bp. This size corresponds to the expected size according to the ITS sequence of other fungi. Amplification of IGS region gave 500bp band size on 1% agarose gel.

PCR was carried out to detect β-tubulin gene using the specifically designed primers. The gel picture analysis revealed that the PCR product obtained was 270bp, which clearly indicated the detection of β-tubulin gene which has got 99 % identity with *P. chlamydosporia* (AY642328.1) and 92% identity with *Eplchloe festucae* β-tubulin gene (AY722412.1). This tells about the presence of β-tubulin gene in *P.*

chlamydosporia. β-tubulin gene of present study was similar and grouped under *Pochonia* [*Verticillium*(AY642328.1)].

From our molecular approaches it was clear that *Pochonia chlamydosporia* strains were known to contain â-tubulin gene which is a target site for thiabendazole and carbendazium and helps during the selection process or selection strategies and design of primers to screen the fungus either benzimidazole resistant strain or susceptible strains and use them in bio- control studies.

As a pathogenic fungus and nematode are involved in producing disease complex it was thought to develop methods for sustainable management of disease complex using a bacterial antagonist *P. fluorescens* and nematode antagonist *P. chlamydosporia*.

Seeds of okra were treated with talc based formulation of *P. fluorescens* ($2x10^8$CFU/g) and *P. chlamydosporia*. ($2x10^6$ CFU/g) @ 25g/kg of seeds. Soil application of 25g/sqm of *P. fluorescens* ($2x10^8$CFU/g) and *P. chlamydosporia*. ($2x10^6$ CFU/g) enriched de-oiled neem cake has proved to be an effective treatment in combating the damage caused by *Meloidogyne incognita* and *Fusarium oxysporum* f.sp.*vasinfectum* to the tune of 68 and 57% respectively. This treatment also increased the yield of the crop by 28% under field conditions.

Impact of Consortia of Bio-inoculants in the Bio-management of Nematode Induced Disease Complex in Tomato (*Lycopersicon Esculentum*)

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Tomato is an important solanaceous crop being rich in antioxidants and forms an integral part of a balanced diet. The crop is highly susceptible to root - knot nematode *Meloidogyne incognita* and *Fusarium oxysporum* f.sp.*lycopercisi*. Both these organisms cause disease complex leading to reduction in its productivity. The present study was undertaken to assess the effect of consortia of inocula of AM fungus (*Glomus fasciculatum*), Fluorescent pseudomonads and *Paecilomyces lilacinus* on growth, population of nematodes, disease incidence and yield of tomato. Studies on the relationship between roots and microbes are essential to design useful consortia of microbes. When these microbes are used as bio-inocuiants in crop cultivation, it can control the nematodes and fungal disease incidence and increase yield. Mycorrhizal fungi are known to enhance productivity on sustainable basis by improving nutrient recycling and inducing synthesis of plant growth hormones. Application of vermicompost enriched with these bio-inoculants enhanced the plant growth parameters and also improved the crop productivity. Application of vermicompost (1000kg) enriched with 2 kg each of *P. fluorescens* ($2x10^8$CFU/g) and *P.lilacinus.* ($2x10^6$CFU/g) and 5 kg of *G. fasciculatum* has proved to be an effective treatment in combating the damage caused by *M. incognita* and *F.oxysporum* f.sp.*lycopercisi* on tomato. One ton bio-pesticide enriched vermicompost was applied in one acre before transplanting the seedlings of tomato. This treatment reduced the root population of *M. incognita* by 70% and infestation of disease by 68%. This treatment also increased the yield of the crop by 30% under field conditions.

Holistic Approach for the Management of Nematodes and other Pathogens on Seedlings of Horticultural Crops

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Large percentage of horticultural crop seedlings raised by the nursery producers in India are generally infested by nematodes and other pathogens. We observed seedlings tomato, brinjal, cauliflower, cabbage, capsicum, carnations, gerbera, acid lime and Papaya infested with one or more species of nematodes such as *Meloidogyne incognita, Rotylenchulus reniformis*, pathogenic fungi such as *Fusarium oxysporum, Phythium sp.* in the nurseries in Karnataka and Andhra Pradesh. Nematode infested seedlings when transplanted would carry the pathogenic nematodes and fungi along with the roots to the main field. These nematodes further multiply and infect the crop. This will reduce the yields considerably. Further the nematode infestation pave the way for the entry other pathogens resulting in the significant reduction in the yield of the crop due to disease complex caused by nematode and fungus. Nematode damage also results into the breakdown of the resistance against pathogenic fungi and bacteria.

Because of all these reasons it was thought to standardize the methods for the management of nematodes on the nursery seedlings of papaya. Seeds were treated with talc based formulations of *Pseudomonas fluorescens* (10^8 cfu/g) and *Paecilomyces lilacinus* (10^6 cfu/g) at the rate of 20g/kg of seed. Treated the soil mixture/ substrate (used for producing the nursery seedlings in plastic bags or trays) with *Bacillus subtilis* or *P. fluorescens* (10^6 cfu/g) and *P. lilacinus* (10^6 cfu/g) each at the rate of 2 kg/ton before using it for seedling production; in addition to this, mixed neem or pongamia cake @ 50 kg/ton of the substrate. Studies on the compatibility of these bio-pesticides among themselves indicated that these bio-pesticides are compatible on the roots of papaya (Arka surya). *Bacillus subtilis, P. fluorescens* and *P. lilacinus* did not affect each other's colonization on the roots of tomato, brinjal, cauliflower, cabbage, capsicum, carnations, gerbera, acid lime and Papaya and they were compatible with each other.

Since the seedlings roots get colonized by the bio-agents, they get carried to the main field and thus the bio-agents reach the main field. Through these methods it was possible to produce the vigorous and bio-agent colonized seedlings of tomato, brinjal, cauliflower, cabbage, capsicum, carnations, gerbera, acid lime and Papaya which could establish well in the main field. Due to presence of bio-agents on the roots of the seedlings they were able to withstand the attack of nematodes and other pathogens after transplanting and resulted in significantly higher yields to the extent of 24 – 32%.

Biotechnological Approaches for Bio-Management of Disease Complex in Carrot (*Daucus carota* L.) by using Fluorescent Pseudomonads and *Paecilomyces lilacinus*

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Carrot (*Daucus carota* L.) is the all time favourite vegetable occupies an important place among the fresh vegetables in India and world. It *is* seriously hampered by disease complex caused by *Meloidogyne incognita* and *Erwinia carotovora.* These two pathogens reduce the productivity up to 20 – 40%. Indiscriminate use of chemical pesticides proved ecologically hazardous. Hence it was thought to standardize a eco-friendly approach to manage the disease complex in carrot using bio-agents.

Single bio-agent cannot be very effective in the management of disease complex in different soil types and agro climatic regions. Hence an attempt was made to evaluate the combined effect of Fluorescent Pseudomonads and *Paecilomyces lilacinus* (nematophagous fungi) against disease complex at Indian Institute of Horticultural Research, Bangalore.

The bio-agents isolated for this study were initially identified at their genus level by the standard biochemical tests and microscopic observations. 16srRNA gene from putative *Pseudomonas* strains were amplified using Universal primers by following standard PCR protocols and were confirmed as *P. putida* by sequencing the 1500bp amplicon. *P. lilacinus* strains were confirmed by the amplification and sequencing of the ITS region by using the universal primers ITS1 and ITS4.

Formulations of identified strains of *P. putida* and *P. lilacinus* were prepared separately. One hundred kgs of deoiled neem cake was enriched for 15 days by mixing 5kg of *P. putida* or *P lilacinus* formulation. Optimum moisture was maintained for better enrichment of neem cake with bio-pesticides. These enriched neem cake with either *P. putida* or *P. lilacinus* was applied @ 20g/m^2. In combination treatment *P. putida* and *P. lilacinus* enriched neem cake was applied @10g /m^2. Seeds of Carrot (cv. kuroda) were also treated with the 10% suspension of *P. putida* and *P. lilacinus* enriched neem cake for 5-10 min. Results indicated that combination treatment of *P. putida* and *P. lilacinus* was more effective when compared with either of *P. putida* and *P. lilacinus* treated individually. This increased the root colonization of both bio–agents and reduced the incidence of *M. incognita* and *Erwinia carotovora* by 66% and 58% respectively. There was also a significant increase in the yield of the crop which was to the tune of 23%. Cost benefit ratio was also calculated. All other relevant details will be presented during the conference.

SESSION IV ABSTRACT

Screening of Banana Hybrids (Phase II Hybrids) Resistant against *Helicotylenchus multicinctus*

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Banana (*Musa* spp.) which is truly a tropical treasure is being targeted by many pests and diseases. Worldwide, nematodes are considered the most important pest of bananas and plantains, for which remedies are the immediate requirement. By damaging the plant root system, nematodes not only hamper the uptake of water and nutrients, but also the growth and fruit production. *Helicotylenchus multicinctus* is considered to be an endoparasite in banana which is able to complete its life cycle within the root cortex where both the sexes, all juvenile stages and eggs can be found and produces shallow superficial lesions in banana roots which resemble pustule like eruptions. The reactions of Eighteen new synthetic banana phase II hybrids to *Helicotylenchus multicinctus* was studied under field as well as artificially inoculated pot conditions. Hybrids H 531 exhibited resistance, while H-02-34, H-03-05, H-03-11, H-03-13, H-03-17, H-04-12, H-04-24 and NPH-02-01showed tolerance. The role of biochemical contents like total phenols, OD phenols, lignin and enzymatic activity like peroxidase, polyphenol oxidase and phenylalanine ammonia lyase revealed that there were more activities in resistant and tolerant genotypes *viz-a-viz* susceptible ones. The hybrids, H 531 (AAB) took the shortest duration of 213.00 days for shooting and recorded moderate bunch weight of 13.50 kg.

Screening under field studies for nematodes in phase II hybrids showed that the hybrids H 531 exhibited resistance, while H-02-34, H-03-05, H-03-11, H-03-13, H-03-17, H-04-12, H-04-24 and NPH-02-01showed tolerance.

Studies on the role of biochemical contents like total phenols, OD phenols, lignin and enzymatic activity like peroxidase, polyphenol oxidase and phenylalanine ammonia lyase revealed that there were more activities in resistant genotypes *viz-a-viz* susceptible ones. Root histological studies also confirmed that the resistant and tolerant hybrids had more number of phenolic cells and lignified cells than the susceptible hybrids.

Among the phase II hybrids, hybrid H 531 was resistant and hybrids, H-02-34, H-03-05, H-03-13, H-03-17, H-04-12, H-04-24 and NPH-02-0 was tolerant to to nematodes (*Helicotylenchus multicinctus)*

Results on phase II evaluation of 18 hybrids developed earlier in this department and one from present studies reveal that, the hybrid H-04-24 recorded the highest pseudostem height and H-02-23 had the maximum girth. Among the hybrids, H 531 (AAB) took the shortest duration of 213.00 days for shooting and recorded moderate bunch weight of 13.50 kg. All hybrids recorded male and female fertility, except hybrid H-04-21, while the hybrid H-03-17 recorded the maximum pollen out put/ anther.

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