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In the coming 50 to 60 years the world population will about double and hopefully also become more prosperous. This demands large yield increases in our food crops, which have to be grown in more sustainable agricultural systems. The need for durable insect and disease resistance, therefore, can be expected to grow enormously. The past decades have brought incredible developments in the field of agriculture. The use of pesticides still dominates the management of insect pests and is a health hazard for humans and the environment. Control of insect pests and plant disease by chemical can be spectacular but the accumulation of harmful chemical residue sometimes causes serious ecological problems. Application of synthetic pesticides is a usual practice to ward off infestation of insect pests and diseases from field crops. The objectives of IPM and Conservation Agriculture are the same: sustain productivity, conserve natural resources, reduce production costs, improve environmental health, maintain biodiversity, and reduce agrochemical use for crop production/protection. The development of resistance and resurgence to these chemical insecticides has made the management of insect pests extremely difficult. The other environmental concerns are also associated with insecticides like residue in food and feed, health hazard to humans reduction in non-target organisms population and environmental pollution. So the entomologists all over the world trying to find eco-friendly and sustainable alternatives for management of insect pests. All these methods of insect pest management are extremely safe to humans and environment. These methods also prevent development of resistance and resurgence in insect. However, much more needs to be done so that farmers are able to adopt these management technologies on large scale to avoid insect pest damage and reduce use of chemical insecticides. Hence, there is need to replace the chemical fungicides by bio-agents, prepared from plant extracts, plant oils and antagonistic microorganisms. It is possible that integration of all these approaches is (use of plant extracts, plant oils and antagonistic microorganisms) an economically viable alternative for crop production system can be developed. So the use of bio-agents proved to be economical alternative that can be implemented at the farm level. Formulation must have adequate shelf life, stability, and titer. Before any formulated product is marketed, it must be thoroughly tested by growers, whose comments, critiques, and suggestions for improvement, must consider. Besides, disease forecasting, gene expressions by pathogens, resistances genes in host and management aspects have also been included as discussed by different researchers which may be useful for future researcher in formulating their research plan.

The basic theme of the Recent Trends in Integrated Pest and Disease Management and this has been emphasized in most of the chapters. The book is structured into 27 chapters and primarily for the degree, post graduate students and for the researchers. We wish to express our deep sense of gratitude to those who helped us directly or indirectly during the preparation of the manuscript of this text. We hope that the book is useful and interesting to readers, teachers and students and would create in them the urge to know more about recent researchers going related to Integrated Pest and Disease Management.

Wajid Hasan
Rashmi Nigam
Joginder Singh
Rajendra Singh
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CONSERVATION AGRICULTURE AND ITS PRINCIPLES IN PEST MANAGEMENT

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Conservation agriculture is a set of soil management practices that minimize the disruption of the soil's structure, composition and natural biodiversity. Despite high variability in the types of crops grown and specific management regimes, all forms of conservation agriculture share three core principles. These include:

1. Maintenance of permanent or semi-permanent soil cover (using either a previous crop residue or specifically growing a cover crop for this purpose);
2. Minimum soil disturbance through tillage (just enough to get the seed into the ground);
3. Regular crop rotations to help combat the various biotic constraints.

Status of conservation agriculture in India and abroad

Globally, CA is being practiced on about 125 M ha (Table 1. The major CA practicing countries are USA (26.5 M ha), Brazil (25.5 M ha), Argentina (25.5 M ha), Canada (13.5 M ha) and Australia (17.0 M ha. In India, CA adoption is still in the initial phases. Over the past few years, adoption of zero tillage and CA has expanded to cover about 1.5 million hectares (Jat et al., 2012; www.fao.org/ag/ca/6c.html. The major CA based technologies being adopted is zero-till (ZT) wheat in the rice-wheat (RW) system of the Indo-Gangetic plains (IGP. In other crops and cropping systems, the conventional agriculture based crop management systems are gradually undergoing a paradigm shift from intensive tillage to reduced/zero-tillage operations.

Prospects of conservation agriculture: (Suraj Bhan and U. K. Behera, 2014)

The direction that Asian countries take to meet their food and energy needs during the coming decades will have profound impacts on natural resource bases, global climate change and energy security for India, Asia and the world. These challenges draw attention to the need and urgency to address options by which threats to Indian/Asian agriculture due to natural resource degradation, escalating production costs and climate change can be met successfully. A shift to no-till conservation agriculture is perceived to be of much fundamental value in meeting these challenges. Asian farmers/researchers will continue to need assistance to reorient their agriculture and practices for producing more with less cost through adoption of less vulnerable choices and pathways. Therefore, business as usual with conventional agriculture practices does not seem a sustainable option for sustainable gains in food-grain production, and hence CA-based crop management solutions adapted to local needs will have to play a critical role in most ecological and socio-economic settings of Asian Agriculture.

The promotion of Conservation Agriculture (CA) under Indian/Asian context has the following prospects:

- **Reduction in cost of production** – This is a key factor contributing to rapid adoption of zero-till technology. Most studies showed that the cost of wheat production is reduced by Rs. 2,000 to 3,000 ($ 33 to 50) per hectare (Malik et al., 2005; RWC-CIMMYT, 2005. Cost reduction is attributed to savings on account of diesel, labour and input costs, particularly herbicides.
- **Reduced incidence of weeds** – Most studies tend to indicate reduced incidence of Phalaris minor, a major weed in wheat, when zero-tillage is adopted resulting in reduced in use of herbicides.
- **Saving in water and nutrients** – Limited experimental results and farmers experience indicate that considerable saving in water (up to 20% – 30%) and nutrients are achieved with zero-till planting and particularly in laser leveled and bed planted crops. De Vita et al. (2007) stated that higher soil water content under no-till than

<table>
<thead>
<tr>
<th>Country</th>
<th>Area (M ha)</th>
<th>% of Global Area</th>
</tr>
</thead>
<tbody>
<tr>
<td>USA</td>
<td>26.5</td>
<td>21.2</td>
</tr>
<tr>
<td>Brazil</td>
<td>25.5</td>
<td>20.4</td>
</tr>
<tr>
<td>Argentina</td>
<td>25.5</td>
<td>20.4</td>
</tr>
<tr>
<td>Australia</td>
<td>17.0</td>
<td>13.6</td>
</tr>
<tr>
<td>Canada</td>
<td>13.5</td>
<td>10.8</td>
</tr>
<tr>
<td>Russian</td>
<td>4.5</td>
<td>3.6</td>
</tr>
<tr>
<td>Federation</td>
<td>3.1</td>
<td>2.5</td>
</tr>
<tr>
<td>China</td>
<td>2.4</td>
<td>1.9</td>
</tr>
<tr>
<td>Paraguay</td>
<td>1.6</td>
<td>1.3</td>
</tr>
<tr>
<td>Kazakhstan</td>
<td>5.3</td>
<td>4.2</td>
</tr>
<tr>
<td>Others</td>
<td>124.8</td>
<td>100.0</td>
</tr>
</tbody>
</table>
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under conventional tillage indicated the reduced water evaporation during the preceding period. They also found that across growing seasons, soil water content under no-till was about 20% greater than under conventional tillage.

- **Increased yields** – In properly managed zero-till planted wheat, yields were invariably higher compared to traditionally prepared fields for comparable planting dates. CA has been reported to enhance the yield level of crops due to associated effects like prevention of soil degradation, improved soil fertility, improved soil moisture regime (due to increased rain water infiltration, water holding capacity and reduced evaporation loss) and crop rotational benefits. Yield increases as high as 200 – 500 kg ha-1 are found with no-till wheat compared to conventional wheat under a rice-wheat system in the Indo-Gangetic plains (Hobbs and Gupta, 2004. Review of the available literature on CA provides mixed indications of the effects of CA on crop productivity. While some studies claim that CA results in higher and more stable crop yields (African Conservation Tillage Network, 2011), on the other hand there are also numerous examples of no yield benefits and even yield reductions particularly during the initial years of CA adoption.

- **Environmental benefits** – Conservation agriculture involving zero-till and surface managed crop residue systems are an excellent opportunity to eliminate burning of crop residue which contribute to large amounts of greenhouse gases like CO2, CH4 and N2O. Burning of crop residues, also contribute to considerable loss of plant nutrients, which could be recycled when properly managed. Large scale burning of crop residues is also a serious health hazard.

- **Crop diversification opportunities** – Adopting Conservation Agriculture systems offers opportunities for crop diversification. Cropping sequences/rotations and agroforestry systems when adopted in appropriate spatial and temporal patterns can further enhance natural ecological processes. Limited studies indicate that a variety of crops like mustard, chickpea, pigeon pea, sugarcane, etc., could be well adapted to the new systems.

- **Resource improvement** – No tillage when combined with surface management of crop residues begins the processes whereby slow decomposition of residues results in soil structural improvement and increased recycling and availability of plant nutrients. Surface residues acting as mulch, moderate soil temperatures, reduce evaporation, and improve biological activity.

**Constraints for adoption of conservation agriculture:**
A mental change of farmers, technicians, extensionists and researchers away from soil degrading tillage operations towards sustainable production systems like no tillage is necessary to obtain changes in attitudes of farmers (Derpsch, 2001. Hobbs and Govaerts (2010) however, noted that probably the most important factor in the adoption of CA is overcoming the bias or mindset about tillage. It is argued that convincing the farmers that successful cultivation is possible even with reduced tillage or without tillage is a major hurdle in promoting CA on a large scale. In many cases, it may be difficult to convince the farmers of potential benefits of CA beyond its potential to reduce production costs, mainly by tillage reductions. CA is now, considered a route to sustainable agriculture. Spread of conservation agriculture, therefore, will call for scientific research linked with development efforts. The following are a few important constraints which impede broad scale adoption of CA.

- **Lack of appropriate seeders especially for small and medium scale farmers:** Although significant efforts have been made in developing and promoting machinery for seeding wheat in no till systems, successful adoption will call for accelerated effort in developing, standardizing and promoting quality machinery aimed at a range of crop and cropping sequences. These would include the development of permanent bed and furrow planting systems and harvest operations to manage crop residues.

- **The wide spread use of crop residues for livestock feed and fuel:** Specially under rainfed situations, farmers face a scarcity of crop residues due to less biomass production of different crops. There is competition between CA practice and livestock feeding for crop residue. This is a major constraint for promotion of CA under rainfed situations.

- **Burning of crop residues:** For timely sowing of the next crop and without machinery for sowing under CA systems, farmers prefer to sow the crop in time by burning the residue. This has become a common feature in the rice-wheat system in north India. This creates environmental problems for the region.

- **Lack of knowledge about the potential of CA to agriculture leaders, extension agents and farmers:** This implies that the whole range of practices in conservation agriculture, including planting and harvesting, water and nutrient management, diseases and pest control etc. need to be evolved, evaluated and matched in the context of new systems.
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- **Skilled and scientific manpower:** Managing conservation agriculture systems, will call for enhanced capacity of scientists to address problems from a systems perspective and to be able to work in close partnerships with farmers and other stakeholders. Strengthened knowledge and information sharing mechanisms are needed (Suraj Bhan and U. K. Behera, 2014).

**Conservation Agriculture and Insects:-**

Insects are dominant animals in the world, so it is not surprising that they interact with humans in more ways than any other groups of organisms. More than 1 million insect species have been described. Most (99%) are innocuous or beneficial to mankind, such as silkworm, honeybees, pollinators, parasitoids and predators. Only a small proportion (1%) is our competitors causing damage to our crops, stored products, other belongings, and acting as vectors for plant and animal pathogens (Grimaldi and Engel 2005; Pedigo and Rice 2009. While the number of insect pest species in small in comparison with the total described species, they still need significant funds, time and efforts to reduce their negative impact on crop production, crop protection, human health and welfare.

Insects provide important ecological services as decomposers, consumers, predators and parasitoids (Swift & Anderson 1989; Miller1993. Decomposition of plant and animal matter by fly maggots (blow flies, muscid flies, small dung flies etc.) and grub and adult beetles (dermestid beetles etc.) is essential for recycling organic matter in ecosystems (Frost 1959. Other predators (green lacewing, lady beetles, predaceous diving beetles and ground beetles etc.) and parasites Encarsia spp. Ichneomids etc play an important role in regulating many phytophagous pest populations (Olembo and Hawksworth 1991. Insect pest management is divided into two parts 1) Natural control & 2) Applied or artificial control.

Natural or automatic control includes abiotic (weather, climate) and biotic (predators, parasites, diseases and other competitors) factors, while applied control includes control measures which are intentionally applied in the filed according to need. Currently, due to the intensification of agricultural practices, only applied control is practiced for crop protection from insect pests.

No doubt, CA is advantages for controlling insect pests by increasing bio-diversity. CA promotes biological diversity below and above ground by making ground cover favorable to the natural biota (Jaipal et al. 2002), which helps to control insect pests. More beneficial insects (predators, parasitoids) have been observed in fields with ground cover and mulch (Kenedall et al. 1995; Jaipal et al. 2002) which keep insect pests in check.

There is no evidence of complete control of insect pests in CA farming systems, which remains a challenge for researchers, farmers, and agriculture policy makers. The best option in this regard is IPM by integrating different techniques to keep insect pest populations at acceptable levels in CA cropping systems. (Jam Nazeer Ahmad, 2015).

**The effect of Conservation agriculture on insects and ecosystem services:**

(Heidi Meyer and DR Annemie Erasmus, 2017)

- Conventional agriculture practices such as continuous tillage lead to the disruption of soil structure and loss of fertile top soil, resulting in a reduction of soil productivity. Conservation agriculture (CA) is recognised as a way to combat soil deterioration brought on by conventional cultivation.

- CA practices in crop production systems may provide different habitats for hosting and supporting pests and may influence beneficial insect populations including underlining biodiversity that supports many ecosystem services.

- Insects play many important roles within an ecosystem such as predators, pollinators, detritivores, herbivores and parasitoids. They are efficient indicators of ecosystem functions, ideal to monitor the quality of a habitat and to observe how a site changes from time to time, and to measure habitat differences.

- There is a general lack of information and statistics concerning the effect of CA on arthropod diversity, and the potential ecosystem services they provide in South Africa.

- To understand the impact of landscape structures on the diversity and abundance of beneficial and harmful arthropods, pest regulation and ultimately crop yield can be of significant help to enhance the management of agricultural landscapes.

- During the growing seasons of 2014/2015, 2015/2016 and 2016/2017 arthropods were sampled via pitfall traps in the Ottosdal, Hartbeesfontein, Sannieshof, Vredefort and Kroonstad areas where well-established CA and conventional farming systems were implemented.

- Arthropods are invertebrate animals of the large phylum Arthropoda, such as insects, spider, or crustacean. A total number of 40,000 soil-dwelling arthropods and 197 mor-aphospecies were collected during this study
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from 14 different orders and 30 different families.

• The term morphospecies is defined as organisms that are classified in the same species if they appear identical by morphological criteria.

• Morphospecies as well as their abundance at a certain time can be tracked to the date of occurrence and what treatment were used along with the type of crop planted (Table 2).

• To prevent crop damage by insects, it is essential to monitor and inspect the crop fields regularly. The identification of morphospecies and data recorded are of great value to monitor the density of the occurrence of pests.

<table>
<thead>
<tr>
<th>Orders</th>
<th>No of morphospecies</th>
<th>CA no. of individuals</th>
<th>Conventional no of individuals</th>
</tr>
</thead>
<tbody>
<tr>
<td>Coleoptera (Beetle)</td>
<td>79</td>
<td>19294</td>
<td>10059</td>
</tr>
<tr>
<td>Hymenoptera (bees, wasps and ants)</td>
<td>24</td>
<td>981</td>
<td>548</td>
</tr>
<tr>
<td>Araneae (spiders)</td>
<td>18</td>
<td>448</td>
<td>128</td>
</tr>
<tr>
<td>Hemiptera (bugs)</td>
<td>22</td>
<td>228</td>
<td>85</td>
</tr>
</tbody>
</table>

• Over the past three growing seasons the number of morphospecies was significantly higher in a CA system as compared to the conventional systems. Therefore, tillage practices may target certain morphological species types (Graph 1).

• The overall total number of individuals was significantly higher in a CA system than conventional farming system. CA contributes in supporting a higher and richer biodiversity (Graph 2).

• To determine if CA can provide ecosystem services by controlling pests with beneficial insects, the Chilo borer (*Chilo partellus*) was used. Larvae and pupae of the Chilo borer (Photo 1) were pinned onto petri dishes to monitor the predator activity between CA and conventional farming systems.

• Results from this experiment can be used to determine if CA provides potential ecosystem services through controlling insect pest with beneficial insects.

• The total percentage predation of the Chilo borer larvae by beneficial predators differs significantly (P = 0.003) between CA and conventional farming (Graph 3). Although no significant difference was observed with the percentage predation on Chilo borer pupae (Graph 4), the predation was still higher in CA compared to the conventional system.

• It is clear from data compiled that arthropods react to the conditions surrounding them. Therefore, the habitat of beneficial insects must be protected and maintained by leaving crop residue on the ground. CA practices support the increase and build-up of natural enemy population.

• Ecosystem services depend on arthropod movement, abundance and diversity across agricultural landscapes at different scales. The biodiversity is higher in CA than conventional practices and a higher diversity relates to increased ecosystem services.
Three core principles of conservation agriculture in insect pests management:

1. **Regular crop rotations:**

   **Biological basis of rotational effects** (Robert J. Wright, 1995)

   Some rotational systems were developed by early agriculturalists through trial and error, without any understanding of the underlying mechanisms responsible for their success. In the last 100 years research has identified the basis for the effects of crop rotation in insect control. Two important factors influencing the impact of a particular rotation on an insect are the host range of the insect and its degree of mobility. Insect species vary in the range of food plants or ovipositional hosts that they will accept. Host selectivity may occur in either the egg-laying behavior of the adult or the feeding behavior of the larva. Some species have very specific requirements and will die or move away in the absence of their required hosts. Other insect species have a broad range of host plant species on which they will feed or lay eggs. Mobility of an insect species is important because it influences how far an insect can travel to search out an acceptable host plant when it is presented with a less preferred plant species due to crop rotation. Depending on the species and stage (adult or immature), the degree of mobility varies from a few inches to several miles. The European corn borer is a good example of a Midwestern insect pest that is not affected by rotating corn with a non-host because of the flight capacity of the adult moth.
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Table 3. Effect of crop rotation of corn on insect populations or potential damage.

<table>
<thead>
<tr>
<th>Pest</th>
<th>None</th>
<th>Soybeans</th>
<th>Pasture &amp; Hay Crops</th>
</tr>
</thead>
<tbody>
<tr>
<td>Seed corn beetles</td>
<td>0</td>
<td>0</td>
<td>+a</td>
</tr>
<tr>
<td>Seed corn maggot</td>
<td>0</td>
<td>0</td>
<td>+</td>
</tr>
<tr>
<td>True armyworm</td>
<td>0</td>
<td>0</td>
<td>+</td>
</tr>
<tr>
<td>Chinch bug</td>
<td>0</td>
<td>0</td>
<td>+</td>
</tr>
<tr>
<td>White grubs</td>
<td>–</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Wireworms</td>
<td>–</td>
<td>–</td>
<td>+</td>
</tr>
<tr>
<td>Corn root aphid</td>
<td>–</td>
<td>–</td>
<td>+</td>
</tr>
<tr>
<td>Billbug</td>
<td>–</td>
<td>–</td>
<td>+</td>
</tr>
<tr>
<td>Grape colaspis</td>
<td>–</td>
<td>–</td>
<td>+</td>
</tr>
<tr>
<td>Northern corn rootworm</td>
<td>+</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>Western corn rootworm</td>
<td>+</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>Black cutworm</td>
<td>0</td>
<td>+</td>
<td>0</td>
</tr>
<tr>
<td>Slugs</td>
<td>–</td>
<td>–</td>
<td>0</td>
</tr>
<tr>
<td>Thrips</td>
<td>0</td>
<td>?</td>
<td>+</td>
</tr>
<tr>
<td>Spider mites</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>European corn borer</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Southwestern corn borer</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Southern corn rootworm</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Corn earworm</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Fall armyworm</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Corn leaf aphid</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

*a + means the practice will increase the population or damage from that insect; – means it will reduce the population or damage; 0 means no effect; ? means effect unknown Source: Luckmann and Metcalf (1975)*

Many of our best examples of pests controlled by crop rotation involve insects, such as white grubs, wireworms and corn rootworms, whose major feeding stage occurs in the soil. The mobility of these larvae is measured in inches or at most a few feet over their life span. Host selectivity may occur through either the egg laying behavior of the adult or the feeding behavior of the larva. Pest management recommendations may focus on either selecting rotations that decrease certain pest populations or avoiding rotations known to favor certain pests.

Crop rotation, other cultural practices, and plant resistance are generally assumed to be compatible with biological controls and to form the basis for nonchemical pest control in Integrated Pest Management systems for many crops. However, there are some cases of incompatibility between plant resistance and biological control, and between some cultural controls (e.g., those involving cultivation) and biological controls. Many studies have documented the beneficial effects of strip-cropping or interplanting, but normally these practices are not considered as crop rotations, and will not be covered.

Table 4. Probability estimates of economic soil insect damage in corn and suggestions for management according to cropping sequence, Illinois.

<table>
<thead>
<tr>
<th>Insect pest</th>
<th>Soybean Use</th>
<th>Corn</th>
</tr>
</thead>
<tbody>
<tr>
<td>Crop before corn</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Wireworm</td>
<td>1:100</td>
<td>1:200</td>
</tr>
<tr>
<td>Cutworm</td>
<td>1:25</td>
<td>1:100</td>
</tr>
<tr>
<td>Corn rootworm</td>
<td>1:1,000</td>
<td>2:3</td>
</tr>
<tr>
<td>White grub</td>
<td>1:500</td>
<td>1:10</td>
</tr>
<tr>
<td>Seed corn maggot</td>
<td>1:150</td>
<td>0:10</td>
</tr>
<tr>
<td>Billbug</td>
<td>1:00</td>
<td>0:50</td>
</tr>
<tr>
<td>Grape colapsis</td>
<td>1:00</td>
<td>1:00</td>
</tr>
<tr>
<td>Need for soil insecticide</td>
<td>very low</td>
<td>Moderate</td>
</tr>
<tr>
<td>Pest management practice</td>
<td>planter box seed treatment scout for cutworms; bait for wireworms.</td>
<td>Scout for rootworm beetles; treat corn if population exceeds 0.75 beetle per plant during August</td>
</tr>
</tbody>
</table>
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In some situations, crop rotation may favor biological control indirectly by reducing insecticide use. Certain rotations can control some crop insect pest species (Table 3) and increased use of crop rotation would be expected to decrease the use of certain broad spectrum soil insecticides (e.g., those used for corn soil insect control. Most of these insecticides are nonselective and toxic to numerous predatory insects and mites in the soil.

**Pests controlled by rotations:**
The western and northern corn rootworms are responsible for most insecticide use in Nebraska crops. The western corn rootworm is the predominant species throughout Nebraska, accounting for 80-95 percent of corn rootworms found in corn. According to the most recent pesticide use survey in Nebraska, in 1987 4.6 million pounds of insecticide active ingredient was applied to corn (96 percent of total insecticide used. Of all the corn insecticide used, most is applied against corn rootworms.

However, it has long been known that corn rootworms can be easily controlled by crop rotation. Corn rootworms have an annual life cycle and a host range restricted to grass species. Eggs are laid in the soil of a corn field during July and August, then overwinter and hatch the next spring. If corn is rotated the next year with a corn rootworm non host, hatching larvae will starve and die. Rotation with a broadleaf crop such as soybeans greatly reduces the need for pesticide use. Based on Illinois estimates, the chance of economic damage from corn rootworms changes from 2/3 for continuous corn to 1/1000 for corn after soybeans (Table 4). Even some grass crops (for example, wheat and sorghum) can be used in a rotation with corn to control corn rootworms. Sorghum roots are toxic to corn rootworm larvae because of the presence of a compound which is converted to hydrocyanic acid when root tissue is injured.

Crop rotation should greatly reduce the use of insecticides against corn rootworms, the major target of most insecticide use in Nebraska. Although not well researched, reduced insecticide use in corn should encourage populations of various insect predators, especially those which spend a portion of their life in the soil (e.g., various beetle or fly larvae and soil mites. Increased biological control of several soil insects might be an added benefit from increased use of crop rotation in corn, although some rotations may slightly increase the occurrence of some soil insect pests (Tables 3 and 4).

### 2. Minimum soil disturbance through tillage: The Effects of Reducing Tillage on Insect Management

Although reducing tillage can shift the number and type of insects in a field, entomologists seldom alter their control recommendations for different tillage systems. However, there are some changes in reduced tillage systems that increase the potential for problems with specific pests. Those pests that overwinter in the soil or in crop residues and become active early in growth of the crop benefit most from tillage reduction. Although lower soil temperatures (2-5 °F cooler) may cause these insects to develop more slowly than in tilled soil, they can be more numerous because they have not been exposed to tillage. Other insects may decrease after a number of years of no-till. This may be due to increased survival of beneficial insects, ants, ground beetles, rove beetles, and spiders, all of which can contribute to insect pest control.

**The effects vary depending on the crop and pest:** Notes on specific insects:

**Wireworm:**
Wireworm numbers have increased in general in the past few years, but research does not suggest a direct link to reduced tillage. They may increase and cause damage after grassy weeds, with reduced soil disturbance, and

<table>
<thead>
<tr>
<th>Small grains</th>
<th>1:100</th>
<th>1:50</th>
<th>1:100</th>
<th>1:250</th>
<th>1:50</th>
<th>1:200</th>
<th>1:5,000</th>
<th>low</th>
<th>Bait for wireworms before planting; scout for cutworms at plant emergence.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Legume</td>
<td>1:25</td>
<td>1:25</td>
<td>1:50</td>
<td>1:150</td>
<td>1:10</td>
<td>1:50</td>
<td>1:4</td>
<td>low</td>
<td>moderate Bait for wireworms before planting; scout for cutworms at plant emergence.</td>
</tr>
<tr>
<td>Grass sod.</td>
<td>1:10</td>
<td>1:25</td>
<td>1:500</td>
<td>1:10</td>
<td>1:25</td>
<td>1:50</td>
<td>1:1,000</td>
<td>Moderate high</td>
<td>Use infurrow soil insecticide for wireworm and white grub; if no till, scout for foliar insect damage as corn emerges</td>
</tr>
</tbody>
</table>

Source: Kuhlman et al. 1988
where germination is delayed by cool soils.

**Slugs:**
Slugs can cause extensive damage to seedlings, especially in low lying wet areas. They are favored by unincorporated crop residues and cool, wet conditions. There are chemical controls but they are expensive.

**Seedcorn maggots:**
Seedcorn maggots are more of a problem where green manures are incorporated than where dead crop residues cover the soil. Winged aphids are more often attracted to barren ground than to residue covered ground. This can limit infestations in new stands but not after canopy closure.

**Corn earworms:**
Corn earworms may increase where planting or crop development is delayed in no-till fields.

**Black cutworms:**
It prefers to lay eggs in fields with unincorporated crop residues. Again, basic pest management practices will be more important when reducing tillage, especially when just starting. Planting corn into a grassy sod or wheat residue requires a high level of insect management. Wireworms and cutworms can be problem. Wheat curl mite can also move from green grass to emerging corn and can infect crops with virus diseases like high plains disease. Killing any grasses with herbicides several weeks before planting can reduce this problem and also limit damage from foliage feeding pests. Insecticide seed treatments should be considered when planting in high risk conditions and are the preferred control method for many insects in no-till systems.

**Tillage practices:**
Reduced tillage systems may increase populations of various predatory insects and mites in the soil. Crop residues encourage the growth of various small insects and mites, and other organisms which feed on decaying organic matter. As these populations increase, predatory insects and mites which feed on them build up, and will feed on various pests present in the soil. The lack of disturbance from cultivation encourages growth of these soil organisms. Studies have documented increased levels of predatory insects and predation on black cutworms in reduced tillage systems. (Robert J. Wright, 1995)

<table>
<thead>
<tr>
<th>Insect</th>
<th>Effect</th>
<th>Notes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Armyworm</td>
<td>0 to +++</td>
<td>Ryegrass and other grass cover crops and hay crops are especially attractive to egg-laying armyworm moths. In no-till systems where the grass cover is not plowed under, larvae move from the grass to feed on corn.</td>
</tr>
<tr>
<td>Black cutworm</td>
<td>+ to +++</td>
<td>Adult black cutworm moths prefer to lay eggs in weedy fields and in fields with unincorporated crop residues. Increased populations of predators and parasitoids also develop, but an increase in black cutworm injury often occurs anyway.</td>
</tr>
<tr>
<td>Corn earworm</td>
<td>0 to +</td>
<td>If planting date or crop development is delayed in no-till fields, corn is usually more attractive to egg-laying moths. This is usually a minor concern except for seedcorn producers.</td>
</tr>
<tr>
<td>Corn leaf aphid</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>Corn rootworms</td>
<td>0</td>
<td>Adults lay eggs in late summer; subsequent tillage has little effect on the survival of eggs during most winters. In harsh winters with subnormal temperatures and subnormal snowfall, egg survival is somewhat greater with reduced tillage.</td>
</tr>
<tr>
<td>European corn borer</td>
<td>0 to +</td>
<td>Conservation tillage favors greater survival of corn borers in crop residue, but effects in specific fields are minor because moths disperse from emergence sites to lay eggs in suitable fields throughout the local area. Where reduced tillage leads to delayed planting or slower germination (cooler soil temperatures), corn may be less susceptible to attack by first generation corn borers and more susceptible to second generation injury.</td>
</tr>
<tr>
<td>Seedcorn maggot</td>
<td>0 to –</td>
<td>Adult flies prefer to lay eggs where crop residue has been partially incorporated into soil. Notill corn stubble may be less attractive to egg-laying flies, but cooler, wetter soils shaded by crop residues slow germination and increase the period of vulnerability to seedcorn maggot injury.</td>
</tr>
<tr>
<td>Slugs</td>
<td>+++</td>
<td>Unincorporated crop residues and cooler, wetter conditions favor increases in slug populations and injury.</td>
</tr>
</tbody>
</table>
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<table>
<thead>
<tr>
<th>Insect</th>
<th>Effect</th>
<th>Notes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Stalk borer</td>
<td>0 to +</td>
<td>Overwintering survival is greatest in conservation tillage systems. In no-till fields, serious injury is most likely where grasses were present to attract egg-laying moths the previous August and September. If corn is no-tilled into soybean stubble where weeds were controlled during the previous year, stalk borers are not a problem.</td>
</tr>
<tr>
<td>Western bean cutworm</td>
<td>0 to +</td>
<td>Conservation tillage favors greater regional survival of western bean cutworms. Effects in specific fields are minor because moths disperse from emergence sites to lay eggs in suitable fields throughout the local area.</td>
</tr>
<tr>
<td>White grubs</td>
<td>+</td>
<td>Increases in grassy weed populations and reduced disturbance of soil favor survival of true white grubs</td>
</tr>
<tr>
<td>Wireworms</td>
<td>+</td>
<td>Increases in grassy weed populations, reduced soil disturbance, and delayed germination caused by cooler soil temperatures may favor wireworm buildup and injury.</td>
</tr>
</tbody>
</table>

*The range of effects notes the possibilities and worst case scenarios. Individual field experience may not confirm these extremes. Weather is directly tied to potential pest problems in to-till. +++=Substantial increase in pest population. + =Some increase. 0=No effect. -=Some decrease in pest population. Modified from Steffey et al. (1992)*

### Table 6. Possible effects of conservation tillage systems on insect pests in soybeans,*

<table>
<thead>
<tr>
<th>Insect</th>
<th>Effect</th>
<th>Notes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bean leaf beetle</td>
<td>0 to +b</td>
<td>Tillage has little effect on foliar feeding by bean leaf beetles, unless planting dates are earlier.</td>
</tr>
<tr>
<td>Grasshoppers</td>
<td>0 to +</td>
<td>Reducing tillage favors the survival of only those grasshopper species that lay eggs within fields. Those that lay eggs in weedy margins are not affected.</td>
</tr>
<tr>
<td>Seedcorn maggots</td>
<td>–</td>
<td>Seedcorn maggot populations are greatest in systems in which a live, green cover crop is incorporated into the soil.</td>
</tr>
<tr>
<td>Slugs</td>
<td>+++</td>
<td>Unincorporated crop residues and cooler, wetter conditions favor increases in slug populations and injury.</td>
</tr>
<tr>
<td>Spider mites</td>
<td>– to 0</td>
<td>Where crop residues help to retard soil moisture loss, plants may be less drought-stressed than in plowed fields; reducing drought stress slows spider mite outbreaks</td>
</tr>
</tbody>
</table>

*The range of effects notes the possibilities and worst case scenarios. Individual field experience may not confirm these extremes. Weather is directly tied to potential pest problems in to-till. +++=Substantial increase in pest population. + =Some increase. 0=No effect. -=Some decrease in pest population. Modified from Steffey et al. (1992)*

### Table 7. Possible effects of conservation tillage systems on insect pests in wheat,*

<table>
<thead>
<tr>
<th>Insect</th>
<th>Effect</th>
<th>Notes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aphids</td>
<td>– to 0</td>
<td>Prior crop residues may decrease the attractiveness of new stands of wheat to airborne aphids in the fall. (Seeding wheat after Hessian fly-free dates avoids most fall infestations of aphids.) By spring, it is unlikely that prior crop residues affect aphid invasion.</td>
</tr>
<tr>
<td>Army cutworm</td>
<td>– to 0</td>
<td>Army cutworm prefers barren or freshly worked soil for oviposition, so surface residues might deter egg laying activities. Oviposition occurs in the fall after planting, so tillage effects would be within-field rather than regional.</td>
</tr>
<tr>
<td>Green bug</td>
<td>– to 0</td>
<td>Fall and early spring infestations are deterred by the presence of surface residues and favored by the presence of volunteer small grains.</td>
</tr>
<tr>
<td>Hessian fly</td>
<td>0 to +++</td>
<td>Hessian fly populations carry over where wheat stubble is not tilled and volunteer wheat is not controlled. Hessian flies from undisturbed stubble move to new wheat that is planted before flyfree dates. Hessian flies that infest volunteer wheat in the late summer and early fall overwinter in the volunteer plants and can move to additional fields in the spring (regardless of those fields’ fall planting dates. No-till seeding of wheat into other crop residues poses no problem.</td>
</tr>
<tr>
<td>Pale western cutworm</td>
<td>– to 0</td>
<td>Similar to army cutworm in that it cannot lay eggs on crusted soil, so other tillage relationships may also be similar. Also oviposit in the fall so tillage effects would be within-field for winter wheat.</td>
</tr>
<tr>
<td>Russian</td>
<td>0 to +</td>
<td></td>
</tr>
</tbody>
</table>
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<table>
<thead>
<tr>
<th>Pest Type</th>
<th>Summary</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Wheat aphid</td>
<td>Favored by presence of volunteer small grains. Adjusting planting dates is a more important cultural practice than modification of tillage. Tillage effects do not seem similar to those observed with greenbug, although moisture conservation from stubble mulch systems may reduce Russian wheat aphid effects substantially.</td>
<td></td>
</tr>
<tr>
<td>Wheat curl mite</td>
<td>0 to + Similar to Russian wheat aphid in that volunteer wheat management and adjustment of planting date are key cultural practices.</td>
<td></td>
</tr>
<tr>
<td>Wheat stem sawfly</td>
<td>Shallow fall tillage may provide up to 90% sawfly control. If only spring tillage operations are performed, approximately 25% of the larvae may be destroyed, depending upon the tillage implement used. Tillage passes that are shallow should expose the larvae in the stubble.</td>
<td></td>
</tr>
</tbody>
</table>

"The range of effects notes the possibilities and worst case scenarios. Individual field experience may not confirm these extremes. Weather is directly tied to potential pest problems in to-till. "++"=Substantial increase in pest population. "+"=Some increase. "0"=No effect. "-"=Some decrease in pest population. Modified from : Steffey et al. (1992)

Impact of conservation tillage on pests:
Incorporation of crop residues by tillage destroys habitat for some pests; inverting soil exposes certain pest species to weather conditions, which can contribute to population suppression of these insect populations (All and Gallaher 1976, Pike and Glazer 1982). When conservation-tillage systems were first introduced, it was thought, a priori, that there would be an accompanying increase in pest severity, especially with soil-inhabiting insects (Gregory, 1974). Many of the early studies on pests associated with conservation-tillage systems in the United States dealt with corn (Zea mays), reflecting the major effort that went into encouraging conservation-tillage practices for this crop (Phillips, 1984). Some of earliest research in the United States linking conservation tillage and pest incidence was reported from Ohio. Crop-seedling emergence and increased populations of soil-inhabiting pests were associated with no-tillage practices (Musick, 1970). Musick & Collins, 1971 found that adult northern corn rootworm beetles, Diabrotica longicornis, a major root-feeding pest of corn, laid three to four times more eggs in no-tillage than in conventionally plowed fields. Yet egg survival and subsequent larval damage to corn roots were equal or significantly less in the no-tillage systems, indicating greater mortality sources under no-tillage conditions. Armyworm populations, Pseudaletia unipuncta, and their damage to crops increased when corn was planted directly into sod fields or with small grain cover crops such as rye and wheat. Adult moths of this pest oviposit on these grasses; herbicides kill the grasses, and then larval armyworms feed on corn (Musick, 1973 and Musick and Suttle, 1973). Increased black cutworm (Agrotis ipsilon) incidence was reported from no-tillage corn fields (Musick and Petty, 1973, Harrison, Bean and Qaiviyy, 1980. Slugs were documented as a serious problem in reduced-tillage fields, especially during excessively wet growing conditions (Musick and Petty, 1973). At Rothamsted, England, from 1964 to 1974, Edwards reported that the only pests to increase significantly when tillage was eliminated were wireworms and slugs. Wireworms were two to three times more abundant in direct drill fields during cropping with continuous cereals, or cereals following sod, than in plowed fields. During mild wet winters, slugs were a particular problem when oil seed rape was included in rotation sequences. (Edwards and Thompson, 1975. In Georgia, All & Gallaher (All and Gallaher, 1976) found that damage to seedling corn by the lesser cornstalk borer, Elasmopalpus lignosellus, was significantly greater in conventionally plowed than in no-tillage fields. They also reported that infestations of European corn borer (Ostrinia nubilalis) populations were over twice as high (32.8%) in conventional tillage as in no-tillage treatments (15.3%). However, in the same study, damage to corn by the com earworm, Heliothis zea, was greater under no-tillage conditions.

More Recent Studies on Pests:
The widespread adoption of conservation-tillage agriculture during the 1980s was accompanied by an increased interest in pest biology and management in these systems. The expanded use of conservation tillage for other row crops besides corn, forage crops and vegetables stimulated research on pest ecology in different cropping systems.

Soybeans:
Adoption of conservation tillage methods for growing soybeans and other crops has been motivated by many of the same reasons as for corn--decreasing the potential for soil erosion, reducing fuel and labor costs, and conserving moisture (King, 1983. Hammond 1987 concluded that conservation tillage farming practices have not resulted in an overall increase of invertebrate pests on soybeans. Rather, shifts have occurred in abundances...
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and the relative economic importance of various phytophagous species. Some of the first reports on soybean arthropods in conservation-tillage systems partitioned the arthropods into guilds. In Georgia, Hammond and Stinner, 1987 observed greater species diversity of foliage arthropods in no-tillage than in conventionally plowed soybeans; they attributed this finding to increased structural diversity in soil litter layers and to higher weed density and diversity in the no-tillage systems. In another study, analysis of no-tillage soybean systems in relation to weed density and community composition revealed that more arthropod taxa inhabit weedy soybeans than weed-free soybeans, and higher arthropod diversity is found in broadleaf than in grass weeds (Shelton and Edwards, 1983. In Kentucky, green clover worm, Plathypena scabra, laid greater numbers of eggs in double-cropped, no-tillage soybean following wheat than in conventionally tilled soybeans (Sioderbeck and Yeargen, 1983. However, subsequent larval populations were lower in no-tillage soybeans, suggesting higher pest mortality in this system. The same study found a trend toward lower populations of foliage feeding beetles in no-tillage treatments. Research in Louisiana indicated higher densities of green clover worm in no-tillage compared with tilled soybeans (Troxclair and Boethel, 1984. From an Ohio study comparing tillage practices in soybeans following winter rye crop, higher populations of green clover worm were observed in no-tillage than in either disk or moldboard plow treatments during one year of a two-year study (Smith, Hammond and Stinner, 1988. Considerable attention has been focused on seedcorn maggots, Delia platura, in relation to conservation-tillage soybeans, particularly on how the ecology of this species is affected by cover-cropping treatments. The dipteran larvae feed on germinating seeds and frequently kill or deform seedlings. Decaying vegetation and high levels of organic matter may attract egg-laying flies and provide a rich habitat for developing larvae (Gregory and Musick, 1976. Funderbuck et al. 1983. found higher populations of maggots in chisel-plowed (surface-tillage) treatments that had only partially buried plant residues than in either no-tillage or moldboard-plowed soils. Subsequent studies supported these findings when soybeans were planted after a winter cover crop of rye or a previous crop of alfalfa was only partially incorporated into the soil with shallow tillage. This positive influence on maggot populations was particularly evident when the previous crop residue was green rather than dried or senescent when incorporated (Hammond and Jeffers, 1983, Hammond, 1984. Other soybean pest species given attention in relation to conservation tillage include Heliothis (Roach, 1981 and Stinner, Reguere and Wilson, 1982.), Mexican bean beetle, Epilachna varivestris, bean leaf beetle, Cerotoma trifurcata (Troxclair and Boethel, 1984.), and slugs (Hammond, 1985. These studies have not indicated economically significant increases of pest damage with conservation tillage practices, with the exception of slugs in eastern portions of the US grain belt. Forage crops: To decrease soil erosion potential, attention has been directed towards establishing pasture and forage crops using conservation tillage methods (Managan et al. 1982. Damage to seedlings by invertebrates was predicted to be a major obstacle to the adoption of reduced- and no-tillage planting of forage crops. In fact, damage by slugs (Grant et al. 1982, Byers and Bierlein, 1984 and Byers and Templeton, 1988), flea hoppers (Managan and Byers, 1987), and crickets (Rogers, 1985) has been associated with poor alfalfa, Medicago sativa, establishment under conservation-tillage conditions. Barney & Pass (Barney and Pass, 1987) compared foliage arthropod communities on no-tillage and conventionally planted alfalfa in Kentucky where they found that pest populations of alfalfa weevil, Hypera postica, clover root curculio, Sitona hispidula, and aphids did not increase under no-tillage treatment. Populations of potato leafhopper, Empoasca fabae, probably the most important pest of alfalfa in this region, were reduced in no-tillage treatments. The authors attributed this last finding to higher grass density among alfalfa in the no-tillage treatments. Other researchers have shown that grasses deter populations of potato leafhoppers (Lamp, 1984. Contributions to a 1986 International Symposium on Establishment of Forage Crops by Conservation Tillage (Hill, 1986) indicated that pest problems with conservation-tillage forage crops are frequent and more severe than damage encountered in row crops under conservation tillage. Still, forage crops harbor substantial populations of natural enemies (Barney and Pass, 1987) whose value should not be discounted and that merit further study. Corn: During the past decade, attention has been given to lepidopterous herbivores on corn in relation to conservation-tillage systems. The stalk borer, Papaipema nebris, serves as an example of a pest that has become more prevalent with the adoption of no-tillage corn, as a result of changes in weed distributions. This moth has a
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broad host-plant range and historically has been associated with damage to corn in limited areas along field edges where grasses are typically abundant (Decker, 1931. Adults oviposit in grass during late summer and early fall (Levine, 1985); the larvae emerge the following spring, and early instars inhabit grasses. Because grasses tend to be more abundant under no-tillage conditions and distributed throughout corn fields, stalk borer population dynamics have been altered concomitantly. The larvae will typically move from grassy weeds to corn when herbicides are applied, and therefore areas within com fields can be damaged (Stinner, 1984. Studies from Ohio (Levine et al. 1984), Iowa (Bailey and Pedigo, 1986, Lasack and Pedigo,1986), and Virginia (Highland and Roberts, 1987.) have examined consumption rates and movement patterns of the stalk borer in conjunction with no-tillage practices.

In a comparative assessment of soil and foliage arthropod communities after 20 years of conventional- (moldboard), reduced- (disked), and no-tillage corn cropping in Ohio, black cutworm damage was least in the conventional tillage treatment. Damage to corn by both European corn borer and com rootworm was not significantly affected by tillage treatments (Stinner, 1988.

Cotton:
Plowing and cultivation have been traditional methods used to reduce populations of the boll weevil, Anthonomus grandis, and Heliothis spp. (Gaylor and Foster,1987.. Tillage reduces overwintering populations of weevils within cotton fields and restricts adults from building fat reserves prior to diapause (Gaylor and Foster,1987. Tillage also is injurious to Heliothis, especially in the pupal stage. Significant injury to these pests can occur in plowed soil from heavy rains (Hopkins et al.1972) despite damaging influences of tillage on these pests, Gaylor & Foster and Gaylor et al 1984 reported that damage by these two species and Lygus spp. did not increase with the use of conservation-tillage systems in Alabama. Furthermore, these researchers have indicated that some of the cultural practices associated with conservation-tillage cotton production, such as delayed planting with double-cropping systems, actually reduce boll weevil damage. In northern Alabama, variegated cutworm populations (Peridroma saucia) increased in conservation-tillage cotton. Damage severity to the cotton was dependent upon cover-cropping practices (Gaylor et al. 1984.

Influence of conservation tillage on natural enemies:
Soil-Inhabiting Macroarthropods
One of most frequent and widespread observations regarding arthropods in conservation tillage is the increase in soil- and litter-inhabiting predatory arthropods, especially ground beetles (Carabidae) and spiders, as tillage is decreased (All, 1978, Raney, 1974 and Stassart and Gregoire-Wibo, 1983. A British study showed that carabid beetles in particular are important predators of cereal pests (Edwards, 1979. Greater carabid beetle abundance was reported from conservation-tillage than from conventionally plowed soybeans in Georgia (House and All, 1981. On some sampling dates, carabid density in the conservation-tillage systems was as much as four times higher than in the conventional treatments. Also in Georgia, greater diversity of soil surface macro arthropods was reported in no-tillage than in conventionally plowed systems (Blumberg and Crossley,1982. In another study comparing no-tillage and conventional-tillage soybeans, a mean density of 17.6 carabid beetles per m2 was documented in no-tillage vs. 0.38 per m2 in plowed treatments (House and Parmalee,1985. Tillage effects on spider populations were less marked, but spider density was higher in the no-tillage treatment, with the exception of once sampling date. In northeastern Italy, Paoletti in 1987 recovered greater numbers of predatory beetles and spiders from pitfall traps in no-tillage and reduced-tillage than in conventionally plowed com systems. A similar trend was reported for com systems in Ohio after 20 years of continuous treatment (Stinner et al. 1988.

To evaluate how these soil macro arthropods affect pest populations from different tillage systems, researchers in Ohio manipulated both macro arthropod predator and black cutworm prey populations in conventional and no tillage com systems (Brust et al. 1985 and Brust et al. 1986. They found four times more com plants destroyed when predators were removed than when they were present in no-tillage treatments. In these studies, tillage significantly reduced both predator density and predation rates on cutworm larvae. The predatory taxa included carabid and staphylinid beetles, phalangids, lycosid spiders, and ants. Across tillage treatments, cutworm damage to com was negatively correlated (r = .90) with absolute density of predators. In cotton systems, removal of soil-dwelling predators from both conventional- and conservation tillage systems significantly increased emergence of adult Heliothis moths (Gaylor et al. 1984. Attacks by ants on prepupa of Heliothis zea in no-tillage com were significantly more frequent than in plowed soils (Landis et al. 1987.

These soil- and litter-dwelling macro rthropods are, for the most part, generalist feeders. For example, carabid
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beetles are known to feed as carnivores, herbivores (including weed seeds), and fungivores (Best and Beegle, 1977, Brust and House, 1988, Rabatin and Stinner, 1988. Although the predatory macrofauna can be very important in reducing specific pest populations, much of these animals' food resources on a year-round basis are provided by non pest sources, including organisms in detrital food chains such as collembola (Schaller, 1968., Ruhendi and Litsinger 1982. Since conservation-tillage systems support greater densities of detrital feeding fauna and microflora involved in decomposition processes (Hendrix, 1986 House and Parmalee, 1985), the linkages between predatory and detrital food webs deserve further attention.

Foliage-Inhabiting Predators and Parasitoids in Conservation Tillage:

In Georgia, a greater abundance of predatory foliage-inhabiting insects, mostly Coleoptera and Hemiptera, was found in no-tillage systems than in conventional tillage, which was attributed in part to greater weed density in the former (House and Stinner,1983. Troxclair & Boethel in 1984 reported that the density of foliage-inhabiting, predacious hemiptera was higher on Louisiana no-tillage soybeans in some sites and lower in other locations, when compared to conventionally tilled systems. Troxclair & Boethel proposed that weed density and species composition were not significant factors affecting foliage inhabiting insects in their study. However, Hammond in 1987, citing the work of Altieri et al and Shelton & Edwards, argued that weeds are important determinants of predator guild distribution and of abundance in conservation-tillage soybean systems. Ferguson et al. 1984, finding numbers of predators more abundant in no-tillage vs. conventionally plowed soybeans, attributed this difference to planting date, row spacing, and previous crop stubble interacting with the tillage systems. Summarizing results of zero tillage in the tropics, van Rijn in 1981 also indicated that crop stubble, as well as weeds, contributes to maintenance of predator populations. House (House, 1989) reported higher densities of soil arthropods, especially predators, in the root systems of weeds under no-tillage than under conventional-tillage management. House thought this indicated that certain weed species provide refugia for predators in no-tillage systems. Foster & Ruesink in 1984 showed that the black cutworm parasitoid Meteorus rubens lived longer, attacked more hosts, and reproduced more successfully when provided with flowering weed species-wild parsnip, wild mustard, chickweed, shepherds purse, and smartweed-typically encountered in minimum-tillage corn fields.

3. Maintenance of permanent or semi-permanent soil cover

Impact of crop residues on pests:

Incorporation of crop residues in conservation agriculture has direct and indirect effects on pests. For example, crop residues directly affect egg laying of beetles and cutworms. Lower soil temperature and higher soil moisture content under crop residues would also affect pest infestation. Indirectly, residues change the type and density of weeds, which in turn influence insects and natural enemies. Crop residues generally increase diversity of useful arthropods and help in reducing pest pressure. The surface residues may ensure survival of a number of insects, both harmful and beneficial. Reduced tillage systems particularly under staggered planting system of crops in monoculture, may contain higher levels of pest inoculums than the conventional system. Further, the decomposition of crop residues along with several inter-related factors like climate, crop geometry, irrigation and fertilization, cultural practices and pesticides may affect the survival of insects in crop residues. The decomposition of residues brings out a chemical change in soil which may affect the host reaction to pests. The decomposition of plant residues may produce phytotoxic substances, particularly during early stages of decomposition. The effects could be severe in reduced tillage systems which incorporate huge amount of crop residues into the soil and an extra application of N is made to hasten decomposition of these residues. A change in weed ecology is expected to influence the survival of several of those insects which tend to develop on weeds, particularly during the fallow period. Since the zero/reduced tillage system reduces the fallow period among crops, it may result in altered incidence of certain insects.

Population of termite and white grubs generally increases under the reduced tillage. However, the effect of crop residues on termite damage is contentious. Under sufficient crop residues, white grubs do not damage the crop even at a very high density. However, at some of the sites, organic mulching has been reported to increase damage of cutworms due to moisture conservation. Also crop residues on the soil surface that conserves the moisture, may favour snails and slugs, causing damage to crops. Increased pest and weed problems during the ‘transition period’ are major hurdles in adoption of conservation agriculture by farmers. Non-judicious application of pesticides under such situations may disrupt the ecosystem and cause pest outbreaks. Therefore, integrated pest management (IPM) should be adopted as a necessary component of a conservation agriculture system.
Impact of conservation agriculture practices on the arthropod population in pigeon pea ecosystem

In majority of the studies, reduction of pests and diseases were well documented through stimulation of biological diversity, mainly by the multiplication of natural enemy activity. The aim of CA is to conserve predators and parasitoids of insect pests and thus to allow the natural control. In general the pest reduction is possible due to interference of pest colonization by limiting dispersal/disrupting the feeding behavior/ inhibiting reproduction, increased multiplication of predators and parasitoids population and break up of food chain caused by crop rotations and crop residue (Raju G. Teggelli).

Effect of zero tillage practices on arthropod dynamics

Stinner et al. (1987) studied the influence of 20 years long-term tillage moldboard ploughed, reduced and no-tillage practices on soil-inhabiting and canopy arthropod communities in corn systems including effects of a soil-applied insecticide (terbufos. There was an increase in total numbers of micro arthropods (mites and collembola) and macro arthropods (Ground beetle, spiders) in no tillage compared to conventional practice. While the cutworm damage was less in conventional tillage. Similarly, Nguyen and Vu (1988) and Bernhard (1989) studied on carabid beetle fauna at three fields of winter wheat (2 biologically farmed, 1 conventionally), two fields of winter wheat (1 biological, 1 conventional) and two fields of sugar beet (1 conventional, 1 conventional without herbicides) and reported that in both the years, abundance were considerably higher in biological winter wheat than in conventional wheat. Abundances in sugar beet was at a similar level as in conventional winter wheat in 1983, both being rather low. Carabid communities of biological winter wheat were characterized by Brachinus explodens (Duftschmidt) and certain species of Amara and Harpalus, mostly facultative phytophagous, which reached higher population densities in biological fields. A review of 45 investigations showed that 28 per cent of the pest species increased with decreasing tillage, 29 per cent showed no significant influence of tillage and 43 per cent decreased with decreasing tillage (Stinner and House, 1990; Cancela da Fonseca, 1993. Marasasa et al. (2001) studied the abundance of different trophic groups under conventional tillage (CT) and no tillage (NT) practices and were compared with a natural field boundary (FB) in a wheat crop and revealed that predators were the most abundant group of all arthropods captured and their number was higher under no tillage than under conventional tillage. In conventional tillage, an increase in predators was observed only in spring, probably associated with a recolonization from the adjacent plots of no tillage. Similarly studies conducted by Bandyopadhyaya (2002) reported that application of organic manures or its combination with mineral fertilizer had higher density of collembola than manured or unmanured soil. The treatments with different doses of organic manures increased the population of collembola compared to treatments with fertilizer alone and the population was higher during September and the density reduced during dry period. Divina et al. (2009) documented on tillage systems not only affected the abundance of arthropod fauna but also proportion between different functional group of arthropods which was carried out using visual observation, destructive sampling, pitfall trap and sweeping methods in two successive years of 2000 and 2001, in a field planted with native ornamental plants, which were also commercially used for landscaping. The field was divided into two areas (sprayed and un-sprayed. Overall results showed that more arthropod taxa were present in the un-sprayed area than in the sprayed area.

Influence of conservation agriculture practices on pests and natural enemies emergence

Gupta and Mukharji (1976) identified 35 genera and 37 species of Acari, 22 genera of Collembola and 16 genera of other soil arthropods. Among the Acari, Meso stigmata was represented by 13 genera and 13 species in cultivated and uncultivated soils. Where in the Cryptostigmata consisted of 13 genera and 15 species. The most abundant and wide spread genera being Epilohamannia and Scheloribates, which inhabited mainly the upper layer of soil. Eight genera and eight species were recorded from Prostigmata, in which Coccotydeus and Tydeus were most common. The Astigmata was represented by genus Tyrophagus. The next group of arthropod recorded were collembolans to the extent of 22 genera where in the, Folsomia, Folsomides, Isotomides and Onychiurus were predominant. Other soil arthropod genera included were Diplura, Pauropoda, Symphyla, Palpigradi, Uropygi, Schizomida and Pseudoscorpionida.

Bhattacharyya and Raychaudhuri (1979) noticed the population fluctuation of Cryptostigmata and Collembola in a wasteland showed two peaks, a pronounced peak during post-monsoon period (September-October) and a less pronounced one during pre monsoon period (May-June). The conservation practices (enriched with organic manures) helped to establish significantly higher soil
invertebrate abundance (7.60) compared to recommended (no manures) method (4.87. Collembola were found to be relatively more abundant (44.9%) in conservation method whereas acari, other minor insects and non-insects fauna were predominant in recommended practices. (Srinivasreddy et al., 1999. Likewise Bandyopadhyaya (2002) reported that application of organic manures or its combination with mineral fertilizer induces higher density of collembola than manured or unmanured soil. The treatments with different doses of organic manures increased the population of collembola compared to treatments with fertilizer alone. The population was higher during September and the density reduced during dry period.

According to Raju G. Teggelli, Raised bed with mulch was found to be an effective conservation agriculture tillage practice with reduced larval population of all the pod borers and also recorded the higher yields. The micro and macro fauna population was higher with respect to the diversity indices values which was found to be good in the raised bed with mulch followed by zero till with mulch. Adult pests emerged from the cages was also low in raised bed with mulch with higher tachnids compared to conventional practice. Pigeonpea intercropped with setaria, maize, sorghum and sunflower were found to be effective in reducing the pod damage and also recorded higher population of natural enemies.

**Conservation Agriculture and its effects on macrofauna diversity especially in reference to White grubs:** (Rabary et al. 2011)

White grubs: Adoption of conservation tillage has reduced soil erosion in many parts of the world. Conservation management practices improve soil quality through the significant amounts of organic residues left on the soil surface, and cover crops also provide many benefits. These supply residues derived from dead plant parts and organic substances released from their living roots, and may foster soil organisms’ abundance and diversity. They may also enhance ecosystem functions such as nutrient cycling (Rabary et al., 2008), soil structure and C sequestration, and pest and disease control (Ratnadass et al., 2006. In some regions of Madagascar, white grubs were found to be more abundant under direct seeding mulch based cropping systems (Ratnadass et al., 2006. White grubs are C-shaped larvae of a large group of beetles (Coleoptera: Scarabaeidae. Several species in this group cause significant damage to cereal crops by feeding on plant roots. The entomopathogenic bacterium *Metarhizium anisopliae* has been proven effective in controlling white grubs in some cases (Razafindrakoto et al., 2010). However, Randriamanantsoa et al. (2010) showed that there are many species of white grubs in Madagascar and most of them are endemic. Furthermore, some species are not pests but show "soil engineering" behaviour. This wide array of white grub species makes their biological control difficult. The fundamental dilemma in pest control with insecticide application is about significant detrimental effects on non-target species in the food web, which may deplete soil diversity and increase the possibility for subsequent pest outbreaks. Researchers report the impact a number of cover crops known for their pest-suppressing on white grub control within upland rice cropping systems. Mutsamba et al., presumed that these plants toxicity would alter the composition of soil macrofauna, but not negatively affect biodiversity overall, due to the positive effects of cover crops compared to those of the conventional tillage system.

**Assessment of Conservation Agriculture Practices by Smallholder Farmers in the Eastern Cape Province of South Africa:** (Muzangwa et al. 2017)

Best management practices by CA farmers and comparison of selected soil quality indicators between CA and CT fields: According to Lindah Muzangwa, 2017, the majority of the fields lacked soil cover (Figure 2. Weed control was a challenge for the farmers, particularly in fields lacking organic mulch. However, disease and insect pest control was above average and most farmers agreed that these were not much of a problem in their fields.
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Figure 2. Assessment of CA fields based on the soil cover and pest control (weeds, insects and diseases.

Current and future directions

The objectives of IPM and CA are the same: sustain productivity, conserve natural resources, reduce production costs, improve environmental health, maintain biodiversity, and reduce agrochemical use for crop production/protection. The past decades have brought incredible developments in the field of agriculture. The use of pesticides still dominates the management of insect pests and is a health hazard for humans and the environment (Nawaz et al. 2013). Agriculture needs the cooperation of international development agencies that often have solutions for agricultural problems. Integration of interdisciplinary projects with new satellite technology available for these agencies can bring positive change to agricultural production. In future, the focus will be on those farming systems which provide high-quality food with low risks to the environment and public health. CA is the best choice in this regard Ahmad Nawaz and Jam Nazeer Ahmad said. Conservation tillage in some form undoubtedly will be a part of this future. Research and farming will be challenged to expand the roles that invertebrates play, not only as pests and natural enemies, but as agents in the regulation of ecosystem processes of decomposition and nutrient cycling. We predict that in lower-input agricultural systems, the emphasis will shift toward this larger role that invertebrates can play in conservation tillage agriculture (Stinner and House, 1990.

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FRUIT FLIES AND THEIR MANAGEMENT

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Abstract

Fruit fly is most serious pest of fruit crops in India and its incidence. It is a major obstacle in production of fruits and vegetables, which causes yield losses mostly during harvest and post-harvest time so that quality as well as quantity of fruits gets deteriorate and ultimately affect market value of produce. Therefore, appropriate intervention is required to avoid the losses by implanting preventive measures. Among the various management strategies for fruit fly, trapping using lures found practical for detection and monitoring as well as proved most effective, eco-friendly and economical tactics.

India has variety of soil and climate where, all kinds of field, horticultural and forest crops can be grown. Among horticultural crops, fruits are of most importance because of their economic value and returns. Fruits are also important due to their nutritional value and are relatively an expensive source of protein, mineral, sugar and vitamins. Besides, fruits are utilized in preparation of pickles, jam, jelly, curries, candies, etc. Their use is recommended for curing blood diseases, rheumatism, diabetes, asthma, etc. In India, the total area under fruit cultivation is 6.4 million hectares with an approximate yield of 91.44 million tonnes, which accounts for 12.8 per cent of the world total production. India is the second largest producer of mango, banana, sapota and acid lime. About 39.5 per cent of the world's mango and 23 per cent of world's banana are produced in India (Anonymous, 2016). The major states producing fruits are Uttar Pradesh, Andhra Pradesh, Bihar, Gujarat, Maharashtra, Karnataka, Punjab and West Bengal.

Cucurbit crops are infested by several insect pests which are considered to be the significant obstacles for economic production. Among them, cucurbit fruit fly is the serious pest responsible for considerable damage of cucurbits (Butani and Jotwani 1984). The cucurbit fruit fly, Bactrocera cucurbitae can attack about 16 different types of cucurbit crops. Although the rate of attack varies among the crop, infestation reduced both the yield and quality of the cucurbit fruits. Yield losses due to fruit fly infestation vary from 19.19 to 69.96 percent in different fruits and vegetables (Kabir et al. 1991). Depending on the environmental conditions and susceptibility of the crop species, the extent of losses varies between 30 to 100% (Gupta and Verma, 1992; Dhillon et al., 2005a, b, c; Shooker et al., 2006).

Fruit crops suffer from various insect pests, among them Fruit fly are considered as most important insect pest due to its host and efficiency of damage. Fruit flies are belongs to the family, “Tephritidae” (Diptera) are one of the most fascinating and diversified. They are commonly called as “fruit flies” or “orchard flies” due to their close association with fruits. These flies are also referred to as “Peacock flies” due to their habit of strutting and vibrating their wings. There are over 4000 species of fruit flies in the world (Norrbom et al., 1998) of which about 5 per cent occur in India (Ramani, 1998. This family is represented in the entire world region except Antarctica. The oriental region comprises nearly 1000 species so far recorded (Kapoor, 1993. Of the three subfamilies under Tephritidae Dacinae, Tephritinae and Trypetinae, the subfamily Dacinae is of economic importance. In this subfamily, the genera Dacus and Bactrocera are important as they include economically important species such as Bactrocera dorsalis (Hendel) and Bactrocera zonata (Saunders. The subgenus Zeugodacus includes economically important species like B. cucurbitae. Nearly 35 per cent of the known fruit fly species attack soft fruits like mango, guava, sapota, citrus, ber, peach, etc. and several cucurbitaceous vegetables (White and Harris, 1992).
Species composition of fruit flies on different hosts

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<th>Host</th>
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<td>B. affinis</td>
<td>Madhura and Viraktamath (2001)</td>
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<td>B. caudate</td>
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<td>B. correcta</td>
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<td>B. diversa and B. cucurbitae</td>
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<td>Cucurbits</td>
<td>B. cucurbitae</td>
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Methyl eugenol (4-allyl-1, 2-dimethoxybenzene) is a kairomone (Metcalf and Metcalf, 1992) that is attractive to many species of the Subfamily Dacinae (Tephritidae) in the South Pacific Region (Hardy, 1979). Methyl eugenol was used for male annihilation or killing of sexually immature males before they were able to mate with females (Steiner, 1952) to eradicate established populations of Oriental fruit fly (Bactrocera dorsalis (Hendel)) in the Mariana Islands (Steiner et al., 1970) and Okinawa (Koyama et al., 1984). Cue-lure is a derivative of anisylacetone (4-[p-methoxyphenyl]-2-butanolone), an unstable synthetic compound that is sensitive to hydrolysis (Metcalf and Metcalf, 1992). It is the most effective attractant available for survey and detection of this insect. Cue-lure also attracts other tephritids (Drew, 1974) and Drew and Hooper (1981) reported over 80 species of Dacini responded to it. Cue-lure is not known to occur naturally, but its hydrolyzed form, raspberry ketone (4-[p-hydroxyphenyl]-2-butanolone) (Boroza et al. 1960; Metcalf and Metcalf, 1992), is known to occur in at least one plant species (Nishida et al., 1993). Pawar et al. (1991) reported that sex attractant cue-lure was more effective than the food attractant tephrtile in traps for monitoring D. cucurbitae on bitter gourd, whereas Hardy (1991) reported that at least 90 per cent of the Dacinae species were strongly attracted to either methyl eugenol or to cue-lure raspberry ketone. Methyl eugenol, when used together with an insecticide impregnated into a suitable substrate, forms the basis of male annihilation technique (MAT). Methyl eugenol especially attracts the males of B. dorsalis, B. correcta and B.zonata (Verghese et al., 2006). Male annihilation is the primary control technique for the Oriental fruit fly as it uses a mixture of methyl eugenol and an
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insecticide to attract and kill males resulting in a decreased mating and subsequent decline in the population over time (USDA-APHIS, 1985; Stark and Vargas, 1992).

Life Cycle

Egg
The female fly found to visit mostly a physiological matured fruit. The fly wonders on the surface of fruit and quickly flies off. After two to three such visits, finally it selects the fruit for oviposition. The fly walks over the fruit and surveys it by up and down movement of ovipositor. After selecting suitable place for oviposition, it bends its abdomen at right angle to the body and then gradually insert its ovipositor in epidermis by forward and backward movement of abdomen. In the laboratory as well as under field conditions it was observed that female fly laid eggs in clusters of 3 to 12 eggs underneath the rind of the fruit at a depth of about 2 to 4 mm (Bansode, 2009. Similar observations were also made by Narayanan and Batra (1960) as well as by Shah et al. (1948. In this way there are about 2 to 6 punctures on a single fruit where in eggs are laid, which can be terminated a “True puncture”. While 7 to 12 punctures do not contain eggs, which termed as “Pseudo-puncture”. In field, the fruit flies were attracted more to the fallen and rotten fruits rather than fruits attached to the tree; besides, cracked and damaged fruits on the tree were more preferred by flies for laying eggs. A typical smell coming out from damaged and rotten fruits probably attracts the flies. It was inferred from such observation that the flies attracted to rotten fruit more in search of food followed by oviposition. Moreover, B. dorsalis preferred to lay eggs in proximal (stalk side) and middle part of fruit rather than distal portion. At the site of oviposition puncture, sticky fluid oozes out that might allow the entry of microorganisms. Later on, a discoloured spot is developed around the true puncture. According to Doharey (1983) eggs period of B. dorsalis on different hosts varied from 1 to 4 days. Lower incubation period (3.00 days) of D. dorsalis on mango and sapota as compared to guava (3.20 days).

Maggot
The freshly emerged maggot are translucent and white with slightly yellowish colour except its mouth parts, which were dark black in colour, Bansode (2009. The head was pointed and slightly bent downward with a pair of mandibular hooks. The maggot was apodous with three thoracic and nine abdominal segments. The cuticle of maggot was so translucent that the internal organs were visible through it. Butani (1979) has also described similar morphological characters. The full grown maggot was longer and broader with cephalic end and blunt at posterior end. The colour of full grown maggot was creamy white to yellowish and more opaque than newly emerged or young maggot. Similar results were found by Narayanan and Batra (1960. The larval period ranged from 6 to 12 days on different hosts. It was recorded as 9.40 ± 1.47 days on sapota, 8.52 ± 0.71 days on banana, 7.84 ± 1.03 days on guava and 7.44 ± 1.66 days on mango. Thus, the study indicated that larva could complete its larval period earlier when fed on mango compared to guava, banana and sapota. The faster maggot development on mango (6.80 days) compared to guava (9.00 days) was also reported by Doharey (1983. Similarly, the findings are more or less in conformity with Kalia (1992) who reported larval period was 6.00 to 7.75 days on mango and 7.50 to 9.00 days on guava.

Pupa
The pupation of B. dorsalis took place at a depth of 0.5 to 5.0 cm in soil under laboratory and field condition. The freshly formed pupae were yellowish white to reddish yellow in colour which later on changed to light golden yellow to honey brown colour. Moreover, pupae obtained from mango and sapota is dark honey brown colour and those developed from guava and banana is light brown or golden brown in colour. The pupae were barrel shaped and having eleven distinct segments with last abdominal segment being little more prominent (Bansode, 2009. The pupal period was varied between 6 to 9 days on different host. The average pupal period recorded was 7.80 ± 0.82 days on sapota, 7.28 ± 0.82 days on banana, 7.20 ± 0.76 days on guava and 7.00 ± 0.71 days on mango. Pupal period of 10 to 12, 8 to 10, 8 to 10 and 8 to 11 days, respectively on mango, guava and Elakli and Robusta variety of banana was reported by Jayanthi and Verghese (2002).
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Adult
The adults are brown to black brown in colour with hyaline wing, yellow legs and the thorax is ferruginous, brownish black in colour. The adult flies possess two prominent compound eyes on the dorso-lateral region of the head and bear aristate type of antennae. The abdominal tergites were free. In thoracic region, pair of yellow coloured lateral vittae is present. A black colour “T” shaped transverse band was observed on dorsum of abdomen. The wings of adult consist of continuous black marking on costal margin, which is a typical character of identifying the *B. dorsalis*. The hind wings were modified into a short tubular structure with rounded end. In male, the abdominal end was bunt while, it was developed into pointed ovipositor in case of female. Moreover, the male flies are slightly smaller than female flies (Bansode, 2009). The average longevity recorded as 11.24 ± 1.92 days on sapota followed by 11.12 ± 1.62 days on banana, 10.32 ± 1.52 days on guava and 9.72 ± 1.17 days on mango. The average longevity of female recorded as 18.08 ± 2.16 days on sapota followed by 17.36 ± 1.75 days on banana, 17.28 ± 2.03 days on guava and 17.16 ± 2.22 days on mango.

Total Life Span
Bansode (2009) observed that the total life cycle starting from egg to death of adult of male varied from 24.50 to 36.50 days. This period recorded as 32.06 ± 2.69 days on sapota, followed by 30.76 ± 1.86 on banana, 29.14 ± 2.21 days on guava and 27.46 ± 1.81 days on mango. While, that of female varied from 30.00 to 46.00 days when, reared on different host. He was also recorded as 39.90 ± 3.32 days on sapota, followed by 37.01 ± 1.84 on banana, 36.10 ± 2.58 days on guava and 34.90 ± 2.07 days on mango. Total life span of male 32.06 ± 2.69 days on sapota, followed by 30.76 ± 1.86 on banana, 29.14 ± 2.21 days on guava and 27.46 ± 1.81 days on mango. While, that of female varied from 30.00 to 46.00 days.

Nature of Damage
The maggots live in liquefied pulp and hang head downward with their posterior spiracles at the liquid surface. The maggots after full feeding move from the centre of the fruit where they had been feeding on the soft and fermented skin of fruit. On completion of full development, the maggots bore holes; exit out through hole and by their jumping movement fall to the ground for pupation. It was further noted that the exit holes made for pupation by full grown maggot on damaged fruit was very clearly visible in case of mango, which was not so in case of sapota, guava and banana. After leaving the fruit, matured maggot wandered on soil in haphazard way and traveled in search of suitable site for pupation. The infested fruits become completely unfit for consumption (Bansode, 2009).

Management of Fruit Fly
Cultural method
According to Verghese et al. (2012) langra and EC-95862 were not infested by fruit fly as the phenol content was higher in peel and pulp. Whereas, genotypes PKM-1, PKM-2, DSH-1, DSH-2, Pilipatti, Singapore and Bhuripatti were least susceptible to *B. dorsalis* in sapota, however Kalipatti, Cricket Ball, Paria collection, Mohangootee and Murabba were found highly susceptible, reported by Nandre and Shukla (2013). Singh (2008) observed that smooth skinned varieties of guava like Red flash, Allahabad safeda and Local were highly susceptible to *B. dorsalis* damage, whereas rough skinned pear shaped variety was least susceptible.

Physical method
In past, Bansode (2009) observed that the hot water treatment at 52 °C for 60 minutes conferred highest result for post-harvest control of fruit fly in mango. While, Post-harvest hot water treatment of mango at 48 °C for 60 as well as 75 min was found to be 100 per cent effective against fruit fly infestation (Vergheese et al., 2006).

Mechanical method
Bansode (2009) noted that highest number of fruit flies were tapped in D.F.I.D water bottle trap (1396 flies) as compared to Patel (Modified) trap (1035 flies) and conventional trap (935 flies. According to Anonymous (2012) recorded highest number of fruit fly were trapped in Modified trap at Paria farm (5285 flies/trap) and Fanaswada location (3161 flies/trap) in mango as compared to conventional trap at both locations i.e. 3012 and 2763 flies/trap, respectively. The lowest number of fruit flies damage (5.00%) was found in Khokha trap as compared to other traps in mango (Anonymous, 2012. Whereas, highest number of adult flies of *B. cucurbitae* were attracted in molasses bait (3.22), followed by fermented date palm juice (3.18), whereas lowest were
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recorded in sugar (0.67), Laskar and Chatterjee (2010). Ali et al. (2010) studied efficacy of different control methods against oriental fruit fly, B. zonata and observed that maximum control of fruit fly (60%) was attained by using male annihilation technique (MAT), whereas minimum in cultural control (10-19%). Male annihilation using methyl eugenol traps has also been found to be very effective in controlling fruit flies @ 4 traps/ acre in mango and guava in different parts of India, in a three-year study under the Integrated Management of Fruit Flies in India (IMFFI) programme (Stonehouse et al., 2005).

Integrated Pest Management

Gupta and Verma (1982) found lowest rate of B. cucurbitae infestation in plots sprayed with fenitrothion (0.025%), protein hydrolysate (0.25%) or molasses (0.5%), which was significantly more effective than the recommended bait spray of malathion (0.25%) and gur (0.5%). Wong et al. (1991) compared both laboratory-reared and wild adults of the melon fly, Dacus cucurbitae Coquillett, for their response to cue-lure at various ages. Virgin laboratory (4, 6, 8, 10, 12, 14 days old) and wild (10, 12, 14, 16, 18, 20, 22 days old) flies were released into outdoor field cages and trapped from 0800 until 1600 hr. Response of males to cue-lure increased with age and corresponded with sexual maturity for each strain, whereas females of both strains were relatively non responsive to cue-lure. They concluded that failure to eradicate male annihilation programs against D. cucurbitae in past may be explained in part by the fact that only older males, which may have already mated with gravid females, respond to cue-lure. The mean persistence period of melolure was highest in acacia wood block (9.0 weeks), followed by plywood block (8.0 weeks). The straw board block absorbed the maximum quantity of melolure mixture (26.07 ml), while the lowest quantity (6.17 ml) was absorbed by sponge block. The plywood block recorded a maximum catch efficiency of 0.65 flies per ml of melolure mixture absorbed, whereas the lowest catch efficiency (0.09 flies/ml) was observed in soft board block. Zaman (1995) reported that the cue-lure baited traps attracted the melon fruit fly, B. cucurbitae males from mid-July to mid-November (peaked in August) and from 2nd week of August to the 2nd week of November (peaked in September) for the two years. However, Jaiswal et al. (1997) opined that 90 per cent farmers used attractant traps of cue-lure along with field sanitation for B. cucurbitae management to be very effective. Patel et al. (2005) while studying the efficacy of different dispenser blocks with methyl eugenol and malathion mixtures against B. dorsalis in mango orchards revealed that acacia, plywood, mango wood, neem wood, straw board, teak wood, salwood and rose wood blocks recorded a half-life of 95, 101, 108, 99, 90, 91, 108 and 91 days, respectively, with a mean daily catch of 7.36, 7.69, 7.00, 7.35, 8.01, 6.92, 5.51 and 5.61 flies/trap, respectively. Verghese et al. (2006) recorded that MAT + Sanitation + Deltamethrin 0.5 ml/l + Azadirachtin 2 ml/l furnished 100 per cent control in pre harvest IPM of mango fruit fly (B. dorsalis). Ahmad et al. (2005) compared different methods to control fruit fly in ber and recorded that Integrated Management and insecticide control showed excellent result with yield potential 35 and 34 kg/plant with minimum damage of 2 and 2.5 per cent, respectively.

Other methods

Bottrell (1979) reported that sex pheromones have been utilized in the insect pest control program through population monitoring, survey, mass-trapping, mating disruption and killing the target pest in the trap. Dhillon et al. (2005) reported that the melon fruit fly can successfully be managed by protein baits, cue-lure traps. Ali (2012) reported that pheromone trap with funnel + Bait trap showed the best performance in controlling cucurbit fruit fly. He reported that highest yield (38.44 t/ha), highest healthy fruit (35.23 t/ha) and lowest infested fruit (3.21 t/ha) were achieved from the treatment. The effects of different doses of gamma radiation on different parameter of peach fruit fly, B. zonata and recorded that by increase in radiation dose to 90 Gray, adult emergence, egg hatching and sex ratio was decreased reported by Mahmoud and Barta (2011). Sohrab et al (2018) management of cucurbit fruit flies among four cucurbits crops Cue lure traps was found most effective in bitter gourd, ridge gourd, and pumpkin and bottle gourd crops them Methyl eugenol. Among four installed Methyl eugenol traps (treated) cucurbits field trials higher yield 10.00, 17.75, 7.7 and 19.6 t/ha were obtained in bitter gourd, bottle gourd, ridge gourd and pumpkin respectively but uninstalled Methyl eugenol traps (untreated) cucurbits field trials 8.00, 14.00, 5.50 and 16.00 tonnes/hectare yield were obtained in bitter gourd, bottle gourd, ridge gourd and pumpkin respectively at Meerut. While among four cucurbits fields installed methyl eugenol traps (treated) cucurbits fields higher yield 11.50, 15.50, 8.00 and 17.50 were obtained (tonnes/hectare) in bitter gourd, bottle gourd, ridge gourd and pumpkin but uninstalled Methyl eugenol traps (untreated) cucurbits fields 8.50, 12.20, 6.65 and 13.50 tonnes/hectare yield were obtained in bitter gourd, bottle gourd ridge gourd and pumpkin respectively
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Conclusion

The cue-lure baited traps attracted the melon fruit fly, *B. cucurbitae* males from mid-July to mid-November (peaked in August) and from 2nd week of August to the 2nd week of November (peaked in September) for the two years. For management of cucurbit fruit flies among four cucurbits crops Cue lure traps was found most effective in bitter gourd, ridge gourd, and pumpkin and bottle gourd crops them Methyl eugenol. It can be concluded that that Integrated Management and insecticide control showed excellent result.

References


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The present chapter elaborates upon the potential of biocontrol fungal endophytes for control and management of various plant-related diseases and their pests. The precise role played by these asymptomatic associates for plant defence against biotic factors (insecticidal, nematicidal, antifungal, antibacterial, and termiticidal) and their bioactive principles responsible for plethora of bioactivities have been a source of investigation for past many years. Further, fungal endophytes like other PGP microbes hold huge promise for commercialisation in agriculture sector.

Agriculture is a highly vital pillar of Indian economy, accounting for 15% of the country's GDP. Approximately, 58% of the rural families are dependent on agriculture as the primary source of living (Srivastava et al., 2016). In wholesome, agriculture consists of crop and animal husbandry (including fisheries) produce providing basic food requirements to mankind. Indian agriculture is hugely dependent on nature’s gamble since it utilizes rain water as main source of irrigation. The exploitative and intensive cultivation of land that emerged following green revolution led to an unfortunate exhaustion of soil resources. Indiscriminate use of pesticides, fungicides and herbicides has led to serious diseases, reduction in arable land and low per hectare yield (Kesavan and Swaminathan, 2006). Self-sufficiency in terms of food security is a matter of immense concern for India.

All microbes which are capable of inhabiting inter and intracellular spaces of plant for at least one period of their life cycle are considered as endophytes (Rodriguez et al, 2009). Most of the endophytic fungi belong to phylum Ascomycota. They are often closely related to fungi known to cause disease, either in healthy tissue or as secondary invaders of damaged tissue; suggesting that the endophytes may have evolved from the pathogens or vice-versa (Schardl et al., 1997). Endophyte assemblage and diversity depends on methods of isolation and type of surface sterilant used. Hypothesis has been developed that fungal endophyte–plant host interaction is characterized by a finely tuned equilibrium between fungal virulence and plant defense as depicted in Figure 1 (Schulz et al., 2002). Geographical and seasonal factors affect the assemblages of endophytic fungi as they are subjected to different selection pressures at each ecological niche (Petrini 1991). Most of the plant species examined to date harbors endophytic fungi within their asymptomatic tissues. Uncovering presence of endophytic fungi inside the host tissue could be done by techniques such as ), direct staining, tissue print immunoassay and transmission electron microscopy (Christansen et al, 2002). Further to verify that a microorganism has an endophytic lifestyle, it must be able to demonstrate Koch’s postulates (it can be successfully reintroduced into disinfected seedlings confirmed by microscopy) (Hyde and Soytong 2008).

Pest and disease management conferred by endophytic fungi

For the very first time Webber (1981) described that the endophyte Phomopsis oblonga protected elm trees against the beetle Physoconemum brevilineum by synthesizing anti-insect metabolites. Considerable increase in plant growth rate was observed in tall fescue plants infected with Neotyphodium endophytic fungi (Faeth and Fagan, 2002). Likewise, Gliocladium catenulatum controlled the witches broom disease of cacao reducing the disease incidence upto 70% at seedling stage (Rubini et al, 2005). Also, Sclerotinia homoeocarpa was effectively controlled by endophytic fungus Epiclloe festucae in turf grasses (Clarke et al, 2006). Morita et al, 2003 described induction of systemic resistance in Chinese cabbage caused by Alternaria sp. in presence of endophytic Heteroconium chaetospira. Fusarium sp. isolated as an endophyte reduced nematode population in soil when tested against Radopholus similis (Vu et al, 2004). Root exudates of endophytic fungi Nigrospora sp. have nematicidal activity by concentrations of 100, 50 and 25 % against root-knot nematode Meloidogyne sp. (Amin, 2015). Endophytic fungus Botryosphaeria sp. P483 isolated from Huperzia serrata produced antinematicidal compound botryosphaerins in (Chen et al., 2015).
Penicillium sp. isolated from Derris elliptica synthesized insecticidal rotenone analogues against aphid Lipaphis erysimi (Hu et al., 2005). Claviceps purpurea (PF-2) endophytically residing within A. inebrians had more than 85% of mortality to cotton aphis (Aphis gossypii) (Zhang et al., 2010). Ethyl acetate concentrate of Pestalotiopsis sp. secreted wilforagine was found antagonist to third instar larvae of Culex pipiens pallens (Han et al., 2013). Similarly, larvicidal action of crude extract of Pestalotiopsis virgulata and Pycnoporus sanguineus depicted growth inhibition against A. aegypti with having LC$_{50}$=101.8 ppm and 156.8 ppm respectively. Extract of endophytic fungus, Mycoleptodiscus indicus harbored inside Echinacea sp. exhibited larvicidal activity against A. aegypti (Rosa et al., 2012). Endophytic fungi from Melia azedarach L. were isolated and their insecticidal activities were studied by Hong et al in 2010. Insecticidal activities of condensed fermentation broth were also examined on Dendrolimus punctatus by touching test and three strains Beauveria KL042, Paecilomyces KL017 and Alternaria KL062 exhibited mortality of 76.67%, 56.67% and 53.33%, respectively (Hong et al., 2010).

Endophytic A. flavus and N. sphaerica isolated from teak leaves (Tectona grandis L.) produced numerous phytochemicals i.e (duroquinone, dodecanoic, pentadecanoic and myristic acid) which reportedly have insecticidal activity (Senthilkumar et al., 2014). Termites are a serious menace to both crops and wooden structures in tropical, semi-tropical and humid places. They cause significant losses to annual and perennial crops, forestry and housing in the semi-arid and sub-humid tropics (Verma et al., 2009). α-terpineol containing fermentation broth of endophytic Aspergillus sp. isolated from termite resistant Port-Orford-Cedar tree exhibited 95.3% mortality against Odontotermes formosanus (Sun et al., 2015). Likewise, endophytic Bacillus subtilis isolated from Juniperus virginiana L. depicted 98% mortality against O. formosanus (Zhao et al., 2011).

Daldinia concentrica isolated endophytically produced mixture of volatile organic compounds which were found safe for post harvest control of dried fruits (Liarzi et al., 2016). Epichloe endophytes were commercialized in perennial ryegrass and tall fescue in New Zealand, Australia, South America and the USA due to presence of metabolites which perform as deterrents to pests and herbivores i.e lolitrem B , ergovaline and peramine (Young, Hume and McCulley 2013). Hexane extract of M. phaseolina from O. sanctum demonstrated highest activity against S. sclerotiorum with IC$_{50}$ value of 0.38 mg/ml due to the presence of 2H-pyran-2-one, 5, 6-dihydro-6-pentyl and palmitic methyl ester (Chowdhary and Kaushik 2015). LC-MS/MS dereplication using accurate mass identified antifungal metabolites sulfamethazine in bioactive fraction of endophytic Acremonium sp (Chowdhary and Kaushik 2018).

Some of the take home points for future plant pathologists are (i) Preservation of endophytic microbiomes of plants dedicatedly, (ii) identify and screen best performing ones from the collections in an agricultural and horticultural context and (iii) investigate novel microbial beneficial secondary metabolites. Few of the limitations of endophytic and plant benefitting microbes are: a) endophytes have demonstrated rather high specificity towards host plant and re-inoculation in plants different from host has been quite challenging. Through study of host specific population dynamics needs to be varied before starting massive production which in turn requires high technology and economical inputs. Multiple attempts are required to optimize dosage inoculum of desired endophytes. Additionally, compatibility of desired endophyte bioinoculant with other biostimulants and chemical synergists needs to be varied before field application. For an effective and workable
solution in terms of integrated pest management, development of “sprayable endophytes usable in presence of other chemical or biochemical pesticides” should be realized by researchers.

Table 1. Endophytic fungi and their bioactivities against plant pathogens and pests

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MECHANISM OF RESISTANCE TO PAPAYA RINGSPOT VIRUS

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Abstract
Papaya ringspot virus (PRSV) is most devastating problem of papaya in several papaya growing areas of India. This chapter concentrates on resistance of papaya to PRSV through different strategies such as cross protection technique and use of transgenic mechanism viz., coat protein mediated resistance, RNA interference-mediated resistance, replicase gene-mediated resistance etc. In different geographical region, there are many PRSV strains which are influenced by respective geographical conditions. Cross protection technique is used for development of resistance in papaya. Pathogen-derived resistance using a coat protein-mediated mechanism, RNA-silencing mechanism and replicase gene-mediated transformation for effective PRSV disease management also followed for the development of resistance and it is used in the development of transgenic papaya. The development of PRSV-resistant papaya via post-transcriptional gene silencing is a promising technology for PRSV disease management. PRSV-resistant transgenic papaya is environmentally safe and has no harmful effects on human health. Recent studies have revealed that the success of adoption of transgenic papaya depends upon the application, it being a commercially viable product, bio-safety regulatory issues, trade regulation and the wider social acceptance of the technology. Here we discusses the genome and the genetic diversity of PRSV, host range determinants, molecular diagnosis, disease management strategies, the development of transgenic papaya, environmental issues, issues in the adoption of transgenic papaya and future directions for research.

Papaya (Carica papaya L.) is an important tropical fruit crop grown in different tropical and sub-tropical region of the world. Papaya is an important fruit crop which is commercially grown in orchards as well as in backyard or kitchen gardens in India. India is the largest producer of papaya in the world and currently it is cultivated in an area of about 136,000 ha with an annual production of about 6,108 million tons during 2016-17 (NHB 2017). The total area under cultivation increases every year but its production and productivity is hampered due to problems of pests and diseases which also affect quality and export of the produce.

Papaya cultivation faces many biotic and abiotic stresses, among them papaya ringspot viral disease type P (PRSV-P) is the most destructive disease (Manshardt, 1992). The term papaya ringspot (PRS) was first coined by Jensen in 1949 to describe a papaya disease in Hawaii. This virus is transmitted by a number of aphid species in a non-persistent manner to a limited host range of cucurbits and papaya. Although papaya is not a preferred host of the PRSV vectors (aphids). Disease management by controlling the vector within the papaya orchard is very difficult as aphid visits and probe papayas but do not colonize papaya orchards.

PRSV is a positive single stranded RNA virus from the potyvirus group (Purcifull et al., 1985). The virions are non-enveloped, flexuous and filamentous particles of 780 x 12 nm. PRSV particles are composed of 94.5% protein and 5.5% nucleic acid. Its genome composed of 10 326 nucleotides with a 5 terminusYeh SD, Gonsalves D (1984). The virus coat proteins have an molecular weight of about 36 k Da as estimated by Western blot analysis. Density of the sedimenting component in purified PRSV preparations was 1.32 g/cm 3 in CsCl. This virus is classified into two types viz., ‘type P’ and ‘type W’ according to the host range. Type P infects papaya and some cucurbits while type W, which infects only cucurbits, although these two types are serologically not distinguishable. PRSV type P can infect papaya plant at any growth stage. The papaya plants after infection with PSRV shows a wide range of symptoms which includes vein-clearing and yellowing of younger leaves, mottling of leaves, distortion and narrowing of leaves, appearance of ringed spots on the fruit and dark green streaks on the petioles and stems. These infected plants show slow and stunting growth and reduction in fruit production with low quality.
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If the transmission occurs before flowering, the flower production will be affected which could cause severe yield loss up to 85-90% (Lokhande et al., 1992; Hussain and Varma, 1994). PRSV was first reported in Hawaii (Jensen, 1949) and was found to occur in tropical and sub-tropical areas where papaya is grown (Purcifull, 1972). In India, PRSV-P was first reported from North India during 1960 (Khurana and Bhargava, 1970), but only during 1995 in South India (Byadgi et al., 1995). Within a span of 5 to 6 years, PRSV had spread throughout South India (Verghesee et al., 2002).

There are some conventional and modern approaches to deal with this problem. Conventional methods adopted are less successful but still adopted by farmers which are mild strain cross protection and conventional breeding programs. In modern approaches coat protein mediated resistance (CPMR) has been successfully used to produce transgenic plants resistant to viruses (Baulcombe, 1996; Beachy, 1997), including transgenic papaya resistant to PRSV from Hawaii (Fitch et al., 1990, 1992).

Classical resistance and tolerance in papaya

Natural resistance to PRS in Carica papaya has not been identified. Resistance to PRSV exists in Vasconcellea species, which are related to Carica papaya. However, crosses between Carica papaya and Vasconcellea species are not fertile. Efforts to transfer the putative resistance gene(s) by crossing of Carica papaya and Vasconcellea species followed by embryo rescue and subsequent back crossing with Carica papaya has been attempted, but so far have not been developed to the stage of commercial cultivation. Efforts to incorporate tolerance to PRS in Carica papaya have been a bit more successful. Some PRS tolerant lines have been commercialized in the Philippines and are sold commercially in Taiwan, and tolerant lines have been released in Florida and Thailand; these lines develop only mild symptoms when infected. PRSV can still spread throughout the orchards of tolerant lines, since they are not resistant to the virus. When these tolerant lines do become infected with PRSV, they are still able to produce respectable crops. However, the quality of fruit from these infected tolerant lines is generally marginal which leads to their not being widely grown.

Cross protection

Cross protection is a technique which is adopted in many fruit crops for developing immunity to certain virus disease by using mild strain of that virus. This mild strain of virus protects the plant against more virulent related strain. Cross protection was practiced since 1980 in Taiwan and to a very limited extent in Hawaii. The first successful attempt to use mild strain of PRSV for large scale protection was initiated in Taiwan, where PRSV was widespread and causing losses to crop. Hypothesis behind cross protection is by using the mild strain of PRSV was used, which was developed by selecting a mild mutant from nitrous acid treatment of tissue extracts of leaves infected with a severe strain of PRSV. The infected mutant strain shows mild symptoms on papaya and protects against the more virulent strain of PSRV. The general procedures for cross protection of papaya were production of inoculums, which consisted of infection of Cucumis metuliferus (horned melon) with the mild PRSV strain, spray inoculation of young papaya seedlings with extracts from infected C. metuliferus plants in the greenhouse or net house, and planting the seedlings in the field 3-4 weeks after establishment of the virus so that these plants would subsequently be protected against damage caused by severe strains of the virus.

Cross protection was practiced on a large scale in Taiwan for several years, but the practice was abandoned for several reasons. The logistics of producing the mild strain inoculums and subsequently ensuring the supply of mild strain inoculated papaya seedlings added a significant step in the production of papaya. This added step would have been worthwhile, but results from the large scale application in Taiwan showed that the mild strain did not protect as well against the Taiwanese PRSV strains as it did against the virulent Hawaiian strain. After awhile farmers were not enthusiastic to continue the management practice of cross protection in Taiwan.

In Hawaii, cross protection worked very well in limited field trials on the island of Oahu where PRS was very severe. Even in Hawaii, however, the practice was not fully commercialized because the mild strain caused quite severe symptoms on some cultivars such as Sunrise and because the logistics for raising the inoculums, uniformly infecting plants with the mild strain and delivery of the mild strain-infected plants to the farmers did not work out economically.
Genetically Engineered resistance in papaya

Development of transgenic papaya to control PRSV: The investigations to develop genetically engineered papaya with resistance to PRS began in 1985. The concept of parasite-derived resistance (PDR) was employed for the controlling PRSV. Parasite-derived resistance or pathogen-derived resistance is a phenomenon whereby transgenic papaya plants containing genes or sequences of a parasite (The coat protein gene of a virus) are protected against detrimental effects of the same or related pathogens. The application of this technique for controlling plant viruses was first demonstrated by Beachy et al. (1990) in protection of tobacco by tobacco mosaic virus by implementation of transgenic tobacco expressing the coat protein gene of tobacco mosaic virus and further investigation reported that this approach is effective in controlling many plant viruses (Powell et al., 1986).

For papaya, our approach was to develop transgenic papaya with the coat protein (CP) gene of PRSV which would protect the transgenic plant against damage caused by PRSV. Briefly, the research to obtain the transgenic papaya involved isolating and sequencing the CP gene of PRSV from Hawaii, transforming embryogenic calli of non-transgenic Sunset papaya, a commercial papaya in Hawaii, selecting and regenerating plants transformed with the CP gene of PRSV, screening transformed plants for resistance to PRSV, field trials, deregulation and commercialization.

Evolution of Transgenic Papaya

Resistance developed through utilisation of coat protein (CP): The investigations to develop genetically engineered papaya with resistance to PRS began in 1985. The concept of parasite-derived resistance (PDR) was employed for the controlling PRSV. Parasite-derived resistance or pathogen-derived resistance is a phenomenon where by transgenic papaya plants containing genes or sequences of a parasite (The coat protein gene of a virus) are protected against detrimental effects of the same or related pathogens. The application of this technique for controlling plant viruses was first demonstrated by Beachy et al. (1990) in protection of tobacco by tobacco mosaic virus by implementation of transgenic tobacco expressing the coat protein gene of tobacco mosaic virus and further investigation reported that this approach is effective in controlling many plant viruses (Powell et al., 1986). The CP mediated protection of PRSV has been adopted throughout the world (Davis and Ying, 1999). Fitch, 1995 reported that scientist have preferred CP genes as the agents utilized to develop PRSV-resistant papaya. Fitch et al. (1992) conducted an experiment for development of transgenic papaya which was resistance to PRSV by utilising CP genes resistant to PRSV by adopting the gene transfer system of immature zygotic embryos with a plasmid construction containing the neomycin phosphotransferase II (npt II) gene.

In 1991, a PRS-resistant GE papaya line designated 55-1, was identified which was used to create the cultivars Sun Up and Rainbow. Sun Up was developed through transformation of somatic embryos with the CP gene of the Hawaiian PRSV strain Sunset and it is homozygous for the CP gene (Fitch et al., 1992). Rainbow is a F₁ hybrid developed through crossing of Sun Up (transgenic) with Kapoho (non-transgenic).

Cheng et al., 1996 used CP gene of Taiwanese strain of PRSV constructed with a Ti binary vector pBGCP through Agro-bacterium mediated transformation and developed PRSV-resistant transgenic papaya. Gonsalves et al. (1998) observed that resistance to homologous PRSV isolates from Hawaii, Australia, Taiwan, Mexico, Jamaica, the Bahamas, and Brazil when using gene gun technology for the development of a PRSV-resistant papaya variety by transferring of untranslatable CP gene. developed transgenic papaya which shows high level of resistance against a severe strain of PRSV (PRV-HA) by expressing the CP gene of the mild PRSV strain from Hawaii (PRV HA 5-1) was developed by Tennant et al. (1994). Bau et al. (2004) created transgenic papaya lines by expressing a CP gene with broad-spectrum resistance PRSV of different strains in different geographical areas in Taiwan. Magdalita et al. (2004) used CP gene of PRSV isolate from Philippine and regenerated putative transgenic R₀ plantlets, which were moderately susceptible while R₁ plantlets were completely resistant. Chen et al., 2001, Lines et al., 2002 and Tennet et al., 2002 reported that in recent decades many scientist from the different countries using different explants with plasmids containing the neomycin phosphotransferase II (nptII) gene trying to develop the PRSV-resistant transgenic papaya. However, the effectiveness of CP mediated PRSV resistance depends upon the origin of PRSV isolates. The untranslatable and translatable constructs of PRSV-CP containing genes utilized in different countries.
### Table 1. Adoption of various technologies for development of transgenic papaya by scientists worldwide

<table>
<thead>
<tr>
<th>Country</th>
<th>Cultivar</th>
<th>Construct</th>
<th>Type of coat protein</th>
<th>Transformation</th>
<th>Transgenic expression</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Australia</td>
<td>Local variety</td>
<td>uidA leader + CaMV35S promoter + PRSV Bridgeman Downs cp gene from Q/S start with stop codon in the middle of sequence</td>
<td>Translatable cp</td>
<td>Biolistics</td>
<td>cp not detected in ELISA and low levels of cp detection in northern analysis</td>
<td>Lines et al. (2002)</td>
</tr>
<tr>
<td>Brazil</td>
<td>Sunrise solo, Sunset solo</td>
<td>CaMV35S + CMV leader + PRSV Bahia cp gene from Q/S start</td>
<td>Translatable cp</td>
<td>Biolistics</td>
<td>Low to high levels cp detected in ELISA</td>
<td>Souza et al. (2005)</td>
</tr>
<tr>
<td>Florida</td>
<td>CV.F65</td>
<td>uidA leader + CaMV35S promoter + PRSV HIK cp gene from Q/S start</td>
<td>Translatable cp</td>
<td>Agrobacterium</td>
<td>cp is not detected in northern analysis</td>
<td>Davis and Ying (2004)</td>
</tr>
<tr>
<td>Florida</td>
<td>CV.F65</td>
<td>uidA leader + CaMV35S promoter + PRSV HIK cp gene from Q/S start in antisense</td>
<td>Untranslatable cp</td>
<td>Agrobacterium</td>
<td>cp is not detected in northern analysis</td>
<td>Davis and Ying (2004)</td>
</tr>
<tr>
<td>Florida</td>
<td>CV.F65</td>
<td>uidA leader + CaMV35S promoter + PRSV HIK cp gene from Q/S start with frame shift mutation</td>
<td>Untranslatable cp</td>
<td>Agrobacterium</td>
<td>cp is not detected in northern analysis</td>
<td>Davis and Ying (2004)</td>
</tr>
<tr>
<td>Hawaii</td>
<td>Sunset solo</td>
<td>CMV leader + 16 aa CMV cp + PRSV HA 5-1 cp gene from Q/S start</td>
<td>Translatable cp</td>
<td>Biolistics</td>
<td>cp RNA detected in northern analysis</td>
<td>Fermin. (2002)</td>
</tr>
<tr>
<td>Hawaii</td>
<td>Sunset solo</td>
<td>CaMV35S + CMV leader + PRSV Caymanas untranslatable cp</td>
<td>Untranslatable cp</td>
<td>Biolistics</td>
<td>cp RNA detected in northern analysis</td>
<td>Tennant et al. (2002), Tennant et al. (2005)</td>
</tr>
<tr>
<td>Jamaica</td>
<td>Sunrise solo</td>
<td>CaMV35S + CMV leader + PRSV Caymanas untranslatable cp</td>
<td>Untranslatable cp</td>
<td>Biolistics</td>
<td>cp RNA detected in northern analysis</td>
<td>Tennant et al. (2002), Tennant et al. (2005)</td>
</tr>
<tr>
<td>Jamaica</td>
<td>Sunrise solo</td>
<td>CaMV35S + CMV leader + PRSV Caymanas cp from Q/S start</td>
<td>Translatable cp</td>
<td>Biolistics</td>
<td>cp RNA detected in northern analysis</td>
<td>Tennant et al. (2002), Tennant et al. (2005)</td>
</tr>
<tr>
<td>Taiwan</td>
<td>Tainung no. 2</td>
<td>uidA leader + CaMV35S promoter + PRSV YK cp gene from Q/S start</td>
<td>Translatable cp</td>
<td>Biolistics</td>
<td>cp transcript detected in northern analysis</td>
<td>Bauet al. (2003), Tripathi et al. (2004)</td>
</tr>
<tr>
<td>Thailand</td>
<td>KhakDum</td>
<td>CaMV35S + uidA leader + PRSV Ratchaburi province cp</td>
<td>Translatable cp</td>
<td>Biolistics</td>
<td>cp detected in western analysis</td>
<td>Ruanjana et al. (2007), Kerkundit et al. (2007)</td>
</tr>
<tr>
<td>Venezuela</td>
<td>Thailand red</td>
<td>CaMV35S + CMV leader + PRSV EV &amp; VE from Q/S start</td>
<td>Translatable cp</td>
<td>Agrobacterium</td>
<td>cp RNA is not detected ELISA and low level cp detected in northern analysis</td>
<td>Fermin et al. (2004)</td>
</tr>
</tbody>
</table>
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RNA-Interference-Mediated Resistance

Waterhouse et al. (1998) was first discovered RNA interference (RNAi) mediated virus resistance by using the Potato virus Y in transgenic tobacco plants. RNA-Interference-Mediated Resistance brings a new area with lots of scope in developing molecular tools for crop improvement (Eamens et al., 2008) and also in various genetical studies, biotic and abiotic stress problems in plants, protection against pathogens and insects. Ramash et al. (2007) reported that this novel technology could also be used for procreating disease resistance by suppressing a specific gene or genes. RNA present in PRSV which has single open reading frame which is translated into a large polyprotein which produces final protein products as reported by Yeh et al. (1992). RNA-mediated protection is effective if the attacking virus has similar transgene. There is different in strains from different geographical region and it is a problem behind development of PRSV-resistant transgenic plants.

The failure of PRSV resistance has frequently involved the silencing by suppressor proteins of viral origin (Ruanjan et al., 2007) and this problem could be overcome by the silencing suppressor protein HcPro, through a RNA-silencing mechanism within transgenic papaya. The helper component proteinase (HcPro) has been shown to be a highly effective suppressor of RNA silencing. Mangrauthia et al. (2008) reported that helper component proteinase is an important component which must be taken into consideration for the development of PRSV-resistant papaya on the Indian sub-continent. The mechanism of RNA-mediated virus resistance is also referred to as homology dependency resistance to reflect the specific mechanism of post-transcriptional gene silencing ‘PTGS’ (Meins, 2000; Wassenegger and Pelissier, 1998). PTGS is the accumulation of 21–25 nucleotide small-interfering RNAs, the sequence-specific degradation of target mRNAs, and the subsequent methylation of target gene sequences. Tennant et al. (2001) reported that mechanisms of transgenic papaya resistance against PRSV are sequence homology dependent and mediated by RNA via PTGS. They found that an untranslatable CP gene was able to confer resistance to the homologous strain of the virus isolate of PRSV by PTGS. On the other hand, the silencing suppressor was the main factor for the suppression of PRSV transgenic resistance as reported by Tripathi et al. (2004). Ruanjan et al. (2007) also reported that transgenic papaya showed resistant to PRSV by suppressing post-transcriptional gene silencing (PTGS).

Replicase Gene-Mediated Resistance

This technology has been successfully utilized for development of resistance in plants against several viral diseases viz., tobacco mosaic virus, pea early browning virus, cucumber mosaic virus and potato virus X (Carr and Zaitlin, 1993). Resistance developed by the introduction of replicase gene was first time done for the tobacco mosaic virus (TMV) in Nicotiana tabacum (Golemboski et al., 1990). Here the host resistance mechanism is protein based which changes upon mutation as the primary structure of the protein encoded by the transgene changes upon mutation. Within genera replicase structures are different. Replicase genes with mutations have been shown to be able to confer virus resistance Nunome et al., (2002). Chen et al. (2001) reported that replicase gene (RP) conferred resistance to PRSV in transgenic papaya. Wei et al. (2007) reported that transgenic papaya with mutated replicase genes (RP) showed high resistance to PRSV.

Summation

The 1998 APSnet Feature was entitled "Transgenic Papaya: A New Hope for Hawaii." It can be said that the transgenic papaya fulfilled the hope of the Hawaii papaya industry to control PRSV and to stabilize and restore the supply of papaya to nearly the level existing before PRSV entered Puna in 1992. There remain challenges to the Hawaii papaya industry, mainly in getting the transgenic papaya approved for sale in Japan. Due to its success, the transgenic papaya has often been referred to as the model for the use of biotechnology to help agriculture without investments by large companies. Indeed, transgenic papaya has been developed and transferred to other countries. In Jamaica, and especially in Thailand the transgenic papaya has performed very well under field trials, and deregulation procedures are progressing. However, it is very likely that the process will take much more time than it did in Hawaii. This is not due to the technical difficulties in product development, but is due to the GMO controversy. Thus, technology has moved along, and the major challenge will be to see how political processes proceed toward decisions on whether this technology will actually be used to fight this very severe problem in Thailand, Jamaica, and other countries (Gonsalves et al., 2004).
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INNOVATIVE APPROACHES IN INSECT PEST MANAGEMENT

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Abstract

As it is clear that chemical used for insect pest management are found to be associated with so many disadvantages like residue in food and feed, health hazard to humans, reduction in population of non-target organisms, development of resistance and resurgence in insects and environmental pollution. The insecticides are also not compatible to biological control management of various insects as it has lethal effect to natural enemies. So, it is need of time to develop some innovative approaches of insect pest management which can overcome the disadvantages associated with chemical insecticides. The biotechnological approaches is the best pest management practice as it have following advantages over chemical insecticides like reduce environmental contamination, avoid operator exposure to pesticides, effective pest control throughout the season, target specific and compatible with natural enemies. In biotechnology introduction of BT cotton is one of the successful example for Bollworms management. Insect growth regulator like insect hormones and chitin synthesis inhibitor as well as Pheromones can become a viable component of management program, since many commercial formulations are now available in market. Both Insect Growth Regulators and Pheromones show low mammal toxicity, biodegradability and specific nature of these compounds make them eco friendly and add to the diverse spectrum of pest control. Therefore, these innovative approaches can be used as important component of integrated pest management program.

In insect pest management the theater arising due to over use of insecticides. The development of resistance and resurgence to these chemical insecticides has made the management of insect pests extremely difficult. The other environmental concerns are also associated with insecticides like residue in food and feed, heath hazard to humans reduction in non-target organisms population and environmental pollution. So the entomologists, all over the world trying to find eco-friendly and sustainable alternatives for management of insect pests. Out of various strategies tried, one is the application of biotechnological approaches/ technology, Insect Growth Regulators (IGRs) and pheromones are innovative approaches of insect pest management. All theses method’s of insect pest management are extremely safe to humans and environment. The selective nature and different modes of action from the conventional insecticides make theses approaches’ safe to natural enemies as well as important component of Integrated Pest Management (IPM). These methods also prevent development of resistance and resurgence in insect. In this chapter three innovative approaches viz biotechnological approaches, Insect Growth Regulators and Pheromones is explained in detail for insect pest management.

Biotechnological approaches in pest management

Biotechnology defined as technique in which living organism or its part is used to produce improved plant which may show resistance or toxicant to the insect pest. New crop cultivars with resistance to insect pests and diseases combined with bio-control agents should lead to a reduced reliance on pesticides, and thereby reduce farmers’ crop protection costs, while benefiting both the environment and public health. The methods used in crop improvement are tissue culture technique and recombinant DNA technique.

Tissue culture technique

Tissue culture methods offer a rich scope for creation, conservation, and utilization of genetic variability for the improvement of field, fruit, vegetable, and forest crops, and medicinal/aromatic plants. Tissue culture technique provides the means to multiply and regenerate novel plants from genetically engineered cells. The promising plant thus produced may be readily cloned in cultures under aseptic conditions. Tissue culture involves various techniques like Micropropagation, Somaclonal variation, Protoplast culture and somatic hybridization.

Micropropagation is a the method of rapidly multiplying stock plant material to produce a large number of progeny plants that has a great potential to develop high quality as well as disease-free plants. Advancements in this field have led to the development of several techniques for rapid multiplication and improvement of a wide
range of crops and their production systems. Plants produce by micropropagation are of true to type, disease free. Micropropagated field grown plants usually exhibited vigorous growth, better quality and higher yield. **Somaclonal variation** is the variations obtain in plants that have been produced by *invitro* somatic cell culture. This variation that occurs due to genetic mutation which caused by in *vitro* conditions or by chimera separation. Somaclonal variation is usually undesirable which lead to new cultivar that may have desirable characteristics or increased pest resistance. The main steps involved in the isolation of somaclones for insect resistance are (1) growing calli or cell suspension cultures for several cycles from a high yielding and well Adapted susceptible variety (2) regeneration of plant from such Long term cell lines (3) evaluation of large population of regenerated plants for insect resistance.

**Protoplast culture and somatic hybridization** is a type of genetic modification in plants by which two distinct species of plants are fused together to form a new hybrid plant with the characteristics of both. Somatic hybridization via protoplast fusion is an important tool for the production of interspecific and intergeneric hybrids. Hybrids have been produced either between different varieties of the same species or between two different species. Complete fusion of the nuclei and cytoplasm of somatic cell from two species leads to the formation of somatic hybrid cell and plant. Likewise, the fusion of cytoplasm from two species and nuclear genes from any one leads to the development of cybrid. Somatic hybridization has been widely exploited in different crops to create novel hybrids with increased yield and resistance to pest and diseases.

**Recombinant DNA technique**

Recombinant DNA technique involves transfer of recombinant DNA into plant cells to generate transgenic plants. With this technology, a gene or multiple genes can be identified, cut, and inserted into the genome of another organism. Recombinant DNA technology involve the use of three main tools: (1) enzymes (restriction enzymes, polymerases, and ligases); (2) vectors; and (3) host organism. The enzymes will help cut (restriction enzymes), synthesize (polymerases), and bind (ligases) DNA. The restriction enzyme will cut at a specific site within the DNA molecule called a restriction site. Usually, the restriction enzyme will produce sticky ends in the DNA sequence that will help it bind specifically to the desired gene. The vector will carry the desired gene. Vectors are considered as the final vehicles that carry genes of interest into the host. A vector must contain the same restriction sites that are found within the desired gene to facilitate gene integration. The host is the cell in which the recombinant DNA is introduced. To introduce vectors into hosts, techniques involving microinjection, biolistics, gene gun, alternate cooling and heating, and calcium phosphate ions have been used. In generally, a recombinant DNA technology has five steps: (1) cutting the desired DNA by restriction sites, (2) amplifying the gene copies by PCR, (3) inserting the genes into the vectors, (4) transferring the vectors into host organism, and (5) obtaining the products of recombinant genes.

The two methods used for gene transfer in plants are: (1) **Vector-Mediated Gene Transfer** and (2) **Direct or Vector-less DNA Transfer** (Fig 1. Vector-Mediated Gene Transfer involves the use of a plant pathogen called Agrobacterium tumefaciens, which causes crown gall disease in many species. This bacterium has a plasmid, or loop of non-chromosomal DNA, that contains tumor-inducing genes (T-DNA), along with additional genes that help the T-DNA integrate into the host genome. For genetic engineering purposes, Agrobacterium must first be “disarmed” so that it does not make the plant sick. This is done by removing most of the T-DNA while leaving the left and right border sequences, which integrate a foreign gene into the genome of cultured plant cells. The Direct or Vector-less DNA Transfer method is a “gene gun,” which fires gold particles carrying the foreign DNA into plant cells. Some of these particles pass through the plant cell wall and enter the cell nucleus, where the transgene integrates itself into the plant chromosome. Because both methods of gene transfer are fairly random, one must screen for the plant cells that contain the foreign gene.

**Insect resistant Transgenic Crops**

Insect-resistant transgenic crops were first commercialized in the mid-1990s with the introduction of GM corn (maize), potato and cotton plants expressing genes encoding the entomocidal δ-endotoxin from *Bacillus thuringiensis* (Bt; also known as Cry proteins).

*Bacillus thuringiensis*, a natural soil bacteria that secretes a deadly endotoxin. Bt toxins are highly effective for many pest organisms, like Lepidopterans, coleopterans, Dipterans and other related species, but not toxic to mammals and most other non-target organisms. The use of genes encoding endotoxins from *Bacillus thuringiensis* is now a well-established technology for producing transgenic plants with enhanced resistance to the larvae of lepidopteran insect pests. Regarding mechanism of bacterial toxin action, when the insect larvae feed on transgenic plant, crystals and spores are ingested into the midgut of the insect. Since the pH is alkaline in nature, so the the crystals become toxic to insect midgut leading to septicamia.
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Fig 1. The methods of gene transfer in plants

**Bt Potato**

The first commercially available Bt crop was potato expressing the cry3A toxin gene (NewLeaf), a product manufactured by an affiliate of Monsanto (NatureMark) that was commercialized in 1995. The expressed cry3A gene derived from Bt subsp. tenebrionis was selected due to its high activity against larvae of the Colorado potato beetle (*Leptinotarsa decemlineata*), one of the most economically relevant pests of potato. NewLeaf potatoes were protected from *L. decemlineata* during all of its life stages and throughout the entire growing season, resulting in significant reductions in insecticide use.

**Bt cotton**

Bt cotton is an insect-resistant transgenic crop designed to combat the bollworm. Bt cotton was created by genetically altering the cotton genome to express a microbial protein from the bacterium *Bacillus thuringiensis*. The Bt cotton plants are genetically modified by the addition of genes encoding toxin crystals in the Cry group of endotoxin. When ingested by insects, the Cry toxins are dissolved and activated by the high pH environment of the animal's gastrointestinal system. In the midgut, the activated Cry molecules bind to cadherin-like proteins on cells comprising the brush border membrane. The epithelium of the brush border membrane has the function of maintaining separation of the body cavity from the gut while allowing absorption of nutrients from the digested food bolus. Cry toxins bind to specific locations on the cadherin-like proteins present on the epithelial cells of the midgut, and form ion channels allowing potassium ions to flow from the cells. As the control of potassium ion concentration is critical to the survival of every living cell, they are tightly regulated under normal function. With the formation of Cry ion channels and the subsequent efflux of potassium ions, the affected epithelial cells lyse and die. This creates gaps in the brush border membrane, allowing bacteria and Bt spores to enter the body cavity. Subsequently, the insect will die of internal infection after ingesting a Bt crop.

The Bt genes that are currently deployed are from two sources. Monsanto developed and deployed the Cry IA(c) gene in its Bollgard varieties, which are the most widely used in all nine countries that grow cotton. The second source is the Bt-fused gene that was developed by the public sector, Chinese Academy of Agricultural Sciences (CAAS) in Beijing, China (Jayaraman *et al*., 2005).

**First generation Bt-cotton**

The most prevalent Bt-gene on a global basis Cry I A(c) was incorporated into Coker 312 cotton designated MON 531 by Monsanto and later named Bollgard cotton (first generation Bt-cotton. The high transformation
efficiency was achieved in Coker 312 with *Agrobacterium tumefaciens*. The transformed Coker was then back crossed with lines from Delta and Pine land and other companies that had necessary agronomic qualities for commercial acceptance.

**Second generation Bt-cotton (Bollgard II cotton)**

The Insect Resistance Management (IRM) strategy for Bt-cotton that Monsanto in conjunction with USDA developed second generation of improved Bt-cotton with two Bt-genes, now designated Bollgard II. The new product bollgard II, Event 15985 was developed using particle acceleration plant transformation procedures to add the Cry II A(b) gene to the cotton line DP 50B that already had the cry 1 A(c) gene. Hence the bollgard II cotton contains Cry 1 A(c) and Cry II A(b). The dual gene cultivars are expected to provide growers with a broader control over a wide variety of insects. In 2002, Dow Agrosciences announced the development of new Bt-cotton with traits that confer broad spectrum resistance to lepidopteran pest of cotton. The new Bt cotton product contains the dual genes Cry IA(c) and Cry IF, transformed with *Agrobacterium tumefaciens* and incorporated through back crossing into several high quality commercial varieties of cotton.

**Bt Maize:** Transgenic Bt maize was the effective to control of the European corn borer (*O. nubilalis*) (Koziel et al. 1996). Larvae of *O. nubilalis* are highly susceptible to Cry1Ab, which was the toxin gene selected for production of Bt maize varieties, which were based on events 176 (KnockOut from Syngenta and NatureGard from Mycogen), Bt11 (Agrisure from Northup King), or MON810 (Yieldgard from Monsanto) (Sanahuja et al. 2011). Ensuing Bt maize products expressing the cry1Fa or cry9C toxin genes were commercialized to target *O. nubilalis* and additional selected species of armyworm (*Spodoptera* spp.).

Expression of multiple toxins in Bt maize achieves increased control of target pests with low susceptibility to single-toxin Bt crops. For instance, pyramiding of the cry2Ab2 gene with a chimeric toxin gene (cry1A.105) composed of portions of the cry1Ab, cry1Ac, and cry1Fa genes, into maize plants expanded the range of control to armyworms (*Spodoptera* spp.) and the black cutworm (*Agrotis ipsilon*). Similarly, Agrisure Viptera maize producing Vip3Aa20 and Cry1Ab toxins can control corn earworm (*Helicoverpa zea*) and the fall armyworm (*Spodoptera frugiperda*), which display low susceptibility to Cry1A toxins.

**Table 1: Major transgenic crops expressing Bt genes for Insect Resistance**

<table>
<thead>
<tr>
<th>Transgenic Crop Plants</th>
<th>Foreign Gene</th>
<th>Target insect pests</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cotton</td>
<td>Cry1A(b), Cry1A(c)</td>
<td>H. armigera, H. zea, Heliothis virescens, Pectinophora gossypiella, S. exigua</td>
</tr>
<tr>
<td>Maize</td>
<td>Cry1A(b), Cry1A(c), Cry9C</td>
<td>Chilo partellus, H. zea</td>
</tr>
<tr>
<td>Tomato</td>
<td>Cry1A(c)</td>
<td>Manduca sexta</td>
</tr>
<tr>
<td>Tomato</td>
<td>Bt(k)</td>
<td>M. sexta, H. zea</td>
</tr>
<tr>
<td>Rice</td>
<td>Cry1A(b), Cry1A(c), CryH(a)</td>
<td>Scirpophaga incertulas, Cnaphalocrosis medinalis</td>
</tr>
<tr>
<td>Potato</td>
<td>Cry 1A(b), Cry1A(b)6, CryIII A, CryIII B</td>
<td>Phthorimaea operculella, Leptinotarsa sp.</td>
</tr>
<tr>
<td>Tobacco</td>
<td>Cry1A(c)</td>
<td>H. virescens, M. sexta,</td>
</tr>
<tr>
<td>Brinjal</td>
<td>CryI AC</td>
<td>Leucinodes orbonalis</td>
</tr>
</tbody>
</table>

**Advantages of BT Crops**

They help in controlling the soil pollution as the use of synthetic pesticides are reduced when the plants begin to produce the toxins by themselves in own tissues. BT Crops help in protecting the beneficial insects. Reduced manpower and labor charges. The pests hiding inside plant parts are controlled effectively. It is cost effective as multiple sprays are not needed.

**Disadvantages of BT Crops**

The BT crops are more costly than the normally grown crops. There is a possibility for allergic reactions while using these crops. BT Crops are not effective for certain pests including spider mites, seed corn, etc.
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Insect Growth Regulators (IGRs)

Insect Hormones

a) Neurohormones: It is also called as brain hormone or prothoracicotropic hormone (PTTH) as it is secreted from the groups of neurosecretory cell present in the insects brain. There are more than 40 neurosecretions on record in the insects but the prothoracicotropic hormone (PTTH) is extensively studied. The application of PTTH and other neuropeptides could induce metabolic disturbances in the target insect species. The metabolic disturbance can lead to derangements, so it could used effectively in insect pest management. The limitations associated with neurohormones are :-

- The synthesis of neurohormones is very costly and required skilled person with sound knowledge of insect physiology.
- They are proteinous in nature due to which the penetration of insect cuticle become difficult
- They are highly unstable under field conditions due to their heat labile nature.

b) Juvenile Hormones (JH) and their analogues (JHAs): The corpora allata is principal organ for secretion of Juvenile hormones (JHs. In lepidoptera corpora allata cells are bilaterally paired whereas in other insects they are fused to form a mass. They are chemically sesquiterpenoids. The six type of JHS found in insects are JH I, JH II, JH III, JH 0, iso JH 0 and JH bisepoxide. The concentration of JHS in the haemolymph is determined by the rate of its biosynthesis in the corpora allata and its degradation in the tissues. They perform differential roles in immature and mature insects. In immature stages the higher concentration of JHAs inhibits metamorphosis, thus affecting morphogenesis. In adults they perform gonadotropic actions, synthesis and uptake of yolk. So exogenous application of JHs will derange the process of metamorphosis producing adultoids, larval-pupal intermediates or supernumerary instars, which rarely moult into proper adults. Williams (1967) was the first to raise the possibility of use of JHs as the ‘Third generation Pesticides’ which led to the synthesis of a large number of JHAs. Farnesol was first synthetic hormone synthesized use in pest control. It does not penetrate insect cuticle which limit its use in pest control. Methoprene (ALTOSID*) was the first, commercially JHA introduced in the market by Zoecon Corporation, California and approved by Environmental Protection Agency, USA for use against the California flood-water mosquito, Aedes nigromaculatus (Linnaeus) USA. It was also registered in UK in 1980 to control pharaoh’s ants under the trade name PHARORID®. Methoprene has also been used in combination with the organophosphate insecticide methamiphos to control grain pests Sitophilus oryzae (L) and S. granaries L. (Thacker, 2002). Hydroprene (GenTrol®/GENCOR®/GENCORPLUS®/ MATRON®/ MATOR® / TURBO=CIDE GOLD®) was another JHA marketed for the control of cockroaches that by inducing sterility in treated insects (Sehnal, 1985). Hydroprene resulted in arrested larval growth, morphological deformities, incomplete emergence and twisted wings in T. castaneum and T. confusum Duval (Arthur, 2003). Hydroprene and another JHA (R-20458) demonstrated appreciable morphogenetic activity by producing adultoids, nymphal-adult intermediates and supernumerary nymphs upon their topical application to mustard aphid, Lipaphis erysimi (Kalt.), apart from affecting reproduction of the aphid, apterising and fumigant effects of the JHAs (Singh & Sidhu, 1992, 1991, 1990a & 1988. Fenoxycarb (INSEGAR®/ PRECLUDE®/ PRECISION®) was registered in Switzerland and Italy for the control of several fruit pests such as Pear psylla, Cacopsylla pyricola, grape moths, Lobesia botrana Schiff and Eupoecilia embigulla Hubner, plum moth, Grapholita funebrana Treitschke; codling moth, Cydia pomonella L.; summer fruit tortrix, Adoxophyes orana (Fischer von Rosslerstamm); fruit tree tortrix, Archips podana (Scopoli), and several other tortricid species (Solomon & Fitzerald, 1990; Charmillot et al., 1987; Pluciennik et al., 1999; Charmillot et al., 1989; Charmillot & Brunner, 1989; De Reed et al., 1985. Pyriproxyfen (ADMIRAL®/ DISTANCE®/ NYLAR®/ NEMESIS®/ TIGER® 10 EC), a phynylethylene juvenile hormone mimic, marketed for the control of public health pests (Estrada & Mullia, 1986 ; Okazawa et al., 1991), field pests and as a feed compound in poultry, cattle, and swine for the control of house fly, Musca domestica L., and the facr fly, M. autumnalis De Geer (Miller, 1989), was also shown to suppress egg hatch in pear psylla, C. pyricola, egg hatch and adult formation in cotton whitefly, Bemisia tabaci (Gennadius) and greenhouse whiteflies, Trialeurodes vaporarium (Westwood) and Haematobia irritans (L.) in Israel (Higbee et al., 1995; Ishaaya & Horowitz, 1992; Bull & Meola, 1993; Ishaaya et al., 1994. It also caused sterility and reduction in adult longevity in cotton aphid, Aphis gossypii Glover on cotton (Kerns & Stewart, 2000. Owing to its
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environmental instability, Pyriproxifen can be effectively used in greenhouse conditions. Epifenobnane, another JHA, was reported to be effective against last larval instars of *S. litura* on cauliflower (Singh & Singh, 1988). Toxic effects along with abnormal morphogenesis were observed after foliar treatment with a JHA, R-20458 and hydroprene against mustard aphid, *Lipaphis erysimi* (Kaltenbach) on cabbage (Singh & Sidhu, 1990b). JH analogue, ZR-777 drastically reduced the fecundity of red rust flour beetle, *T. castaneum* (Singh, 1994).

**Plants with JH factors:** A large number of JHAs of plant origin have also been identified from a number of plant species. The compound juvocimence I and II, isolated from sweet basil, *Ocimum basilicum* showed JH activity against milkweed bug, *Oncopeltus fasciatus* Dallas. Roots of the American coneflower, *Echinacea angustifolia* has a compound called echinolone, which showed JH activity against *O. fasciatus* and *T. molitor* (Jacobson *et al.*, 1975). Biological activity of some plant oils, especially the terpenoid lactones has also been noted against the mustard aphid, *L. erysimi* (Arora *et al.*, 1982; Singh & Kalsi, 1996).

**Anti-Juvenile Hormones (AJHAs):** The knowledge that some plants produce juvenile hormone mimics has stimulated the search for antagonists of JHs within plants. The anti-JHs is accomplished by competing with JH in binding to the JH receptors of to the JH-carrier proteins, killing the corpora allata cells, or interfering with JH biosynthesis (Leighton *et al.*, 1981). There are JHAs that compete with JH at the receptor site and become feedback inhibitors of JH biosynthesis. An example is ETB [ethyl 4- (2-pivaloyloxybutyloxy)-benzoate], which showed JH agonist and antagonist activities in *M. sexta* larvae (Staal, 1986).

**c) Moulting Hormone Analogue (MHAs):** Ecdysones or moulting hormones are secretions of prothoracic glands of insects. The Prothoracic glands sequester cholesterol from the circulating haemolymph and convert it into ecdysone, which is considered a prohormone and is secreted into the haemolymph as it is produced, which is soon converted into the active form i.e. 20-hydroxyecdysone (ecdysone) by 20-monooxygenase enzyme by adding a hydroxyl group at c-20 position. Several other ecdysteroids i.e. 26-hydroxyecdysone and 20, 26-dihydroxyecdysone with hormonal activity are known from different insects. Exogenous application of moulting hormone analogues (MHAs) or ecdysone agonists result in hormonal imbalance since these can not be metabolized or excreted rapidly and consequently precocious metamorphosis is ensued. Rohm and Haas Company in 1983 serendipitously discovered the first bisacylhydrazine ecdysteroid against. Soon after, a simple and more potent MH analogue, RH-5849 was synthesized that target many lepidopteran, coleopteran and dipteran insect pests (Aller & Ramsay, 1988. It indicated high effectiveness on *Leptinocarsa decemlineata* (Say), *Mamestra brassicae* (L.), *Pieris brassicae* (L.) and *O. fasciatus*. RH-5849 also induced premature initiation of moulting at all stages of larval development of the tobacco hornworm, *M. sexta* (Wing *et al.*, 1988. It was superseded by a group of some cost effective MHAs. The best known representatives of this group comprise the chemical compounds tebufenozide, methoxyfenozide and halofenozide. Both methoxyfenozide and halofenozide were developed by the joint venture of Rohm and Haas and American Cynamid Company. These are similar to tebufenozide, but differ in respect that both are plant systemic, thus also effective against soil-dwelling insects. These compounds appear to work by binding to the ecdysone receptor protein, causing a precocious and unsuccessful moulting to take place.

**Anti-Moulting Hormone Analogue (AMHAs):** The AMHAs are the compounds antagonizing the action of moulting hormones. Chemically, the AMHAs are azosterols and non-steroidal compounds which act by antagonizing the conversion of phytosterols to cholesterol, which is further required as a base for the synthesis of ecdysone in insects. As a result, the moulting hormone titer is affected and consequently, the moulting process is delayed (Walker & Svoboda, 1973). Certain plants producing AMHAs have been identified as *Ajuga decumbens*, *Ajuga remota*, *Azadirachta indica* and *Plumbago capensis*, which can be used in the near future for their synthesis in pest control programmes.

**2. CHITIN SYNTHESIS INHIBITORS (CSIS)**

The chitin synthesis inhibitors (CSIs) are chemically benzoylphenlures, which act by inhibiting the enzyme chitin synthetase required for polymerization of N-acetylglucosamine in the bio-synthesis of chitin during the process of moulting of insects. The insecticidal activity of these compounds was first discovered by Philips-Duphar Company around 1970 for the first time and now multiple numbers of commercial formulations are
available for the control of insect pests of field crops. The first chitin synthesis inhibitor introduced into the market as a novel insecticide was a benzoylphenylurea named diflubenzuron (Miyamoto et al., 1993. Diflubenzuron (DIMILIN® / ADEPT® 25 WP/MICRO®) is toxic to a wide range of pests, has a low mammalian toxicity, a low toxicity to most beneficial species, possesses a high degree of environment persistence and has been registered in India (CIBRC, 2005. It is an effective tool for controlling fungus gnats, shore flies, and foliar feeding pests (armyworms, leaf miners) on ornamental crops grown in greenhouses, shadehouses, and interiorscapes. Lufenuron (MATCH® 5 EC/PROGRAM®), another CSI has been found quite effective against several insect pests of field crops. Sublethal concentrations of lufenuron affected various morphological, biological and physiological parameters of P. xylostella (Josan & Singh, 2000. Lufenuron was found to be very effective against several leaf-rollers such as Ctenopseustis obliquana Walker, C. herana Walker, Planotortrix octo Dugdale, P. exscessana (Walker) and Epiphyas postvittana (Walker) infesting apples and kiwi fruit in New Zealand (Follas et al., 1994. Butter et al., (2003) reported a reduction in pupal weight, adult emergence and fecundity of Helicoverpa armigera on cotton treated with lufenuron. It has also been registered in India against phytophagous mites. Another CSI, Hexaflumuron (SENTRICON®) has been used a bait against subterranean termite species. Triflumuron and Teflubenzuron have been found to be promising against C. molestus on apple and pear and against soft scale C. floridensis (Pollini & Bariselli, 1993; Eisa et al., 1991. Triflumuron, when used against red rust flour beetle, T. castaneum, resulted in arrested spermatogenesis and production of abnormally large sperm bundles (Parween, 2001. Novaluron, another very potent CSI, was evaluated against S. exigua, S. littoralis Boisduval and B. tabaci on cotton under field conditions and was very effective against these pests, besides being safe for non-targets and exhibiting no cross resistance to other groups (Ishaaya et al., 2003. Teflubenzuron, another CSI, significantly reduced the population of black scale, Saissetia oleae (olivier. Teflubenzuron resulted in ovicidal action, interference with all development stages and mortality of eggs at blackhead stage in rice grain moth, Corcyra cephalonica Stainton (Chakraborti & Chateree, 2001. Teflubenzuron and Hexaflumuron were also found to reduce adult emergence in pulse beetle, C.maculates (Su et al., 2003. Buprofezin (APPLAUD®/ACCOLADE®/KNACK®/TALUS®), another chitin synthesis inhibitor, is a Thiodiazine insect growth regulator. It acts by inhibiting cuticle deposition. It also suppresses egg laying in female adults with inhibition of prostaglandin synthesis and has effects on levels of hormones associated with moulting in nymphs. APPLAUD® has been registered in over 40 countries around the world. Buprofezin was found to be potent against homopteran pests including nymphs of brown planthoppers, N. lugens, leafhoppers, Nephrotettix cincticeps Uhler, whiteflies B. tabaci, T. vaporariorum, and scale insects attacking fruit crops and certain species of Coleoptera and Acarina (Asai et al., 1985; Ellsworth & Martinez, 2001; Palumbo et al., 2001)

Pheromones Control

Pheromones:

The pheromones are chemical or a mixture of chemicals released by an organism in the environment that cause a specific reaction in a receiving organism of the same species. They secreted outside the body so called as ectohormones. It is derived from two Greek words Pherein = to carry and Horman = to excite.

They are of two types: Primer and Releaser pheromones

The primer pheromones operate through gustatory (taste) sensilla and trigger a chain of physiological changes in the body. In insects they regulate caste determination and reproduction in social insects like ants, bees, wasps and termites. They are of no practical value in pest control. The releaser pheromones operate through olfactory (small) sensilla and regulate the behavior of the insects. The pheromones of this category are of the four types: Sex pheromones, Aggregation pheromones, Alarm pheromones and Trail pheromones. The sex pheromones have successfully employed in insect pest management. The sex pheromones are generally produced by females to attract males for mating, but some male also produce sex pheromones to attract females. About 150 and 50 species of insects are known which produce females and males sex pheromones, respectively. The property of both sex pheromones is different from each other. The female sex pheromones act for longer range as compared to male sex pheromones. The female sex pheromones are species specific in nature. These pheromones are produced by glands which are ectodermal in origin so that they release their products outside the body. They found in body-head, thorax abdomen, legs or wings but location vary from insect species to species. The sex
pheromones are highly developed in lepidopteron and are frequently produced by glands at the tip of abdomen. The pheromonal communication system consists of three components: exocrine glands, medium (air or water) and pheromone receptors. The exocrine glands play important role in pheromone production, medium help in travel from point of release to the point of receptor and receptors could be olfactory (smell) or gustatory (taste) sense organs. The olfactory are majorly involved in communication which found on antennae, the maxillary and labile palps in insects. The bombykol is first female sex pheromone which is produced by female silkmoth, *Bombyx mori*. The female of pink bollworm, *Pectiophoa gossypiella* produce gossyplure. The queen substance acts as sex attractants is produced by the mandibular glands of the queen of honeybees. It also ovarian inhibitor for worker bees. As compared to female few male produce sex pheromones like queen butterfly, *Danaus berenice* and cabbage looper, *Trichoplusia* etc. The effective distance of a sex pheromone will depend on a amount of pheromone released by the female, the threshold concentration of male stimulation and weather conditions (wind speed, rainfall etc.)

**Pest Management with Pheromones:**

More than 250 pest species are registered for synthetic pheromones. The major contribution is of Lepidoptera (80%) and Coleoptera (10%). The remaining 10% is contributed by Diptera, Orthoptera and Hymenoptera collectively. These products are commonly used in various techniques of insect pest management like sampling and detection, disrupt mating as well as attract for killing insects. About 90 per cent of synthetic pheromones products are used in sampling and detection.

**Sampling and detection**

The oldest practical is application of sex pheromones as attractants in traps. Sex pheromones play major role in monitor insect activity as well as relative density under field conditions. Pheromones traps are used to get information about economic injury level and economic threshold level. This information found helpful in decision making of effective management of insect pest with judicious use of chemicals. The application of pheromone traps reduce use of pesticide and increased grower’s profit. Some successful example of pest management programme by using pheromone traps were reported in various places. In a New York five major Lepidoptera pests were managed by using pheromone traps and also reduce 50 per cents pesticide. In the Netherlands similar programme used in apple orchard found helpful in reducing two to three insecticide sprays. Pheromone traps are used in various crops to monitor insects like Codling moth (*Cydia pomonella*), red-banded leaf roller (*Argyrotaenia velutinava*) and other leaf rollers in decidues in fruits, pink bollworm and corn earworms (*Helicoverpa Zea*) in cotton; black cutworms (*Agrota ipsilon*) in corn; and California red scale (*Aonidiella auvant*) in citrus. Pheromone traps can detect insect even in low density of insect pest present in field.

**Pheromones used as attract and kill programmes**

Pheromones used as attractant and killed by one means or another. The conventional approach to attract and kill has been to use traps lined with sticky material, and others have allowed slow release formulations of small particles that emit boll pheromone and an insecticide. The sex pheromones affect half the population as only one sex is attracted or kill by this method. If a large portion of one sex is attracted and killed, there individuals are eliminated, mating success is reduced and numbers in the next generation fall. In species whose members mate more than once, lures that attract females have the greatest effect on the subsequent generation. When males are attracted and killed, a very high percentage must be removed to have an effect. For instance, if each male mates with ten females, 90 per cent of the males would need to be removed at high population densities to cause a significant reduction in the next generation. When the pheromone traps are used in attract and kill programme it referred as mass trapping. The principal used in mass trapping is same as in sampling and detection programmer. The mass trapping programmer may or may not have toxicant. The number of traps used in mass sampling is 100 traps per hectares which is only 5 to 10 traps per hectare in sampling and detection programmers. Mass trapping has been used in fruit-trees, field and forest crops, as well as stored products and household pests.
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**Mating disruption**

This approach attempts to permeate the air with sex pheromone. The insects entering the area cannot locate mate emitting natural pheromone because synthetic pheromones permeate the whole environment. This would cause a reduction of reproductive rates and achieve crop protection without the use of insecticides. The mechanism of confusion includes covering up the natural pheromone scent of females, misdirection of males and male antennal receptions fail to detect pheromone produced by female. The potential of this approach was first evaluated in 1967 with the cabbage looper, *Trichoplusia ni*. The various type of controlled-release dispensers were developed for successful pest control. The Hercon flake is one such dispenser developed by Hercon Environmental Company. The other controlled-release dispensers include ropes, microcapsules and flowables. Ropes are twist ties consisting of hollow, sealed polythene tubes that contain a synthetic pheromone and are reinforced with aluminum wire. It is commonly used against pink bollworm. Microcapsules and flowables also represent recent formulation developed and can be sprayed on foliage in liquid forms with conventional equipments. Because they can be mixed with other chemicals, such as fertilizations and insecticides their application can be very cost effective. However, multiple applications are required as compared to single rope applications. All types of dispensers have been used to permeate atmosphere for mating disruption of several insect species. Often mating disruption is most successful when population levels are low and the synthetic pheromones is applied over a broad area.

**Advantage:**

Minute quantities of pheromones are needed to attract and kill large number of insects and so they are economical. They are non-pollutant and ecologically acceptable. They are species specific, so non-targets are spared. Pheromonal method in labor saving since large numbers of insects could be brought from long distances right at the door for being destroyed. They offer an easy mean to monitor the build-up of pest populations. They are the only means to keep surveillance on foreign pest entry into a country through its ports and airports with the help of which development of new pest problems could be checked at the point of entry itself.

**Disadvantages:**

Pheromones for all insect are not yet known. Sex pheromones can attract only one sex, the other sex could still be there to do the damage. The pheromonal control methods demands knowledge and expertise which are not within reach of our farmers, only Govt. agencies can provide them. Quick results cannot be obtained from pheromones so they cannot be employed in short term control measures; they best suit in IPM.

**Conclusion**

Biotechnological technology, Insect growth regulators and pheromones can become an important component of IPM programme. The many commercial formulations of these management technologies are now available. Low mammalian toxicity, biodegradability and specific nature of these make them eco-friendly and add to the diversity spectrum of chemical control agents. The novel modes of action also make them less prone to development of cross-resistance. However, much more needs to be done so that farmers are able to adopt these management technologies on large scale to avoid insect pest damage and reduce use of chemical insecticides. All these technologies are safer to human beings and environment.

**References**


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HOST PLANT RESISTANCE

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Abstract

Due to these stresses plant shows many diseases which ultimate reduces the production of plants. But in the initial stages of plants are resistant to most of pathogens because they have the many defenses system to protect them from pathogen attacks or damages but due to the environment conditions and pathogen selection pressure produces the new races which may damages the crops.

Plants are the only sources on the earth which provide direct or indirect food for all living being like humane, animal, insects and microorganisms (fungi, bacteria, viruses etc.). These plants were converted the solar energy into the chemical energy by the process of photosynthesis and released the oxygen to the environment.

![Chemical Reaction]

$$6CO_2 + 6H_2O \xrightarrow{Sunlight} C_6H_{12}O_6 + 6O_2$$

But plants are faces challenges from their surrounding environments like the biotic (herbivores and microbes) and abiotic (temperature, nutrients deficiency etc.) stresses which effects their survival and production or productivity which ultimately affects human beings.

Plants are in a continuous evolutionary battle against a multitude of microbial and other pathogens. Pathogens usually access the plant interior either by penetrating the leaf and root surfaces directly or by entering through wounds and natural openings such as leaf stomata. During the invasion process, plant pathogens degrade the cell wall by synthesizing and liberating cell wall-degrading enzymes, then deliver pathogen effectors by specialized infection structures, and eventually interfere with the normal activities of the host.

### Systemic acquired resistance (SAR)

The systemic acquired resistance (SAR) is a "whole-plant" resistance response that occurs following an earlier localized exposure to a pathogen. It is characterized by an activation of a broad spectrum of host defense mechanisms, locally at the site of the initial pathogen attack as well as systemically, in tissues untouched by the pathogen. SAR can provide resistance against widely diverse organisms such as fungi, bacteria, and viruses. Plants use pattern-recognition receptors to recognize conserved microbial signatures. SAR requires the signal molecule salicylic acid (SA) and is associated with accumulation of pathogenesis related proteins which are thought to contribute to resistance. This recognition triggers an immune response. Once pathogens penetrate the cell wall, the plant two-tiered innate immune system is activated to counter-attack pathogen invasion.

1. **PAMP-triggered immunity (PTI)**, which is based on the sensitive perception of pathogen- or microbe-associated molecular patterns (PAMPs or MAMPs),
2. **Effector-triggered immunity (ETI)**, which perceives effectors produced by pathogens that have evaded PTI
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The first plant receptors of conserved microbial signatures were identified in rice (XA21, 1995) and in Arabidopsis (FLS2, 2000).

Mechanisms Underlying Plant Resistance to Pathogens

The upper part of the diagram is the defense response to necrotrophic pathogens, conferred by RLKs, defensin, phytoalexin, and JA/ET signaling. The lower part of the diagram is the two-tiered immune system of plant resistance to biotrophic pathogens. The first tier of defense (PTI) is triggered on perception of P/DAMPs by membrane-anchored PRRs, followed by activation of MAPK cascade and downstream transcription factors, leading to immune responses. The second tier of defense is elicited by pathogen effectors via an interaction with R protein (ETI), in which the interaction between R protein and pathogen effectors oscillates between compatible and incompatible reactions over time. The R gene-mediated resistance to biotrophic pathogens usually results in hypersensitive response (HR), and meanwhile activates SA-dependent signaling, leading to systemic acquired resistance (SAR). Abbreviations: HSTs, host-selective toxins; PRRs, pattern recognition receptors; TTSS, type III secretion system; ROS, reactive oxygen species; HR, hypersensitive response; P/DAMPs, pathogen/damage-associated molecular patterns.

Biotrophic and necrotrophic pathogens

Plant pathogens are broadly divided into biotrophs and necrotrophs, according to their lifestyles. Biotrophic (Downey mildew and Powdery mildew, Rust) pathogens gain nutrients from living host tissue, whereas necrotrophic (Botrytis spp and alternaria spp.) pathogens kill host tissue and feed on the remains. There are, however, many hemi-biotrophic pathogens that behave as both biotrophs and necrotrophs, depending on the conditions or the stages of their life cycles. Many fungi that are commonly considered necrotrophs but have a biotrophic stage early in their infection process, and may thus be better described as hemibiotrophs (Pieterse et al., 2009). Generally it was assumed that no gene-for-gene resistance functions in resistance to necrotrophic pathogens, but it was indicated in a number of reports that several plant pattern recognition receptors (PRRs) are involved in the perception of necrotrophic fungi, such as receptor-like protein kinases (RLKs) (Berrocal-Lobo and Molina, 2008). One of the putative RLKs is encoded by BIK1 (Botrytis-induced kinase 1) gene, which is predicted to be involved in early stages of plant defense against B. cinerea and A. brassicicola (Veronese et al., 2006). Host-selective toxins (HSTs) are considered as efficient weaponry of necrotrophic fungi and the diseases caused by necrotrophs are manifested by the appearance of necrotic lesions.

Plant resistance to biotrophs

Previous studies suggest that plant defense against biotrophic pathogens is largely due to gene-to-gene resistance (Glazebrook, 2005. R gene-mediated resistance usually results in hypersensitive response (HR), which is thought to be very important for plants to combat biotrophic pathogens, such as Peronospora parasitica, Pseudomonas syringae, and Erysiphe spp., by restricting their access to water and nutrients (Glazebrook et al., 1997; Aarts et al., 1998; Feys et al., 2001. R gene-mediated resistance also activates SA-dependent signaling, leading to an activation of a string of presumed defense effector genes. This activation of SA signaling occurs throughout the plant to develop systemic acquired resistance (SAR) against subsequent pathogen infections (Glazebrook, 2005. During SAR, deposition of callose and lignin occurs in the plant cell
walls, and plants acquire the ability to mount a rapid HR. Analysis of Arabidopsis mutants with defects in various defense-related signaling pathways provides support for the idea that SA signaling can result in resistance to biotrophic pathogens. For instance, both EDS1 and PAD4 play important roles in SA signaling, and mutations in these two loci weaken resistance to some *P. parasitica* isolates. The NPR1 (Nonexpressor of pathogenesis-related genes 1) gene is a master regulator for SA signaling. The npr1 mutant is more susceptible to a variety of pathogens (Cao et al., 1994; Bi et al., 2011.

**Hypersensitive Response (HR)**

The HR is a localized induced cell defense in the host plants at the site of infection by a pathogen. It is responsible for limiting the growth of the pathogen by this way it provides resistant to host against pathogen. Hypersensitive response involve only single cell or very few cells The HR occurs only specific host - pathogen combination and its combination are incompatible. HR leads to necrosis of infection points of plants it collapse and leads to large necrotic area of infection.

Hypersensitive response leads to

- ROS produced e.g. \( \text{O}_2^- \), \( \text{H}_2\text{O}_2 \) etc.
- Membrane disruption.
- Activation of ion exchange and hormones activation.
- Accumulation of phenolics and salicylic acid.
- Programmed cell death (PCD).

**Phytoalexins**

It is a low molecular weight antibiotic like and produces by many plants in response to infections. They serves as antibiotic compound but not antibiotic. These are biotic elicitors or 350 phytoalexins are known for 100 plant species. They are produce in any parts of plants and usually appeared surrounding the infected cells. These are viz Pterocarps, Sesquiterpen, Cryptophenoles, Isocoumarins besides Isoflaveloides are major chemicals.

**Signal transduction in SAR**

A primary local infection can triggered not only effector-triggered immunity (ETI) which is often associated with PCD of the infected cells, but also production of the immune signal salicylic acid in the chloroplasts through the activity of isochorismate synthase. Accumulation of SA effects the cellular redox and NPR1 nuclear translocation through S- nitroglutathione and thioredoxins. The nuclear NPR1 concentration is controlled by SAR levels through the receptor proteins NPR3 and NPR4. A high concentration of SA in the local infection sites promotes NPR1-NPR3 interactions and NPR1 degradation to allow PCD, whereas in the neighboring cells, the intermediate level of SA disrupts NPR1-NPR4 interaction, resulting in the accumulation of NPR1. NPR1 cat interact with transcription factors (TFs) to activate the expression of ER genes to facilitate protein secretion; the expression of antimicrobial PR-proteins such as PR1, PR2, and PR5; and resistant to secondary infection.

An avirulence pathogen not only triggers defense responses locally but also induce the production of signals such as salicylic acid (SA), methyl salicylic acid (MeSA), azelaic acid (AzA) etc. These signals then leads to systemic expression of the antimicrobial PR (pathogenesis related) genes in the uninoculated distal tissues to protect the rest of plant from secondary infection. This phenomenon is called systemic acquired resistance (SAR). SAR can also induce by exogenous application of the defense hormones SA. SAR provides broad spectrum resistance against pathogenic fungi, virus and bacteria etc.
Phenylalanine ammonia lyase (PAL)

It is an enzyme that catalyzes a reaction converting L-phenylalanine to ammonia and trans-cinnamic acid. Phenylalanine ammonia lyase (PAL) is the first step in the phenyl propanoid pathway and is therefore involved in the biosynthesis of the polyphenol compounds such as flavonoids, phenylpropanoids, and lignin in plants is found widely in plants, as well as some yeast and fungi. The activity of PAL is induced dramatically in response to various stimuli such as tissue wounding, pathogenic attack, light, low temperatures, and hormones.

Pathogenesis related (PR) proteins

Pathogenesis-related (PR) proteins are proteins produced in plants in the event of a pathogen attack. They are induced as part of systemic acquired resistance. Infections activate genes that produce PR proteins. Some of these proteins are antimicrobial, attacking molecules in the cell wall of a bacterium or fungus. Others may function as signals that spread “news” of the infection to nearby cells.

Functions of PR-proteins

The most common function of most PRs is their antifungal effects. Some PRs also exhibit antibacterial, insecticidal or antiviral action. Function as signals that spread “news” of the infection to nearby cells. Infections also stimulate the cross-linking of molecules in the cell wall and the deposition of lignin, responses that set up a local barricade that slows spread of the pathogen to other parts of the plant. Chitinase activity also stimulates Peroxidase, ribonuclease and lysozyme activities. Their hydrolytic, proteinase-inhibitory and membrane-permeabilizing ability.

<p>| Table 1: Classification of pathogenesis related proteins |
|---------------------------------|----------------------------------|</p>
<table>
<thead>
<tr>
<th>Families</th>
<th>Type member</th>
<th>Properties</th>
</tr>
</thead>
<tbody>
<tr>
<td>PR-1</td>
<td>Tobacco PR-1a</td>
<td>Antifungal</td>
</tr>
<tr>
<td>PR-2</td>
<td>Tobacco</td>
<td>PR-2 β-1,3-glucanase</td>
</tr>
<tr>
<td>PR-3</td>
<td>Tobacco P. Q</td>
<td>Chitinase type I,II, IV, V, VI, VII</td>
</tr>
<tr>
<td>PR-4</td>
<td>Tobacco ‘R’</td>
<td>Chitinase type III</td>
</tr>
<tr>
<td>PR-5</td>
<td>Tobacco S</td>
<td>Thaumatin-like</td>
</tr>
<tr>
<td>PR-6</td>
<td>Tomato Inhibitor I</td>
<td>Proteinase-inhibitor</td>
</tr>
<tr>
<td>PR-7</td>
<td>Tomato P69</td>
<td>Endoprotease</td>
</tr>
<tr>
<td>PR-8</td>
<td>Cucumber chitinase</td>
<td>Chitinase type III</td>
</tr>
<tr>
<td>PR-9</td>
<td>Tobacco ‘lignin forming peroxidase’</td>
<td>Peroxidase</td>
</tr>
<tr>
<td>PR-10</td>
<td>Parsley ‘PR1’</td>
<td>Ribonuclease like</td>
</tr>
<tr>
<td>PR-11</td>
<td>Tobacco ‘class V’ chitinase</td>
<td>Chitinase, type I</td>
</tr>
<tr>
<td>PR-12</td>
<td>Radish Rs- AFP3</td>
<td>Defensin</td>
</tr>
<tr>
<td>PR-13</td>
<td>Arabidopsis THI2.1</td>
<td>Thionin</td>
</tr>
<tr>
<td>PR-14</td>
<td>Barley LTP4</td>
<td>Lipid-transfer protein</td>
</tr>
<tr>
<td>PR-15</td>
<td>Barley OxOa (germin)</td>
<td>Oxalate oxidase</td>
</tr>
<tr>
<td>PR-16</td>
<td>Barley OxOLP</td>
<td>Oxalate oxidase-like</td>
</tr>
<tr>
<td>PR-17</td>
<td>Tobacco PRp27</td>
<td>Unknown</td>
</tr>
</tbody>
</table>

References


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HEAT UNITS AND CROP MANAGEMENT

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Abstract

Growth and development of are strongly dependent on temperature. For an example, Corn develops faster when temperatures are warmer and more slowly when temperatures are cooler. A string of warmer than normal days in late spring will encourage faster leaf development than normal. Temperature controls the developmental rate of many organisms. Rates of most biological processes are affected markedly by temperature. Growth and development of whole organisms show a temperature response which results from the integrated effect of temperature on the many individual physiological processes involved. Plants require a certain amount of heat to develop from one point in their life cycles to another. This measure of accumulated heat is known as physiological time. The amount of heat required to complete a given organism’s development does not vary; the combination of temperature (between thresholds) and time will always be the same. Physiological time is often expressed and approximated on hourly or daily time scales using units of degree-hour (°hr) or degree-day (°D. Despite general acceptance of this close relationship, the use of temperature indices has not been generally extended to growth analysis. Growth analysis has been a valuable tool in the quantitative analysis of plant and crop growth. The growth functions; AGR, RGR and NAR, increase with temperature and light flux within a range specific for a given crop. Growth functions calculated in the traditional manner will necessarily include the effect of controlled and uncontrolled environmental variables.

Growth analysis has been a valuable tool in the quantitative analysis of plant and crop growth since the suggestion by Blackman in 1919 that growth generally follows the compound interest law. He used absolute growth rate (AGR), relative growth rate (RGR), leaf area ratio (LAR), net assimilation rate (NAR), and other similar functions to describe plant growth. Growth analysis can be approached on an individual plant or areal basis. The growth functions; AGR, RGR and NAR, increase with temperature and light flux within a range specific for a given crop. Growth functions calculated in the traditional manner will necessarily include the effect of controlled and uncontrolled environmental variables.

The purpose of calculating growth functions is generally to describe or explain how one or more plant species respond to a given environmental situation. In many experiments, environmental conditions will vary considerably among years and will vary within any one year for different treatments, such as planting date or location. These environmental variables confound comparisons of growth functions for crops having the same treatment regime over two or more years or for crops having different treatments in the same season. Calculations based on time may be appropriate for an experiment as long as it is recognized that environmental conditions are confounded with species and treatment. However, in experiments designed to make comparisons of physiological response, growth functions ideally should be independent of environmental variables. Comparisons of growth functions within and among different experiments would be less ambiguous if sources of variation other than imposed treatments could be eliminated. Hence it is suggested that growth function be calculated using a temperature index as divisor, rather than using time.

Plant growth and development are certainly affected by factors other than temperature, such as flux and duration of photosynthetically active radiation, availability of nutrients and water, and loss of photosynthetic tissue. Day length plays a well-known, integral part in induction and initiation of flowering in many species. However, even with maize (Zea mays L.) grown under field conditions, for example, temperature indices alone can often explain over 95% of the variability in development. Estimates of leaf stage development are only that, just estimates. One of the factors that most influences the accuracy of these estimates is the existence or not of other growth-limiting stresses. Stressed plant will generally retard stem and leaf sheath elongation, thus delaying the appearance of leaf collars. In the absence of extreme conditions such as unseasonal drought or disease, plants grow in a cumulative stepwise manner which is strongly influenced by the ambient temperature. Growing degree days take aspects of local weather into account and predict the plants’ pace toward maturity. Unless stressed by other environmental factors like moisture, the development rate from emergence to maturity for many plants depends upon the daily air temperature. Because many developmental events of plants and insects depend on the accumulation of specific quantities of heat, it is possible to predict when these events should occur during a growing season regardless of differences in temperatures from year to year.

Concept

Heat units are a method of quantifying a biological organism’s thermal environment. Research conducted over the past several decades has proven that proper use of heat units can provide a reliable means of predicting the growth and development of important crop species as well as many crop pests.
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Commonly used terms
Growing Degrees (GDs): is defined as the number of temperature degrees above a certain threshold base temperature, which varies among crop species. GDs are calculated each day as maximum temperature plus the minimum temperature divided by 2 (or the mean temperature), minus the base temperature.
Growing Degrees Days (GDDs): It is the amount of heat energy an organism accumulates over a period of time.
Base Temperature (\( T_{\text{BASE}} \)): The base temperature is that temperature below which plant growth is zero.
Growing Degree Units (GDUs): are accumulated by adding each day’s GDs contribution as the season progresses.
Crop Heat Units (CHUs): Crop specific indices that employ separate equations for the influence of the daily minimum (nighttime) and the maximum (daytime) temperatures on growth are called crop heat units.
Baselines: 10 °C is the most common base for GDD calculations; however, the optimal base is often determined experimentally based on the lifecycle of the plant or insect in question.
For example;
- 5.5°C: wheat, barley, rye, oats, flaxseed, lettuce, asparagus
- 6°C: Stalk Borer
- 7°C: Corn rootworm
- 8°C: Sunflower, potato
- 9°C: Alfalfa weevil
- 10°C: Maize, sorghum, rice, soybeans, tomato, Black cutworm, European corn borer, Coffee, insect and mite pests of woody plants
- 11°C: Green Clover worm
- 12°C: many other crop calculations
- 30°C: In GDD above 30 °C; for many plants this is significant for seed maturation, e.g. reed (Phragmites) requires at least some days reaching this temperature to mature viable seeds.

Heat Units (HUs)?
A proper thermal environment is essential to the survival of all biological organisms. The thermal environment is especially critical to “cold-blooded” organisms such as plants and insects because their internal temperatures are dictated by the surrounding environment. If the outer temperature exceeds some upper limit or decline below some lower limit, growth and development of “cool-blooded” organism are impaired or halted (Fig 1). However, when these organisms are exposed to temperature within some optimal range, growth and development typically increase with temperature. Heat unit is a daily estimate of the amount of contributory heat-heat that will contribute to growth and development.
The heat unit concept is best understood with the use of diagrams (Fig 2). Fig 2A shows a typical daily temperature cycle for Arizona. The heat unit concept utilizes this daily temperature information along with knowledge about a plant’s or pest’s thermal limits to quantify the “contributory” heat. Fig 2B shows the temperature cycle with two horizontal lines representing the upper and lower thermal limits of cotton (and pink bollworm) superimposed on the diagram. As previously mentioned, temperatures below the lower limit are too cold for proper growth and development, while temperature above the upper limit do not contribute to additional growth and development. The hatched area in fig 2C, bounded by daily temperature curve and the lines designating the upper and lower temperature limits, represents the daily amount of “contributory” heat. This area can be calculated with the help of mathematics to give a number that is the daily accumulation of heat units or growing degree days. The accumulation of heat units is very dependent on daily temperature conditions.
Growing Degree Days (GDDs)?
Growing Degree Days are used to estimate the growth and development of plants and insects during the growing season. Insect and plant development are very dependent on temperature and the daily accumulation of heat. The amount of heat required to move a plant or pest to the next development stage remains constant from year to year. However, the actual amount of time (days) can vary considerably from year to year because of weather conditions. Each organism has a minimum base temperature or threshold below which development does not occur. These base temperatures have been determined experimentally and are different for each organism. GDD information can be very useful for predicting crop and insect development. Growing degree days are sometimes referred to as “degree days” or the “degree days averaging method.” Some jurisdictions also use the term “heat units” interchangeably with “degree days.”
The basic concept is that development will only occur if the temperature exceeds some minimum development threshold, or base temperature ($T_{\text{BASE}}$). The base temperatures are determined experimentally and are different for each organism.

Procedure to calculate GDDs:
First find the mean temperature for the day. The mean temperature is found by adding together the high and low temperature for the day and dividing by two.
If the mean temperature is at or below $T_{\text{BASE}}$, then the Growing Degree Day value is zero.
If the mean temperature is above $T_{\text{BASE}}$, then the Growing Degree Day amount equals the mean temperature minus $T_{\text{BASE}}$. For example, if the mean temperature was 75° F, then the GDD amount equals 10 for a $T_{\text{BASE}}$ of 65° F.

In equation form:
$$\text{GDD} = T_{\text{MEAN}} - T_{\text{BASE}} \text{ if } T_{\text{MEAN}} \text{ is greater than } T_{\text{BASE}}$$
$$\text{GDD} = 0, \text{ if } T_{\text{MEAN}} \text{ is less than } T_{\text{BASE}}$$

Where:
$T_{\text{BASE}} = \text{Growing Degree Day base temperature}$
$T_{\text{MEAN}} = \text{mean temperature, } (T_{\text{MAX}} + T_{\text{MIN}}) / 2$

There are four factors to consider when comparing GDD accumulations from various sources or regions:
1. Are the base temperatures used in the equations the same?
Different organisms have different base temperatures used to calculate GDDs: 150 GDD at base 10 does not equal 150 GDD at base 0.
2. Are the start dates for the accumulations the same?
Generally, GDD accumulations start on April 1 each year, but some insect GDD models start at the emergence of a specific life stage. This is referred to as a biofix.
3. Are the equations used to calculate the daily GDD the same?
Many modifications to the simple GDD calculation have been developed over the years and may be referred to generally as degree days.
4. Are the temperatures used in degrees Celsius or Fahrenheit?
GDD accumulations will vary significantly, depending on whether they are being tracked in Celsius or Fahrenheit. GDD models have been designed specifically for use in one or the other and cannot be interchanged without making conversions. The ECB GDD model was based on measurements in Celsius. However, the terms "growing degree days" (GDD) and "crop heat units" (CHUs) are used independently since they represent two very different, temperature dependent, development models.

Modified Growing Degree Days?
Modified growing degree days are similar to GDD with several temperature adjustments. If the high temperature is above 86° F, it is reset to 86° F. If the low is below 50° F, it is reset to 50° F. Once the high / low temperatures have been modified (if needed), the average temperature for the day is computed and compared with a base temperature, which is usually 50° F. Modified Growing Degree Days are typically used to monitor the development of corn, the assumption being that development is limited once the temperature exceeds 86° F or falls below 50° F. For example, if the high for the day was 92° F and the low 68° F, the average for use in the modified GDD calculation would be $86 + 68 = 154 / 2 = 77$, for a value of 27 Modified GDD with a base of 50°F.

How are Crop Heat Units determined?
Different methods exist for calculating heat units depending on; a) the crop or biological organism of interest and b) the whim or personal preference of the researcher. Temperature data must be available to determine heat unit accumulations. Typically, daily maximum and minimum temperature are used to reconstruct the daily temperature cycle since detailed temperature data (e.g. hourly) are rarely available. Reconstructing a daily temperature cycle involves forcing a “sine curve” (Fig. 2) through the day’s maximum and minimum temperatures. The sine curve closely approximates the daily temperature cycle on clear days. With the temperature cycle reconstructed, the next step in calculating heat units is to select the upper and lower temperature limits. These limits, or thresholds, differ between various crop and insect species, and individuals calculating their own heat units must take care to choose the thresholds appropriate for their crop or pest in question. Finally, the daily heat unit total is obtained by determining the area under the daily temperature curve that resides within the temperature thresholds. A computer and appropriate software, or a set of heat units tables...
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(Table 1) are typically employed to make this area determination. Daily crop heat units are calculated by using the average of the two daily values from the equations below.

Below equation is used to calculate a daily CHU for a site:

\[
\text{Daily CHU} = \frac{Y_{\text{MAX}} + Y_{\text{MIN}}}{2}
\]

Where,

\[
Y_{\text{MAX}} = \{3.33 \times (T_{\text{MAX}}-10)\} \quad \{0.084 \times (T_{\text{MAX}}-10.0)^2\} \quad \text{if value are negative, set to 0}
\]

Heat unit efficiency (HUE) or thermal time use efficiency (TTUE) - amount of dry matter produced per unit of thermal time or growing degree day. It is expressed in g/m² °C.

\[
\text{HUE} = \frac{\text{Dry matter production}}{\text{Growing degree day base temperature of crop}}
\]

Or can be read from the matrix in Table 1. “Daily Crop Heat Unit Accumulations Based on Maximum and Minimum Temperatures”. Producers who record high and low temperatures can use Table, to calculate CHUs for their own farm. Many crops especially Corn development is driven primarily by temperature and this is especially true during the planting-to-silking period. Unlike soybeans, day length has little effect on the rate at which corn develops. The Crop Heat Unit System has been developed to calculate the impact of temperature on crop development.

The calculation method most commonly used throughout the U.S. for determining heat unit accumulation relative to corn phenology was first evaluated by Gilmore & Rogers (1958) and termed "Effective Degrees". Barger (1969) later proposed that the same method, which he termed "Modified Growing Degree Days", be adopted as the standard heat unit formula by the National Oceanic and Atmospheric Administration. This method calculates daily accumulation of GDDs as the average daily temperature (degree F) minus 50. In late April to early May, normal daily GDD accumulations for central Indiana are about 10 GDDs. By late July, the normal daily accumulation rises to about 23 GDDs. For a typical corn growing season in central Indiana, say from late April to late September, the total seasonal accumulation of GDDs is about 2800 GDDs.

The USDA-funded Useful to Usable (U2U) multi-state research and Extension project developed a GDD decision support tool that is now hosted by the Midwestern Regional Climate Center at http://mrcc.isws.illinois.edu/U2U/gdd/. The “GDDTool” estimates county-level GDD accumulations and corn development dates based on current and historical GDD data plus user selected start dates, relative hybrid maturity ratings, and freeze temperature threshold values. The GDD and corn development predictions are displayed graphically and in tabular form, plus the GDD accumulation estimates can be downloaded to work in farmers’ own spreadsheet programme.

Application

The heat units or thermal time are involved in the physiological process of plants. The specific amount of heat units required for the plant at each stage from its germination to harvest of the crop viz., growth and development, growth parameters, nitrification, biomass, physiological maturity, yield etc. would vary. Following are the application of heat units or growing degree units in crop management:

- Heat units used as a guide in planting schedules for an orderly harvest of canning crops.
- As an index in making crop zonation maps for extension of undeveloped agricultural lands and multiple cropping system for effective land use.
- It has been adopted to flowering time of parent variety in cross pollination crops for synchronizing the flowering time for breeding and seed production.
- GDUs can be used to assess the suitability of a region for production of a particular crop.
- By heat unit can estimate the growth-stages of crops, weeds or even life stages of insects. So can be used in pest management. For example, both the development of cotton and annual spring emergence cycle of pink bollworm can be predicted using heat units. By tracking late winter and early spring heat unit accumulation, grower can select planting dates to avoid or minimize infestation.
- Predict maturity and cutting dates of forage crops.
- Predict best timing of fertilizer or pesticide application especially where farm size is so large to be difficult to visit regularly.
- Estimate the heat stress on crops.
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- Plan spacing of planting dates to produce separate harvest dates.
- Assess the suitability of a region for production of a particular crop
- Tool for managing growth regulators or harvest aids
- Ideal unit in crop weather model.

References
INTINTEGRATED MANAGEMENT OF CHILLI DAMPING-OFF CAUSED BY PYTHIUM SPECIES

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Abstract

Chilli is one of the most important vegetable cum spice crop grown in India. This crop is being subjected to be infected by various fungal diseases viz., damping-off, leaf spot, anthracnose or fruit rot and wilt. Of these, damping-off incited by *Pythium aphanidermatum* (Edson) Fitzp. is an important nursery disease in major chilli growing tracts of Tamil Nadu and causing 60% mortality of the seedlings both in field and green house grown plants. Conventionally, this disease can be controlled by using systemic fungicides. But incriminate use of fungicides leads to accumulation of toxicity in the soil are major problems. Development of fungicide resistance by *Pythium* spp. further discourages its use for disease control. Other control measures like host resistance has not yet become a viable measure. Till today, no resistant variety has been developed for the control of soil-borne *Pythium* species. Accordingly, the researchers switch over for alternate strategies for controlling damping-off disease. One of the key elements of such sustainable agriculture is the application of biological controlling strategies (use of plant extracts, plant oils, antagonistic micro-organisms-*Trichoderma, Pseudomonas* and *Bacillus*) for plant protection. Integration of all these bio-agents applied through either seed treatment or soil application can be effective and alternative way for the control of *Pythium* species.

Chilli (*Capsicum annuum* L.) is an Universal spice of India. It belongs to the genus *Capsicum* and the family solanaceae which is popularly known as “Red Pepper”. It has its origin at New Mexico with secondary origin of Guatemala and later it was introduced into Spain during 1943. Subsequently chilli was cultivated in other parts of European countries. The same crop was introduced to India by Portuguese during the early 16th century. In India, chilli occupies an area of 844 thousand hectares with an annual production of 2106 thousand metric tons (Anonymous, 2017. Other chilli producing countries include China, Mexico, Turkey, Indonesia, Spain and United States. Indian chilli is mainly exported to Asian countries like Vietnam, Thailand, Sri Lanka, Bangladesh and U.A.E. In India, major chilli producing states are Andhra Pradesh, Telangana, Tamil Nadu, Karnataka and Madhya Pradesh.

The major chilli growing areas in Tamil Nadu are Ramanathapuram, Virudhunagar, Sivagangai, Salem, Tiruchirapalli, Cuddalore, Madurai and Dindigul. Chilli fruit is used fresh, cooked, pickled and canned in sauces and as powder for hot spices (Parey et al., 2013. Nadkarni (1927) has reported many medicinal value of chilli. Its paste is externally used as rubefacient and as local stimulant for the tonsils in tonsillitis. It is irritant internally and produces gastroenteritis. It is used with many ingredients for local remedies. Though chilli plays a vital role in increasing the National economy, still the productivity and foreign exchange realized through chilli can be increased by the management of various diseases caused by pathogens of fungal, bacterial and viral origin. Among the fungal diseases, damping-off incited by *Pythium* cause more than 60 per cent mortality of seedlings both in nursery and main field (Jadhav and Ambadkar, 2007.

Etiology

General account of the disease

Damping-off disease in nursery seedlings of chillies, tomato, brinjal, papaya, turmeric, tobacco, cucumber, sugar beet, ginger and several other plants caused by species of *Pythium, Phytophthora* and *Rhizoctonia* is known for the past about 100 years. The diseases caused by *P. aphanidermatum* varies with the host plant, it is the causal agent of pre and post emergence damping-off of various seedlings. It also causes seedling rots, root rot, cottony-leak, cottony blight, stalk rot etc.

Causal organism

The *Pythium* species are fungal-like organisms, commonly referred to as water molds, (kingdom Straminopila; phylum Oomycota; class Oomycetes; subclass Peronosporomycetidae; order Pythiales and family Pythiaceae)
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are worldwide in distribution and associated with a wide variety of habitats ranging from terrestrial or aquatic environments, in cultivated or fallow soils, in plants or animals, in saline or fresh water. The genus *Pythium* is one of the largest Oomycetes genus and consists of more than 130 recognized species which are isolated from different regions of the world (Dick, 1990; de Cock and Lévesque, 2004; Paul et al., 2006. *Pythium aphanidermatum* (Edson) Fitzp. is one of the most aggressive species in the genus and has a wide host range causes many economically important diseases and is one of the most pathogenic species in the genus. *P. aphanidermatum* is cosmopolitan in distribution and one of the most common plant pathogenic of a number of other crop plants in warmer parts of the world. *P. aphanidermatum* is also known to cause infection on a wide range of plant species, belonging to different families viz., Amaranthaceae, Amaryllidaceae, Araceae, Basellaceae, Bromeliaceae, Cactaceae, Chenopodiaceae, Compositae, Coniferae, Convolvulaceae, Cruciferae, Cucurbitaceae, Euphorbiaceae, Gramineae, Leguminosae, Linaceae, Malvaceae, Moraceae, Passifloraceae, Rosaceae, Solanaceae, Umbelliferae, Violaceae, Vitaceae, Zingiberaceae (Waterhouse and Waterston, 1964.

**Symptomatology**

Damping-off symptoms occurs in two phases, i.e. the pre-emergence and the post-emergence phase. In the pre-emergence phase the seedlings are killed just before they emerge out from the soil. The young radical and the plumule are killed and there is a complete rotting of the seedlings occur. The post-emergence phase is characterized by the infection of the young, juvenile tissues of the collar at the ground level. The infected tissues become soft and water soaked. The seedlings topple over or collapse (Agrios, 2005).

**Inclination**

The pathogen is soil-borne in nature and practice of continuous cultivation make the crop more vulnerable to attack by plant pathogens. The cultivars grown are not resistant to the pathogen which also helps to spread the disease. *P. aphanidermatum* is an aggressive plant pathogen at high temperature (Gold and Stanghellini, 1985. The disease is highly dependent on a favourable environment and appears only when conditions of moisture, temperature and soil reaction are suitable (Chester, 1948.

**Perpetuation**

*Pythium* species are most commonly identified as causal agents of pre and post-emergence damping-off, leading to poor stands and low crop vigor (Agrios, 2005. Some *Pythium* species can infect mature plants, causing significant damage to yields, such as the species that cause carrot cavity spot (Suffert and Guibert, 2007. *Pythium* species can survive well in the form of oospores and sporangia for long time period in the soil (Hendrix and Campbell, 1973; Plaats-Niterink, 1981. The oospore, a sexual spore or diploid spore or resting spore is the primary survival structure of many *Pythium* species because of the thick-wall enables the spore to survive in the absence of a host, even in the presence of unfavorable soil conditions (Dick, 1990. The sporangia and hyphal swellings of *Pythium*, on the other hand, serve as the asexual reproductive structures for many of species (Hendrix and Campbell, 1973; Plaats- Niterink, 1981.

**Biological management**

**Cultural methods**

Cultural management practices are the primary strategies used to manage disease problems (Sullivan, 2004. Selection of site such as choosing land without history of any soil- borne diseases, without undulation, and planting in fields with well-drained and aerated soil, is important for preventing damping-off (Koike et al., 2000. Abawi and Widmer (2000) reviewed the impact of some cultural practices on soil health management practices in controlling soil-borne pathogens and concluded that the cultural practices such as cover cropping and green manure incorporation, use of composts, crop rotation, and tillage practices improve soil compaction, increase drainage, and increase soil temperature, all of which have an impact on the physical soil characteristics as well as increasing diversity of the soil biota and ultimately reducing the pathogen inoculum. The use of resistant cultivars can be the most important weapon for damping-off disease control. However desirable cultivars with resistance to important soil- borne diseases are not always available (Koike et al., 2000.

**Physical method**

Soil solarization has been used for managing diseases caused by *Pythium* species. Soil solarization is a hydrothermal process that occurs in moist soil when the soil is covered by plastic film and heated by exposure to sunlight during the warm months. This process can changes soil physical, chemical, and biological properties and thereby helps to improve soil health. Efficacy of soil solarization against *Pythium* species has been reported by Katan (2000) reported the efficacy of soil solarization in reducing the disease caused by *Pythium*. Usman et al. (1996) reported the significance of soil solarization against *Pythium* spp. Besides the physical method of soil
solarization, organic materials were also used against disease caused by *Pythium* by many workers. Kao and Ko (1986); Paulitz and Baker (1987); Shuler et al. (1989); Matsuzaki et al. (1998); Boehm and Hoitink (1992); Theodore and Toribio (1995); Craft and Nelson (1996); Boehm and Hoitink (1992); Ringer et al. (1997) Erhart and Burian (1997) reported that there are many factors present in organic material which affects the suppression of disease, these factors include the quality and quantity and associated levels of microbial activity. However, solarizing soils along with suitable organic materials gives good result as compared to individual treatment of either soil solarization or use of organic materials. It has been reported that organic materials when added in the soil then it actuate a chain reaction of chemical and microbial degradation, which enhance toxicity against soil-borne plant pathogens. The addition of organic manure probably contributed to the higher nutrient contents and increases the yield (Gamhiel et al., 2000. Satya et al. (2005) reported that composted chicken manure alone at 5381 kg/ha reduced populations of *Pythium* sp. significantly and when combined with heat (42°C), eradicated the *Pythium* population from the soil.

### Chemical management

According to Haware and Kannaiyan (1992) the management of seed borne and soil-borne diseases has always been problematic. Satija and Hooda (1987) evaluated fungicides belonging to different chemical groups for their efficacy in controlling damping-off tomato and chilli caused by *Fusarium solani* and *P. aphanidermatum*. As seed treatment, copper oxychloride was found to be best fungicide in controlling damping-off of the both the tests plants caused by *F. solani* whereas for the control of damping-off due to *P. aphanidermatum* MEMC (methoxy ethyl mercuric chloride) was found the best fungicide. However, for controlling damping-off caused by both the fungi MEMC and captan were very promising on tomato and captafol on chill. Hickman and Michailides (1998) reported that fallow field soil, solarized soil, methyl bromide treated soil and metalaxyl soil drench treatments were also effective against damping off caused by *P. aphanidermatum* in cucumber. Although fungicides have shown promising results in controlling the damping-off disease, phytotoxicity and fungicide residues are major problems leading to environmental pollution and human health hazards. For this reason, a number of restrictions are imposed in the licensing, registering and using of each chemical (Cuthbertson et al., 2010. Hence use of environmental friendly bio-control agents can more effectively control the soil-borne plant pathogens (Nam et al., 1988 and Saleem et al., 2000.

The systemic fungicides were evaluated at 1000 and 1500 ppm. The non systemic and combo fungicides were evaluated at and at 2000 and 2500 ppm concentration. Among all the tried chemical fungicides, Mancozeb (64%) +Metalaxyl (8%), Propiconazole and Tubaconazole were found most effective and inhibited 100% radial growth of *P. aphanidermatum* (Ratan et al., 2017. Chavan et al. (2017) reported that all the fungicides tested significantly inhibited mycelial growth of *P. aphanidermatum* over untreated control. Average mycelial growth inhibition recorded with the test systemic fungicides was ranged from 73.32 (Propiconazole) to 100 (Metalaxyl) per cent. However, it was 100 per cent with Metalaxyl (100 %), followed by arbidazim (97.67 %), Azoxystrobilin (94.55 %), Thiophanate methyl (94.15 %), Fosetyl-AL (86.64 %), Hexaconazole (85.76 %) and Difenconazole (82.85. Recently, Apet et al. (2018) recorded that Thiophenate methyl and Copper oxychloride were found to be most fungistatic and recorded significantly highest mean mycelia inhibition (93.28%) of *P. aphanidermatum* in ginger.

### Biological management

Sanford and Broadfoot (1931) were the pioneers to introduce the term biological control in Plant Pathology and conducted the first experiment on biological control of plant pathogens with antagonists in Canada. Subsequently, the importance of biological control was realized and attempted by several workers in various crops. In the light of the present day environment and economic constraints encountered with the use of fungicides, biological control by using antagonistic microorganisms is gaining importance as a practical strategy for the management of several soil-borne pathogens (Cook and Baker, 1983. The control of plant pathogens with microorganisms was carried out during the early part of the twentieth century. Members of the genus *Pseudomonas* and *Trichoderma* have long been known for their potential to reduce the plant diseases caused by fungal pathogens and they have gained considerable importance as potential antagonistic soil microorganisms (Papavizas, 1985; Pant and Mukhopadhyay, 2001. *Trichoderma* and *Pseudomonas* are the two mostly used bio-control agents as they have antifulgal, plant growth promoting and plant defense inducing activities (Zaidi et al., 2004.

### Fungal antagonist

Growth inhibition of the plant pathogens by the *Trichoderma* metabolites has been well researched (Dennis and Webster, 1971. Successful management of damping-off caused by *Pythium* species in various crops by application of *Trichoderma* has been previously reported (Jayaraj et al., 2006. Gomathi et al. (2011) evaluated the antibiotic ability of local isolates of *Trichoderma spp.* against damping off disease. Subash et al. (2013) evaluated *T. harzianum* against damping-off disease caused by *Fusarium oxysporium*, *Rhizoctonia solani* and *Alternaria alternata* and found *T. harzianum* efficient not only to inhibit the growth of all the above pathogens
but also to enhance the growth of the plants. One of the mechanisms observed to be adopted by *Trichoderma* species to parasitize pathogenic organisms is by competition. *Trichoderma* suppress the growth of these phytopathogenic fungus through the over growth. The *T. harzianum* was found effective in inhibition of mycelium (80.03 %) against *P. debaryanum in vitro* in chilli (Rajendraprasad et al., 2017). Recently, Majeed et al. (2018) reported that *T. viride* was found most effective with highest mycelial growth inhibition (85.81 %) of *P. aphanidermatum* (Majeed et al. 2018).

**Bacterial antagonist**

The phylloplane bacteria (*P. fluorescens*) isolated from the rambutan leaf surface can be effective against some soil-born pathogens, especially *P. aphanidermatum* (Yenjit et al., 2004). Kavitha et al. (2005) reported that the production of phenazine derivatives which was effective against *P. aphanidermatum* (chilli damping-off) which disorganized the hyphal morphology by inducing the formation of vacuolation in hyphal cells, degeneration of cell content followed by hyphal lysis. The bacterial antagonist *P. chlororaphis* strain PA23 and *B. subtilis* strain BSCBE4 showed the maximum inhibitory effect on the mycelial growth of *P. aphanidermatum* causing chilli damping-off (Nakkeeran et al., 2006). Muthukumar and Bhaskaran (2007) reported that 12 isolates of *P. fluorescens* were tested against the growth of *Pythium* sp. Of these, *P. fluorescens* three and four were highly effective in inhibiting the mycelial growth of *Pythium* sp. Muthukumar (2008) reported that the endophytic bacterial isolate 5.6 and 7 (isolated from stem and root) showed highest inhibition on the mycelial growth of *P. aphanidermatum* (51.4, 41.7 and 40.0 %) inciting damping-off of chilies. Muthukumar et al. (2010) reported that among the isolates, EBS (endophytic bacteria stem 20) produced largest inhibition zone and the least mycelia growth of *P. aphanidermatum*. The same isolate produced more amounts of salicylic acid, sideropore and HCN. Chili seeds treated with *P. fluorescens*, Thiram 75WS and Captan 50WP and formulations reduce damping-off incidence and enhanced yield in chilli (Saha et al., 2011). Dar et al. (2012) reported that seed treatment and soil application with t alc based formulations of *P. fluorescens* reducing the damping-off of papaya caused by *P. debaryanum* and increased plant growth parameters of papaya seedlings. Further, cucumber seeds treated with peat and t alc based formulations of these antagonistic bacteria effectively controlled the disease incidence and increased the plant growth (Khabbaz and Abbasi, 2014). Of the bacterial antagonist tested in *vitro*, *Pseudomonas fluorescens*-3 were found effective in inhibition of mycelia growth of (80.03, 58.50) % of *P. aphanidermatum* in chilli (Rajendraprasad et al., 2017).

**Plant extracts**

The antifungal effects of 66 medicinal plants belonging to 41 families were evaluated against *P. aphanidermatum*, the causal agent of chilli damping-off. Of these, Zimmu (*Allium sativum* L. *× Allium cepa* L.) leaf extract at 10 % concentration had the highest inhibitory effect (13.7 mm) against mycelial growth of *P. aphanidermatum* (Muthukumar et al., 2010). Komathi et al. (2011) reported that the methanolic extracts of various plants such as *Muraya Koenigii* (Karuveppilei), *Pithecellobium dulce* (Kudukkapuli), *Vitex negundo* (Karunocchi), *Aleo vera* (kattalai) individually tested for antifungal activity against *P. debaryanum* by agar well diffusion method. The results revealed that the methanolic extracts of *vitex negundo* showed considerably highest antifungal activity against *P. debaryanum* than other plant extracts tested in the present study. Parveen and Sharma (2014) stated that among the 20 plants tested, *Jacaranda mimosifolia*, *Moringa oliefera*, exhibited 27.7 % inhibition of mycelial growth of *P. aphanidermatum*. Whereas, *Polyalthia longifolia* and *Terminalia arjuna* showed 22.2 % inhibition of *P. aphanidermatum*. Besides these, *Lawsonia inermis*, *Aegle marmelos*, *Nigella sativa*, *Azadirachta indica*, also exhibited maximum inhibitory activity against *P. aphanidermatum*. Among the plant extract tested, the leaf extract obtained from *Lantana camara* at (20%) concentration showed maximum inhibition of mycelia growth of *P. aphanidermatum* (98.87 %) followed by *Eucalyptus globulus* (20%) with (93.61%) inhibition (Pandey et al., 2016).

**Plant oils**

Among the plant oils tested, *Eucalyptus* oil recorded the maximum mycelia growth inhibition of *P. aphanidermatum* causing chilli damping-off. This was followed by Citrodara oil and Plamarosa oil in the decreasing order of merit (Muthukumar and Sangeetha, 2008). The essential oil extracted from *T. vulgaris* was the most effective and caused complete mycelial growth inhibition of *P. aphanidermatum* and *R. solani*. The main antifungal compound present in *T. vulgaris* is thymol (Amini et al., 2012, Fonseca et al. (2015) reported that the antifungal activity of *Origanum vulgare*, *O. majorana*, *Mentha piperita* and *R. officinalis* against *P. insidiosum*. The results revealed that the essential oil obtained from *O. vulgare* showed highest efficacy on *P. insidiosum*.

**Conclusion**

The management of *Pythium* is very difficult due to its wide host range, soil-borne nature and prolonged survival of propagules in the soil. Though management of *Pythium*, the causal agent of damping-off was a difficult task, attempts were made to manage the same using different fungicides. But the indiscriminate use of fungicides resulted in the accumulation of residual toxicity, environmental pollution and altered the biological balance in the soil by over killing the non-targeted microorganisms besides developing resistance among the
pathogen. Hence, there is need to replace the chemical fungicides by bio-agents, prepared from plant extracts, plant oils and antagonistic microorganisms. Bio-agents will also be economical to the farmers and besides this the use of bio-agents will not leave any residue in the soil, water as well as in the environment. It is possible that integration of all these approaches is (use of plant extracts, plant oils and antagonistic micro-organisms) an economically viable alternative for crop production system can be developed. So the use of bio-agents proved to be economic alternative that can be implemented at the farm level. Formulation must have adequate shelf life, stability, and titer. Before any formulated product is marketed, it must be thoroughly tested by growers, whose comments, critiques, and suggestions for improvement, must consider.

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APPLICATIONS OF FUNGICIDE

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Abstract

The fungicidal application varies according to the nature of the host part diseased and nature of survival and spread of the pathogen. The object of spraying or dusting is to cover the entire susceptible surface of host with a thin covering of a suitable concentration of the fungicide before the pathogen has come into contact with the host.

Proper selection of a fungicide and its application at the correct dose and the proper time are highly essential for the management of plant diseases. The basic requirement of an application method is that it delivers the fungicide to the site where the active compound will prevent the fungus damaging the plant. This is mostly achieved by spray, fog, smoke, aerosol, mist, dust, or granules applied to the growing plant or by seed or soil treatment. In addition, some trees and shrubs can be protected by injection of fungicide liquid into the trunk or by brushing wounds with fungicide paints or slurries. In the case of sprays, mists, aerosols and fogs, the fungicide is in of droplets of water of another fluid. In the case of smokers, the solid particles of the fungicide are carried by the air. In the case of dusts and granules, the fungicide is straightly mixed with an inert carrier, impregnated into it coated on the particles, which are applied mechanically.

The object of spraying or dusting is to cover the entire susceptible surface of host with a thin covering of a suitable concentration of the fungicide before the pathogen has come into contact with the host. However, these practices may not effectively eradicate the inoculum present on the surface of the seeds or deep-seated in the seed. So, the application of chemicals as seed dressing is highly essential. In addition, soil harbours several pathogens which cause root diseases in several crop plants. So treatment of soil with chemicals is also highly useful in reducing the inoculum load present in the soil. The method which are commonly adopted in the application of the fungicides are discussed.

SEED DRESSING

The seed treatment with fungicides is highly essential because a large number of fungal pathogens are carried on or in the seed. In addition, when the seed is sown, it is also vulnerable to attack by many common soil-borne pathogens, leading to either seed rot, seeding mortality or produce diseases at a later stage. Seed treatment is probably the effective and economic method of disease control and is being advocated as a regular practice in crop protection against soil and seed-borne pathogens. Seed treatment is therapeutic when it kills pathogens that infect embryos, cotyledons or endosperms under the seed coat, eradicative when it kills pathogens that contaminate seed surfaces and protective when it prevents penetration of soil-borne pathogens into the seedling. There are various types of seed treatment and broadly they may be divided into three categories (a) Mechanical, (b) Chemical and (c) Physical.

A. Mechanical method

Some pathogen when attack the seeds, there may be alteration in size, shape and weight of seeds by which it is possible to detect the infected seeds and separate them from the healthy ones. In the case of ergot diseases of cumbu, rye and sorghum, the fungal sclerotia are usually larger in size and lighter than healthy grains. So by sieving or flotation, the infected grains may be easily separated. Such mechanical separation eliminates the infected grains may be easily separated. Such mechanical separation eliminates the infected materials to a larger extent. This method is also highly useful to separate infected grains in the case of ‘tundu’ disease of wheat. Ex. Removal of ergot in cumbu seeds.

Dissolve 2 kg of common salt in 10 litres of water (20% solution. Drop the seeds into the salt solution and stir well. Remove the ergot affected seeds and sclerotia which float on the surface. Wash the seeds in fresh water 2 or 3 times to remove the salts on the seeds. Dry the seeds in shade and use for sowing.

B. Chemical methods

Using fungicides on seed is one of the most efficient and economical methods of chemical disease control. On the basis of their tenacity and action, the seed dressing chemicals may be grouped as (i) Seed disinfectant, which disinfect the seed but may not remain active for a long period after the seed has been sown and (ii) Seed protectants, which disinfect the seed surface and stick to the seed surface for sometime after the seed has been sown, thus giving temporary protection to the young seedlings against soil-borne fungi. Now, the systemic fungicides are impregnated into the seeds to eliminate the deep-seated infection in the seeds. The seed dressing chemicals may be applied by (i) Dry treatment (ii) Wet treatment and (iii) Slurry.
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**Dry Seed Treatment**
In this method, the fungicide adheres in a fine from on the surface of the seeds. A calculated quantity of fungicide is applied and mixed with seed using machinery specially designed for the purpose. The fungicides may be treated with the seeds of small lots using simple Rotary seed Dresser (Seed treating drum) or of large seed lots at seed processing plants using Grain treating machines. Normally in field level, dry seed treatment is carried out in dry rotary seed treating drums which ensure proper coating of the chemical on the surface of seeds.

In addition, the dry dressing method is also used int pulses, cotton and oil seeds with the antagonistic fungus like Trichoderma viride by mixing the formulation at the rate of 4g/kg of the seed. Ex. Dry seed treatment in paddy.

**Wet seed treatment**
This method involves preparing fungicide suspension in water, often at field rates and then dipping the seeds or seedlings or propagative materials for a specified time. The seeds cannot be stored and the treatment has to be done before sowing. This treatment is usually applied for treating vegetatively propagative materials like cuttings, tubers, corns, setts rhizomes, bulbs etc., which are not amenable to dry or slurry treatment.

a. **Seed dip / Seed soaking**
For certain crops, seed soaking is essential. Seeds treated by these methods have to be properly dried after treatment. The fungicide adheres as a thin film over the seed surface which gives protection against invasion by soil-borne pathogens.

Ex. Seed dip treatment in paddy.
Prepare the fungicidal solution by mixing any of the fungicides viz., carbendazim or pyroquilon or tricyclazole at the rate of 2g/litre of water and soak the seeds in the solution for 2 hrs. Drain the solution and keep the seeds for sprouting. Ex. Seed dip treatment in Wheat.

Prepare 0.2% of carboxin (2g/litre of water) and soak the seeds for 6 hours. Drain the solution and dry the seeds properly before sowing. This effectively eliminates the loose smut pathogen, Ustilago nuda tritici.

b. **Seedling dip / root dip**
The seedlings of vegetables and fruits are normally dipped in 0.25% copper oxychloride or 0.1% carbendazin solution for 5 minutes to protect against seedling blight and rots.

c. **Rhizome dip**
The rhizomes of cardamom, ginger and turmeric are treated with 0.1% emisan solution for 20 minutes to eliminate rot causing pathogen present in the soil.

d. **Sett dip / Sucker dip**
The setts of sugarcane and tapioca are dipped in 0.1% emisan solution for 30 minutes. The suckers of pine apple may also be treated by this method to protect from soil-borne diseases.

**Slurry treatment (Seed pelleting)**
In this method, chemical is applied in the form of a thin paste (active material is dissolved in small quantity of water. The required quantity of the fungicide slurry is mixed with the specified quantity of the seed so that during the process of treatment slurry gets deposited on the surface of seeds in the form of a thin paste which later dries up.

Almost all the seed processing units have slurry treaters. In these, slurry treaters, the requisite quantity of fungicides slurry is mixed with specified quantity of seed before the seed lot is bagged. The slurry treatment is more efficient than the rotary seed dressers.

Ex. Seed pelleting in ragi.
Mix 2.5g of carbendazim in 40 ml of water and add 0.5g of gum to the fungicidal solution. Add 2 kg of seeds to this solution and mix thoroughly to ensure a uniform coating of the fungicide over the seed. Dry the seeds under the shade. Treat the seeds 24 hrs prior to sowing.

**Special method of seed treatment**
Ex. Acid - delinting in cotton
This is follows in cotton to kill the seed-borne fungi and bacteria. The seeds are treated with concentrated sulphuric acid @ 100 ml/kg of seed for 2-3 minutes. The seeds are then washed 2 or 3 times thoroughly with cold water and shade dried. After drying, they are again treated with captan or thiram @ 4g/kg before sowing.

C. **Physical methods**
Some of the seed treating procedures do not involve the use of fungicides, the physical agents like hot water or hot air or steam is used to eliminate the seed-borne infection. These methods are successfully used in controlling certain internally seed-borne disease like loose smut of Wheat and systemically infected diseases caused by virus and MLOs. Some of the commonly followed physical methods are discussed.
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Hot water treatment (HWT)
The seeds are soaked in cold water at 20-30°C for 5 hrs to induce the dormant mycelium to grow. Then the seeds are immersed in hot water at 50-54°C for 10 minutes to kill the mycelium. It is very effectively used to eliminate loose smut of wheat. The setts of sugarcane can be treated at 50°C for 2 hrs to eliminate grassy shoot pathogen.
The main drawback in the hot water treatment is that the seeds may be killed or lose its germinability, if the period of treatment exceeds the specified time. So this method is replaced by other physical methods like Hot air and Aerated steam treatment wherein the seeds are exposed only to hot air/aerated steam.

Hot air treatment (HAT)
Sugarcane setts are treated with hot air at 50°C for 2 hrs to eliminate mosaic virus.

Aerated steam therapy (AST)
Sugarcane setts are also exposed to aerated steam at 50°C for 3 hrs to eliminate mosaic virus.

Moist hot air treatment (MHAT)
This method is effectively used in sugarcane to eliminate grassy shoot disease. Initially the setts are exposed to hot air at 54°C for 8 hrs, then exposed to aerated steam at 50°C for 1 hr and finally to moist hot air at 54°C for 2 hours.

Solar heat treatment (SHT)
A simplest treatment has been devised in India to eliminate the pathogen of loose smut of wheat. Previously the hot water treatment was followed to eliminate loose smut. As the thermal death point of the fungus and the embryo are very close. The extensive care should be taken to avoid killing of the embryo. Luthra in 1953 devised a method to eliminate the deep seated infection of *Ustilago nuda*. The method is popularly known as solar heat or solar energy treatment.

Luthras solar energy treatment: The seeds are soaked in cold water for 4 hours in the forenoon on a bright summer day followed by spreading and drying the seeds in hot sun for four hours in the afternoon. Then, the seeds are again treated with carboxin or carbendazin at 2g/kg and stored. This method is highly useful for treating large quantities of the seed lots.

II. SOIL TREATMENT
It is well known that soil harbours a large number of plant pathogens and the primary sources of many plant pathogens happens to be in soil where dead organic matter supports active or dormant stages of pathogens. In addition, seed treatment does not afford sufficient protection against seedling diseases and a treatment of soil around the seed is necessary to protect them. Soil treatment is largely curativ in nature as it mainly aims at killing the pathogens in soil and making the soil ‘safe’ for the growth of the plant.

A. Physical methods

Soil Solarization
Soil solarization is generally used for controlling soil-borne pathogens like *Pythium*, *Verticillium*, *Rhizoctonia*, *Fusarium* etc. and nematodes in small areas like nurseries. Irrigate the nursery bed to moisten the soil to a depth of 10cm. Cover the bed after 2 days with thin transparent polythene sheets for 4-6 weeks and then irrigate the beds once in a week. The purpose of irrigation is to increase the thermal sensitivity of resting structures of fungi and to improve heat conduction.

Steam Sterilization
Steam is passed through perforated pipes at a depth of 15 cm to sterilize the upper layers of soil. It is mostly practised under glass house and green house conditions.

Hot air Sterilization
Hot air is also passed through pipelines to sterilize the soils in the nursery areas.

Hot water treatment
It is mainly done in pot culture studies to kill the fungi and nematodes. The pots containing soil are immersed in boiling water at 98°C for 5 minutes or drenching boiling water @ 20 litres/ Sq.m.

B. Chemical methods
Chemical treatments of the soil is comparatively simple, especially when the soil is fallow as the chemical is volatile and disappears quickly either by volatilization or decomposition. Soil treating chemicals should be non-injurious to the plants in the soil adjacent to the area where treatment has been carried out because there may be standing crop in adjacent fields. The soil treatment methods involving the use of chemicals are (i) Soil drenching, (ii) broadcasting, (iii) furrow application, (iv) fumigation and (v) chemigation.

Soil drenching
This method is followed for followed for controlling damping off and root rot infections at the ground level. Requisite quantity of fungicide suspension is applied per unit area so that the fungicide reaches to a depth of atleast 10-15 cm.

Ex. Emisan, PCNB, Carbendazim, Copper fungicides, etc.
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Broadcasting
It is followed in granular fungicides wherein the pellets are broadcasted near the plant.

Furrow application
It is done specifically in the control of some diseases where the direct application of the fungicides on the plant surface results in phytotoxic. It is specifically practiced in the control of powdery mildew of tobacco where the sulphur dust is applied in the furrows.

Fumigation
Volatile toxicants (fumigants) such as methyl bromide, chloropicrin, formaldehyde and vapam are the best chemical sterilants for soil to kill fungi and nematodes as they penetrate the soil efficiently. Fumigations are normally done in nursery areas and in glass houses. The fumigant is applied to the soil and covered by thin polythene sheets for 5-7 days and removed. For example, Formaldehyde is applied at 400 ml/100 Sq. m. The treated soil was irrigated and used 1 or 2 weeks later. Vapam is normally sprinkled on the soil surface and covered. Volatile liquid fumigants are also injected to a depth of 15-20 cm, using sub-soil injectors.

Chemigation
In this method, the fungicides are directly mixed in the irrigation water. It is normally adopted using sprinkler or drip irrigation system.

III. FOLIAR APPLICATION

Spraying
This is the most commonly followed method. Spraying of fungicides is done on leaves, stems and fruits. Wettable powders are most commonly used for preparing spray solutions. The most common diluent or carrier is water. The dispersion of the spray is usually achieved by its passage under pressure through nozzle of the sprayer.

The amount of spray solution required for a hectare will depend on the nature of crops to be treated. For trees and shrubs more amount of spray solution is required than in the case of ground crops. Depending on the volume of fluid used for coverage, the sprays are categorised into high volume, medium volume, low volume, very high volume and ultra low volume.

The different equipments used for spray application are: Foot-operated sprayer, rocking sprayer, knapsack sprayer, motorised knapsack sprayer (Power sprayer), tractor mounted sprayer, mist blower and aircraft (aerial spray).

Dusting
Dusts are applied to all aerial parts of a plant as an alternative to spraying. Dry powders are used for covering host surface. Generally, dusting is practicable in calm weather and a better protective action is obtained if the dust is applied when the plant surface is wet with dew or rain drops.

The equipments employed for the dusting operation are: Bellow duster, rotary duster, motorised knapsack duster and aircraft (aerial application).

IV. POST-HARVEST APPLICATION

Fruits and vegetables are largely damaged after harvest by fungi and bacteria. Many chemicals have been used as spray or dip or fumigation. Post harvest fungicides are most frequently applied as aqueous suspensions or solutions. Dip application has the advantage of totally submerging the commodity so that maximum opportunity for penetration to the infection sites. Systemic fungicides, particularly thiabendazole, benomyl, carbendazim, metalaxyl, fosetyl-AI have been found to be very effective against storage diseases. In addition, dithiocarbamates and antibiotics are also applied to control the post-harvest diseases. Wrapping the harvested products with fungicide impregnated wax paper is the latest method available.

V. PAINTING (SWABBING)

This is practiced normally in most of the ornamentals and fruit trees after pruning. The fungicidal solution/paste is painted on the cut ends to prevent the entry of pathogens. Sometimes, the swabbing is done after removing the diseased portion of the plants.

Eg. Swabbing of Bordeaux paste in stem bleeding disease of coconut.

SPECIAL METHODS

Trunk Application / Trunk Injection
It is normally adopted in coconut gardens to control Thanjavur wilt caused by Ganoderma lucidum. In the infected plant, a downward hole is made to a depth of 3-4” at an angle of 45° at the height of 3’ from the ground level with the help of an auger. The solution containing 2g of Aureofungin soil and 1 g of copper sulphate in 100 ml of water is taken in a saline bottle and the bottle is tied with the tree. The hose is inserted into the hole and the stopper is adjusted to allow the solution in drops. After the treatment, the hole is covered with clay.
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Root Feeding
Root feeding is also adopted for the control of Thanjavur wilt of coconut instead of trunk application. The root region is exposed; actively growing young root is selected and given a slanting cut at the tip. The root is inserted into a polythene bag containing 100 ml of the fungicidal solution. The mouth of the bag is tied tightly with the root.

Pseudostem Injection
This method is very effective in controlling the aphid vector (Pentalonia nigronervosa) of bunchy top of banana. The banana injector is used for injecting the insecticide. Banana injector is nothing but an Aspee baby sprayer of 500 ml capacity. In which, the nozzle is replaced by leurlock system and aspirator needle No. 16. The tip of the needle is closed and two small holes are made in opposite direction. It is for free flow of fluid and the lock system prevents the needle from dropping from the sprayer. One ml of monocrotophos mixed with water at 1:4 ratio is injected into the pseudostem of 3 months old crop and repeated twice at monthly intervals. The same injector can also be used to kill the bunchy top infected plants by injecting 2 ml of 2,4-D (Femoxone) mixed in water at 1:8 ratio.

Corn Injection
It is an effective method used to control Panama will of banana caused by Fusarium oxysporum f. sp. cubense. Capsule applicator is used for this purpose. It is nothing but an iron rod of 7 mm thickness to which a handle is attached at one end. The length of the rod is 45 cm and an iron plate is fixed at a distance of 7 cm from the tip. The corm is exposed by removing the soil and a hole is made at 45) angle to a depth of 5 cm. One or two gelatin capsules containing 50-60 mg of carbendazim is pushed in slowly and covered with soil. Instead of capsule, 3 ml of 2% carbendazim solution can also be injected into the hole.

Paring and Pralinage
It is used to control Fusarium wilt and burrowing nematode (Radopholus similis) of banana. The roots as well as a small portion of corm is removed or chopped off with a sharp knife and the sucker is dipped in 0.1% carbendazim solution for 5 minutes. Then, the sucker is dipped in clay slurry and furadan granules are sprinkled over the corm @ 40 g/corm.

References
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MAJOR DISEASES OF MAIZE AND THEIR MANAGEMENT

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Abstract

Diseases are destructive to the maize production due to fact that they occur widespread in maize growing areas. Proper disease identification will be important and help to understand more about the diseases prior the intervention. This chapter provides information which will lead to development of management practices and therefore improve maize production in disease affected areas.

Maize (Zea mays L.) is one of the most important cereal crops in world agricultural economy as food, feed and industrial products. Maize is known as ‘Queen of Cereals’ because of its highest yield potentiality among cereals. India ranks sixth in global maize production, contributing to 2.4 per cent of world production with almost 5 per cent share in world harvested area (Dash et al., 2015. In India, maize is the third important cereal crop after wheat and rice. Maize growing areas includes Madhya Pradesh, Bihar, Haryana, Punjab, Rajasthan, Uttar Pradesh, Gujarat, Andhra Pradesh and Karnataka (Joshi et al., 2005). Despite very high yield potential and one of the major deterrents to high grain yield is its sensitivity to several diseases. Almost all parts of the maize plant are susceptible to numerous diseases that considerably reduce the grain yield and quality of the crop (Shurtleff, 1980. From different maize growing regions of the world, about 112 diseases of maize have been reported, of these, 65 are known to occur in India (Saxena, 2002; Rahul and Singh, 2002. Brown spot, maydis leaf blight, Seed rot, downy mildews, stalk rots, banded leaf and sheath blight, Turcicum leaf blight, smut and downy mildews are the most important diseases of maize crop (Hafiz, 1986; Payak and Sharma, 1980. Globally, losses due to maize diseases have been estimated to the tune of 9.4 per cent, annually, for the countries of Asia is 12 per cent, while for African countries the estimate as high as 14 per cent (Cramer, 1967; James, 1981. Even for the developed countries like U.S.A., 12 per cent of the produce is lost due to diseases, annually. For India a per cent loss of 13.2 has been estimated (Payak and Sharma, 1985. Hence, this document aimed to provide comprehensive management strategy to reduce the losses caused by different major diseases of maize.

Maydis leaf blight (Bipolaris maydis syn. H. maydis)

Symptoms: Maydis leaf blight is also known as southern corn leaf blight of maize. The disease appears as young small, diamond shaped lesions and later on elongate with maturity within the veins. The center of lesions straw colored to light brown, surrounded by a dark brown margin. Growth of lesions limited by adjacent veins and finally lesions shape become larger and rectangular. Lesions of disease coalesce, producing complete burning of large surface of leaf and indirect losses by decreasing the grain yield.

Disease Cycle: The disease cycle of B. maydis is polycyclic and releases either asexual conidia or sexual ascospores to infect maize plants. The main route of maydis leaf blight infection is asexual via conidial infection or asexual cycle is known to occur in nature. Upon favorable moist and warm conditions, conidia (primary inoculum) released from lesions of an infected maize plant and carried to nearby plants via wind or splashing rain. Once conidia have landed on the leaf or sheath of a healthy plant, pathogen will germinate on the tissue by way of polar germ tubes (Singh and Srivastava, 2012.

Management:

Grow resistant variety namely HQPM-4, HQPM-5, HQPM-7, HM-10 and HM-11. Seed treatment with Captan or thiram (4g/kg). Two sprays of 0.2 per cent mancozeb (2g/l) in 200 litre water per acre and second 10-15 days after.

Common smut (Ustilago maydis)

Symptoms: The symptoms appear by producing the galls on all the above ground plant parts and even on leaves or tissue if mechanically injured. The mycelium developing between thin walled embryonic tissues induces hyperplasia, hypertrophy and excessive development of phloem bundles. The extent of damage depends upon size and sites of the galls. Initially the galls are light colour which becomes dull white or grey. The outer membrane breaks and exposes the black powdery masses of spores.
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Disease Cycle: Diseased plant debris and manure heaps primary source of inoculum as pathogen grow saprophytically. The chlamydospores on germination produce sporidia which are disseminated by wind and initiate the infection on the plants. The secondary infection takes place through the spores released by the breaking the galls.

Management:

Grow maize crop in fields with no history of common smut. Deep tillage can bury the fungus, which might reduce the level of inoculum available for the following year. Seed treatment with Bavistin 2g/kg seed. Crop rotation is not much effective. Excess nitrogen tends to increase the incidence and severity of the disease, so maintain balanced fertilization.

Downy mildew *(Peronosclerospora sorghi)*

Symptoms: Disease is characterized by development of chlorotic streaks on the leaves. Plants exhibit a stunted and bushy appearance due to shortening of the internodes. Whitish downy growth is seen on lower surface of leaf. Downy growth may also occur on bracts of green unopened male flowers in the tassel. Small to large leaves are noticed in the tassel. Proliferation of auxillary buds on the stalk of tassel and cobs.

Disease Cycle: The primary infection is through oospores in soil and dormant mycelium present in the infected seeds. Secondary spread is through airborne conidia. The initial source of inoculum can be oospores that over winter in the soil or conidia produced in infected, over wintering debris and infected neighboring plants. At the onset of the growing season, at soil temperatures above 20°C, oospores in the soil germinate in response to root exudates from susceptible maize seedlings.

Management:

Deep ploughing and Crop rotation with pulses. Grow resistant varieties/ hybrids and rogue out infected maize plants. Seed treatment with metalaxyl at 6g/kg and spray with Metalaxyl + Mancozeb on twenty days after sowing.

Brown spot *(Physoderma zea maydis)*

Symptoms: Brown spot is common in low lying or poor drained fields. It is appear on leaves and leaf sheath as oval shaped, light green or yellow water soaked lesions which become reddish brown to brown with lighter margin. Several spots may coalesce giving the leaf blades or leaf sheath a rusty brown appearance. Spots are more concentrated at the basal portion of leaves. Spots are irregular and bigger in shape on stem, leaf sheath and mid rib. Diseased leaf may die prematurely. Stem infection weakens the tissue and breaking of stem occurs at infection point.

Disease cycle: The pathogen is seed and soil borne. The sporangia can survive inside the host tissue even after harvest and remain viable about more than four years in soil. Next season sporangia germinate by opening of lid like structure and which gives rise to sporangium which attaches to host surface by means of rhizoidal system. The sporangia germinate and give rise to planospores which are smaller and function as gamets and produce infection hypha. On germination which expands within host cell to form several enlarged intercellular vegetative cells called "Sammelzellen". These sammelzellen form more enlarged cell which are directly converted into sporangia or special hyphae for infection. Secondary spread is through the dissemination of spores by means of wind. High temperature 28-29°C and high RH are ideal for growth of host as well as for disease development. Teosinte (*Euchlena maxicana*) is also known as other host.

Management:

Field sanitation and crop rotation are effective. Spray of Bavistin, Benlate or Plantvax @ 0.2% is effective. Captan spray at whorls, twice a week for 4 weeks before silking is effective in managing the disease.

Banded leaf and sheath blight *(Rhizoctonia solani)*

Symptoms: The disease appears on leaves and sheath on 40-45 days old plant and later on spread to the ear. Disease is characterized by lesions appear as concentric bands and rings on lower leaves and sheath. Infected plant produces large, grey or brown discolor areas alternating with dark brown bands *(Saxena, 1997.* Later on sclerotia are formed in diseased areas. It causes direct loss due to premature death of early infected plants and stalk breakage and ear rot in the older plants. Losses in grain yield from 11 to 40 per cent, even to 100 per cent on some cultivars in some warm and humid regions, where
the conditions are favourable for the pathogen (Madhavi et al., 2011, Mehra et al., 2012, Izhar and Chakraborty 2013; Gao et al., 2014.

Disease Cycle: High relative humidity and rain fall significantly favors spread and development of BLSB. An optimum temperature about 28°C, high relative humidity (88 to 90 per cent) in the first week of infection favor rapid disease progress and high crop densities impact disease severity (Sharma, 2005.

Management:

Stripping of 2-3 lower leaves along with leaf sheath. Seed treatment with Trichoderma harzianum.

Application of 0.2 per cent validamycin or 0.2 per cent carbendazim.

Head smut (Sphacelotheca reilianum)

Symptoms: This disease appears only at the time of flowering stage. Infection appears when young panic is still enclosed in the boot and is completely replaced by large smut sorus enclosed in a whitish membrane. When membrane rupture, exposed large mass of black spores intermingles with a network of long thin dark broom like structures. Sometime, bunch of small rolled leaves (witch's broom) protrude from the heads of suckers of infected plants.

Disease cycle: The pathogen is both seed and soil borne. The chlamydospores germinate in soil or infection hypha produced after germination of sporidia infects the meristematic tissue of the growing seedlings. The mycelium become systemic and produces the symptoms at the emergence of panicle. The black powder mass fall on the ground survive till next season. Optimum temperature range for germination of spores is 27-31 °C during moist soil period.

Management:

Field sanitation and crop rotation with pulses. Avoid waterlogging and poor drainage. Seed treatment with Bavistin (2g/kg seed) lower sowing depth large seed size, frequent irrigation after sowing use of clean seed and raising the crop in the soil having high water holding capacity helps in reducing the incidence.

Stalk rot (Macrophomina sp., Cephalosporium sp.)

i) Fusarium stalk rot: (Fusarium spp.; F. graminearum)

Disease appears when the crop enters senescence phase. The pathogen affects roots crown region and lower internodes. On splitting stalk shows pink to purple discoloration. Sometimes perithecia develop on the rind. The pathogen causes permanent wilting and drying of whole plant. After that browning of nodal tissue with little shredding of pith takes place. The pathogen is soil borne, high plant population density cause favours the disease and more prevalent in dry and warm climate.

ii) Charcoal rot: (Macrophomina phaseolina):

The disease is more prevalent in drier maize growing regions of tropical and sub-tropical countries. The disease is more conspicuous apparent at the time of maturity. The disease is more in temperature range of 30-40 °C at low soil moisture. The affected plants dry prematurely. Affected internodes become straw colored pith become disintegrated and small pin head like black colored sclerotia appear on the stalk. Water stress at flowering predisposes the plants to infection. The pathogen has wide host range, perpetuates soil and diseased debris for longer periods. The infection takes place through the roots or stem.

iii) Bacterial Stalk rot (Erwinia chrysanthemi)

Symptoms: The disease symptom starts from the lower nodes of the plants. The upper leaves show wilting, internodes are discolored, leaf sheath and leaves covering the stem become yellow and discolored. Occasionally the rot causes soft dark decay of rind but interior of stalk rotted. The rind become pale yellow and gives a corky appearance. The affected tissues become soft and later on turn into dry mass of shredded and disjoined fibers. Ear shoots and cobs are not infected but these drop down and keep hanging. Ultimately stalk breaks and plant collapses: Bacterium is facultative anaerobic and induces various enzymes which cause into necrosis and vascular wilt.
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Disease cycle: The pathogen is soil borne and lives saprophytically on debris. Neutral or slightly acidic soils are favourable. In next crop season, bacterium causes infection at the base of the stem through injured areas, insect injury or stomata or hydathodes. Accumulation of water around the stem base and relatively high temperature (30-35°C) favours the infection.

Management:

Grow resistant cultivar Sona and Surya-72 and avoid water logging. Two applications of Bleaching powder (33 % chlorine) and Calcium hypochlorite) @ 25 kg/ha first before flowering and second 10 days after one quite effective. Drenching of the basal stalk region at knee high stage with bleaching powder (33%) is also effective.

Late wilt: *(Cephalosporium acremonium)*:

It is important disease in maize growing areas in light soils. The pathogen is soil borne and affects plants through roots. Affected plants show wilting and dry up after flowering stage. The lower internodes become shunken, soft and discolored. The infected plant part may further attacked by other stalk rot fungi.

Management:

Cultivar Hybrid is resistant and water stress avoid at flowering stage. Phosphate amendments and lime reduce the disease.

*Turcicum leaf blight* *(Exserohilum turcicum)*

Symptoms: Turcicum leaf blight also commonly known as northern corn leaf blight. Disease appears as lesions are gray-green and elliptical, beginning 1-2 weeks after infection. In a susceptible reaction, fungal sporulation will begin within a few days. These lesions become pale gray to tan as they enlarge or longer. Distinct cigar shaped lesions unrestricted by leaf veins. Under moist or favorable conditions, lesions may produce dark gray spores, usually on the lower leaf surface, giving the lesions a dirty appearance. Lesions may enlarge and coalesce, leaf areas may be covered. Lesion coalescence and heavy blighting give leaves a burned appearance.

Disease Cycle: Pathogen survives in debris and builds up over time in high residue. Disease favored by heavy dews, frequent showers, moderate temperatures and high RH. Spores are spread by rain splash and air currents. Infection occurs when free water is present on the leaf surface for 6 to 18 hours and temperatures are 65 to 80°F. Infections begin on lower leaves and progress up the plant, but infections may begin in the upper plant canopy when spore loads are high.

Management:

Crop rotation to reduce the disease inoculum. Tillage to help break down crop debris and reduce inoculum load. Fungicide application to reduce yield loss.

References


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**Recent Trends in Integrated Pest and Disease Management**

**ECONOMICALLY IMPORTANCE LIFE CYCLE OF MAIZE CYST NEMATODE AND THEIR MANAGEMENT IN MAIZE PRODUCTION**

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**Abstract**

Maize suffers from number of nematodes in India (Koshy and Swamp, 1971; Mehta et al., 2015). Plant parasitic nematodes viz. Heterodera, Pratylenchus, Tylenchorhynchus, Meloidogyne, Hoplolaimus, Helicotylenchus, Rotylenchus, Longidorus, Trichodorus, Xiphinema and Belonolaimus) were found to be associated with rhizosphere of maize. These nematodes cause annual losses of 10.2% in maize world over. Integrated nematode management is the key to manage naemtodes in Maize.

Maize (Zea mays L.) is considered as the queen of cereals and cultivated under the wide range of agro-climatic conditions all over the world including India. Maize has multidimensional utilization and mainly used as food, feed and fodder and as an industrial raw material for starch and processed food industries. It is primarily cultivated as a kharif crop but presently, it is also being popularized as an important Rabi and summer crop in certain parts of the country depending upon the environmental conditions and irrigation facilities. Maize (Zea mays L.) is one of the most important cereal crop of the world, ranking third after rice and wheat in terms of area as well as production. It is extensively grown in Rajasthan, Andhra Pradesh, Bihar, Gujarat, Haryana, Karnataka, Madhya Pradesh, Maharashtra, Punjab and Uttar Pradesh in India. In Rajasthan, it occupies 9.16 lakh hectare of area with a production of 16.01 lakh tonnes resulting in productivity of 1771 kg/ha during 2015-16 (Anonymous 2016 b).

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**BIOLOGY AND LIFE-CYCLE**

The nematode is mainly active during the kharif season. In comparison to the other species of Heterodera, because these univoltine nature. The life cycle of maize cyst nematode is very short five to six generation are completed in crop seasion (Parihar, A. and Yadav, B.S.1992, Parihar, A. and Siddiqui, A.U. 1995. Emergence because these univoltine nature. The life cycle of maize cyst nematode is very short five to six generation are completed in crop season (Parihar, A. and Yadav, B.S.1992, Parihar, A. and Siddiqui, A.U. 1995. Emergence of multiplication.

Normally cysts are completed life cycle in 20-25 day at 20°C temperature (Lauritius, et.al.1983.

**Second stage larval:** Cysts are second stage larval penetration was observed in root at 48 hours after inoculation (Srivastava, A.N. and Sethi, C.L. 1985. The penetration was observed in zone of elongation and larvae were found to penetrate both in the tap as well as in lateral roots. The larvae were observed fully embedded in root just after penetration.

**Third stage larvae:** The third stage larvae were observed on 6th day after inoculation. At this stage, development of gonad initiated but sex differentiation was not detectable. The head region of these larvae were observed to have three annules, tapering anteriorly, stylet prominent, median bulb large with a conspicuous valve. In the later stage , the larvae developing into females showed bifurcated development of the genital cells.

**Fourth stage larvae:** The evidence of 4th stage larvae was observed on the 13th day after inoculation. The female larvae were observed to have thin cuticle and swollen posteriorly. Female larvae at this stage were embedded inside the stellar region and the rest of the body was completely outside the roots.

**Adult female:** The adult female was observed on the 19th day after inoculation. Adult female was lemon shaped with reflexed and coiled ovaries. The adult females have well developed neck and vulva with thin wall cuticle.

Maize only (2) Vativer only (3) Both maize and Vativer

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Mature cyst: The adult female started turning yellow and change into mature cyst after 25th days of inoculation. The cyst formed were lemon shaped and the eggs retained in cyst are easily visible from outside due to thin body cuticle of cyst. The vulval cone was generally prominent and sub crystalline layer was observed on new cyst.

Table 1. Life cycle of maize cyst nematode, *Heterodera zeae* on maize.

<table>
<thead>
<tr>
<th>Developmental Stage</th>
<th>Time after inoculation of second stage larvae</th>
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<tbody>
<tr>
<td>Penetration (2nd stage larvae)</td>
<td>48 hour</td>
</tr>
<tr>
<td>3rd stage larvae</td>
<td>6th day</td>
</tr>
<tr>
<td>4th stage larvae</td>
<td>13th day</td>
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<tr>
<td>Adult female</td>
<td>19th day</td>
</tr>
<tr>
<td>Mature cyst</td>
<td>25th day</td>
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</tbody>
</table>

Symptoms
The nematode thrives well in light soil and under optimum condition. Symptoms are given below. The infected plants show poor, stunted growth of plants. Yellowing of the leaves & Defoliation. Unthrifty growth with discolored foliage. Roots are poorly developed and cysts can be observed adhering to plant root. Less formation of cobs & grains. Visibility of white females on the clean roots & later on converted in to cysts.

Management
**Crop rotation and mixed crops:**
Guar, Moong, Urd, Cowpea, Soybean, Til & groundnut crops can be sown with Maize in intercropping manner for reducing the nematode effects. By growing the crop in following rotation can reduce the nematode population by 35-65 percent:
- a. Soybean-Garlic-Maize-Mustard
- b. Maize-Mustard-Til-Gram
- c. Maize-Pea-Groundnut-Mustard
- d. Guar-Wheat-Maize-Mustard

**Summer Ploughing:**
Light irrigation with 2-4 deep summer ploughings in the month of May & June (15 days intervals) reduced the population up to 60.

**Chemical control:**
1. Soil application of carbofuran @ 2kg a.i./ha and neem cake @ 5q/ha can avoided yield losses of maize to the tune of 44.00 and 22.00 % and reduced nematode population 56.50 and 32.70 %, respectively.
2. Seed treatment with neem seed kernel @ 10 % w/w found most effective with respect to management of maize cyst nematode on maize. However, maximum increase in maize yield (15.56 %) was noticed with karanj seed kernel @ 10 % w/w.

**Biological control:**
Seed treatment and soil application with some available formulations of bio-agents like *Paecilomyces lilacinus*, *Verticillium chlamydosporium*, *Trichoderma viride* and *Pseudomonas fluorescens* can also help in reducing nematode population and increase in plant growth.

**Use of plant products:** Seed treatment and soil application with powder form of seeds and leaves of some plant like Neem, Karanj, Mustured jatropha and Mahua can be used successfully against this nematode.

**Reference**
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BREEDING FOR DISEASES RESISTANCE IN FIELD CROPS

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Abstract

In the coming 50 to 60 years the world population will about double and hopefully also become more prosperous. This demands large yield increases in our food crops, which have to be grown in more sustainable agricultural systems. The need for durable disease resistance, therefore, can be expected to grow enormously. This need can be met technically by exploiting two sources that are largely untapped at present. These sources are the QR already present in our crops and the possibilities of transforming genes or gene constructs encoded for resistance into our crops. Quantitative Resistance at present is poorly exploited. If the same large effort that went into breeding for the hypersensitive, major gene type had gone into QR, most cultivars of our major crops would now carry high levels of it. With respect to sustainable agriculture and integrated forms of crop protection quantitative, durable resistance is a more desirable form of resistance than the non-durable type. Much of the resistance obtained after transformation is of a quantitative nature. This view should be consequential in modern genetic engineering activities. A considerable part of successful molecular manipulation leads to the type of resistance in which there is no shortage in most crops to most pathogens, and which is poorly used by the breeders.

In nature organisms are classified as producers, green plants, consumers organisms exploiting other organisms), and decomposers (organisms using dead organisms. Green plants, including our crops, are used by a multitude of consumers of almost every kind, from various types of herbivores (mammals, snails, insects) to typical parasites (insects, mites, fungi, bacteria. In order to survive green plants developed a broad range of defence mechanisms to ward off most of these consumers. These defence mechanisms are principally based on avoidance, resistance or tolerance.

Measuring Resistance

Selection for resistance implies measurements of plant resistance. Ideally one should measure the amount of pathogen present at a given moment compared with the amount present on or in a extremely susceptible cultivar. The quantitative or partial resistance of a host cultivar cannot be assessed in absolute terms; it is always a relative measure compared with that of a well-known standard cultivar. This standard cultivar is often the most susceptible cultivar available (Parlevliet, 1989. The amount of tissue affected is, in general, a good estimator of the amount of pathogen present. The amount of pathogen present, however, is not just dependent on the level of resistance of the host cultivar. Other factors may and do interfere with it such as:

Interplot interference
Van der Plank (1963) stated: “Plots in the experiment are meant to represent farmers’ fields receiving the same treatment as these fields receive”. But plots represent fields only when the plots within an experimental area do not interfere with one another. The representational error - the error of taking plots to represent fields when they do not can be large.

Relation between disease symptoms and amount of the pathogen
True disease symptoms are observed with several pathogens such as wilting caused by vascular pathogens, and leaf rolling, mottling, stunting, etc., caused by viruses. These symptoms tend to be rather unreliable for assessing resistance, since the relationship between the amount of pathogen present and the severity of symptoms is often poor. In other cases the pathogen itself is observed, making assessment much easier and far more reliable. The ecoparasitic powdery mildews are good examples; their mycelia remain on the surface of the host epidermis and are visible as white to grey spots.

Inoculum Density
This factor may obscure real differences in quantitative resistance. In order to prevent escape of genotypes from infection, there is a tendency to apply high inoculum densities. Complete resistance in such cases is easily detectable, but small differences in susceptibility tend to disappear. The optimal inoculum density is the density whereby escapes are largely prevented while only the most susceptible cultivars are strongly affected (Parlevliet, 1989.

Earliness
If the entries differ considerably in earliness the period of exposure to the pathogen varies greatly as the assessment is usually done at the same moment for all entries. Resistance to head blight caused by Fusarium in wheat is considerably overestimated in late cultivars due to this aspect (Parlevliet, 1993.)
Plant Habitat
In dense crops and short plants the amount of tissue affected tends to increase. In loose crops and tall plants it tends to decrease. This is probably due to micro-climatic effects. Short wheat cultivars are more affected than tall cultivars by *Septoria* leaf and glume blotch (Parlevliet, 1993).

GENETICS OF RESISTANCE
If published research is representative of the resistance present it is most often controlled by major genes. These major genes are often inherited dominantly, less frequently recessively. Polygenic inheritance of resistance has been reported as well, but its much lower frequency is most likely due to the more difficult nature of the research than to a truly lower frequency. Major resistance genes often occur in a surprisingly high numbers. In coffee (*Coffea arabica* L.) – *Hemileia vastatrix* Berk. & Br.

The gene-for-gene concept
Many major resistance genes operate in a gene-for-gene way. For each resistance gene in the host there is a corresponding avirulence gene in the pathogen (Flor, 1971), and only the corresponding avirulence gene can initiate the hypersensitive reaction (HR) leading to incompatibility. Resistance and avirulence inherit in most cases in a dominant manner, susceptibility and virulence in a recessive way. The HR is now known to result from the specific interaction at the cellular level of the product of the resistance gene and the product of the avirulence gene. If one of the two products is absent, there is no incompatibility; the normal pathogenicity of the pathogen results in a compatible reaction (the host appears susceptible. What is normally meant with virulence is actually the normal pathogenicity shown in the absence of avirulence. Virulence is absence of avirulence, it is genetically seen as an empty concept; there are no virulence genes.

QUANTITATIVE RESISTANCE
Resistance, like other traits, occurs in a qualitative or in a quantitative way. With the former the different genotypes in a population occur as discernible phenotypes; it is usually controlled by a major gene. Quantitative resistance (QR) is defined as a resistance that varies in a continuous way between the various phenotypes of the host population, from almost imperceptible (only a slight reduction in the growth of the pathogen) to quite strong (little growth of the pathogen). This resistance is often indicated with other terms such as partial, residual and field resistance or even (wrongly) with tolerance

Components of partial resistance
QR is expressed as a reduced amount of tissue in the invaded or affected host compared with that of a highly susceptible standard. When the total amount of disease is the collective result of a large number of discrete lesions, it is possible to identify a number of components contributing to the amount of tissue affected, as in the case of the cereal rusts (Parlevliet, 1992). QR may reduce the chance of infection, resulting in fewer lesions. It may reduce the growth of the pathogen once the infection is successful, causing smaller lesions that may sporulate less. It is possible to discern at least three components of QR against pathogens that are not systemic; infection frequency, lesion size and sporulation rate per lesion.

NON-DURABLE RESISTANCE
In nature there is a constant race of arms between the attacking parasite and the defending host, and in the evolutionary sense, all resistance is transitory. But large differences exist in the ease by which parasites can overcome a resistance. In agriculture, too the durability of a resistance varies greatly. Resistance may already be neutralized in the last stages of the breeding program (at zero years) and may, still be effective after more than 130 years and wide exposure, as the case of the *Phylloxera* aphid resistance of grape (*Vitis vinifera* L.) rootstocks (Niks *et al*. 1993.

DURABLE RESISTANCE
Resistance is considered durable when it remains effective for a considerable time, despite wide exposure. In this sense, it is a quantitative concept. The Rpg1 gene discussed above was durable, but did not last forever. And in the evolutionary sense, no resistance will last forever. It is possible to discern three groups of resistances that are predominantly durable.

1 - Resistance to pathogens with a wide host range, generalists
Are usually of a quantitative nature (Bruehl, 1983) and nearly always durable (Parlevliet, 1993. But there are exceptions, such as the major resistance genes Mi in tomato and Rk in cow pea against the root knot nematode, *M. incognita* (Roberts, 1995."

2 - QR against specialists and based on some to several genes with additive effects seems durable.
In the few cases where reported QR broke down, the resistance appeared to be monogenic, like the field resistance against rice blast, *M. grisea*, in the rice cultivar St-1. The resistance became ineffective within a few years and appeared to be based on a single dominant gene Pi-f (Toriyama, 1975."

3 - Monogenic resistance against specialists of a non hypersensitive nature.
Such resistance is often quite durable. The non hypersensitive resistance genes Rpg2 (sr-2) and Rpr34 (Lr- 34) of wheat to stem rust and leaf rust respectively and the mlo-gene of barley to powdery mildew have already lasted for a considerable time. Usually, the presence of race-specific resistance effects is considered as evidence of non-durable resistance
BREEDING FOR RESISTANCE
In order to reduce costs and to increase the efficiency of identifying resistant plants or lines in segregating populations, breeders developed screening methods in which plants as young as possible were exposed to high concentrations of, preferably, a specified inoculum. This efficiently identifies complete resistance based on major genes but is inadequate for recognizing small differences in resistance. These screening approaches, together with the belief that polygenic resistance is difficult to select for and might not give a good level of resistance, led to the present situation where major gene resistance has been exploited very well, while QR has been used only sparingly. This is unfortunate as there is so much QR available. Quantitative Resistance occurs to most of our important pathogens at various levels in nearly all our crops as discussed in the chapter “quantitative resistance”. Since this QR does occur in the cultivars grown, it is genetic material that is related to what the breeders’ desire. For this type of resistance breeders do not need to look for primitive genotypes from centres of diversity nor to related wild species. The resistance is near at hand in adapted cultivars, a fortunate situation as it makes breeding easier. McIntosh (1997) concluded that the ideal sources of resistance are those present in closely related, commercial genotypes, and any effort to transfer resistance from related species and genera should be considered long term. To select for QR means accumulating QR in much the same way as selecting for higher yields. The breeder selects the plants or lines with the lower levels of disease severity and by doing that continuously over the seasons, the level of QR will increase fairly rapidly as Parlevliet and Van Ommeren (1988) showed. There is, however, one complication. If there is also non-durable major gene resistance around, it has to be taken into account. The QR is not visible when such an effective major gene is present. By using, preferably, local material, the frequency of such non-durable still effective major genes is often low, as the local pathogen population has adapted to these genes. Introducing plant material from elsewhere, especially from the centres of diversity, increases the frequency of such non-durable effective major genes considerably, as the local pathogen population has not yet adapted to the newly introduced resistance. Therefore, to select QR stick as much as possible to local material as they will almost certainly carry QR. One can also avoid ending up with non-durable major resistance in the selected material by selecting against susceptibility, i.e. removing the most susceptible plants and lines all the time (Parlevliet and Van Ommeren, 1988. Plants or lines with complete resistance should also be removed in case of resistance breeding against specialized fungal pathogens, as such resistances can be assumed to be non-durable. In case of non-specialized pathogen and viruses one may use any resistance.

Reference
Recent Trends in Integrated Pest and Disease Management

INTEGRATED DISEASE MANAGEMENT OF MUNGBEAN AND URDBEAN

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Abstract

Of late, mungbean and urdbean production has shifted from a largely subsistence farming to market oriented commercial production. However, it has not been possible to exploit the full genetic potential of high yielding mungbean and urdbean varieties because of immense losses due to diseases and pests. Management of one single disease in isolation would prove counterproductive; hence it calls for the management of all diseases in integrated manner. An integrated disease management to combat the disease is the right approach.

Mungbean (Vigna radiata) and Urdbean (Vigna mungo) are important pulse crops. Both are important short duration grain legume crops with wide adaptability, low input requirement and have the ability to improve soil fertility by fixing atmospheric nitrogen. There is often confusion between mungbean and urdbean, the two being different only at the species (radiata and mungo) level. What our farmers traditionally grow in the southern foothills is actually Urdbean, but we are used to calling it Mungbean (the same way we say Orange for Mandarin. There are many local varieties of Urdbean but none in Mungbean. Mungbean is also known as green gram and serve are a major source of dietary protein. The nutritive value of both urdbean and mungbean lies in its high and easily digestible protein, and contain approximately 25-28% protein, 1.0% oil, 3.5-4.5% fiber, 4.5-5.5% ash and 62-65% carbohydrates on dry weight basis. Methionine concentration is larger in urdbean than in mungbean. Urdbean is also known as black gram. The seed colour can be either black or yellow. High values of lysine make urdbean and mungbean an excellent complement to rice in terms of balanced human nutrition. The mungbean resembles the black gram but there are differences: the corolla of Vigna mungo is bright yellow while that of Vigna radiata is pale yellow; mung bean pods are pendulous (hanging) whereas they are erect in black gram. Mung bean is slightly less hairy than black gram. Mungbean and Urdbean roots fix atmospheric nitrogen through symbiosis with nitrogen-fixing rhizobia, this crop is valuable both economically as well as nutritionally and is widely used in different cropping systems (Yaqub et al., 2010. The area, production and productivity of mung and urd bean during 2016-17 at national level have been mentioned below in Table-1 which is disappointingly low.

Table 1. Crop-wise Scenario (2016-17)

<table>
<thead>
<tr>
<th>Crop</th>
<th>Area</th>
<th>Production</th>
<th>Productivity</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mung</td>
<td>4.30</td>
<td>2.07</td>
<td>481</td>
</tr>
<tr>
<td>Urad</td>
<td>4.49</td>
<td>2.93</td>
<td>651</td>
</tr>
</tbody>
</table>

A- Million ha, P- Million tones, Y-kg/ha

The major factor for low production of mungbean & urdbean in India are ecological factors, lack of appropriate pulse production and protection technologies, poor post harvest technologies, less thrust on basic research, inadequate supply of quality seed to farmers and socio economic constraints etc. Apart from these, the pest and disease problems are the major bottlenecks in realizing the higher yields. Through there are several diseases, which attacks pulse crops, the major ones are mentioned in table-2.

Table-2. Diseases of National and Regional Significance

<table>
<thead>
<tr>
<th>Type of Diseases</th>
<th>Name of Disease</th>
<th>Causal Organism</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fungal</td>
<td>Anthracnose</td>
<td>Colletotrichum lindemuthianum</td>
</tr>
<tr>
<td></td>
<td>Cercospora leaf spot</td>
<td>Cercospora canescens</td>
</tr>
<tr>
<td></td>
<td>Powdery mildew</td>
<td>Erysiphe polygoni</td>
</tr>
<tr>
<td></td>
<td>Root rot and leaf blight</td>
<td>Rhizoctonia solani</td>
</tr>
<tr>
<td></td>
<td>Rust</td>
<td>Uromyces phaseoli</td>
</tr>
<tr>
<td></td>
<td>Macrophomina blight</td>
<td>Macrophomina phaseolina</td>
</tr>
<tr>
<td></td>
<td>Wilt</td>
<td>Fusarium oxysporum</td>
</tr>
<tr>
<td>Bacterial</td>
<td>Bacterial leaf blight</td>
<td>Xanthomonas phaseoli</td>
</tr>
<tr>
<td>Viral</td>
<td>Yellow mosaic</td>
<td>Mungbean yellow mosaic virus</td>
</tr>
<tr>
<td></td>
<td>Leaf crinkle</td>
<td>Leaf crinkle virus</td>
</tr>
</tbody>
</table>
Recent Trends in Integrated Pest and Disease Management

<table>
<thead>
<tr>
<th>Nematode</th>
<th>Cyst nematode</th>
<th>Heterodera cajani Koshi</th>
</tr>
</thead>
<tbody>
<tr>
<td>Root knot nematode</td>
<td>Meloidogyne incognita</td>
<td></td>
</tr>
<tr>
<td>Stunt nematode</td>
<td>Telenchorhynchus sp</td>
<td></td>
</tr>
<tr>
<td>Lesion nematode</td>
<td>Pratylenchus sp</td>
<td></td>
</tr>
</tbody>
</table>

**Major diseases and their diagnostic symptoms**

**Anthracnose**
The characteristic symptoms of this disease are circular brown sunken spots with dark centers and bright red orange margins on leaves and pods. In severe infection affected part withers off. Infection just after germination causes seedling blight. Five species of *Colletotrichum* are known to attack mungbean and urdbean. The pathogen survives from one crop season to the next on infected seeds and crop residue. Intermittent rains at frequent intervals favor the epidemic development of the disease. The optimum temperature and relative humidity for disease development 17-24°C and 100%, respectively.

**Cercospora Leaf Spot**
Leaf spots with brown to greyish centre and reddish brown border are its characteristic symptoms. The petioles, stems and pods also get affected by the pathogen. During favourable condition the spots increase in size and at the time of flowering and pod formation lead to defoliation. Five species of Cercospora infect mungbean and urdbean with slight variation in their symptoms. The fungus survives on the infected seeds and crop debris.

**Powdery Mildew**
Powdery mildew is a major problem for urdbean and mungbean cultivation and causes severe yield loss. This disease produces conidiophores carrying chains of white conidia. The disease appears on all the part of plants above soil surface. Disease initiates as faint dark spots, which develop into small white powdery spots, coalescing to form white powdery coating on leaves, stems and pods. At the advance stages, the color of the powdery mass turns dirty white. The disease induces forced maturity of the infected plant causing heavy yield losses and its intensity increases in stress condition.

**Macrophomina Blight**
It is caused by the fungus *Macrophomina phaseolina* causing root rot, collar rot, seedling blight, stem rot, leaf blight, pod and seed infection. In pre-emergence stage, the fungus causes seed rot and mortality of germinating seedlings. In post-emergence stage, seedling blight disease appears due to soil or seed-borne infection. Decay of secondary roots and shredding of the cortex region of the tap root are prominent symptoms. The fungus attacks the stem at ground level, forming localized dark brown patches which coalesce and encircle the stem. Black dot like sclerotia are formed on the surface and below the epidermis on the outer tissue of the stem and root.

**Rust**
On the lower surface of leaf reddish brown pustules are seen in abundance, representing the uredosori of the fungus. Affected leaves turn yellow. The uredospores are brown, echinulate and single celled. Teliospores are elliptical and papillate. The fungus is autoecious, macrocyclic rust.

**Root rot**
Affected plant exhibits drooping and drying of leaves and branches. The basal portion of stem turns brown and the bark of the roots become shredded. Large number of spherical to irregular black sclerotia can be seen in shredded tissues.

**Yellow Mosaic**
This disease is transmitted by the whitefly (*Bemisia tabaci*). This viral disease is found on several alternate and collateral host which acts primary sources of inoculums. The tender leaves show yellow mosaic spots, which increase with time leading to complete yellowing. Yellowing leads to less flowering and pod development. Early infection often leads to death of plant.

**Leaf Crinkle**
This disease is caused by *Urd Bean Leaf Crinkle Virus* (ULCV) belonging to Tospovirus. The virus is transmitted by aphids, whitefly and leaf hoppers and through sap. Disease symptoms include, crinkling, curling, and puckering of leaves often coupled with stunting and malformation of floral organs. Enlargement in size followed by crinkle surface of laminae are the characteristics symptoms on affected trifoliate leaves. Pollen production, fertility and subsequent pod formation is severely reduced with affect on seed weight, and size of seeds in infected plants leading to decrease in yield.

**IDM Component**
Sowing a crop variety with resistance to a disease. Modified farm management practices that result in reduction of disease loss. The enhancement of natural control processes. Need based application of pesticides

Based on the above components, a package has been developed for the management of Mungbean and Urdbean diseases depicted below:-

**Summer ploughing**
Due to summer ploughing, most of the crop residue which harbor the dormant mycelium/ spores of the pathogen get exposed to the temperature above 40°C for several days. Thus most of the inoculums of soil borne pathogens (*Fusarium spp.*, *Rhizoctonia spp.*, and nematodes) automatically get killed under hot condition and their numbers are reduced.
Soil Solarization:
In this practice, ploughed fields are covered with a thin polythene sheet for a period of 6-7 weeks during April – May, which results in rising of the soil temperature up to 60°C. The resultant high temperature is lethal to soil inhabiting pathogen, nematode and insects. This practice is very effective in controlling the *Heterodera cajani* and *Fusarium spp.* provided the soil is irrigated before polythene covering.

Plant nutrients:
Mungbean and Urdbean is a legume crop so it is believed that it requires less amount of nitrogen. Therefore, di-ammonium phosphate (DAP) @ 100 Kg/ha is applied in the field which provides nitrogen and phosphorus. Application of 20 kg of K/ha reduces the incidence of Mungbean and Urdbean. Manures and oil cakes reduces wilt and nematodes incidence. It increases the population of beneficial microorganism and reduces the population of fungal and bacterial propagules.

Varieties:
The improved Mungbean and Urdbean varieties having good yield potential as well as resistant/tolerant to diseases are mentioned in Table-3.

### Table-3. Disease resistant/tolerant varieties of Mungbean and Urdbean

<table>
<thead>
<tr>
<th>Disease</th>
<th>Mungbean: Narendra Mung1, Pant Mung 3, PDM 139 (Samrat), PDM-11(Spring Season), ML 131, ML 267, ML 337, Pusa 105, MUM 2</th>
<th>Urdbean: Narendra Urd1, IPU 94-1 (Uttara), PS 1, Pant U 19, Pant U 30, UG 218, WBU 108, KU 92-1 (Spring season), KU 500 (Spring Season)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Yellow Mosaic Virus</td>
<td>Mungbean: Narendra Mung1, Pant Mung 3, PDM 139 (Samrat), PDM-11(Spring Season), ML 131, ML 267, ML 337, Pusa 105, MUM 2</td>
<td>Urdbean: Narendra Urd1, IPU 94-1 (Uttara), PS 1, Pant U 19, Pant U 30, UG 218, WBU 108, KU 92-1 (Spring season), KU 500 (Spring Season)</td>
</tr>
<tr>
<td>Leaf Crinkle</td>
<td>Mungbean: D-3-9, K 12, ML 26, RI 59, T44 KII</td>
<td>Urdbean: HUP 27, 102, 164, 315</td>
</tr>
<tr>
<td>Powdery Mildew</td>
<td>Mungbean: LM 223, LM 24, P115, ML 131, MI 322, ML 337, ML 395 SS1, JRUM 1, TARM 1 and AVRDC 1381</td>
<td>Urdbean: COBG10, LBG 648, 17, Prabha, IPU 02-43, AKU 15 and UG 301.</td>
</tr>
<tr>
<td>Macrophomina blight</td>
<td>Mungbean: RMG 62</td>
<td>Urdbean: TAU 2</td>
</tr>
</tbody>
</table>

Seed treatment:
For the Prevention of soil and seed borne diseases and better yield, seed should be treated with antifungal bioagents, Rhizobium and Phosphorus Solubilising Bacteria seed should be treated with Trichoderma formulation @ 5-10g or *P. fluorescens* @ 10 g/kg seed (or) Carbendazim 2 g/kg or Captan/Thiram @ 3 g/kg of seed before sowing of seed reduces the incidence of wilt, Anthracnose, Macrophomina Blight and Cercospora Leaf Spot. After seed treatment, the seed should be mixed with Rhizobium culture. One packet of Rhizobium culture (250g) is sufficient for the seed required for one acre.

Sowing time and season:
Date of sowing and season also plays an important role in disease management. Removing root rot infected mungbean plants reduced sclerotia loads in the field and delayed sowing and maintaining wider spacing between the plants reduced powdery mildew incidence (Satyagopal et al., 2014). Mittal (1999) recommended second fortnight of June as the optimum time for sowing blackgram in Uttar Pradesh as it reduced the incidence of Cercospora spp. and anthracnose.

Crop Geometry:
The crop geometry influences the crop growth as well as microclimate of the area. The closely spaced plants are more weak and prone to attack of Cercospora spp., Yellow mosaic and Leaf crinkle diseases, while widely spaced plant suffer less. The optimum row to row and plant to plant spacing recommended for mungbean and urdbean is 30 x 10 cm for early, medium and late maturing varieties.

Inter/mixed cropping:
Inter/mixed cropping provides diversity in the field and ultimately creates sequential and temporal discontinuity to the pathogen. Crops like sorghum, maize, and sesame are grown as inter/mixed crop during cultivation. These crops are of short duration and mature at different intervals and perform better under such conditions. Vishwa Dhar et al., (2004) reported that yellow mosaic caused by *Mungbean Yellow Mosaic Virus* (MYMV) is the most serious limiting factor in mungbean and urdbean cultivation. The pathogen is transmitted by the white fly *Bemisia tabaci* Genn. Cultivation of resistant varieties, manipulation in sowing dates, inter/mixed cropping of mungbean and urdbean with non-host crops like sorghum, pearl millet and maize. Roots of sorghum exudates hydrogen cyanide (HCN) which inhibit the growth and reproduction of the pathogen.

Crop rotation:
Crop rotation with non Host crop helps in reduction of soil borne diseases. A three to four year rotation with tobacco, cotton and cereal crops reduces intensity of wilt and Cercospora Leaf Spot diseases reported by Sharma et al., 2011.
Sanitation
The pathogens Cercospora Leaf Spot, wilt, powdery mildew and several others survive in the field on the infected plant debris. Therefore, removal and destruction of the infected crop debris and collateral hosts after harvesting or at the time of field preparation, help in reducing the severity of diseases reported by Singh et al., 2001.

Chemical Management
Yellow mosaic virus: In order to prevent whitefly (*Bemisia* spp.) infestation sprays with triazophos 40 EC @ 2.0 ml/l or malathion 50 EC @ 2.0 ml/l or oxydemeton methyl 25 EC @ 2.0 ml/l at 10-15 days intervals if required.

Cercospora leaf spot: On appearance of the symptoms spray with carbenzadim 50 WP @1.0 g/l or mancozeb 45 WP @ 2.0 g/l. Subsequent spray should be done after 10 to 15 days, if required. Spraying with copper oxychloride @ 3 to 4 g /liter water has also been found effective in management of the disease.

Powdery Mildew: Spray NSKE 5% or Neem oil 3% twice at 10 days interval from initial disease appearance. Spray Eucalyptus leaf extract 10% at initiation of the disease and 10 days later also if necessary. Spray with water soluble sulphur 80 wp @ 4 kg/l or carbenzadim 50 WP @ 1 g/l (0.05%), benlate (0.05%) and topsin-M (0.15%).

Anthracnose: Spray the crop with 0.2% zineb 80% WP @ 2 g/l or ziram 80% WP @ 2 g/l with first appearance of symptoms on the crop and repeat after 15 days (if necessary).

Rust: Spray Mancozeb 1000g or wetttable sulphur 1500g /ha at initiation of the disease and 10 days later.

Leaf spot: Spray Carbendazim 500 g/ha or Mancozeb 1000g /ha at initiation of the disease and 10 days later.

Root rot: Basal application of zinc sulphate 25 kg/ha and neem cake @ 150 kg/ha. Soil application *P. fluorescens* or *T. viride* – 2.5 kg / ha + 50 kg of well decomposed FYM or sand at 30 days after sowing. Spray with carbenzadim 50 WP @ 1.0 g/l at an interval of 15 days with the appearance of the symptoms.

Leaf Crinkle: Give one foliar spray of insecticide (dimethoate 30 EC @ 1.7ml/ha) on 30 days after sowing.

Integrated Disease Management Packages:
- Selection of disease-free fields,
- Cultivation of disease tolerant varieties such as Pant Mung-3, Pant Mung-4, Pant Mung-5 which are resistant or relatively less susceptible.
- Soil solarization or summer ploughing
- Long crop rotations for 3-4 year with non host crop like tobacco, sorghum, pearl millet, and cotton.
- Wide row interspacing
- Intercropping with sorghum and maize
- Amendment of soil with oil cakes, appliances of trace elements such as boron, zinc and manganese.
- Soil amendment with Trichoderma @ 1.0 kg + 100 kg FYM at the time of field preparation to reduce the incidence of wilt disease.
- Seed treatment with hexaconazole + captan @ 2.5g/kg seed or metalaxyl @ 2.0 g per kg of seeds (or) carbenzadim or thiram @ 2 g/kg of seed 24 hours before sowing (or) with talc formulation of *Trichoderma viride* @ 4 g/kg of seed (or) *Pseudomonas fluorescens* @ 10 g/kg seed .
- Preventive sprays of mancozeb, wetttable sulphur or carbenzadim at 15-20 days interval starting from 15 days after germination reduces the incidences of diseases.
- Eradication of self sown plants in and around mungbean and urdbean fields is very helpful to manage viral disease.

**DO’S AND DON’TS IN IDM**

<table>
<thead>
<tr>
<th>DO’S</th>
<th>DON’TS</th>
</tr>
</thead>
<tbody>
<tr>
<td>Deep ploughing is to be done on bright sunny days during the months of May and June. The field should be kept exposed to sun light at least for 2-3 weeks.</td>
<td>Do not plant or irrigate the field after ploughing, at least for 2-3 weeks, to allow desiccation of weed’s bulbs and/or rhizomes of perennial weeds.</td>
</tr>
<tr>
<td>Adopt crop rotation.</td>
<td>Avoid mono cropping</td>
</tr>
<tr>
<td>Grow only recommended varieties.</td>
<td>Do not grow varieties not suitable for the season or the region.</td>
</tr>
<tr>
<td>Sowing early in the season</td>
<td>Avoid late sowing as this may lead to reduced yields and incidence of white grubs and diseases.</td>
</tr>
<tr>
<td>Always treat the seeds with approved biopesticides/chemicals for the control of seed borne diseases.</td>
<td>Do not use seeds without seed treatment with biopesticides/chemicals.</td>
</tr>
<tr>
<td>Sowing in rows at optimum depths under proper moisture conditions for better establishment.</td>
<td>Do not sow seeds beyond 5-7 cm depth.</td>
</tr>
<tr>
<td>Use NPK fertilizers as per the soil test recommendation.</td>
<td>Avoid imbalanced use of fertilizers.</td>
</tr>
<tr>
<td>Apply short persistent pesticides to avoid pesticide residue in the soil and produce.</td>
<td>Do not apply pesticides during preceding 7 days before harvest.</td>
</tr>
</tbody>
</table>
Reference


Dubey, SC and Singh, B. 2013. Integrated management of major diseases of mungbean by seed treatment and foliar application of insecticides, fungicides and bioagent. Crop Protection 47:55-60.


Recent Trends in Integrated Pest and Disease Management

STUDIES ON PHYTOPHTHORA AND ITS ASPECTS- OVERVIEW

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Abstract
Phytophthora is one of the most serious and devastating pathogen of world including India and is an important limiting factor for yield reduction. Management of Phytophthora is always a challenge for both scientists as well as growers because it’s close association with several fungal and bacterial saprophytes. This chapter deals with symptoms, isolation techniques, identification of Phytophthora spp on different selective media. Besides, disease forecasting, gene expressions by pathogens, resistances genes in host and management aspects have also been included as discussed by different researchers which may be useful for future researcher in formulating their research plan.

Phytophthora (from Greek (phyton),"plant" and (phthora), "destruction"; "the plant-destroyer") is a genus of plant-damaging Oomycetes (water molds), whose member species are capable of causing enormous economic losses to crops worldwide, as well as environmental damage in natural ecosystems. The genus was first described by Heinrich Anton de Bary in 1875. Approximately 100 species have been described, although 100–500 undiscovered Phytophthora species are estimated to exist. Although, Phytophthora spp. are mostly pathogenic to several dicotyledons but many are relatively host-specific parasites. Phytophthora cinnamomi can infects thousands of species ranging from club mosses, ferns, cycads, conifers, grasses, lilies, to members of many dicotyledonous families. Many species of Phytophthora are plant pathogens of considerable economic importance. Phytophthora infestans was the infective agent of the potato blight that caused the Great Irish Famine (1845–1849), and still remains the most destructive pathogen of solanaceous crops, including tomato and potato. The soya bean root and stem rot agent, Phytophthora sojae, has also caused longstanding problems for the agricultural industry. In general, plant diseases caused by this genus are difficult to control chemically, thus the growth of resistant cultivars is the main management strategy.

Phytophthora is represented by 48 species (Water house, 1973) which are cosmopolitan in distribution. Most of the species attack higher plants, mostly angiosperms and cause diseases of economic significance. Some species are facultative parasites and others as facultative saprophytes. One of the most common and well known species of Phytophthora is P. infestans that causes the disease called late blight of potato or Potato blight. Cool temperature (between 22-23°C) and excess of water favours the growth of this fungus. In India, P. infestans has been reported from Nilgiri Hills and Darjeeling. It also occurs periodically in the plains particularly of northern India. However, in the Indo-Gangetic plain blight of Colocosia antiquorum (Vern. arvi) caused by P. colocasiae is quite common.

Symptoms produced by different Phytophthora species on different crops in India

Phytophthora sojae

Seed rot and damping-off: Phytophthora sojae causes seed decay, and pre- and post-emergence damping-off of soybeans under wet and warm soil conditions. Fields with extensive seed rot and pre-emergence damping-off often require replanting. A light brown soft rot may develop on roots or the hypocotyl as seedlings emerge from the soil. As the roots and or hypocotyls become colonized, the seedlings may die.

Root and stem rot: The severity of the infection on soybean plants in the vegetative and reproductive stages of growth is directly related to the level of resistance in the plant. In highly susceptible cultivars The roots and stem turn chocolate brown in color, the leaves of the plant turn yellow, and the whole plant turns reddish-orange to orange-brown in color. Occasionally a lesion will only occur on one side of the plant, but it is continuous from below the soil line up the plant. The yellow wilted leaves cling to the plant as it dies. Roots colonized by pathogen turn light brown in color, and in few cases the plants may be stunted. There are usually no visible symptoms, other than reduced yields, under field conditions for cultivars with high levels of partial resistance.

Phytophthora spp on citrus

Damping-off of citrus: Damping-off of seedlings in nursery bed is a widespread problem of citrus industry and frequently occurs in citrus orchards where phyto-sanitary conditions are difficult to maintain. More than 20%
Recent Trends in Integrated Pest and Disease Management

seedling mortality has been observed in central India due to *Phytophthora* infection. (Naqvi Samh 2000. Damping-off of Citrus seedlings is caused by *Phytophthora parasitica*, *P. citrophthora* and *P. palmivora*. Typical symptoms of damping-off result when the soil or seed-borne fungus penetrates the stem just above the soil line and causes the seedling to topple. *Phytophthora* spp. also cause seed rot or pre-emergence rot. Infected seedlings are killed rapidly when moisture is abundant and temperatures are favourable for fungal growth (Klotz, 1969).

**Brown rot of fruit**: *Phytophthora* infected fruits get decayed in which the affected area turns light brown, leathery but not sunken compared to the adjacent rind. Under humid conditions, white mycelium growth on the rind surface can be observed. In the orchard, fruits near to ground become infected when splashed with soil containing the fungus. The disease spreads to fruits throughout the canopy, if favourable conditions of optimum temperature (75-82°F) and long periods of wetting (18 plus hours) continue. Most of the infected fruits soon absicile, but harvested fruits may not show symptoms until they have been kept in storage for a few days. If infected fruit is packed, brown rot may spread to adjacent fruits in the container. In storage, infected fruit have a characteristic pungent, rancid odour. Brown rot epidemics are usually restricted to areas where rainfall coincides with the early stages of fruit maturity (Timmer et al., 2003).

**Foot Rot and Gummosis**: Foot rot and gummosis are serious diseases caused by *Phytophthora* spp. Foot rot results from an infection of the scion near the ground level produce lesions which extend down to the bud union on resistant rootstocks (Fawcett, 1936) while on susceptible rootstocks scaffold root rot or crown rot below ground may occur. Infected bark remains firm with small cracks through which abundant gum exudation occurs. Citrus gum disappears after heavy rains but remain persistent on the trunk under dry conditions. Lesions spread around the circumference of the trunk, slowly girdling the tree. Pale green leaves with yellow veins are a typical symptom of severe infection of *Phytophthora*. If the lesions cease to expand or the fungus dies, the affected area is surrounded by a callus tissue. Nursery trees and young orchard trees of small trunk circumference can be rapidly girdled and killed. Large trees may be killed likewise, but typically the trunks are partially girdled and the tree canopy undergoes defoliation, twig dieback and short growth flushes. On susceptible rootstocks, lesions may occur on the crown roots below the soil line and symptoms in the canopy develop without obvious damage to the trunk above grown (Alvarez, 2008).

**Fibrous root rot**: Fibrous root rot is caused by infection of *Phytophthora* spp to the root cortex which starts decaying of fibrous roots. The cortex turns soft, becomes somewhat discolored and appears water soaked. After severe infection cortex of fibrous roots get destroyed leaving only the white thread-like stele, which gives the root system a stringy appearance. Root rot can be especially severe on susceptible rootstocks in infested nursery soil. Root rot also occurs on susceptible rootstocks in bearing orchards where damage causes tree decline and yield losses. In advanced stages of decline, the production of new fibrous roots cannot keep pace with root death. Under these conditions tree is unable to maintain adequate water and mineral uptake and nutrient reserves in the root as roots are depleted by repeated severe attack of fungus. This results in the reduction of fruit size and production, loss of leaves and twig dieback of the canopy, (Timmer 2003).

**Phytophthora capsici on Cucurbits**: *Phytophthora* blight symptoms may be observed on all above–ground parts of a cucurbit plant. However, cucumbers and watermelons usually have symptoms on fruit, but not on foliage. On the other hand, lesions readily form on the leaves and stems of pumpkins and squash. Cantaloupe is perhaps less sensitive to *Phytophthora* blight than the other hosts listed here, may have symptoms on foliage or fruit.

Lesions on leaves often marked as light green, sunken area that may be wedge shaped, becoming wider toward the margin of the leaf and become necrotic with advancement of disease. Stem lesions may cause the vine to wilt from the constricted area toward the end of the vine. Lesions on fruit vary depending on the host. Lesions on watermelon are often round, water-soaked and may appear as a bruise. Under moist conditions, these lesions are covered with a white mycelial growth of *P. capsici*. Lesions on fruit may be more common on the underside of the fruit where moisture accumulates. Fruit lesions on pumpkin may be large and occur in no particular shape. *Phytophthora* blight may also cause damping-off of seedlings (Dan egel, 2017).

**Phytophthora nicotinae**

This pathogen can cause root rot, crown rot, fruit rot, leaf infection, and stem infection. Root rot symptoms are observed on tobacco, poinsettia, tomato, pineapple, watermelon, and African violet. Fruit rots occur on tomato, papaya, and eggplant. Onion shows a leaf and stem infection. In tobacco, Black Shank affects the roots and basal stem area but other parts of the plant can also get infected. Damping off symptoms can be observed in young seedlings. The first above ground symptom that will be observed is the wilting of plants, which leads to stunting. Roots will be blackened and decayed. In final stages of the disease the stem begins to turn black, hence
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the name Black Shank. As this happens, tobacco leaves turn brown and loose its marketable value. Another symptom is disk-like appearance of the pith, although this is not a definitive symptom as it may also be the result of lightning strikes. On onion, it causes the disease on different stages of crop and known as *Phytophthora* neck and bulb rot. Initially, tips of newly infected plants get yellow and dry followed by softening of the “neck” of the plants that eventually fall off. Infected leaves may show grey lesions while roots may become necrotic in later stage of disease development (www.extento.hawaii.edu 2016).

*Phytophthora palmivora*
Although the common name of disease caused by *Phytophthora palmivora* is bud rot of palms, it affects many tropical plants and has a moderately broad host range. One common symptom of *P. palmivora* is fruit rots which are found in papaya, citrus, coconuts, durian, and cacao. Root rots are another symptom of *P. palmivora* and can be seen in red maples, citrus, papaya, mango, durian, and black pepper. Appearance of cankers on red maple, papaya, rubber, mangos, and cacao is also associated with the attack of *P. palmivora*. Bud rots can also be seen in papaya and coconuts infected with *P. palmivora*. Bud rots are also found in Palmyra palms and coconut palms. Collar rots are found on citrus, mango, and black pepper infected with *P. palmivora*. The signs of *P. palmivora* are microscopic and can be differentiated from other oomycetes by the presence of oval shaped papillate sporangia with short pedicles and spherical oogonia with narrow stalks (Butler 2008).

Vegetative Structure of *Phytophthora*
The mycelium is coenocytic, aseptate, hyaline and profusely branched (monopodial branching. The septa are formed at the time of reproduction or at maturity. The cell wall consists of glucan. Cytoplasm contains many nuclei, mitochondria, endoplasmic reticulum, ribosomes, dictyosomes, vacuoles and many oil globules. The mycelium is intracellular, and directly kills the invaded cells. However, in some cases it is intercellular, present in the intercellular spaces of the host tissue. Some of the species develop haustoria to absorb their food material. In *P. infestans* the haustoria are slender and finger like haustoria develop as lateral outgrowths from the intercellular hypha. Young haustorium invaginates the host cell its remains surrounded by an extra-haustorial sheath, an extra haustorial membrane and the cytoplasm of the host cell. Cytoplasm of haustoria contains mitochondria, ribosomes, endoplasmic reticulum and nuclei.

Reproduction of *Phytophthora*:
The fungus reproduces by vegetative/ asexual method and rarely by sexual methods.

(i) Vegetative Reproduction:
Many species of *Phytophthora* (*P. colocasiae* and *P. parasitica*) reproduce by means by Chlamydospores. These vegetative reproductive bodies may be terminal or intercalary. They germinate by giving rise to 3-11 germ tubes which generally develop sporangia at their tips.

(ii) Asexual Reproduction:
The asexual reproduction takes place by means of sporangia which are borne on aerial sporangiophores. Low temperature (12-20°C) and high relative humidity (91-100%) favours the growth of sporangia. The sporangiophores arise directly from the internal mycelium and emerge out of the host singly or in clusters through stomata or by piercing through the epidermal wall. Each branch of sporangiophore bears, sporangium at its tip. With the growth of the hypha below, the sporangium is shifted to lateral position and another sporangium is formed at the tip. The process may be repeated several times. Thus, the sporangiophore in *Phytophthora* is sympodialy branched. The sporangia may vary in shape (i.e.lemoni form, ovoid or elliptical. It is hyaline to light yellow in colour, terminally papillate and has a basal plug. In *P. colocasiae* the sporangia are 38-60 µ long and 18-26 µ broad. The sporangia are deciduous (fall off) and are disseminated by water or are blown by the wind. At the place of detachment of sporangia, the sporangiophores bear nodular swellings which are typical for this fungus. On falling upon a suitable host, the sporangia germinate. The germination of sporangium is governed by two main factors i.e., moisture and temperature. At high temperature (20-30°C), germinates directly by a germ tube. However, lower temperature (12°C) and presence of moisture favours indirect germination i.e., by zoospore formation. The sporangia are also susceptible to dessication. They lose their viability above 20°C temperature in 1-3 hours in dry air and 5-15 hours in moist air.

Direct Germination:
In the absence of moisture and high temperature (25 °C), sporangia germinate directly by germ tube and behave as conidia. The germ tube enters through stomata and infects the leaf.
Indirect Germination:
In the presence of moisture and lower temperature (12°C) it behaves as zoosporangium and produces zoospores. The protoplasm of the sporangium is cut off into many uninucleate polyhedral pieces in *P. infestans* and about 20 in *P. colocasiae*. Each polyhedral piece later rounds up and metamorphoses into zoospore. Zoospores are kidney shaped, biflagellate and possess flagella on lateral side of the two flagella one is of whiplash type and the other of tinsel type. The zoospores are liberated by the bursting of the sporangial wall. After swimming for some time they come to rest, encyst and germinate by a tube.

The germ tube adheres on the epidermis of the host and produces a flattened pressing organ i.e., appresorium, at its tip. From the appresorium a fine tubular, peg like outgrowth arises. It is the infection hypha. It penetrates the host tissue through stomata or epidermal cells. After penetration it develops into a profusely branched mycelium. The mycelium is intercellular and develops haustoria in the host cells. Under favourable conditions numerous sporangiophores emerge from the stomata and give rise to large number of sporangia. They are again disseminated by the wind and infect new plants. Thus, under favourable conditions the pathogen can reproduce several times by asexual method in one growing season.

(iii) Sexual Reproduction:
Clinton (1911) reported for the first time the sexual stages (oospore) in *P. infestans*. The sexual reproduction in *Phytophthora* is highly oogamous. The fungus is heterothallic i.e., requires two opposite strains, + and – for sexual reproduction. The male and female reproductive organs are called antheridia and oogonia, respectively.

The antheridium is of following two types:
(a) Amphigynous: Attached to oogonium as a collar e.g., *P. infestans*.
(b) Paragynous: Attached laterally to the oogonium e.g. *P. cactorum*. The antheridium arises earlier than the oogonium showing a protandrous condition. It develops as a terminal, more or less club shaped structure on a short lateral hypha of one strain. In young stages, it is thin walled with non-vacuolar cytoplasm and possessing only one or two nuclei. The mature antheridium is funnel shaped and forms a collar like structure at the base of the mature oogonium. The two nuclei divide mitotically and forms 12 nuclei. All nuclei disintegrate except one in mature antheridium.

Oogonium:
It is initiated laterally or below the antheridium on a hypha from other strain. The young oogonium pierces the developing antheridium from below and swells above it into a pear shaped or spherical structure. When young, it is multinucleate (up to 40 nuclei) and contains dense cytoplasm. On maturity it becomes vacuolated and differentiated into an outer multinucleate periplasm and a central uninucleate ooplasm. Nucleus of the ooplasm divides mitotically and out of the two one survives and it functions as an egg or oosphere nucleus.

Fertilization:
The oogonial wall bulges out at one point inside the antheridium and forms the receptive papilla. Later on the wall at the receptive spot dissolves and the antheridium pushes a short fertilization tube towards the oogonium. It penetrates the periplasm and passes into the ooplasm. Its tip opens and liberates a male nucleus and some of the cytoplasm. However, in *P. himalayensis* 2 to 3 papilla like outgrowths develops from the antheridium. One of these grows upwards and establishes a connection with the oogonium. The male nucleus passes into the oogonium through papilla and brings about fertilization. The oospore may also develop parthenogenetically in some cases.

Oospore: During fertilization, first of all the plasmogamy takes place. The fertilized oospore secretes a wall and undergoes rest. Fusion of the two nuclei is very late and occurs even until after the oospore walls are laid down. A mature oospore consists of an outer thick wall called exospor and an inner thin wall endospor. Exospor is made up of pectic substances and endospor is composed of cellulose and proteins.

Germination of Oospore: It is of rare occurrence and observed in a few species like *P. cactorum, P. palmivora* etc. The fusion nucleus divides meiotically and later on successive divisions result in the formation of few or many nuclei in the oospore. The exospor cracks and the endospor comes out in the form of a germ tube which
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devels a sporangium at the tip. The contents of a sporangium may divide to form zoospores or sometimes may directly develop into a mycelium (P. cactorum. Thus, it completes its life cycle only within its host tissue. It has no saprophytic existence. It lives as dormant mycelium in the dead host remains lying in the soil.

**Phytophthora biology:** Phytophthora species may reproduce sexually or asexually. In many species, sexual structures have never been observed, or have only been observed in laboratory as mating types. In homothallic species, sexual structures occur in single culture. Heterothallic species have mating strains, designated as A1 and A2. When mated, antheridia introduce gametes into oogonia, either by the oogonium passing through the antheridium (amphigyny) or by the antheridium attaching to the proximal (lower) half of the oogonium (paragyny), and the union producing oospores. Asexual (mitotic) spore types are chlamydospores, and sporangia which produce zoospores. Chlamydospores are usually spherical and pigmented, and may have a thickened cell wall to aid in their role as a survival structure. Sporangia may be retained by the subtending hyphae (non caducous) or be shed readily by wind or water tension (caducous) acting as dispersal structures. Also, sporangia may release zoospores, which have two unlike flagella which they use to swim towards a host plant.

**Techniques for detection of Phytophthora**

The Oomycetes are not true fungi, and therefore special techniques are required for their isolation. Most species of Phytophthora grow rather slowly in vitro compared with saprophytic fungi and bacteria. In addition, bacterial populations need to be kept low because they may suppress the growth of Phytophthora by direct competition, by antagonism caused by antibiotic production, or by direct parasitism. The use of selective media usually overcomes these problems. Antibiotics are added to isolation media in order to suppress the growth of bacteria because Phytophthora spp. are out-competed by many fungi. It is desirable to choose media which are “weak” in nutritional terms. This reduces the growth rate of fungal contaminants, allowing colonies of Phytophthora to become established. Synthetic cornmeal agar (manufactured by Difco) is the most frequently used basic medium for isolation of Phytophthora from infected plant tissue. However, other desirable basal media include: water agar, or 2% or 4% (v/v) V8 juice agar. Suitable antibiotics that are effective against bacteria include ampicillin, penicillin, rifampicin, and vancomycin, alone or in combination can be added to media. Suitable antibiotics with antifungal activity include nystatin and pimaricin. Nystatin is usually inexpensive and more readily available than pimaricin.

**Lupin baiting of Phytophthora from soil samples**

This method is a modification of a method from Pratt and Heather (1972) where lupins can be substituted by soybean seeds. Surface sterilized lupin seeds in 70% alcohol for 2 minutes, rinse in sterile distilled water and then soak for 1 hour in sterile distilled water. Four to five seeds will be required for each sample, depending on germination. Pre-germinated lupin seeds in sterile vermiculite at room temperature and regularly watering with sterile distilled water to ensure that the water drains from the seeds. Lupin radicals of 2-3 cm length will be suitable as baits (this will take 2 to 3 days. Punch or melt 5 holes, 5 mm in diameter in lids of plastic cups. When lupin radicals are of desired length, place a layer of soil sample approximately 3 cm deep in the bottom of plastic cup. Fill cup with distilled water to 1cm from top and cap with lids with pre-punched holes. Place pre-germinated lupins on top of lid, with radical inserted through hole and into water. Do not overfill the cups with soil. The volume of water should be 5-10 times compared to the volume of soil for best results. Check baits after 2 days upto 7 days. Brown, discoloured lesions on the lupin radicle should be surface sterilised and plated onto a selective medium. Phytophthora spp. are usually associated with those soft rots.

**Lupin baiting:** In Australia, radicals of young New Zealand blue lupin seedlings (Lupinus angustifolius) are used extensively in diagnostic laboratories because they detect many Phytophthora species. This is an excellent technique where Phytophthora infect the radical within five days; seedlings either do not grow or die completely. Lupins with obvious symptoms can be placed in a Petri dish with water and examined under a dissecting microscope; sporangia will be visible on the root lesion. Sometimes soil bacteria and Fusarium spp can also infect lupin baits making it difficult to observe the sporangia of the target Phytophthora. Chalara spores and rotifers may sometimes be observed on lupin roots but do not cause rotting symptoms. In addition, Pythium may also be detected but rarely cause significant rot symptoms. Pythium is more likely to cause a superficial root tip rot; spherical sporangia are sometimes able to be observed.

**Leaves of the infected host**

In most cases, the Phytophthora can be isolated from leaves of the symptomatic host by method described above. The method is similar to lupin baiting where instead of placing lupin seedlings in holes of the cup lid,
leaves are placed directly in the water. Leaves should be clean and healthy without any evidence of any pathogen infection. Prick or make a small cut in the leaf before placing in the water. Infection should occur within 1-5 days and include rotting tissue, most often around the artificially damaged areas. Sporangia may be observed under a dissecting microscope.

**Baiting water sources for Phytophthora:** The methods for lupin baits and using leaves of the infected host can be easily modified to bait water sources for *Phytophthora*. For lupin baits, fill up three cups entirely with the water source. If there are no plants showing signs of infection, umbrella tree leaves (*Schefflera actinophylla*) can be suspended in water, as per the method described above. Many other host plant species can be used if necessary.

**Fishing for Phytophthora species**

In order to bait for *Phytophthora* from streams, ‘fishing’ was conducted in 2011. Bait bags were prepared using polyvinyl chloride (PVC) coated fiber glass insect screen mesh (Cyclone Industries, ITW Australia Pty Ltd) into an A4 envelope. Fishing for *Phytophthora* was conducted as described by Huberli *et al.* (2013) by placing leaves of different baits such as *Metrosideros excelsus* (New Zealand Christmas tree), *Prunus armeniaca* (plum), *Pittosporum undulatum* and *Quercus* spp. inside the bags. Each bag was attached to a rope tied to the riverbank. Bags were placed in six locations along the Warren and Donnelly Rivers or tributaries (two bags per location). Buoyant polyurethane material was placed along one side of the bait bags to ensure they floated just below the surface of the water. Baits were collected after 6 days and lesions were plated onto the two *Phytophthora* selective media. Potential isolates were maintained under long-term storage.

**Baiting techniques for isolating Phytophthora from soil**

*P. citrophthora* Bait material: Apple, lemon or orange fruit can be used to isolate the pathogen. Insert soil or citrus tissue into fruit as per Campbell (1949) for *P. cinnamomi*. Alternatively, place lemon or orange on the surface of soil for 4 or more days (Klotz and DeWolfe 1958.

*P. nicotianae*: Bait material: Cocoa pods are used for isolation. Insert soil or diseased rubber tissues into unripe green pods as per the method of Campbell (1949. Incubate at 26-30°C for 4-5 days (Chee and Foong 1968)

**Basic media for isolation from diseased tissue**

**V8 juice Media**

Empty the contents of two 665 ml well-shaken cans of V8 juice into a 2 litres beaker. Add 10 g calcium carbonate (analytical grade), and stir for 20 minutes to adjust acidity. Unused V8 juice can be stored at –20°C. It must be completely thawed prior to use. Dilute media as described below.

**V8 juice agar – for routine growth and maintenance of Phytophthora cultures**

Dilute amended V8 juice to 20% (v/v) final concentration (100 ml CaCl2-amended V8 juice, 400 ml distilled or deionised water and 7.5 g agar. Autoclave the prepared medium at 121°C for 20 minutes, cools it to 55-60°C before pouring into plates.

**Diluted V8 juice agar – for isolation of Phytophthora from infected plant material**

Amended V8 juice can also be diluted to 2% or 4% (v/v) final concentration. Autoclave as above and cool before adding antibiotics.

**Cornmeal agar (CMA)**

CMA is not rich in nutrients so it is very suitable for isolation of *Phytophthora* from infected tissue. Amend the medium with antibiotics as medium cool down to 50-55°C. If commercially produced cornmeal agar is not available, fresh CMA can be prepared (Cornmeal (polenta) 60 g and 15 g agar in one litre distilled or deionised water) and autoclaved at 121°C for 15 minutes before use.

**Water agar**

Add 15 g agar to one litre distilled or deionised water. Autoclave, and add antibiotics once the media has cooled to 50-55°C.

**Selective media for isolation from diseased tissue**

Plates of selective media used for isolations should not contain any free or condensed water on the lids as water encourages the growth and spread of bacterial contaminants. Therefore, dry the plates with the lids half-off in the laminar flow for 20-30 minutes. Store the plates wrapped in plastic in the refrigerator. The plates should be stored upside-down. Note that pimaricin nystatin are light-sensitive. Media containing these anti-fungal agents should be wrapped in foil or black plastic and stored in the refrigerator. Ideally, selective media containing antibiotics should be made fresh before use or maximum within 2-4 weeks of preparation.

**3-P Medium (Eckert and Tsao 1960, 1962)**

This medium can be prepared by adding 17 g cornmeal agar (for example, Difco brand) to 1 litre distilled or deionised water followed by Autoclaving at 121°C for 20 minutes and cool to 50-55°C in a water bath. Add any
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of the antibiotics of the following concentrations (μg/ml=mg/l=ppm): pimaricin (100), penicillin (50), and polymyxin B (50). Plates should be stored in a refrigerator (4°C), wrapped in foil or black plastic as pimaricin is light sensitive.

**P10VP Medium (Tsao and Ocana 1969)**
Cornmeal agar is prepared as above, autoclaved, cooled to 50-55°C and amended with the any of the following antibiotics (μg/ml): pimaricin (10), vancomycin (200), and penta-chloronitrobenzene (PCNB; 100. This medium is suitable for isolating Phytophthora from the soil and infected plant tissue. Hymexazol can also be added to a final concentration of 25-50 μg/ml.

**P10ARP Medium (Kannwischer and Mitchell 1978)**
Cornmeal agar is prepared as above, autoclaved, cooled to 50-55°C and amended with the following (μg/ml): pimaricin (10), ampicillin (250), rifampicin (10), penta-chloro nitrobenzene (PCNB; 100. Hymexazo can also be added to a final concentration of 25-50 μg/ml.

### Culturing and storage of Phytophthora

Most Phytophthora species grow well on a range of media including V8. Cultures of Phytophthora should be grown at 15-25 °C in a dark incubator. Cultures should be transferred every two to four weeks to maintain vigour. For long-term storage, water storage has been recommended. The pathogenicity of Phytophthora cultures is known to decrease after prolonged storage media. In case of pathogenicity studies, serial passage through the host plant required. Another alternative is storage of cultures in liquid nitrogen which seems to overcome the problem of loss of pathogenicity.

**Long term storage in liquid nitrogen**
Cryopreservation of Phytophthora cultures in liquid nitrogen at −196°C is a good way to store most Phytophthora species. The cultures are maintained in their original genetic state and do not seem to lose their pathogenicity or aggressiveness. In addition liquid nitrogen storage is very effective and time efficient since the only requirement is maintaining the supply of liquid nitrogen.

**Long term storage in sterile water**
Phytophthora strains should be maintained as living cultures for two reasons. (i) To provide other researchers and ourselves with reference strains for various studies involving pathogenicity, virulence, mating type etc. (ii) As a source of DNA for genetic diversity and evolutionary studies. Phytophthora cultures can be stored in two ways, either in sterile water or in liquid nitrogen. To store cultures of Phytophthora, cut 8-10 small blocks from an actively growing plate culture, and place in small screw capped glass bottles containing autoclaved distilled water. The caps should be tightened during storage and the vials placed at room temperature in the dark. Most species of Phytophthora can be stored this way but keep in mind that the isolates will lose pathogenicity and aggressiveness during storage and cannot be used for studies in that area after prolonged storage. Ideally cultures should be revitalised once in a year or every second year. For some species, a soybean seeds or corns seed can be added prior to autoclaving the water which seems to induce oospore formation in homothallic species.

### Identification of Phytophthora

Many species of Phytophthora can be easily identified. However, the morphological differences among some species are few and variable, making it difficult to classify the species accurately. Identification of Phytophthora is based on the taxonomic keys of Waterhouse (1963) and Stamps et al. (1990). Characters which are used to classify species of Phytophthora include: sporangium morphology; morphology of sexual structures such as antheridia, oogonia and oospores; presence or absence of chlamydospires, and morphology of hyphae.

#### Morphological characters

There are a number of morphological characters upon which identification of Phytophthora species is based. These include: sporangium shape, papillation, and caducity; sporangiophore morphology; presence of chlamydospires and hyphal swellings; antheridal attachment, and whether sexual reproduction is heterothallic or homothallic.

### Sporangia

Sporulation in Phytophthora cultures provides important clues for species identification.

Important characters to observe are:

1. Sporangium morphology (shape, size, length:width ratio).
2. Papillation of the sporangium.
3. Caducity (shedding of the sporangium at maturity).
4. Length of the pedicel on the sporangium.
5. Proliferation of sporangium (production of new sporangium within a sporangium that has germinated directly. Branching of the sporangiophores on which the sporangia are borne.

However, many species need to be cultured in water, mineral salt solutions or dilute soil extracts before they will produce sporangia. It is important to remember that sporangia production in Phytophthora is dependent on light (Schmitthinner and Bhat 1994).
Sporangia production on solid media: Examples: P. heveae, P. capsici, P. megakarya, P. nicotianae, P. palmivora.

Sporangia produced in liquid media: P. cambivora, P. cinnamomi, P. citricola, P. cryptogea, P. drechsleri.

Rapid Diagnostic Field Test Kit for Identification of Phytophthora spp

Lateral Flow Device: Phytophthora LFD kits designed to recognise all species of Phytophthora including P. ramorum and P. kernoviae, were supplied by Forsite Diagnostics Ltd, York, UK. In summary, several small pieces of leaf showing symptoms were broken up between the thumb and fingers before placed in a plastic bottle containing 5 small (approximately 3mm) ball bearings and extraction buffer. Pieces of suspected diseased tissue transfer into the extraction bottle. The bottle was shaken vigorously for 60 seconds and then the extract taken up in a small disposable dropper. Two to four drops were placed onto an absorbent pad within the kit and left for at least 2 minutes but no longer than 10 minutes before reading. A single blue line developed to indicate the test was working (control line) whilst the development of a second blue (target line) indicated the presence of Phytophthora spp. A larger sample from the same part of the plant with identical symptoms was submitted for laboratory testing according to a protocol developed at CSL and now part of the EPPO Diagnostic protocol (Anonymous, 2006).

PCR is the most important and sensitive technique presently available for the detection of plant pathogens (Ward et al., 2004. This technique has the potential to detect single copies of the target gene contained in single propagules (Lee and Taylor, 1990) and is widely reported as an effective detection method for a number of Phytophthora species including those prevalent in forest ecosystems. However, in some circumstances, a nested PCR approach is used to improve the sensitivity and/or specificity of the assay. This involves two consecutive PCR reactions, in which the use of the first primer pair is followed with a second pair recognising a DNA region within the PCR product amplified by the first set (Ippolito et al., 2002). The use of nested PCR increases the risks of false positives due to cross contamination and involves more time and effort (Kwok, 1990. However, the availability of Phytophthora genus specific primers for a common first amplification could be useful to reduce the number of required amplifications with beneficial effects on the costs of the analysis and, to some extent, on the risks of false positives. A primer amplifying the ITS1 and ITS2 regions from all members of the Peronosporales in combination with the universal primer ITS4 was reported by Cooke et al. (2002) reported a primer combination (Ph2-ITS4) amplifying DNA from all Phytophthora species but did not, however, test their specificity against the abundant soil-borne genus Pythium. A pair of Phytophthora-specific primers amplying a fragment of the ras-related protein gene has also been reported (Schena et al., 2006a). Finally a set of primers amplifying the ITS1 and ITS2 regions from all Phytophthora species and not cross reacting with Pythium species has been utilised to develop a new method for the monitoring of Phytophthora diversity in soil and water environments.

Multiplex PCR, based on the use of several PCR primers in the same reaction, can be used to detect several pathogens simultaneously and reduce time and costs. In conventional PCR, multiplex assays are difficult to develop because different targets need to be differentiated by product size on agarose gels and yet the efficiency of amplification is strongly influenced by amplicon size. Consequently, it is difficult to identify good amplification conditions for all amplicons and shorter amplicons may be amplified preferentially over longer ones (Henegariu et al., 1997). Several attempts have been made to use conventional PCR as a quantitative detection method. However, conventional PCR amplification product is no longer proportional to the amount of original template present. A method, called competitive PCR, has been developed to quantify DNA of a number of target pathogens including P. infestans (Judelson and Tooley, 2000); however the method is very laborious and not sufficiently accurate (Mahuku and Platt 2002.

Real-time PCR

With emerging diseases such as sudden oak death caused by P. ramorum (Hayden et al., 2004), which have brought with them a realisation of the threat to entire ecosystems and industries, real-time PCR approaches have been adopted to provide a more rapid means of screening water, plant, and soil samples (Schaad et al., 2003. Several reviews covering the most common real-time PCR chemistries and their application to the study of plant pathogens have been recently published (Mumford et al., 2006). In particular, real-time PCR chemistries applied to the study of Phytophthora species include amplicon sequence non-specific (SYBR Green) and specific (TaqMan, Molecular beacons and Scorpion PCR) methods. Compared to conventional PCR, real-time PCR eliminates the requirement for post-amplification processing steps thus saving time and labour. Without ethidium bromide, health risks for operators and environmental contamination are reduced and the throughput of PCR testing as an automated diagnostic system suitable for large-scale applications increases. This is
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particularly true when, as reported for *P. ramorum*, specific on-site methods are developed to extract and amplify DNA in the field using a portable real-time PCR platform (Cepheid Smart Cycler) (Tomlinson et al., 2005. The ability to test plant samples for specific infections rapidly and at the point of sampling is likely to have a number of useful applications. Epidemiological studies in the field or at remote locations, for example, could greatly benefit from the ability to perform molecular testing without the need to return samples to a laboratory. Also, decisions regarding control strategies including eradication measures, which may need to be taken rapidly, could be better informed by the availability of reliable real-time PCR data on-site and within few hours of inspection. Another significant advantage of real-time PCR is the increased sensitivity in comparison to other detection methods including conventional PCR. For a number of forest *Phytophthora* spp. detection sensitivity ranging from 1 to 100 fg of target DNA has been reported for methods based on different target genes including the ITS regions (Hughes et al., 2006), the ras-related protein gene (Schena et al., 2006a) and an intergenic region of the mitochondrial genome (Tooley et al., 2006).

**Target DNA regions for the development of diagnostic assays**

**Internal Transcribed Spacer (ITS) regions**: The ITS regions of the nuclear ribosomal DNA (rDNA) array are the most commonly sequenced regions for *Phytophthora* and have been widely utilised for phylogenetic studies (Kroon et al., 2004) and diagnostic assay development (Silvar et al., 2005. The ITS regions provide attractive targets because they are highly stable. They can be easily amplified and sequenced with universal primers, occur in multiple copies, and possess conserved as well as variable sequences (White et al., 1990. ITS-based PCR primers have been reported and are available for the detection of a number of *Phytophthora* species including those important for their impact on natural ecosystems. Furthermore, the same regions have been utilised to design specific oligonucleotide arrays for a number of plant pathogens (Anderson et al., 2006. However, in recent years, the discovery and ITS sequencing of many new *Phytophthora* species have raised concerns about the specificity of some ITS-based molecular detection methods. This is due to cases where the ITS sequences are not sufficiently variable, making the design of primers to identify and detect closely related taxa that are very difficult or impossible to detect. Important forest *Phytophthora* spp. such as *P. nemorosa*, *P. lilis*, *P. psychrophila*, and *P. pseudosyringae* have very similar ITS regions and the design of effective and robust specific primer set is very challenging (Schena and Cooke, 2006. Similarly *P. alni*, *P. cambivora*, *P. fragariae*, and *P. europaea* are phylogenetically closely related and challenging to distinguish via ITS sequences (Brazier et al., 2004. The PCR assay used widely for *P. ramorum* detection was recently found to cross-react with *P. foliorum* a newly discovered and closely related species of unknown importance (Donahoo et al., 2006. The discrimination of *P. lateralis* from *P. ramorum* requires two lengthy procedures such as single strand polymorphism (SSCP) analysis (Kong et al., 2004) or a double amplification with two different primer pairs (Hayden et al., 2004. For the same reason in the real-time PCR detection method developed by Hughes et al. (2006) it was necessary to introduce a base substitution in one primer to increase specificity but at a cost of decreased sensitivity.

**Intergenic spacer (IGS) region**: The intergenic (IGS1 and IGS2) regions of rDNA seem to have great potential as alternative to the ITS regions. Like the ITS regions, they are multicopy (up to 200 copies per haploid genome) but their length (4000-5000 bp) also provides considerable scope for primer development. Specific primers to detect *P. medicaginis* were developed on the IGS2 region because of difficulties in discriminating the related species on the basis of ITS (Liew et al., 1998. However, the utilisation of the IGS regions as targets to develop specific molecular markers has been limited, primarily due to the difficulties in amplifying a long fragment (4000-5000 bp) and the lack of effective universal primers. The complete sequence of the IGS region of *P. megasperma* has been recently determined and utilised to develop a primer set to identify *P. megasperma* isolates (Nigro et al., 2005. Recently, a set of universal primers has been developed and utilised for the amplification of a PCR fragment of approximately 450 bp in a region of the IGS very close to the 28S rDNA gene (Schena and Cooke, 2006. The alignment and comparison of this region from 28 different *Phytophthora* species showed a level of genetic variability comparable to those of the ITS regions. However the new sequences do allow the discrimination of some additional *Phytophthora* species with very similar ITS regions and should facilitate the amplification and characterization of the potentially more variable flanking regions.

**Forecasting model for Phytophthora spp**

Forecasting model: Forecasting is applied epidemiology, this needs complete knowledge about factors affecting disease developments. A disease forecasting model predicts out-break or changes in intensity of one or more diseases on the basis of information about weather, crop, pathogen(s) or some combination of the three. Forecasting includes all the activities in ascertaining and notifying the farmers that -The weather conditions are sufficiently favorable for certain disease. The application of control measures will return in economic gain. The amount of disease expected is unlikely to be enough to justify the expenditure of time, energy and money for control. Forecasting assists the growers for spray schedule and reduces the costs involved by eliminating the
Role of weather on disease development: The congenial conditions for appearance and build up of disease are 10-22°C temperature, humidity above 75%, cloudy or foggy weather (Bhattacharyya et al., 1983; Deweille, 1964. Ambient temperature, relative humidity, light, fogginess, rainfall, dew, wind velocity etc. have a strong relationship with the late blight pathogen and the disease. The role of environment in the development of late blight has been well documented (Harrison, 1992; Rotem et al., 1971. Usually infection by zoospores take less time as zoospore remain motile up to 22 hr at 5-6°C temperature whereas high temperature i.e. 24-25°C decrease motility of zoospores. Sporangia are formed at high humidity and disperse at high temperature and low relative humidity. The detachment of sporangia is mainly due to changes in humidity (Singh, 2007).

Temperature below 23.3°C is more favourable for disease development. Epidemic conditions are mainly favoured by humidity i.e. prolonged survival of sporangia require high relative humidity. Disease development is also depending on the presence of free water on the surface of foliage. In absence of surrounding water film, air borne sporangia lose their viability. Host susceptibility depends on the distribution and duration of saturated or near saturated air within haulm (foliage. Wind is one of the important factors for spreading of disease. Photoperiod, light intensity etc. has direct impact on pathogen development and host susceptibility.

Bourke (1953) developed model known as the Irish rules which currently used in Ireland. For germination and infection, sporangia of Phytophthora infestans require a wetness period of at least 12 hrs, air temperature not below 10°C and relative humidity at least 90%. Finally, potato growers are warned that weather conditions are conducive for late blight spread and are favourable for spraying.

Sharma (2000) concluded that late blight development in the North-Western region especially in Jalandhar region was positively correlated with maximum relative humidity, rainfall, dew and cloudy days while negatively correlated with minimum temperature. late blight disease occurrence have been favoured by air temperature less than 26°C and relative humidity more than 85%. Henshall et. al. (2006) developed a new model i.e. Shitenberg model for late blight risk in which they combined inoculums index with the infection index to produce a late blight risk index (0-3) corresponding to nil, light, moderate and severe risk of disease. Singh et al. (2000) modified JHULSACAST and developed a modified model for late blight forecasting at Pantnagar as is follows: i. 7-days moving > 85% cumulative relative humidity period e” 85 hours.

Van Everdingen (1926) pioneered in using ‘Dutch Rules’ based on four weather parameters, viz. dew periods, night temperature, cloudiness and rainfall to predict initial appearance of potato late blight disease in Holland were not universal in adoption but have impetus for several workers. Subsequently, Dutch Rules were modifies as Beaumont rules based on minimum temperature of 100C or more and RH periods of 75% or above for two consecutive days (Beaumont, 1938. They successfully forecast late blight under UK conditions.

Resistance genes in host against Phytophthora spp.

Dominant plant disease resistance (R) genes confer resistance to specific pathogens encoding matching Avrulence (Avr) genes (Dangl and Jones, 2001. R gene mediated resistance is an active process whereby recognition of pathogens results in the activation of multiple signaling pathways and often culminates a form of programmed cell death known as the hypersensitive response (HR) (Mur et al., 2008. Most Avr genes encode pathogen effector proteins and this phenomenon is now more commonly referred as effector-triggered immunity (ETI) (Jones and Dangl, 2006. Plant R genes are highly polymorphic both within and between populations ( Yang et al., 2008), and effective R genes are often introgressed into crop plants from wild relatives. Avr genes are also frequently polymorphic and R genes may confer resistance to most pathogen or all pathogen races, isolates, or strains or they may only be effective against a small subset. As such, some R genes are durable over long periods of deployment, whereas others are quickly rendered ineffective due to newly emerged or introduced “resistance-breaking” strains (Parlevliet, 2002. The majority of plant R genes encode for NB-LRR (Nucleotide Binding Site–Leucine-Rich Repeat) proteins. There are three major classes of NB-LRR proteins, distinguished by the protein domain encoded at the N terminus; either a Toll and Interleukin-1 Receptor homology domain (TIR; TIR-NB-LRRs), a loosely predicted coiled-coil domain (CC; CC-NB-LRRs) or a CC domain with homology to RPWS (CCG; CC-K-NB-LRR) (Shao et al., 2016. NB-LRR proteins recognize effector/Avr proteins from all types of pathogens and several R genes conferring resistance to members of the Phytophthora species have been described including Rhs/RplBlb, R1, R3b, and R3a, which confer resistance to P. infestans (Oh et al., 2014), and Rps1k, Rps4, and Rps6 which confer resistance to P. sojae (Gao et al., 2005. The potato resistance protein R3a was shown to recognize AVR3a from P. infestans (Bos et al., 2006; Engelhardt et al., 2012. Silencing of Avr3a compromises P. infestans pathogenicity, indicating an important role of Avr3a in virulence (Bos et al., 2010).
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Soybean cultivars and germplasm accessions differ in their responses to isolates of *P. sojae* (Kaufmann et al., 1958). The use of resistant soybean cultivars is the most economical and effective method of controlling this pathogen. Two distinct types of host resistance to *P. sojae* have been described: (i) race-specific resistance conditioned by single dominant genes (*Rps*); and (ii) broad-spectrum partial non-race-specific resistance conferred by several minor genes (Sugimoto et al., 2012). When novel *Rps* genes are introduced through the release of new cultivars *P. sojae* isolates evolve to overcome the introduced resistance genes (Grau et al., 2004). Over 200 known pathotypes of this pathogen have been reported (Stewart et al., 2014), presumably due to selection pressure on the *P. sojae* population for new pathotypes that can overcome *Rps* genes (Mac Gregor et al., 2002). The rapid evolution of new *P. sojae* virulent pathotypes limits the effectiveness of an *Rps* gene to 8–15 years. Consequently, there is a constant need for novel *Rps* genes that can effectively manage the disease. The first *Rps* gene was identified in the 1950s (Bernard, 1957) and 27 *Rps* genes have been identified and mapped till date. The *Rps* genes encode receptors that presumably recognize *P. sojae* effectors and induce effector-triggered immunity (Dong, 2011). The *Rps* genes mapped to Chromosome 3 include *Rps1, Rps7, Rps9, RpsYu25, RpsYD29, RpsYD25, RpsUN1* and *Rps1?* (Sugimoto et al., 2011, Zhang et al., 2013). The *Rps1* locus is complex and contains at least five functional alleles, *Rps1a, 1b, 1c and 1d* and 1k (Weng et al., 2001). High resolution genetic and physical maps were constructed for the *Rps1*-*k* region and two functional nucleotide binding site-leucine rich repeat (NBS-LRR) containing *Rps* genes, *Rps1*-*k*-1 and *Rps1*-*k*-2, were cloned from the *Rps1*-*k* locus (Gao et al., 2008).

**Gene expression in Phytophthora**

Gene expression approaches constitute a starting point from which to determine the best strategy for building a computational model of a plant disease. Host-expressed molecules give insights into the underlying defense mechanisms, whereas identification of the pathogen counterparts allows us to ascertain possible mechanisms of attack and/or avoidance mechanisms used to establish a disease. Differential expression of particular genes is a common strategy in gene expression analysis to identify a particular gene of interest, and then to study or characterize its expression profile in different hosts or treated tissues. For instance, based on the findings that during the early phases of the interaction between *P. infestans* and potato, the genes *ipiB* and *ipiO* are expressed at high levels, Pieterse et al. (1994) hypothesized that these genes played an important role in the early stages of the infection process. Both genes were isolated and their expression studied in various host tissues and different host plants. The results showed that the expression of these genes was activated in compatible, incompatible and non-host interactions. In the case of *ipiO*, it was revealed that a motif on the promoter region functioned as a variable nutrient environment that governs expression of particular genes. The expression profiles of these genes were also correlated with disease resistance in cultivars with different resistance levels. The authors concluded that perhaps a variable nutrient environment could trigger the expression of *ipiO* and *ipiB* depending on the host and/or the expressing tissue.

**Protein-protein interactions in Phytophthora**

One approach to study protein-protein interactions is by using yeast two hybrid screening, co-immunoprecipitation or surface plasmon resonance. This is arguably the most important approach towards a broad understanding of any plant pathogen interaction. It enables some mechanisms for the suppression of host defense in several organisms, such as the fungal pathogen *Septoria lycopersici* (Bouarab et al., 2002) or the Oomycete *Phytophthora sojae* (Darvill et al., 2003) to be revealed. In the case of *P. infestans*, relevant host defense suppression molecules have been also identified by this approach, such as the extracellular protease inhibitors EP11 (Tian et al., 2004), EP110 - the first protease inhibitor reported in any plant-associated pathogen, which suppresses tomato defense by targeting - the P69B subtilisin-like serine protease (Benedetti et al., 2005), and the EPIC family of secreted proteins that target the extracellular cysteine protease PIP1 (Phytophthora Inhibited Protease 1) (Tian et al., 2007). Protein-protein interactions play an important role in recognition between plant pathogens and their hosts. This recognition has been studied at two levels: recognition of the host by the pathogen and recognition of the pathogen by the host. During an interaction, host resistance (*R*) and pathogen avirulence (*Avr*) proteins interact in a gene-for-gene manner. Proteins encoded by *R* alleles recognize the products of corresponding *Avr* alleles, thus triggering disease resistance. Using an association genetics approach the *P. infestans* *Avr3a* effector was shown to be recognized in tomato cytoplasm by R3a (a member of the R3 complex locus on chromosome 11. R3a was isolated by positional cloning the same year. Together, these and other studies along with computational chemistry and/or computational modeling and prediction of protein-protein interactions, provide valuable information about the recognition mechanisms in *S. tuberosum* - *P. infestans* R-*Avr* interactions and could lead to the identification of metabolic and/or signaling pathways underlying incompatible interactions.
Phytophthora spp management aspects

Since Phytophthora spp. are an oomycetes, their simplest management technique is to control the amount of water present in the soil. Techniques for controlling moisture include: monitored watering, pruning to increase airflow and decrease humidity in the soil, as well as making sure that areas where potential hosts are planted are not prone to flooding, often times this includes planting on an incline.

Chemical control methods for P. palmivora include: protectant fungicides such as the Bordeaux mixture, phosphonates which control the mycelial growth of the pathogen, dithiocarbamates such as Mancozeb, and phenylamides which control the spread of the pathogen from the roots of the host. Host resistance is also a method of controlling the pathogen; resistant plants generally have thicker cuticles which inhibits the ability of the pathogen to enter the host.

Non-chemical control in papaya

Root rot of papaya seedlings, caused by P. palmivora, in replant fields can be controlled with the virgin soil technique. Virgin soil (soil in which papaya has never been grown in before) is placed in planting holes about 30 cm in diameter and 10 cm deep with a mound about 4 cm high. Roots of papaya plants are protected by the virgin soil during the susceptible stage, and become resistant to the pathogen when they extend to the infested soil. Trees established with the virgin soil method in the replant fields produce fruit as abundantly as those growing in the first planting fields. The virgin soil method has the advantages of being relatively inexpensive, very effective and nonhazardous.

Phytophthora sojae

Host resistance is the primary method of control for Phytophthora sojae. There are three types of resistance: R gene mediated resistance, root resistance, and partial resistance. Currently there are 14 Rps genes, meaning 14 different single-resistance genes, which have been identified for R-gene mediated resistance and mapped in the soybean genome. Phytophthora sojae can also be controlled using fungicides. For example, Metalaxyl, a fungicide that is specifically used for oomycetes, is used for treating soybean seeds. It’s used to prevent seed decay and pre-emergence damping off.

Phytophthora infestans

P. infestans is still a difficult disease to control. There are many chemical in agriculture for the control of both damage to the foliage and infections of the tuber. A few of the most common foliar-applied fungicides are Ridomil, a Gavel/Super Tin tank mix, and Previcur Flex. All of the aforementioned fungicides need to be tank mixed with a broad-spectrum fungicide such as mancozeb or chlorothalonil not just for resistance management but also because the potato plants will be attacked by other pathogens at the same time.

Phytophthora citrophthora

Irrigation water has been treated. Electrolytically generated chlorine injected into citrus micro-irrigation systems effectively killed propagules of P. citrophthora, but soil populations under citrus trees in the field were unaffected by chlorinated water and no chlorine-induced phytotoxicity was observed (Grech and Rijkenberg, 1992. Fungicides have also been applied to trees via sprinkler irrigation systems.

Resistant rootstocks have been used for many years to control root, foot and stem infections on citrus caused by P. citrophthora. Selection of these depends on cultural factors and their resistance to some viruses, such as citrus tristeza closterovirus (CTV), and to the nematode Tylenchulus semipenetrans.

Citrus cultivars and related species which exhibit resistance are used in breeding programmes to produce hybrid rootstocks .Stocks with resistance include trifoliate (Poncirus trifoliata), sour orange (Citrus aurantium), mandarin (C. reticulata), and several types of citrus such as Benton, Carrizo and Troyer citrange.

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**BOTANICALS: A NATURAL APPROACH TO CONTROL PLANT PARASITIC NEMATODES**

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**Abstract**

Despite the usefulness of nematicidal compounds in agricultural practices, their excessive use has led to their reduced efficacy under field conditions.Biopesticides and specifically bioinsecticides constitute a desirable component of pest management. Botanical pesticides are derived from plants and their use in pest management is as old as agricultural practice. Growth of botanical pesticides constitutes around 7% of the total market share (Isman, 2015). Botanicals can have insecticidal activities, repellence to pest, modifier to insect behavior, antifeedent activity, toxicity to mites, snails, slugs, nematodes, bacteria, fungus and other crop pests (Chitwood, 2002. It has been estimated that nearly 2400 species of plants in India possess pesticidal properties (Singh and Saratchandra, 2002). Botanical pesticides are recognized as safe in agriculture. Hence, they have been regarded as attractive alternatives to synthetic chemical pesticides for the pest management (Dang et al., 2012). Researchers prefer to use botanicals in pest management because of their target specific nature and less phytotoxicity. They are degraded faster in sunlight and moisture. Neem based formulations, Azadirachtin (Achook® 0.15% EC and Nimbecidine® 0.03% EC) have proved highly active against Meloidogyne incognita in tomato plants. They are good alternative to synthetic pesticides because diverse groups of phytochemicals act as a major barrier to herbivory (Miresmailli and Isman, 2014). Identification of new effective pesticidal compounds is essential to contest increasing resistance (El-Wakeil, 2013). Future research work should concentrate on screening of new plants and isolation of new bioactive molecules. The use of botanicals is emerging in crop protection to save the environment from pesticide pollution, which is a worldwide problem.

Nematodes are roundworms, invertebrate multicellular animals, have well developed organs/systems like higher animals. They occupy all conceivable habitats of this earth, they may be freeliving or parasitic, visible or invisible to the eyes. They can be categorized into (i) Free living - they feed on dead organic matter or microorganisms, and are present everywhere in terrestrial or aquatic habitats; they are microscopic. (ii) Parasitic – they are parasitic on human beings, invertebrate and vertebrate animals and plants. Plant parasitic nematodes are microscopic, mostly live in soil water, are obligate parasites, and have a small hypodermic movable needle-like structure (stylet) in their snout with which they puncture, enter and feed on plant cell sap and attack underground plant parts like roots, tubers, bulbs etc., rarely they attack shoots also. Some remain in soil only and feed on roots from outside (ectoparasites). Others enter the roots partially (semiendoparasites) or completely (endoparasites). While inside the roots, they may keep on migrating within the plant tissues (migratory endoparasites), but some of them do not move after penetration (sedentary endoparasites). The latter category is most damaging as they modify plant cells to feed them continuously and become swollen like sac. Parasitically, PPNs could be differentiated into above ground feeder (Aphelechides besseyi, Anguina tritici, Ditylenchus dipsaci) and below ground feeder (Meloidogyne, Heteroderda, Globodera). Below ground feeders are called as “invisible enemy” due to lack of conspicuous diagnostic symptom and their hidden mode of life cycle. Below ground feeder includes ectoparasites like Xiphinema, Trichodorus, Belonolaimus are migratory in nature, parasitizes externally either on root epidermis or cortical tissue and serves as potential virus vector for many crops and endoparasites are either migratory or sedentary in nature. Migratory endoparasite (Pratylenchus, Radopholus, and Hirschmaniella) moves to cortical cell for fulfilling their nutritional demand and causes severe necrosis. Sedentary group are highly evolved group of parasite develops an intimate relationship with their host modifying feeding cell in pericycle or vascular regions. Sedentary group further can be differentiated as sedentary semi-endoparasites (Rotylenchulus, Tylenchulus) and sedentary endoparasites (Globodera, Heteroderda, Meloidogyne). Parasitism leads to damage of root cells, reduction of root system resulting reduction of nutrient and water supply to above ground parts ultimately lowering the yield of crops.

Plant parasitic nematodes are most devastating pathogens for major food, fiber, fodder, oilseeds, vegetable, ornamental and horticultural crops. More than 4100 species of PPNs have been reported. Economic damage attributed by PPNs is 173 billion dollar annually (Elling et al., 2013). A wide range of strategies are available to combat the PPNs problem such as cultural practices like land falling normally practiced due to farmers willing in multiple cropping scheme in a year. Crop rotation not very adaptive due to non-availability of economically important crop. Physical method like Soil solarisation involves covering of the soil with clear plastic, entrapping the radiation and generation of heat, which kills the nematode larvae in soil. Soil solarization limited for use in...
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small area and tropical climatic regions. Deep summer ploughing exposes the nematode larvae to heat and moisture stress, thus killing the infective larvae. Chemical method includes use of fumigants (Methyl bromide) and non-fumigant (Organophosphate, carbamate) are in phase of withdrawal due to ground water contamination and severe non targeted effect. However, use of resistant cultivar is most economically feasible, adaptive, and novel strategy to overcome PPNs. But due to drawback of (1) limited availability of vertical R-gene sources, (2) break down of resistance under high temperature and against races, (3) species barrier during transfer among crops (4) transfer of undesirable trait, (5) long duration to breed resistant cultivar lowers the effectiveness of strategy.

Frequent application of synthetic organic pesticides resulted into pest resistance and outbreak. Most pesticidal compounds have four major classes, the organochlorines, organo-phosphates, the carbamates and pyrethroids. Out of these organophosphates and carbamates are the major classes in use today. Problems of pesticides are resistance and negative impacts on non-target organisms including man and the environment. Proper use of the active ingredients present in the plants may be reduced many environmental problems such as development of resistance in pests to pesticides, resurgence of target and non-target pests, destruction of beneficial organisms and pesticides residue in host plants (Singh and Saratchandra, 2002. The alternative methods of insect pest control are being investigated and use of organochlorine insecticides has been banned in developed countries. Botanicals are the most promising source and under extensive trails for their biological activity against various pests. Hence, plants are wealthy sources of bioactive compounds need to be documented in detail.

Botanicals vs. Synthetic Chemicals

Many plants generate chemicals that are toxic to different pests for their self-defense purposes. Because these naturally occurring pesticides are derived from plants, they are called botanical pesticides or botanicals. Botanical pesticides were commonly used all over the world to defend against insect pests before World War II. However, just before the war, a highly effective “synthetic” (man-made) insecticide called DDT was introduced which changed the nature of pest control worldwide. Because these chemicals were cheaper, easier to apply and longer lasting, other synthetic pesticides soon followed, which quickly displaced botanicals in the marketplace and greatly slowed the research and development of natural, botanical compounds. Unluckily, these synthetic pesticides target a nervous system common to people and mammals, and can be toxic to fish and the environment. Many of the chemicals persist for long periods and cause residual problems. Insect pests have also developed resistance over time to many of the synthetic chemicals. Awareness of the potential health and environmental hazards of many residual synthetic pesticides increases, and pests become resistant to more synthetic compounds, interest in plant-derived pesticides is increasing (Basu et al, 1998. Degrade rapidly in sunlight, air and moisture and by detoxification enzymes are the properties of botanicals. Rapid breakdown means less persistence and reduced risk to non target organisms. Though specific timing and/or more frequent applications may be necessary.

Botanical pesticides are fast acting. Although death may not occur for several hours or days, most botanicals have low to moderate mammalian toxicity. Some botanicals quickly breakdown or are metabolized inside bodies of their target pests by enzymes. Breakdown may occur rapidly. To inhibit certain detoxification enzymes in insects synergist may be added to a compound. The insecticidal action of the product can enhances by this action. Synergists have low in toxicity, low or no inherent pesticidal properties, and very short residual activity. Pyrethrins are often mixed with a synergist such as piperonyl butoxide (PBO) to increase their effectiveness of the products. Rapid breakdown and fast action make botanicals more selective to certain plant feeding pests and less harmful to beneficial insects. Most botanicals are not phytotoxic (toxic to plants. However nicotine sulfate may be toxic to some vegetables and ornamentals. Botanicals are more expensive than synthetics and some are no longer commercially available (e.g. Nicotine. The strength of some botanicals may vary from one source or batch to the next. Data on effectiveness and long term (chronic) toxicity to mammals are unavailable for some botanicals. Tolerance for residues of some botanicals on food crops has not been established. Botanical pesticides include nicotine from tobacco, pyrethrums from chrysanthemums, derris from cabbages, rotenone from beans, sabadilla from lilies, ryania from ryania shrub, limonene from citrus peel, and neem from the tropical neem tree. Nearly all, other than nicotine have low levels of toxicity in mammals and birds and create few unfavorable environmental effects.

Botanicals as Fungicide, Insecticides and Nematicides:

Plant extracts or phytochemicals provide attractive alternative to presently used synthetic fungicides as regards controlling phytopathogenic fungi, since they constitute a rich source of bioactive molecules (Castillo, 2003. Monoterpene isolated from essential oil of Carum carvi exhibited fungicidal activity against potato tubers from rotting (Anonymous, 1994. The essential oil and methanol extract and derived fractions of Metasequoia glyptostroboides showed vast potential of antifungal activity against Fusarium oxysporum, Fusarium solani and Sclerotinia sclerotiorum. Dongre et al., 2013.a-cedrol isolated from essential oil of Thuja orientalis have antifungal activity for controlling Alternaria alternata.

The use of botanicals as insecticides against crop pests is gaining importance in recent years. The organic synthetic insecticides are more hazardous, leave toxic residues in food products, and are not easily
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biodegradable; besides their influence on the environment and public health is harmful. Unlike synthetic chemicals that kill both pests and predators outright, the natural insecticides are relatively inactive against the later. Most of the botanical insecticides are easily biodegradable and their supply can be made at cheaper rate by regular cultivation. Among the natural insecticides rotenone from *Derris elliptica*, nicotine from tobacco leaf, pyrethrins from pyrethrum flowers (*Chrysanthemum cinerariaefolium*) and azadirachtin from neem (*Azadirachta indica*) have attained commercial importance. Essential oils of cumin (*Cuminum cyminum*), anise (*Pimpinella anisum*), oregano (*Origanum syriacum var. bevanii*) and eucalyptus (*Eucalyptus camaldulensis*) were effective as fumigants against the cotton aphid (*Aphis gossypii*) and carmine spider mite (*Tetranychus cinnabarinus*) (*Trifolonofo & Atahasov, 2009*). Contact, fumigant and antifeedant effects of a range of essential oil constituents (cinnamaldehyde, and -pinene) have been demonstrated against the maize weevil (*Sitophilus zeamais*) and the red flour beetle (*Trichobium castaneum*) (*Huang et al., 2011*). *Mentha spicata* (*Lamiaceae*) root extracts which contains flavonoids and phenolic compounds found effective against the juveniles of *Meloidogyne javanica* (*Alakabi et al., 2016*).

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<td><strong>Differentiators</strong></td>
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<td>Active ingredients</td>
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<td>Manufacturing</td>
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<td>Shelf life</td>
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<td>Production cost</td>
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<td>Applications</td>
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<td>Regulatory hurdles</td>
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**Methods of Application of Botanicals:**

**Organic Soil Amendments**

Amending the soil with organic materials entails the incorporation of such into the soil. This type of treatment has been found promising and / or effective, in the control of several plant parasitic nematodes. Earlier reports on the use of organic soil amendments to control plant parasitic nematodes were not encouraged because of the inherent problems in their uses such as their bulkiness and slow activity.

**Dried Leaves**

The addition of dried leaves to soil for the control of plant parasitic nematodes has been extensively documented. Akhtar and Alam (2001) presented *Calotropis procera* as the most effective against freshly hatched juveniles of *Meloidogyne incognita*. It was better than *Azadirachta indica* (neem), *Ricinus communis* and a host of other plant leaves evaluated against root knot nematodes on tomato and chilli. Ek-Wakei et al., 2013 applied dried leaves of *Azadirachta indica*, *Cymbopogon citratus*, *Acacia alata*, *Ocimum gratissimum* and *Acalypha ciliata* in the control of the root knot nematode, *Meloidogyne incognita* on tomato in the screen house. The striking observation was that *A. ciliata* and *C. citratus* supported higher production of males than female nematodes, revealing that there were substances deleterious to the development of the nematodes. Moreover, damage potential of the nematode is reduced as the male nematodes do not feed.

**Seed Powders and Cakes**

Das (2014) made comparative studies on the nematicidal properties of *Typhonium trilobatum* and *Melia azedarach* corn and powders respectively. They found *T. trilobatum* at a dose of 0.75 g / kg soil to control 18% more nematodes than *M. azedarach* in pre-plant application. The effects of seed cakes on root-knot nematodes were also documented. Suppression of nematode population (*Shadunget et al., 2015*), improved plant yield reduction in total number of egg masses, males and females per plant (Ranjit, 2009) were among the effects recorded. Shaukat *et al.* (2001) reported castor, mustard, neem mahu and groundnut seed cakes to have exhibited suppressive activity on the *Meloidogyne* spp. used in their investigation. Maximum reduction in the number of root-knot nematode was also recorded from neem cake-amended soil in Mukhtar and Pervaz (2003) study.

**Green Manure**

The use of green manure in suppressing the population of *Meloidogyne* spp. has also been reported. Manju *et al.,* (2015) obtained greatest reduction in *M. chitwoodi* population density by cropping rapeseed (*Brassica rapa*) for two months and then incorporating it into the soil as green manure. Muniasamy *et al.,* (2010) went ahead to support this earlier finding and also reported wheat green manure to be inferior to the rapeseed green manure treatment. However, Jourand *et al.,* (2004) could not support this report this report when they observed that
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incorporating 30-60 t/ha green biomass of rapeseed into the soil six months after planting did not affect the population densities of *M. incognita* and *M. javanica.*

**Plant Extracts**

Literature is replete on the nematicidal efficacy of plants and plant products. *Mentha spicata* (Lamiaceae) root extracts which contains flavonoids and phenolic compounds found effective against the juveniles of *Meloidogyne javanica* (Al Saba *et al.*, 2001. Mint aqueous extracts exhibited significant nematicidal activity against *Meloidogyne incognita* (Chitwood, 2002. menthol (28.8 %), α-pinene (16.4 %), methylene (12.7 %), α-terpineol (6.3 %) and limonene (5.5 %), from *M. canadensis* exhibited nematicidal activity against *Heterodera avenae* and *Meloidogyne incognita* with LC₅₀ values of 385.7μg/ml and 139.0μg/ml, respectively (Qamar *et al.*, 1989. Methanol extracts from *Dryobalanops aromatica* (DA) and *Mentha haplocalyx* var. *piperascens* (MH) and their constituents found effective against *Meloidogyne incognita* (Guleria *et al.*, 2009. Different extraction methods have been employed by investigators and their effects on egg hatching and freshly hatched juveniles were extensively documented. Different modes of application were also employed by different investigators.

**Compatibility of Organic Soil Amendments with Other Cultural Practices**

There are reports to support the recommendation of organic soil amendments with other cultural practices. Akhtar and Alam (2001) presented results indicating that combining organic amendments with inter-cropping in integrated method gave better control than either treatment singly. Gabhe *et al.* (2006) also reported synergistic results in applying different rates of wheat straw in combination with soil solarization and fumigant nematicides.

**Mode of Application**

**Soil Drench**

Permanathan (2000) applied water extracts of neem leaves as soil drench and they obtained effective control of *Pratylenchus zeae* on maize. In an investigation of the influence of seaweed concentrates (Kelpak 66) on the reproduction of *Pratylenchus zeae* on maize, Sono *et al.* (2000) applied different concentrations of the product as a soil drench. Result obtained showed 47-60% suppression in reproduction of *P. zeae* compared with the control. In view of this report, Pavaraj *et al.* (2010) investigated the role of seaweed extracts, *Ascophyllum nodosum* in the reduction of fecundity of *M. javanica* and applied their dilutions as a soil drench. They obtained reduction in the infestation of tomato roots by the nematodes compared with the untreated control.

**Root Dip**

Effects of rhizome dip of pared plaitain in crude water extracts of *Acalypha wilkesiana* leaves and at different duration of exposure in comparison with the conventional hot water dip, in protecting against nematode damage under field conditions(Jourando *et al* 2004. The most effective treatment in terms of reduction in nematode population density and related damage was obtained with 15 min. exposure to the extract on the field.

**Foliar Application**

Literature is restrictive on the foliar application use of the extracts from botanicals. Abolusoro *et al.* (2005) compared among other applications, the foliar application and soil drench when they tested *Vernonia amygdalina* (bitter leaf) leaf extract in the control of *M. incognita*. Their results showed that foliar application did not show phytotoxicity on the soybean test plant, but soil drench treatments were phytotoxic on the test plants. When they compared their extract treatments with the recommended dose of carbofuran, they observed that carbofuran reduced the egg hatch of *M. incognita* while the bitter leaf extract treatments completely suppressed egg hatch of the nematode.

**Geographic patterns: the dominance of India, China, and Brazil:**

Examination of the country of origin for these studies indicates some clear trends. India dominates the category: from 2000 to 2010, numbers of published papers on botanical insecticides increased from around 125 per year to just over 300 per year. Same period, papers from China increased from around 30 per year to around 100 per year, and in Brazil the numbers increased from around 20 per year to around 80 per year. In fact, India, China, and Brazil accounted for 494 (40.9%) of the 1207 botanical insecticide articles published in 2012. Recent growth in this type has been more modest in some African (Egypt and Nigeria) and Middle Eastern countries (Iran and Turkey), where the numbers of articles have remained essentially constant in most developed countries (UK, USA, Germany, and Japan.

**Phytonematicides Used on a Commercial Scale**

Effective application of phytonematicides in large commercial and smallholding farming systems in SA involves the use of crude extracts in granular and liquid formulations (Nath *et al.*, 2002. Plant materials commonly used in the phytonematicide Nemalan include lantana (*Lantana camara*) shoots and wild garlic (*Tulbaghia violacea*) (Daneel *et al*. 2014a. Fruits from *C. africanaus* and *C. myriocarpus* (Mashela 2014) are used in the production of nemafric-BL and nemarioc-AL, respectively (Mashela. 2014. The centre of
biodiversity of the two *Cucumis* spp. is the Limpopo Province (Khurma *et al.* 1997), whereas lantana is an invader plant (Daneel *et al.* 2014a. Active substances in lantana leaves are saponins and cucurbitacins in fruits of the two *Cucumis* spp. (Ujvary,2002. Combining plant organs with different a.s., for example, *C. myriocarpus*, *L. javanica* and *R. communis*, resulted in synergistic effects on the suppression of nematode population densities (Mashela *et al.* 2007).

Plant parts (powdered leaf meals) of non-crop plant species used in traditional medicine in SA were selected and examined for their nematicidal activity as soil amendments on *M. incognita* race 2 (Moosavi ,2012. Parts of these non-crop species are locally known as ‘muti’ as they are considered to have certain medicinal properties. Traditional healers in SA frequently use these mutis to treat human and domestic animals for various ailments. Living specimens of these plant species, as well as supplies of dried and finely ground material made from them, can be found in abundance in SA in the rural areas and communities of the lowveld in the Mpumalanga, Limpopo and KwaZulu-Natal provinces. These plant species contain chemicals such as alkaloids, diterpenes, diterpenoids, esters, fatty acids, ingenol, oxalic acid and terpenoids. The observed general effects of these traditional medicines on humans at prescribed dosage rates suggested that they might be toxic to small multicellular organisms such as plant-parasitic nematodes. Nine plant species were identified, collected and examined for their nematicidal activity and plant growth enhancement in glasshouse trials. Five of these plant species, namely, cactus vine (*Cissus cactiformis*), Candelabra tree (*Euphorbia ingens*), Bushveld head-bean tree (*Maerua angolensis*), Dead-man’s tree (*Synadenium capulare*) and Toad tree (*Tabernaemontana elegans*) were further tested under field conditions (Manju *et al.* 2014a. Soil amendments of powdered leaf meals of *C. cactiformis*, *M. angolensis* and *T. elegans* were shown to reduce the number of eggs and J2 of *M. incognita* race 2 in tomato (Lalita, 2012).

**Perspective of botanical pesticides**

BP shows a number of positive aspects that cannot be ignored even by strict advocates of synthetic products. Their environmental safety is one of their key positive aspects. Though their opponents often object that BPs may contain non-selective substances that may have a negative impact on non-target organisms, many tests have shown that upon properly targeted application, the active substances of BPs are very friendly to many non-target organism. Known that the active substances are natural secondary metabolites of the plants, BP residues are degraded easily and rapidly through natural degradation mechanisms (Turner, 19980. This fact is furthermore improved in farming products or basic sub- substances where no irrelevant carriers and emulsifiers are used. One more undoubted positive aspect is that BPs (apart from exceptions) have extracts from plants which do not contain any substances toxic to homeothermic animals. Products based on plant extracts generally have synergistically acting mixtures of active substances that show various mechanisms of action , which prevents the development of resistant pest populations (Isman 2006; Miresmailli & Isman 2014. A further finding that is not only interesting but also important is that much advanced biodiversity as well as occurrence of pollinators and natural enemies of pests are seen in crops treated with plant extracts unlike crops which are treated using synthetic insecticides (Abolusoro*et al.* 2005.

These positive aspects confirm the strong belief that BPs should play an important against harmful insects. This is why every year scientists come up with new information about the pesticidal effects of plant metabolites. Based on the number of scientific studies focused on the research of plant substances with insecticidal effects, it seems that commercial BPs should occupy an important position in the market. Although research of secondary plant metabolites is increasing and has seen its renaissance (Isman 2015), there is very little scientific knowledge which has been applied in practice. This is due to some reasons.

**Lack of suitable plant material**

Many perspective plants are very difficult to grow in such a way that they could provide a sufficient amount of high-quality material appropriate for the isolation of active substances. Hence, nearly all commercial products are manufactured only from a few plant species that provide suitably high yields (Isman 2015; Pavela *et al.* 2016).

**Toxicity of certain active ingredients of some botanical pesticides (in mg/kg)**

<table>
<thead>
<tr>
<th>Generic name</th>
<th>Oral LD$_{50}$</th>
<th>Dermal LD$_{50}$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nicotine</td>
<td>50–60</td>
<td>50</td>
</tr>
<tr>
<td>Rotenone</td>
<td>60–1,300a</td>
<td>940–3,000</td>
</tr>
<tr>
<td>Sabadilla</td>
<td>4,000</td>
<td>–</td>
</tr>
<tr>
<td>Rynia</td>
<td>750–1,200</td>
<td>4,000</td>
</tr>
<tr>
<td>Pyrethrins</td>
<td>1,200–1,500</td>
<td>&gt; 1,800</td>
</tr>
<tr>
<td>d-Limonene</td>
<td>&gt; 4,000</td>
<td>&gt; 5,000</td>
</tr>
<tr>
<td>Linalool</td>
<td>2,440–3,180</td>
<td>3,578–8,374</td>
</tr>
<tr>
<td>Neem oil</td>
<td>&gt; 5,000</td>
<td>&gt; 2,000</td>
</tr>
</tbody>
</table>
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**Insufficient support from the government**
The greatest problem encountered by potential as well as presented BI producers is the costly and lengthy registration process of developed products, often criticized by specialists (Isman 2015). Especially severe criteria for the registration of pesticides have been set in some countries including EU states; the criteria have been tightened up in order to avoid environmental and health problems (Shaukat et al. 2002). BIs are thus viewed as critically as synthetically produced substances, even though they are products that often have the same substances as, for example, commonly used herbal teas or spices. Although, biofoods are supported and their popularity has been rising among consumers worldwide. On the other hand, sufficient rules are missing that would enable small and medium-sized companies to quickly market a sufficient spectrum of botanical pesticides, which would make it possible to strengthen environmental production of plant products (Isman & Machial, 2006). In contrast, various plant essential oils (e.g. clove, spearmint, citronella) have been approved as pesticides, which would enable small and medium-sized companies to quickly market a sufficient spectrum of botanical pesticides, which would make it possible to strengthen environmental production of plant products (Isman & Machial, 2006).

**The quality of BI formulations**
Quality should be improved so that an adequate persistence of their effect, quality and stability of the products are guaranteed. A number of scientific sites are currently focused on this type of research. In an attempt to increase biological efficacy, various so-called green syntheses of nanoparticles seem to be perspective, where the insecticidal efficacy of plant extracts is extensively improved. In the past few years, micro- and nano-encapsulation trials have been examined to see if they can provide a controlled release of botanical insecticides (Facchini, 2001). These technologies can prolong the efficiency of botanical insecticides over extended periods of time. Despite of these formulation developments, the controlled release remained at the entire formula mixture level without calling for changes in the volatilisation and biological features of specific elements in the botanical materials used in the production of botanical insecticides. A better understanding about the behavior and bioactivity of individual components of botanical insecticides in addition to new advanced methods of compartmentalization and formulation is needed to allow us better degrees of control over the accessibility and activity of specific elements of intricate botanical mixtures. As a result, this should assist get better efficiency of botanical insecticides (Chitwood et al., 2003; Miresmailli & Isman 2014).

**Drawbacks of botanical nematicides**
- Unavailability of the basic materials from which the amendments are derived.
- The rather long period needed for the microbial decomposition of the amendments before they can be applied on the field.
- The need for large quantities of the amendments (ranging from 10-500 metric tonnes (MT) / ha.
- The high transportation costs to haul the amendments to the field where they will be used.
- Excessive lowering of soil pH.
- Inconsistency of the results.

**Challenges for Improving the Use of Botanicals**
- Literature is replete on the activities of botanicals in the control of plant parasitic nematodes while only limited information is available on the active components of these materials.
- Efforts should be directed at identifying active components with the aim to formulating new environmentally friendly nematicides from them.
- Mode of application of botanicals needs to be harmonized while the application rate should be established.
- A way forward would be to first standardize procedures in order to allow comparison of results.
- Domestication of some of these plants would be necessary as several of them are only in the wild while others are given less attention since they are not food crops.

**Future Perspective**
Application of synthetic pesticides is a usual practice to ward off infestation of insect pests and diseases from field crops. Though, as these conventional chemicals are reported to cause environmental load and menace to public health, the world trends in pesticide research now a day calls for discovery of safer and eco-friendly chemicals for pest control. Plants are prosperous resource of chemicals that are toxic to pests. When extracted from plants these chemicals are called botanicals. Botanicals are endowed with a spectrum of properties like insecticidal activities, repellence to pest, insect behavior modifier, antifeedent activity, toxicity to mites, snails, slugs, nematodes and other agricultural pests (Digrak, 1999). Separately, they also have antifungal, antiviral and antibacterial properties. They have variable toxicity to non target organisms, even though as a group they tend to be less toxic to mammal (with + ve exception to nicotine), than non botanicals. The utilization of botanicals is
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now emerging as one of the major means to protect crops and their products and the environment from pesticide pollution, which is a global problem. Since most of them usually degrade within few days, and sometimes within a few hours, these pesticides must be applied more frequently. More frequent application coupled with very high cost of production generally makes botanicals more expensive to use than their synthetic counterparts. In spite of the wide acknowledgment that many plants have insecticidal properties, only a handful of pest control products directly obtained from plants including pyrethrum (pyrethrin), neem (Azadirachta indica), rotenone, quassia and tomato leaf extract are in use because the commercialization of new botanicals can be held up by a number of issues. In addition regulatory protocols being designed, keeping in view the synthetic chemicals constitute a obstacle to the commercialization of potentially useful botanicals, mostly due to the presence of complex mixtures of active ingredients. Though, in vision of the negative property of the synthetic chemicals on human health, environment and ecosystem the regulatory authorities are likely to look more favourably on the alternative products so that these new products can be mobilized into the market with less hurdles (Isman, 2006). Future research efforts, consequently, should be heading for not only towards the development and application of known botanicals but also on screening more plants and isolate new and new bioactive molecules which have pest controlling properties or can provide as leads for the development of eco-friendly pesticides. Reasons for limited commercial development of botanical pesticides are their comparatively slow action, variable efficiency, lack of persistence and inconsistent availability when compared to synthetic insecticides. Other barriers to commercialization of botanical pesticides are lack of the natural resource, standardization, quality control and registration.

Reference


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PSEUDOMONAS SPP. IS ANEmerging BIOCONTROL AGENT

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Plant diseases cause 13 - 20 % losses in crop production worldwide (Agrios, 2005). Control of plant disease by chemical can be spectacular but the accumulation of harmful chemical residue sometimes causes serious ecological problems (Aktar et al., 2009). Biocontrol agents are economical; suppress the inoculum load of the target pathogen, long lasting and free from residual side effect. Fungi in the genus Trichoderma and Bacteria in the genera of Pseudomonas and Bacillus are increasing interest as bioproducts against plant diseases (Janarthanam, 2013). Biocontrol agents in general and Pseudomonas fluorescens in particular have gained importance as a component of Integrated Pest Management for sustainable agriculture. Pseudomonas fluorescens belong to Plant Growth Promoting Rhizobacteria (PGPR), the important group of bacteria that play a major role in the plant growth promotion, induced systemic resistance, biological control of plant pathogens etc. Pseudomonas is a genus of Gram-negative, Aerobic Gamma proteobacteria, belonging to the family Pseudomonadaeae and containing 191 validly described species (EUZeBY, J.P., 1997). The members of the genus demonstrate a great deal of metabolic diversity and consequently are able to colonize a wide range of niches. And easy to culture in vitro and availability of an increasing number of Pseudomonas strain genome sequences has made the genus an excellent focus for scientific research; the best studied species include P. aeruginosa in its role as an opportunistic human pathogen, the plant pathogen P. syringae, the soil bacterium P. putida, and the plant growth-promoting P. fluorescens.

Pseudomonas fluorescens as biocontrol agent and their properties

The bacteria P. fluorescens possess many traits that make them well suited as biocontrol and growth-promoting agents. These include the ability to Grow rapidly in vitro and to be mass produced. Rapidly utilize seed and root exudates. Colonize and multiply in the rhizosphere and spermsphere environments and in the interior of the plants. Produce a wide spectrum of bioactive metabolites. Compete aggressively with other micro-organisms. Adaptive to environmental stresses Inexpensive.

Mode of action against plant pathogens

Antibiotic Production: The P. fluorescens is very effective antibiotic producer. Many secondary metabolites of P. fluorescens acts as antibiotics against plant pathogens. The P. fluorescens produces antifungal compounds which are fungistatic, inhibiting spore germination and lysis of fungal mycelia. Phenazine-1-Carboxylic Acid (PCA), 2, 4-Diacetylphloroglucinol (DAPG), Pyocinine, Pyrrolnitrin, Pyoluteorin, Oomycin-A etc.

Siderophores Production: Siderophores are extra cellular, low-molecular weight compounds with very high affinity for ferric iron. As siderophore sequester the limited supply of iron in the rhizosphere, they limits it’s availability to pathogens and ultimately suppress their growth. Ferribactin, Ferriochrome, Ferroxamine B, Pseudobactin, Pyochelin, Pyoverdine.

Induced Systemic Resistance: The P. fluorescens induce systemic resistance in plants that is phenotypically similar to Systemic Acquired Resistance (SAR). Induction of resistance by P. fluorescens is mainly through the Production of phytoalexins, increased lignifications, Production of PR-protein in the induced plants.

Competition: The P. fluorescens preempt the establishment of other rhizosphere microorganisms through competition for favored sites on the roots and in the rhizosphere.

Hydrogen cyanide production: Hydrogen cyanide (HCN) is representative of class of volatile inhibitors. The P. fluorescens produces HCN which can check growth of phytopathogen although the producer bacterium itself resistance.

Plant Growth Promotion Antibiotic Production: The P. fluorescens promotes plant growth by production of phytohormones such as Auxins and Gibberelmins and also by Phosphate solubilization.

Ecology: Pseudomonas fluorescens are commensal species with plants, allowing plants to attain key nutrients, degrading pollutants, and suppressing pathogens via antibiotic productions. These microbes produce secondary metabolites that suppress plant disease and signal gene expression to neighboring cells inhabiting the rhizosphere. Pseudomonas also uses siderophores from other microorganism to obtain iron which increases their survival in iron-limited environments. Plants provide these organisms with nutrients and shelter against stressful environments. One of many byproducts of plant cells includes active oxygen such as superoxide which are toxic.
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to microbes. Rhizosphere bacteria such as *P. fluorescens* possess superoxide dismutases to convert superoxide to hydrogen peroxide and catalases to convert peroxide to water. The presence of these enzymes contributes to *Pseudomonas fluorescens*’s tolerance to oxidative stress.

**Pathology:** Despite their commensal nature, *Pseudomonas fluorescens* are nonpathogenic and lack virulence factors of other plant pathogens. In *Pseudomonas fluorescens* Pf-5, enzymes that degrade plant cell walls and their components such as cellulase, pectinase, or pectin lyase are not present. However, it is capable of breaking down some plant-derived carbohydrates, fatty acids, and oils and can hydrolyze proteins causing the spoilage of milk, meat, and fish. It has also been found to be an opportunistic pathogen in immune compromised fish like Koi which are commonly kept in backyard garden ponds. There are other species of *Pseudomonas* that are pathogens. *Pseudomonas syringae* is a plant pathogen that impacts food and biomass production. Diseases include bacterial specks on tomatoes and leaves which result in stunted growth and halo blight of beans. These microbes can spread by rain and are seed-borne pathogens.

Although *Pseudomonas fluorescens* typically has a low level of virulence, in 1997 four patients at the National Taiwan University Hospital developed *Pseudomonas fluorescens* bacteremia. These patients had been treated in the chemotherapy room and had begun exhibiting symptoms such as fevers and chills. Eight cultures were isolated from catheters and from the blood of the four patients. All isolates were identified as *Pseudomonas fluorescens*. *Pseudomonas aeruginosa* are opportunistic human pathogens that are one of the main causes of human infections. These microbes live in diverse environments including soils, marshes, as well as plant and animal tissues, which show their nutritional versatility. Their resistance to antibiotics has made them dangerous pathogens. *Pseudomonas aeruginosa* are commonly seen in the lungs, especially those with cystic fibrosis. They are also present in urinary-tract infections, burn victims, and patients on respirators with hospital-acquired pneumonia.

**Application to biotechnology:** Studies done on *Pseudomonas fluorescens* have shown the microbe’s potential benefit in bioremediation against several strains of plant pathogens. The results of the experiment showed that at high concentrations all five strains of *Pseudomonas fluorescens* tested inhibit spore production by pathogenic plant fungus. Fungi such as *Alternaria cajani* and *Curvularia lunata* grow on plant surfaces causing disease and death of the plant. Plant treatment with *Pseudomonas fluorescens* can prevent these fungi from growing and spreading through spore production. *Pseudomonas fluorescens* grows at an optimum temperature of 25°C but can also survive in temperatures as low as 0°C. Therefore, it is rarely pathogenic in humans making it an effective microbe for treating crops since it is not able to survive in the human body. *Pseudomonas* species are effective against mold causing disease in produce such as apples and pears. This and further studies of *Pseudomonas fluorescens* will determine its effectiveness an alternative to chemical fungicides. Productions of secondary metabolites play an important role in plant disease suppression. Antibiotics such as pyrrolnitrin, pyoluteorin, and 2, 4-diacetylphloroglucinol that inhibit phytopathogen growth are produced by *Pseudomonas fluorescens* Pf-5. Diseases from *Rhizoctonia solani* and *Pythium ultimum* that affect cotton plants are inhibited by this strain. *Pseudomonas fluorescens* produces hydrogen cyanide and the siderophores pyochelene and pyoverdine which it uses to outcompete with many pathogenic bacteria for iron necessary for growth and suppress pathogens in the rhizosphere. The bacteria’s degrading ability has been applied to pollutants such as styrene, TNT and, polycyclic aromatic hydrocarbons. (4-6) *Pseudomonas fluorescens* produce exopolysaccharides which are used for protection against bacteriophages or dehydration as well as for defense against the host immune system. Polysaccharides are being used within the food, chemical, and agricultural industries.

**References**


ROLE OF BIOTECNOLOGICAL TECHNIQUES IN PLANT DISEASE MANAGEMENT

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Abstract

The future of sustainable agriculture will increasingly depend on the integration of biotechnology with traditional agricultural practices. Plant biotechnology is a precise process in which scientific techniques are used to develop molecular and cellular based technologies to improve plant productivity, quality and health, to improve the quality of plant products, or to prevent, reduce or eliminate constraints to plant productivity caused by diseases, pest organisms and environmental stresses. Plant biotechnology involves the modification of plant performance for a particular purpose. Genome segments from plant pathogenic fungi are widely used as vectors into which genes are inserted to make transgenic plants. This is of paramount importance to ensure efficacy and genetic integrity of the product and to protect intellectual genetic modification. Diseases can be caused by a variety of plant pathogens including fungi, bacteria, viruses and others. Plant pathogens represent real threat to world agriculture. Their management requires the use of techniques in transgenic technology, molecular biology, and genetics. These include, genes that express proteins, peptides, or antimicrobial compounds that are directly toxic to pathogens or that reduce their growth in situ, gene products that directly inhibit pathogen virulence products or enhance plant structural defense genes, that directly or indirectly activate general plant defense responses, and resistance genes involved in the hypersensitive response and in the interactions with a virulence factors. Biotechnology will enhance our understanding of the mechanisms that control a plant ability to recognize and defend itself against disease causing fungi.

Plant pathogens represent real threat to world agriculture. More than 70% of all major crop diseases are caused by fungi (Agrios, 2005). Crops of all kinds often suffer heavy losses. Fungal plant diseases are usually managed with applications of chemical fungicides. For some diseases, chemical control is very effective, but it is often non-specific in its effects, killing beneficial organisms as well as pathogens, and it may have undesirable health, safety, and environmental risks (Manczinger et al., 2002 and Cavrilesca and Chisti, 2005). Control of disease is a subject of great interest for biotechnologists. Biotechnology will enhance our understanding of the mechanisms that control a plant ability to recognize and defend itself against disease causing fungi (Punja, 2007). The future of sustainable agriculture will increasingly rely on the integration of biotechnology with traditional agricultural practices.

Biotechnology is a technology based on biology, especially when used in agriculture and food science. Various definitions are given for the term biotechnology. Biotechnology, an abbreviation of “biological technology”, has been defined as “the application of scientific and engineering principles to the processing of materials by biological agents to provide goods and services” (Alan, 1985). Elsewhere, Jones (1990) described biotechnology as comprising a continuum of technologies, ranging from traditional to modern biotechnologies. It is in the context of technologies that the OTA (1989) described biotechnology as “any technique that uses living organisms, or substances from these organisms, to improve plant for specific uses”. According to Persley (1992) traditional biotechnology covers well established and widely used technologies based on the commercial use of living organisms. These include the biotechnologies currently employed in breeding, fermentation, and many others (Amalu, 2004). Modern biotechnology on the other hand encompasses the uses of more recently developed technologies, particularly those based on the use of recombinant DNA technology, monoclonal antibodies, and new cell and tissue culture techniques, including novel bio-processing (Monti, 1992). The major techniques of biotechnology are tissue or cell culture, cell fusion, embryo transfer, recombinant DNA technology, etc and the age old techniques of fermentation. In recent years, bioinformatics is an interdisciplinary field which addresses biotechnology and biological problems using computational techniques, and makes the rapid organization and analysis of biological data possible. The field may also be referred to as computational biology, and can be defined as, “conceptualizing biology in terms of molecules and then applying informatics techniques to understand and organize the information associated with these molecules, on a large scale (Baxevanis et al., 2007). Bioinformatics and computational biology involve the use of techniques including applied, informatics, statistics, computer science, and biochemistry to solve biological problems usually on the molecular level. Major research efforts in the field include sequence alignment, gene finding, protein structure alignment, protein structure prediction, prediction of gene expression, and the modeling of evolution (Baxevanis and Ouellette, 2005).

Plant biotechnology is a precise process in which scientific techniques are used to develop molecular and cellular based technologies to improve plant productivity, quality and health, to improve the quality of plant products, or to prevent, reduce or eliminate constraints to plant productivity caused by diseases, pest organisms and environmental stresses.
products, or to prevent, reduce or eliminate constraints to plant productivity caused by diseases, pest organisms and environmental stresses (Azhaguvel et al., 2006. Plant biotechnology involves the modification of plant performance for a particular purpose. Genome segments from plant pathogenic fungi are widely used as vectors into which genes are inserted to make transgenic plants. This is of paramount importance to ensure efficacy and genetic integrity of the product and to protect intellectual genetic modification. This can be achieved in a number of ways include increasing or decreasing the activity of genes that are naturally present in an organism or transferring genes between individuals of the same species from one organism to another of different species across different types of living things.

Applications of Plant Biotechnology

Natural Transfer of Genes between Species

Natural movement of genes between species, often called horizontal gene transfer or lateral gene transfer, can occur because of gene transfer mediated by natural processes. This natural gene movement between species has been widely detected during genetic investigation of various natural mobile genetic elements, such as transposons, and retrotransposons that naturally translocate to new sites in a genome, and often move to new species over an evolutionary time scale. There are many types of natural mobile DNAs, and they have been detected abundantly in food crops such as rice, a species resistant to blast disease (Feschotte 2005).

Synthetic Transfer of Genes between Species:

(a) Callus and Suspension Cultures

Callus is an unorganized, proliferative mass of differentiated plant cells, and usually occurs naturally as wound response (Dhekney et al., 2007. It can be induced through culture of plant tissue on a medium usually containing relatively high levels of auxin, especially 2, 4-D (Gamborg, 2002). As a general rule, mutation breeding is difficult to control and frequently yields mosaic and deleterious changes. However, in some cases, beneficial changes can occur, for example, in sugarcane, plants were obtained that had resistance to the “Fiji” disease, which was a significant disease in the production of sugarcane (Pierik, 1997. Suspension cultures can be produced from non-embryogenic or embryogenic callus, and are commonly used these days by molecular biologists for transformation research. With many species the development of regenerable embryogenic cell suspensions has provided the opportunity for the production of transgenic plants. One of the most promising developments is the expression in banana and plantain of antimicrobial peptides, some of which have been shown to exert in vitro fungistatic activity to Mycosphaerella fijiensis and Fusarium oxysporum, the causal agents of black sigatoka and Panama wilt disease, respectively (Pierik, 1997).

(b) Protoplast

Protoplasts are produced by the enzymatic removal of the cell wall, through the use of mixtures of fungal cellulases, pectinases and hemicellulases in a solution of high osmotic potential. Removal of the cell wall then facilitates genetic manipulation of some kind. Fusion of protoplasts can be induced chemically or by electrofusision. After fusion has occurred the resulting culture usually contains a mixture of fusion products and parental types, and so some method has to be adopted to distinguish between these two. Plants with a crown and root system from different genotypes frequently form commercial plantations of Citrus. Recently there have been reports of somatic hybrids formed from protoplasts isolated from Citropsis reticulata and C. gilletiana, a species resistant to collar rot and tolerant to a number of diseases including tristeza (Pierik, 1997).

(c) Embryo Culture

In vitro culture techniques have important applications for the collecting, exchange and conservation of plant germplasm (Dhekney et al., 2007. Production of transgenic groundnut germplasm resistant to rust and late leaf spot diseases has been reported (Mace et al., 2006. Because of its large size, and its immediate germination after seed maturation, pant-collecting missions can be problematic. For germplasm exchange, the FAO/IBPGR technical guidelines for the safe movement of coconut Germplasm recommend that coconut germplasm be distributed as zygotic embryos in vitro. In the mid-90s the crop was wiped out with the introduction of Phytophthora colocasiae, (taro leaf blight. With the importation of taro leaf blight resistant material to Samoa, taro is now being grown again (Tien et al., 2000.

(d) Plant Breeding

Production of transgenic plants in wide-crosses by plant breeders has been a vital aspect of conventional plant breeding for about a century. Without it, security of our food supply against losses caused by crop diseases such as peanut Sclerotinia blight, rusts, and mildews would be severely compromised (Chenault et al., 2005 and Huang et al., 2005. In the 20th century, the introduction of alien probing into common foods was repeatedly achieved by traditional crop breeders through artificially overcoming fertility barriers. Novel genetic rearrangements of plant chromosomes, such as insertion of large blocks of rye genes into wheat chromosomes (“translocations”), have also been exploited widely for many decades and rust disease management (Huang et al., 2005).
Genetically Engineered Plants
Genetic engineering refers to artificial techniques capable of transferring genes from other organisms directly to recipient organisms (Gold, 2003). The techniques of genetic engineering can be used to manipulate the genetic material of a cell in order to produce a new characteristic in an organism. Genes from plants and microbes can be recombined and introduced into the living cells of any of these organisms (Azhaguvel et al., 2006). Transgenic recombinant plants are generated by adding one or more genes to a plant's genome, and the techniques frequently called transformation (Newell, 2000). Transgenic recombinant plants are identified as a class of genetically modified organism (GMO), usually only transgenic plants created by direct DNA manipulation are given much attention in public discussions (Osusky, 2004). Genetic engineering has the potential to provide a cornucopia of beneficial plant traits, particularly an enhanced ability to withstand or resist attack by plant pathogens (Chenault et al., 2005 and Punja, 2007. New approaches to plant disease control are particularly important for pathogens that are difficult to control by existing methods. Genetic engineering can help farmers increase crop yields and feed even more people (Amalu, 2004. The percentage of GMO plant resistant to diseases is approximately about 2% of total cultivated GMO plants (Gold, 2003).

Use of Molecular Markers
In molecular biology, DNA molecular-marking techniques have been used for some time. Genetic fingerprinting is used to identify particular desirable genes required for specific breeding plants (Azhaguvel et al., 2006. The DNA of the offspring from an F1, a segregating population, is examined using different marker technology (AFLPs, RFLPs, and RAPDs. Since the gene markers also segregate in these segregating populations, researchers try to find out which gene marker correlates best to disease resistance, allowing the location of resistance genes on a genetic map of the plant (Azhaguvel et al., 2006. Further germ plasmslines can be evaluated for resistance by using the marker and without putting the plants into a field trial. Existing successes include identifying markers for powdery mildew (Erysiphe graminis) and Rhyhchosporium secalis in barley and apple, Fusarium wilt, ascochyta blight in chickpea (Weeden, 1993) and peanut sclerotinia blight (Chenault et al., 2005).

Genes transfer in crop plants
Gene transfer by Agrobacterium tumefaciens is another powerful tool for plant genetic engineering (Azhaguvel et al., 2006 and Vande Valde, 2003). Transformation is usually achieved using gold particle bombardment or a soil bacterium (Agrobacterium tumefaciens) carrying an engineered plasmid vector, or carrier of selected extra genes. The process is used routinely to move genes into dicotyledonous plants. The “Gene Gun” is a popular tool used world-wide for genetically engineering plant cells (Kikkert and Reisch, 1996).

Tissue Culture
Tissue culture is a major component of plant genetic engineering. For example, banana tissue culture is a first-generation plant biotechnology tool used to curtail the spread of diseases, especially fungal wilt disease, by making disease-free planting material available for the propagation of new crops. Simple tissue culture techniques such as shoot-tip and embryo culture are well-developed in Africa and have greatly improved banana breeding, whereby the shoot meristem is extracted from the male flower and aseptically multiplied into hundreds of shoots for eventual planting in fields (Gamborg, 2002. Traditional breeding combines many genes from two parents at once. Several backcrosses may be needed to remove desired genes. Breeding by biotechnology, a single new gene is added to the genome. Since the late 1960s, all varieties of wheat released from one or more genes for resistance to stripe rust (Line, 2002).

Two kinds of resistance have been in use:

a. Race specific, single-gene, immunity expressed at all stages of plant development, in which the genetics of the host-pathogen interaction follows the gene-for-gene model.

b. Race nonspecific, multiple-gene partial type resistance expressed largely or entirely in adult plants, and in response to high temperatures. Race-specific, single gene immunity is readily defeated by the pathogen, with the result that each new gene deployed in a new variety selected eventually, and sometimes quickly, for a new race of the pathogen. Approximately ninety races of the stripe-rust pathogen now exist whereas only one was known in the 1960s. Nevertheless, through a combination of varieties with high-temperature adult-plant resistance, the use of several sources of single-gene resistance in isolines mixed to provide a multi-line and deployment of combinations of single genes as stacked genes, stripe rust remains largely under control through plant breeding (Line, 2002. The cells are then grown out to whole plants to make sure that the trait is actually expressed as wanted. The methods to incorporate the spliced genes are not precise in fact they are such that the DNA can be inserted in rather random ways. The insertion of the genetic material may cause other genes to be turned on or off which can cause major problems within the plant (Azhaguvel et al., 2006).
Transgenic crop varieties resistant to Fungal Diseases

Genes can be identified that confer resistance to fungal pathogens. For instance, many genes have been found that provide resistance to specific races of each pathogen (Gold, 2003. In many cases, resistance genes are available in the gene pool of cultivated plants and can be transferred to them. In the African region, two initiatives for fungal resistant transgenic varieties are reported, a field trial of transgenic strawberry with phytoalexin synthesis genes on transgenic maize for resistance to cob rot, both in South Africa (Amalu, 2004. The three initiatives for development of fungal resistant transgenic varieties reported in Eastern Europe were all in Bosnia and Herzegovina where laboratory testing of transgenic potato for resistance to Fusarium, Verticilium and Rhizoctonia has been initiated. In the Asian region, there is a field trial underway in China for transgenic cotton with resistance to Verticilium and Fusarium. A few countries in Latin America, mainly Argentina, Brazil and Cuba, are carrying out a number of activities on transgenic resistance to fungi, particularly on tropical fruit trees, with some results already being tested in the field. In this region, most of the activities for transgenic fungal resistance are reported in Cuba, in particular involving field trials of transgenic potato for late blight resistance, and fungal-resistant sugarcane. Other field trials in the Latin America region for transgenic fungal resistance are reported for maize, sunflower and wheat in Argentina, and tobacco in Mexico. Other countries involved in transgenic fungal resistance research are Argentina on alfalfa, Brazil on rice, barley and cocoa, Chile on grape and apple, Colombia on tree tomato, and Peru on potato for late blight resistance and Venezuela on sugarcane (Gold, 2003).

Late Blight resistant (GM) Potato as a noble approach

Late blight, caused by the oomycete pathogen Phytophthora infestans, is the most devastating potato disease in the world. All plant-breeding attempts to increase the plant’s resistance have so far had little success and have repeatedly been threatened by Phytophthora itself because the fungus is incredibly flexible and has developed lots of different strains. Today, in order to prevent the spread of Phytophthora, potato plants must be treated several times with various chemical fungicides as a precaution. In Europe, research to develop a genetically engineered potato has been carried out in Germany and Switzerland. In a German study, potatoes were transformed with the addition of genetic material from a soil bacterium. The transformed potatoes produce a fungal inhibitor, which leads to the death of the plant cell, thus preventing further spread of the disease (Strittmatter, 1995. In a Swiss experiment, potatoes were transformed by introducing a wheat gene that encodes for an enzyme that degrades oxalic acid into carbon dioxide and hydrogen peroxide. Hydrogen peroxide in the plant tissue is a defense against the blight fungus. One focus of current research is a wild plant species related to potato from Mexico. This species, Solanum bulbocastanum, coevolved with the late blight fungus and has exhibited durable race non-specific resistance to the late blight fungus (Naess, 2002. However, S. bulbocastanum is largely sexually incompatible with potato due to differences in endosperm balance numbers. One alternative to sexual crosses is the uniting of diverse genomes via somatic fusion, which has been effectively used to capture the late blight resistance from S. bulbocastanum and has then been passed on to potato breeding lines (Helgeson, 1998. The progeny of the somatic hybrids were grown in Mexico where nearly every race of the fungus is found. The resistance to late blight of S. bulbocastanum has been mapped to chromosome 8.

Development of Dominant Rice Blast Resistance Gene

Rice blast disease caused by the fungus is one of the most devastating diseases worldwide. Twenty resistance genes have been identified by extensive genetic studies (Jia et al., 2002. Pi-b and Pi-ta, two major resistance genes, introgressed from indica cultivars, have recently been molecularly characterized. Both Pi-b and Pi-ta encode predicted nucleotide binding site type proteins that a recharacteristics of products of major resistance genes. Molecular markers linked to resistance genes have been used for selection at the early seedling stages, and genotypes can be easily raised. A PCR-based Pi-ta gene marker is useful in marker-assisted selection breeding since it is the part of resistance gene, and is simple, rapid and inexpensive and can be used for analyzing large numbers of samples (Jia et al., 2002.

Development of Rust Resistance in Wheat

Diseases of wheat are mostly caused by fungal pathogens. Leaf rust caused by Puccinia recondita f. sp. tritici is the most widespread and regularly causing rust on wheat. Various wheat breeding programs throughout the world have had mixed results in producing cultivars with long-lasting, effective resistance to leaf rust (Huang and Gill, 2001. Genetic resistance is the most economical method of reducing yield losses due to leaf rust. Genetic resistance to leaf rust can be most fully utilized by knowledge of the identity of resistance genes in commonly used parental germplasm and released cultivars. Identification of the leaf rust resistance genes allows for efficient incorporation of different genes into germplasm pools, thus helping to avoid the release of cultivars that are genetically uniform. Resistance gene expression is dependent on the genetics of host-parasite interaction, and interaction between resistance genes with suppressors or other resistance genes in the wheat genomes. Genes expressed in seedling plants have not provided long-lasting effective leaf rust resistance. Adult-
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plant resistance genes Lr13 and Lr34 singly and together have provided the most durable resistance to leaf rust in wheat throughout the world.

Benefits and Risks
The list of plants and plant-derived products made as a result of modern biotechnology is ever-increasing. There is public concern over the safety of eating GM foods, there is no scientific reason to suspect that food from GM crops which have been licensed by the various regulatory bodies, presents any more of a safety problem than that from non-GM crops. Many of the potential hazards, such as food allergies, are also associated with traditional breeding techniques. The concerns that are being raised of environmental and food safety risks of biotechnology through gene exchange and evolution of new pathogens, or from putative increased or unexpected allergenicity are legitimate risks that will be addressed as have similar potential risks with any new plant or plant product. Assessment and management of these and other risks of new technologies in a formal process is appropriate, and must be conducted in a science-based manner and also include economic, human and animal health, and ecological consequences. However, these risks and concerns are not limited to plants and plant products produced through biotechnology and thus must be placed in perspective. The consequences of foregoing the use of biotechnology for improving plant health and sustainable plant productivity must also be considered. Potential risks associated with transgenic plants include: Introduction of allergenic or otherwise harmful proteins into foods. Transfer of transgenic properties to viruses, bacteria, or other plants.

Benefits of plant biotechnology
The application of biotechnology in agriculture has resulted in benefits to farmers, producers and consumers. Plant biotechnology has helped make diseases control safer and easier. Some of the potential benefits from using transgenic plants include, Reduced crop production costs and increased yields. Reduced environmental impact from farming and industry. Increased food availability for underdeveloped countries.

Future Prospects
Achievements today in plant biotechnology have already surpassed all previous expectations, and the future is even more promising. The future of biotechnology will depend on how the consumers accept this technology and those to come. As the technology evolves toward the use of tissue-specific or pathogen-inducible promoters, the expression of engineered traits that are effective against a broad range of pathogens, and the utilization of synthetically derived peptides and of R genes, the impact on disease management will be enhanced. Evaluation of these transgenic plants for response to disease will need to be extended to field trials and appropriate agronomic data collected to ensure that this technology can be successfully implemented in farmer’s fields to augment on-going disease management practices. Transgenic plants with enhanced disease resistance can become a valuable component of a disease management program in the future. The next three or four of years will provide an answer to how biotechnology will continue to be used in crop production. The possibility, in the short term is that there will be some sort of labeling of the foods that have been created with GMO material and the consumers will make their determination of what they think by the buying of the product or not. The farmers/producers will have the ability of trying to determine whether it will be more profitable to produce the traditional or biotech types of crops. They may have many reasons to want to use the biotech varieties.

Conclusion
Agricultural biotechnology is the application of molecular biology and, especially, molecular genetics to solving agricultural problems and exploiting agricultural opportunities. Biotechnology research leads to new and improved agricultural inputs. Biotechnology provides new opportunities to build pest-resistance into plants. Such developments may reduce the demand for fungicides. Biotechnology research will produce crop varieties with special characteristics that increase their value as grain and forage. Growers can justify greater inputs of chemicals when producing higher value crops. This, in turn, will increase the demand for biotechnology.

References
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ROLE OF GENETIC ENGINEERING IN DEVELOPMENT OF DISEASE RESISTANCE IN PLANTS

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Abstract
Genetic engineering is also called genetic modification or genetic manipulation. The simple addition, deletion or manipulation of a single trait in an organism to create a desired change. Or “It is the artificial manipulation or alteration of genes.” Genetic engineering involves removing a gene (target gene) from one organism, inserting target gene into DNA of another organism and ‘cut and paste’ process. Alternative names for genetic engineering are Genetic Manipulation, Genetic Modification, Recombinant DNA Technology, Gene Splicing, and Gene Cloning.

Recombinant DNA: The altered DNA is called recombinant DNA (recombines after small section of DNA inserted into it).

Genetically Modified Organism (GMO): It is the organism with the altered DNA” or Transgenic plant.

Alternative names for genetic engineering: Genetic engineering is also known as Genetic Manipulation, Genetic Modification, Recombinant DNA Technology, Gene Splicing and Gene Cloning.

Tools Used in Genetic Engineering: Restriction Enzymes- “are special enzymes used to cut the DNA at specific places”. Different enzymes cut DNA at specific base sequences known as a recognition site. e.g. - i) one restriction enzyme will always cut DNA at the base sequence: GAATTC. ii) Another restriction enzyme only cuts at the sequence: GATC.

Process of Genetic Engineering: Five steps involved in this process: Isolation, Cutting, Insertion (Ligation), Transformation and Expression

There are two main approaches for transferring genes into plant cells

Direct gene transfer: Delivers DNA directly to the nuclear or plastid genome of the plant cell through various techniques such as electroporation or chemical treatment, which stimulate the passive uptake of DNA through protoplast membranes, and biolistics, which uses acceleration of DNA-coated microparticles to carry DNA directly into plant cells.

Indirect gene transfer
It is based on a gene-transfer mechanism mediated by Agrobacterium. During natural infection, these phytopathogenic bacteria transfer and integrate oncogenes into the plant genome. The transferred DNA from Agrobacterium tumefaciens is a discrete segment of DNA from a small plasmid (Ti plasmid) resident in the cell. This Ti plasmid has been genetically engineered to produce efficient nonpathogenic vectors for plant-cell transformation.

Given the low transformation efficiencies of both direct and indirect methods, the selection of transgenic plants is achieved through the combined transfer of a selectable marker gene together with the gene interest. Since 1970’s rapid progress has been done in developing tools for the manipulation of genes in plants using recombinant DNA technology. Genetic transformation has led to the possibility of transforming crops for enhanced resistance to insects and pathogens.


Most commonly used methods are: Chemical Method, Electroporation, Particle Bombardment and Agrobacterium-mediated Transformation

Vector based (Dicotyledons) as well as the direct DNA transfer methods (biolistics) for monocots. It’s Depends upon the stable introduction of transgene into the genome of the plant. Are widely used as methods to understand how plants work and to improve crop plant characteristics.

Chemical Procedures: Plant protoplasts treated with polyethylene glycol, which help readily take up DNA from their surrounding medium, This DNA can be stably integrated into the plant’s chromosomal DNA
Protoplasts are then cultured under conditions that allowed them to grow cell walls, start dividing to form a callus, develop shoots and roots. Regenerate whole plants.

**Electroporation:** Plant cell electroporation generally utilizes the protoplast because thick plant cell walls restrict macromolecule movement. Electrical pulses are applied to a suspension of protoplasts with DNA placed between electrodes in an electroporation cuvette. Short high-voltage electrical pulses induce the formation of transient micro pores in cell membranes allowing DNA to enter the cell and then the nucleus.

**Particle Bombardment:** It includes the following steps are Isolate protoplasts from leaf tissues, Inject DNA-coated particles into the protoplasts using particle gun, Regenerate into whole plants and Acclimate the transgenic plants in a greenhouse.

"Gene gun" method: The "Gene Gun" method is also referred to as "biolistics" (ballistics using biological components. This technique is used for in vivo (within a living organism) transformation and has been especially useful in transforming monocot species like corn and rice. This approach literally shoots genes into plant cells and plant cell chloroplasts. DNA is coated onto small particles of gold or tungsten approximately two micrometres in diameter. The particles are placed in a vacuum chamber and the plant tissue to be engineered is placed below the chamber. The particles are propelled at high velocity using a short pulse of high pressure helium gas, and hit a fine mesh baffle placed above the tissue while the DNA coating continues into any target cell or tissue.

**Characteristics of an ideal vector:** Crown gall formation in plants depends on the presence of Ti plasmid (Tumour inducing plasmid. When the plants (like grapes, walnuts, apples and roses) are wounded or damaged, causes "crown gall" disease. *Agrobacterium tumefaciens* is a soil borne, gram-negative bacterium, rod shaped motile bacterium found in the rhizosphere region. *Agrobacterium* mediated gene transfer: (vector based).

**Ti plasmid:** Opine biosynthesis is catalyzed by specific enzymes encoded by genes contained in a small segment of DNA (known as the T-DNA, for 'transfer DNA'), which is part of the Ti plasmid, inserted by the bacterium into the plant genome. The opines are used by the bacterium as an important source of nitrogen and energy. Each strain of Agrobacterium induces and catabolizes a specific set of opines.

**Cloning vectors for higher plants:** Cloning vectors for higher plants were developed in the 1980s and their use has led to the genetically modified (GM) crops that are in the headlines today. We will examine the genetic modification of crops and other plants in. Here we look at the cloning vectors and how they are used. Three types of vector system have been used with varying degrees of success with higher plants:

- Vectors based on naturally occurring plasmids of *Agrobacterium*
- Direct gene transfer using various types of plasmid DNA

**Agrobacterium tumefaciens—nature’s smallest genetic engineer:** Although no naturally occurring plasmids are known in higher plants, one bacterial plasmid, the Ti plasmid of *Agrobacterium tumefaciens*, is of great importance. A. *tumefaciens* is a soil microorganism that causes crown gall disease in many species of dicotyledonous plants. Crown gall occurs when a wound on the stem allows *A.tumefaciens* bacteria to invade the plant. After infection the bacteria cause a cancerous proliferation of the stem tissue in the region of the crown. The ability to cause crown gall disease is associated with the presence of the Ti (tumor inducing) plasmid within the bacterial cell. This is a large (greater than 200 kb) plasmid that carries numerous genes involved in the infective process. A remarkable feature of the Ti plasmid is that, after infection, part of the molecule is integrated into the plant chromosomal DNA. This segment, called the T-DNA, is between 15 and 30 kb in size, depending on the strain. It is maintained in a stable form in the plant cell and is passed on to daughter cells as an integral part of the chromosomes. But the most remarkable feature of the Ti plasmid is that the T-DNA contains eight or so genes that are expressed in the plant cell and are responsible for the cancerous properties of the transformed cells. These genes also direct synthesis of unusual compounds, called opines, that the bacteria use as nutrients using the *Ti plasmid to introduce new genes into a plant cell.* It was realized very quickly that the Ti plasmid could be used to transport new genes into plant cells. All that would be necessary would be to insert the new genes into the T-DNA and then the bacterium could do the hard work of integrating them into the plant chromosomal DNA. In practice this has proved a tricky proposition, mainly because the large size of the Ti plasmid makes manipulation of the molecule very difficult. The main problem is, of course, that a unique restriction site is impossibility with a plasmid 200 kb in size. Novel strategies have to be developed for inserting new DNA into the plasmid. Two are in general use:

- The binary vector strategy is based on the observation that the T-DNA does not need to be physically attached to the rest of the Ti plasmid.
- A two-plasmid system, with the T-DNA on a relatively small molecule, and the rest of the plasmid in normal form, is just as effective at transforming plant cells. In fact, some strains of *A. tumefaciens*, and related agrobacteria, have natural binary plasmid systems. The T-DNA plasmid is small enough to have a unique restriction site and to be manipulated using standard techniques.
The marker is used to find the organism which has successfully taken up the desired gene. Tissues of the organism are then transferred to a medium containing an antibiotic or herbicide, depending on which marker was used. The Agrobacterium present is also killed by the antibiotic. Only tissues expressing the marker will survive and possess the gene of interest. Thus, subsequent steps in the process will only use these surviving plants. In order to obtain whole plants from these tissues, they are grown under controlled environmental conditions in tissue culture. This is a process of a series of media, each containing nutrients and hormones. Once the plants are grown and produce seed, the process of evaluating the progeny begins. This process entails selection of the seeds with the desired traits and then retesting and growing to make sure that the entire process has been completed successfully with the desired results.

Electroporation: In molecular biology, the process of electroporation is often used for the transformation of bacteria, yeast, and plant protoplasts. In addition to the lipid membranes, bacteria also have cell walls which are different from the lipid membranes and are made of peptidoglycan and its derivatives. However, the walls are naturally porous and only act as stiff shells that protect bacteria from severe environmental impacts. If bacteria and plasmids are mixed together, the plasmids can be transferred into the cell after electroporation. Several hundred volts across a distance of several millimeters are typically used in this process. Afterwards, the cells have to be handled carefully until they have had a chance to divide producing new cells that contain reproduced plasmids. This process is approximately ten times as effective as chemical transformation.

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PLANT GROWTH PROMOTING RHIZOBACTERIA (PGPR) IN PLANT DISEASE SUPPRESSION AND PLANT GROWTH

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Abstract

Bacteria (Gram positive & negative) that colonize plant roots and promote plant growth are referred to as plant growth-promoting rhizobacteria (PGPR) such as *Streptomyces* sp., *Arthrobacter* sp., *Azospirillum* sp., *Burkholderia* sp., *Enterobacter* sp., *Serratia* sp., *Pseudomonas* sp., *Bacillus* sp. etc. PGPR are highly diverse in nature and in this presentation we focus on rhizobacteria as suppressors of plant diseases. In recent years, PGPR and their interactions with plants are exploited commercially. Their effects can occur via local antagonism to soilborne pathogens or by induction of systemic resistance (SAR) against pathogens. Several metabolic-substances produced by antagonistic rhizobacteria have been correlated to pathogen inhibition and consequently indirect promotion of growth in plants, such as siderophores and antibiotics. The mechanism of resistance induced may differ in the pathways. Both types of induced resistance (ISR & SAR) render uninfected plant parts greater resistant to pathogens in several plant species. Rhizobacteria induce resistance through the salicylic acid-dependent SAR pathway, or require JA and ET perception from the plant for ISR. Rhizobacteria belonging to the genera *Pseudomonas* and *Bacillus* are known for their antagonistic effects and ability to trigger ISR. Resistance-inducing and antagonistic rhizobacteria promising and are being useful in formulating new inoculants with combinations of different mechanisms of action, leading to a more efficient application for pathogen suppression and thereby to improve cropping systems.

The rhizosphere is the narrow zone of soil specifically influenced by the plant root system. This zone is comparatively rich in nutrients due to the accumulation of a variety of plant exudates, including amino acids and sugars, that provides a rich source of energy and nutrients for bacteria. The rhizosphere is populated by a diverse range of microorganisms and the bacteria colonizing this ecological condition are called rhizobacteria. Bacteria that colonize plant roots and promote plant growth are referred to as plant growth-promoting rhizobacteria (PGPR) (Kloepper et al., 1981). Beneficial free-living soil bacteria are referred to as plant growth-promoting rhizobacteria (PGPR). Plant-associated bacteria can be classified on the basis of effects on plant growth (i) beneficial, (ii) deleterious and (iii) neutral groups. These colonize the rhizosphere and the rhizoplane (root surface), or the root itself. Bacteria of diverse genera have been identified as PGPR. PGPR affect plant growth in two different ways, indirectly or directly. The direct promotion of plant growth by PGPR entails providing the plant with a compound that is synthesized by the bacterium, for example phytohormones. It also facilitates the uptake of certain nutrients from the environment. The indirect promotion of plant growth occurs when PGPR reduces or prevent the deleterious effects of phytopathogens. *Bacillus* and *Pseudomonas* spp. are predominant PGPR. Plant Growth Promoting Rhizobacteria (PGPR), have ability to suppress plant diseases caused by soil borne pathogens (Thomashow & Weller, 1990). These suppress plant disease through at least one mechanism, production of antibiotics or siderophores and induction of systemic resistance (Tenuta, 2003).

Mechanism of action of PGPRs

**Through nitrogen fixation in atmosphere:** Nitrogen fixing bacteria are designated as urea factories. PGPR bacteria have specific nitrogenous enzyme that converts atmospheric nitrogen to ammonium making available for plants. Symbiotic Nitrogen fixation in root nodules of legumes e.g. *Rhizobium* and *Bradyrhizobium*. Nitrogen fixation by free-living soil bacteria and cyanobacteria in non-leguminous plants e.g. *Azotobacter* and *Azospirillum*

**Through solubilization of mineral phosphates:** Phosphorus is the major essential plant nutrient. Most of the soil phosphorus is in unavailable form, which is converted to readily available form by different soil microorganisms. PGPR possesses the ability to solubilize the inorganic phosphate and can mineralize organic phosphoric compounds. *Bacillus megaterium* & *Pseudomonas fluorescens* are two of the PGPR bacteria decomposing organic phosphates, increase crop yield. Organic acid produced by bacteria i.e. Citric acid, Lactic acid, Gluconic acid, 2-Ketogluconic acid, Oxalic acid, Tartaric acid, Acetic acid, etc. Organic acid work as chelators. Organic acids also solubilize nutrient containing minerals (e.g. apatite mineral containing P).

**Through production of phytohormones:** PGPRs can also produce phytohormones like IAA, GA and cytokinins etc. These phytohormones produced by various PGPRs influence physiological and developmental
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processes of plants like that cell division, seed germination, root development, accumulation of chlorophyll, leaf expansion etc. e.g. *Azotobacter, Azospirillum, Rhizobium, Bacillus* and *Pseudomonas* spp.

Through microbial antagonism: According to Beattie (2006), bacteria/microorganism that reduces the incidence or severity of plant diseases are referred to as biocontrol agents. Whereas those exhibit antagonistic activity toward a pathogen are defined as antagonists. The following rhizospheric environment and bacterial antagonistic activities are observed: Synthesis of hydrolytic enzymes, such as chitinases, glucanases, proteases, and lipases (Neeraja *et al.*, 2010; Maksimov *et al.*, 2011); Competition for nutrients and suitable colonization of niches at the root surface (Ste-phens *et al.*, 1993; Kamilova *et al.*, 2005); Regulation of plant ethylene levels through the ACC-deaminase enzyme (Glick and Bashan, 1997; Van Loon, 2007) and Production of siderophores and antibiotics. Siderophores are amongst the strongest soluble Fe³ binding agents. Siderophores produced by *Pseudomonas* sp., *Alkaligenes, Bacillus, Enterobacter* are implicated in the biological control of plant diseases (Kloeper, 1980. The active transport system through the membrane begins with the recognition of the ferric-siderophore by specific membrane receptors of Gram-negative and Gram-positive bacteria (Bou-khal-fa and Crumbliss, 2002). Siderophores produced such as pyoverdin which Inhibit growth of bacteria & fungi in iron depleted media in vitro, *Pseudobactin* from *P. putida* (B10 strain) which suppress *Fusarium oxysporum* in soil deficient in iron (Kloeper *et al.*, 1980).

Colonization of Rhizosphere through PGPRs: PGPR have ability to survive onto the seed. PGPRs can multiply in the spermosphere (region surrounding seed) in response to seed exudates. Attach to the root surface, subsequently colonize the root system. This ultimately enhances the growth of the plant. It also, produces phytohormones, which helps plant system to combat soilborne pathogens.

Production of antimicrobial substances by PGPRs: The process where two organisms in which one is harmed or killed by the other is called as antibiosis (Agrios, 2015), PGPR has the ability to produce antibiotics e.g. *Pseudomonas* spp. produce secondary metabolites with antimicrobial properties, phenazine-1-carboxylic acid (PCA), 2,4-Diacetylphloroglucinol (DAPG), oomycin-A, pyocyanine, pyolutein and pyroin (Thomasow & Well, 1996. Lipopeptide bio-surfactants produced by *Pseudomonas* and *Bacillus* species have been implied in biocontrol due to their potential positive effect on competitive interactions with organisms including bacteria, fungi, oomycetes, protozoa, nematodes and plants. Many of these have been implicated in suppression of soilborne pathogens such as *Fusarium, Rhizoctonia, Sclerotium, Ralstonia and Pythium* (de Bruijn *et al.*, 2007; Raajmakers *et al.*, 2010).

Production of bacteriocins: Almost all bacteria may produce at least one bacteriocin, antimicrobial metabolite (Riley 1993. Bacteriocins were first discovered by André Gratia in 1925. They commonly have a relatively narrow killing spectrum and are only toxic to bacteria closely related to the producing strain (Riley and Wertz 2002. Some examples of bacteriocins derived from bacteria are Colicin derived from *Escherichia coli*, Pyocins from *Pseudomonas pyogenes*, Megacins from *Bacillus megaterium* etc. (Cas-cales *et al.*, 2007); Abriouel *et al.*, 2011.

Induction of ISR and SAR: Non-pathogenic rhizobacteria have been shown to suppress disease by inducing a resistance mechanism in the plant called "Induced Systemic Resistance" (ISR) Van Loon *et al.*, (1998. Induced resistance is the state of an enhanced defensive ability developed by plants when appropriately stimulated. ISR was formerly described by Van Peer *et al.*, (1991) in carnation plants that were systemically protected by the *P. fluorescens* strain WCS417r against *F. oxysporum* f. sp. *dianthe*. Specifically, *Pseudomonas* and *Bacillus* spp. are the rhizobacteria most studied that trigger ISR. It can induce systemic biochemical and ultra structural changes in the roots that lead to a greater ability of the host plant to defend itself against root infecting pathogens (Klopper *et al.*, 2004; Van Wees *et al.*, 2008. ISR and SAR act through different signaling pathways. Induction of SAR is through salicylic acid (SA) and ISR requires jasmonic acid (JA) and ethylene (ET) signaling pathways (Van Loon *et al.*, 1998. However, ISR and SAR together provide a better protection than each of them alone, indicates that they can act additively in inducing resistance towards the pathogens Van Wees *et al.*, (2000. In SAR, the first infection predisposes the plant to resist further attacks. Development of tissue necrosis used to be considered a common and necessary feature for SAR activation Vleeschauwer and Höfte (2009. SA -activates specific sets of defense-related genes called pathogenesis related proteins (PRs). Generally, ISR is not accompanied by the activation of PR genes VanLoon (2007). Treatment of tobacco roots with *P. fluorescens* CHA0 triggers accumulation of SA-inducible PR proteins in the leaves Maurhofer *et al.*, (1994. Some of these PRs are 1,3-glu-canases and chitinases capable of hydrolyzing fungal cell walls. Infected plants increased their levels of JA and ET as a sign of active defense De Laat and Van Loon (1982. These signaling molecules coordinate the activation of a large set of defense responses and when applied exogenously, can induce resistance themselves.

Conclusion: The ability of bacterial siderophores and antibiotics to suppress phytopathogens could be of significant agronomic importance. Both mechanisms have essential functions in microbial antagonism but also
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are able to elicit induced resistance. Resistance-inducing and antagonistic rhizobacteria might be useful in formulating new inoculants, offering an attractive alternative of environmentally friendly biological control of plant disease and improving the cropping systems into which it can be most profitably applied. These new PGPR will require a systematic strategy designed to fully utilize all these beneficial factors, applying combinations of different mechanisms of action allowing crop yields to be maintained or even increased while chemical treatments are reduced. Although there are challenges in the form of nonculturability of AMF and therefore mass multiplication for agricultural crops, there is promise for non direct sown crops, which is currently undervalued and underexploited. The AMF, by increasing crop productivity using existing resources, avoiding resistance development to chemicals, maintaining pollution and risk-free disease control, and conforming to sustainable agricultural practices, offers more than mere plant disease control. In the future, mycorrhizosphere management must become one of the viable and ecosystem friendly solutions to managing plant diseases and reducing pathogen inoculums.

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SYMPTOMATOLOGY AND BIOLOGY OF MAJOR VIRUSES INFECTION ALLIUM SPECIES

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Abstract
The genus Allium is one of the largest plant genera and most of the plant species in the genus are bulb forming, and several of them are important edible crops all over the world. These cultivated species are greatly hampered by biotic and abiotic stress. Among biotic stress disease caused by fungi, bacteria and viruses are important. Viruses belonging to genera Potyvirus, Allexivirus and Carlavirus are common in Allium species. Yellow mosaic, chlorotic stripe, and stunting are common symptoms caused by potyviruses. Carlaviruses and allexiviruses are latent and don't produce distinct symptoms. Members of potyviruses have a genome of single-stranded, positive-sense RNA and 10kb length, while Allexivirus is linear, single-stranded, positive-sense RNA and 8 kb long. The genome of carlaviruses is linear, single-stranded, positive-sense RNA was 8363 nt long.

For many years, mosaic type symptoms have been observed on the leaves of garlic, onion and shallot crops and were attributed to some diseases of unknown etiology (Van Dijk 1993a, Walkey 1990. In addition, a yellow stripe disease has also been observed on leek crops in different countries (Bos et al 1978a. The identification of the causal agents of these diseases was difficult and for a long time incomplete. Problems related with the isolation and purification of the implicated viruses was mainly due to their limited host range, the absence of specific indicator plants for their differentiation, and the simultaneous infections with many viruses in garlic. This resulted in difficulties in their identification and confusion in the bibliography. Alliumviruses were found worldwide in onion, garlic, leek, and shallot. They have been recorded from Europe, Newzealand, North Africa, South America, and Asia. OYDV is one of the most important viruses affecting Allium species (Van Dijk 1993a. The virus has spread worldwide and a high incidence has been found in many countries, including Greece, Argentina, The Czech Republic, Egypt, Brazil, India and Sudan (Dovas et al 2001, Conci et al 2003, Klukackova et al 2004, Wahab et al 2009, Fayad-Andre et al 2011, Katis et al 2012, Kumar et al 2010, Mohammed et al 2013. This chapter describes diagnostic symptoms, genome organization, and morphology of major viruses of Allium crops.

Symptomatology
Symptoms depend on virus strain and host genetic background. In onion, OYDV causes yellow striping or yellowing, dwarfing of plants. In seed crops, flower stems remain turgid and round but show yellowing, distortion, and curling. In addition, they are shorter than normal and produce smaller flowers with reduced numbers of seeds that are often of poor quality (Kumar et al 2010. Severely affected plants do not develop flower and bud resulting in a heavy loss to seed production. In garlic, symptoms of OYDV infection include yellow stripes covering most of the leaf surface. Diffused chlorotic stripes with little yellow stripes, yellowish dots on leaves and whitish leaf margin or twisting of leaves on a few cultivars (Van Dijk 1993a. Generally, symptoms are mild on young leaves than on mature leaves. Bulbs harvested from mosaic affected mature plants are much smaller in size and cloves are fewer in number (Ghosh and Ahlawat 1997. Lot et al (1998) reported that on Messidrome cultivars, plants exhibited severe yellow and light green striping on most leaves with premature drying of the leaf tips. Stripping was clear on older leaves but less distinct on young leaves. In combination with infection by other viruses, symptoms may be aggravated (Diekmann 1997. LYSV in leek causes chlorotic stripes in leaves, which initiate from their base, whereas yellowing of the whole leaf has been reported. Affected plants are stunted and less juicy. The stem is lusterless, and the storing quality of the harvested product is impaired. Infected leek plants in autumn and winter crops are more susceptible to low temperatures and may be killed (Walkey 1990. Symptoms are more severe when leeks are coinfected with SLV. In garlic, LYSV-G (garlic strain) causes mild streaking up to severe yellow streaking and reduction of bulb size. SLV occurs apparently symptomless, in shallot, onion, and garlic, but it acts synergistically with potyviruses. In leek, mild chlorotic streaking appears in singly infected plants and severe chlorotic or white streaking and even plant death in plants coinfected with LYSV (Paludan 1980. GarCLV is latent in garlic, leek, and onion, but it acts synergistically with potyviruses. Generally, allexiviruses are latent or very rarely they are causing very mild
chlorotic stripes and mild mosaic in leaves of *Allium* species (Van Dijk and Van der Vlugt 1994, Van Dijk *et al* 1991, Yamashita *et al* 1996. It should be noted that often, stripes or other mild leaf deformations are caused by *Aceria tulipae*, the vector of alllexiviruses.

**Potyviruses affecting *Allium* species**

Members of the family *Potyviridae* have a genome of single-stranded, positive-sense RNA. The viruses of five genera (*Potyvirus*, *Macluravirus*, *Ipomovirus*, *Rymovirus*, and *Tritimovirus*) have a monopartite genome that contains only one RNA molecule. Viruses of the genus *Bymovirus* have a bipartite genome which contains two RNA molecules, RNA-1, and RNA-2 (Shukla *et al* 1998. Due to exclusive vegetative propagation, all commercial onion cultivars tend to accumulate multiple viruses, which are passed on to the following generation. They are producing chronic infections, even though they do not cause plant death. OYDV and LYSV are two most important well-known potyviruses responsible for the deterioration of quantity as well as the quality of onion crop.

**Onion yellow dwarf virus (OYDV)**

OYDV is a member of the Family *potyviridae*. OYDV have restricted host range that is mainly limited to several species of *Allium* and *Narcissus*. The virus has a flexuous, rod-shaped particle reported to be between 720-833 nm and 13-16 nm in width (Bos *et al* 1978a. This virus is monopartite, linear, positive sense, single-stranded RNA. The genome of OYDV is 10,538 nucleotides long and is predicted to encode a 3403 amino acid polyprotein (Chen *et al* 2003. OYDV is having a P3 that is significantly larger than any other member of the genus, or indeed the entire family *Potyviridae*. The function(s) of the potyvirus P3 gene is not very well understood. Because as P3 antibodies reacted with inclusions, so it has been suggested that the P3 might be involved in the replication of the RNA (Langenberg and Zhang 1997, Rodriguez *et al* 1993) and this is supported by *in vitro* studies of protein-protein interactions (Guo *et al* 2001. There is also evidence that the P3 has a role in pathogenicity and symptom determination (Dallot *et al* 2001. OYDV is usually very serious and often reaches an epiphytotic level that leads to considerable yield losses (Conci *et al* 2003. The disease has been reported to cause an injurious and colossal effect on the growth of onion plants and consequently on bulb or seed production. Yield losses have been reported to be heavy on dry bulb and grave on seed crop. The quality of the bulb and seeds produced can be significantly affected by infection of mother plants with OYDV. It produces pinwheel and scroll-shaped inclusion bodies that may be seen in the ultrathin section in the electron microscope (Edwardson 1974. Information regarding the variability of populations of OYDV has been obtained through molecular techniques, and genetic studies have provided data mainly on garlic isolates, whereas less information is available on onion isolates (Celli *et al* 2013. Complete nucleotide sequences of OYDV isolates showing mild and severe symptoms in onion were determined and genomes consisted of 10,459 and 10,461 nt (nucleotide) and were 92.2 % identical. In both isolates, the AUG initiation codon and the stop codon (UGA) are likely to be located at nt position 109-111 and 10252-10254, respectively. Therefore, the predicted ORF for each isolate was 10,143 nt, encoding a polyprotein of 3,381 aa (amino acid). Comparison of the individual protein regions of the two onion isolates showed that the CP-encoding region was the most conserved (nt and aa sequence identities of 95.8 % and 97.7 %, respectively. In the entire CP sequence (257 aa), there were six aa changes, four of which were located in the C-terminal region (Celli *et al* 2013. The DAG motif that is involved in transmission by aphids (Atreya *et al* 1991) was found in the N-terminal region of both isolates at the same position (3,150-3,152. By contrast, the P1-encoding regions were the least conserved, sharing only 86.2 % nt identity. P1 was the most variable protein (80.8 % identity), with 84 aa changes being distributed along the protein and corresponding to 45 % of all changes in the whole polyprotein. P1 protein would affect the development of symptoms caused by potyvirus (Lee and Wong 1998.

**Leek yellow stripe virus (LYSV)**

LYSV, a member of the genus *Potyvirus* in the family *Potyviridae*, is a flexuous, filamentous virus containing a single positive sense RNA genome. The virus is known to be a causal agent of mosaic disease in garlic and leek. Symptoms depend on virus strain and host genetic background. The mosaic disease is quite common in garlic producing areas worldwide, and diseased plants exhibit mosaic (or yellow stripe) symptoms on leaves, are often dwarfed and may produce smaller, malformed, brownish colored bulbs resulting in severe losses of yield and quality (Takaki *et al* 2005. Takaki *et al* (2005) determined the complete nucleotide sequence of three isolates of LYSV, the main causal agent of the garlic mosaic disease that is prevalent in Japan. They contained 10,296-
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10,297 nucleotides and encoded a deduced polyprotein of 3,215 amino acids. Sequence variation among the three isolates was 1.5 percent at both the nucleotide and amino acid levels.

**Allexiviruses affecting Allium species**

The genome of *Allexivirus* is linear, single-stranded, positive-sense RNA was 8 kb long (excluding the poly-A tail) and had the expected ORFs which, by comparison with other published sequences, are believed to produce the six virus proteins. In the early 1990s, new types of highly flexuous filamentous virus particles were observed in tissues of different *Allium* species (Barg *et al* 1994, Kanyuka *et al* 1992, Sumi *et al* 1993, Van Dijk *et al* 1991, Vishnichenko *et al* 1993. Unlike the other groups of viruses infecting *Allium* species, these ones are transmitted by mites (*Aceria tulipae*) (Barg *et al* 1994, Van Dijk *et al* 1991), while usually they are latent or they cause very mild symptoms. Based on host range and serological reactions, Van Dijk *et al* (1991) described three types of these viruses namely Onion mite-borne latent virus (OMbLV), OMbLV-Garlic strain (in onion and garlic, respectively) and Shallot mite-borne latent virus (SMbLV. Initially, they were erroneously classified as *Rymoviruses* of the family *Potyviridae* based on their transmission by mites, the morphology and the length of their virus particles (700-800 nm), and the presence of granular inclusion bodies in infected tissues. In Russia, another virus named ShVX was described based on its properties and partial nucleotide sequence (Kanyuka *et al* 1992, Vishnichenko *et al* 1993. The particle morphology of ShVX resembled that of OMbLV, SMbLV and Mite-borne filamentous virus (MbFV). All these viruses are serologically closely related. In addition, Van Dijk and Van der Vlugt (1994) have shown that ShVX could be re-identified as OMbLV, SMbLV or a complex of the two using antiserum against ShVX. The determination of the genome organization of ShVX revealed that it combines characteristics of carlaviruses and potexviruses, except in possessing an unusual ORF4. The identification of genome organization and nucleotide sequence of four other similar viruses (GarV-A, GarV-B, Garlic virus-C (GarV-C) and GarV-D) isolated from mosaic-diseased garlic, and their phylogenetic analysis showed that they are related to ShVX and probably constitute a new virus genus closely related to the carlaviruses (Sumi *et al* 1993. The characterization of these viruses and their species differentiation has largely been hampered by the fact that they often occur in multiple infections and their separation for further studies is difficult. Van Dijk *et al* (1991) isolated some of them through successive local lesion passages onto *Chenopodium* spp. or mite transmission. Nevertheless, the biological data concerning these viruses are limited. Therefore, sequencing data have been mainly used to justify the creation of several species namely GarV-A, GarV-B, GarV-C, GarV-D, Garlic virus E (GarV-E), GarV-X (Sumi *et al* 1993, Ryabov *et al* 1996, Song *et al* 1998, Sumi *et al* 1999, Chen *et al* 2001), Garlic mite-borne filamentous virus (GarMbFV), ShVX (Kanyuka *et al* 1992, Vishnichenko *et al* 1993. Western blot analysis revealed that GarV-D and GarV-E are closely related serologically, while weaker relationships exist between GarV-E and GarV-A, GarV-X and GarV-A, GarV-X and GarV-B, and GarV-X and GarV-C (Lu *et al* 2008. Very often allexiviruses persist in the infected plants as multiple infections with carlaviruses and potyviruses. This coexistence may have a synergistic effect and lead to even higher yield losses. A recent study indicated that garlic yield decreases more rapidly in plants previously infected with at least one allexivirus and then reinfected with other naturally occurring viruses than in plants that are initially virus free (Perotto *et al* 2010). Allexiviruses are thought to be transmitted by mites both in the field where the vectors are moved passively by the wind and in the bulbs storage rooms (Barg *et al* 1997, Van Dijk *et al* 1991. The only vector known is the dry bulb mite, *Aceria tulipae* Keifer which was proved to transmit GarV-C and GarV-D (Zavriev 2008. Allexiviruses are not transmitted by aphids (Van Dijk *et al* 1991, Yamashita *et al* 1996) but are manually transmissible by sap under laboratory conditions. Allexiviruses cause chlorotic local lesions onto *Chenopodium murale* L., *C. amaranticolor* L., *C. quinoa* L., and *Atriplex hortensis* L. However, not all allexiviruses infect all indicator plants (Van Dijk and Van der Vlugt 1994, Van Dijk *et al* 1991.

**Carlavirus affecting Allium species**

The genome is linear, single-stranded, positive-sense RNA was 8363 nt long (excluding the poly-A tail) and had the expected six ORFs which, by comparison with other published sequences, are believed to produce the six virus proteins (Chen *et al* 2001. In Allium species, the first reports of carlaviruses are those of *Shallot latent Virus* (SLV), isolated from *A. cepa* var. *ascalonicum* L.” in Holland (Bos *et al* 1977b), and Garlic latent virus (GLV), isolated from garlic in Japan (Lee *et al* 1979. More specifically, SLV has been reported to affect garlic in the United Kingdom (Walkey *et al* 1987) together with another unknown carlavirus. In Germany, a carlavirus identified as GLV was different from GLV reported from Japan (Van Dijk 1993b. For this reason, Van Dijk renamed this new virus as Garlic common latent virus (GarCLV. GarCLV and SLV have been reported to infect garlic crops in various countries (Barg *et al* 1994, Nieto *et al* 2004, Tsuneyoshi *et al* 1998, Van Dijk 1993b. Van Dijk (1993b) described another carlavirus, namely Sint-Jan’s onion latent virus (SjoLV) which reacts with the
antisera prepared against SLV and GarCLV in electron microscopy decoration tests. Tsuneyoshi et al (1998) suggested that SjoLV might be a strain of SLV or GarCLV, but sequence data of virus isolates previously identified as SjoLV based on biological characteristics (host range, symptomatology) are required to confirm this. In garlic, Narcissus latent virus another carlavirus was also detected in the United Kingdom (Walkey 1990) even though this isolate needs further comparison with SLV (Van Dijk 1993b. Furthermore, a carlavirus from garlic serologically related to Carnation latent virus (CaLV) was reported in Argentina (Conci et al 1992), and another carlaviruses with a similar reaction and closely related to GarCLV was reported from garlic crops in Slovenia (Mavric and Ravnikar 2005. It is not clear whether this is due to a cross-reaction of CaLV antiserum with GarCLV. Carlaviruses have a wide host range within members of the genus Allium. They cause latent infections and, although the effect on crop yields has not been determined (Perotto et al 2010), it seems that they cause rather limited crops losses. However, they can cause significant yield losses when the plants are coinfected with potyviruses due to synergistic effects (Paludan 1980, Sako 1989. Carlaviruses are transmitted nonpersistently by aphids, but probably less efficiently than potyviruses (Van Dijk 1993b. The main source of carlaviruses in the field is probably the infected propagative planting material as weeds do not contribute to their epidemiology (Van Dijk 1993b. Natural transmission of SLV and GarCLV from shallot and garlic crops to leek shows that carlaviruses have the lower degree of host specificity than potyviruses. Nevertheless, failures of SLV transmission from shallot to garlic (Van Dijk 1993b) and of SLV-G (garlic strain) from garlic in onion (Lee et al 1979) show some degrees of specificity.

Conclusion

Nowadays, intensive studies conducted by a great number of researchers have shown that mainly members of the genera Potyvirus, Carlavirus, and the recently ratified genus Allexivirus often found forming viral complexes, prevail in allium crops, and are implicated in the etiology of the above-mentioned diseases. Several of these viruses are often found in high incidence in almost all allium-cultivating regions of the world and are responsible for severe yield losses, while others may be localized in a few geographical regions. Members of the genus Potyvirus are usually the most abundant and cause most of the damage induced, while Carla- and allexiviruses are mainly latent.

References


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Abstract

Insect are major biotic factor which cause significant losses in various crops. The insecticides are used by farmers for their management but over use of chemical insecticides results in many problems such as development of insecticide resistance, reduction in population of natural enemies, resurgence, environmental pollution, human health hazards etc. All these problems can overcome by development of Insect resistance plant varieties. The basic mechanisms of resistance are non-preference, antibiosis and tolerance. The resistance can be due to morphology, physiological, biochemical and genetical factors of plants. The genetic factors are mainly of three type monogenic, oligogenic and polygenic. For development of resistant plant varieties breeder should know the major sources of resistance. The main sources of resistance found in nature are cultivated varieties, wild species, germplasm collections, mutations and introduction of microorganism. The method of breeding to develop insect resistant plants are introduction, selection and hybridization. The field screening is done before released of varieties for commercial purpose. This technology of insect pest management is found to be associated with following advantages like compatible to all other management techniques, much cheaper as compared insecticides and it is the simplest seed based technology do not need any extra skill for application.

Mechanism of resistance

In 1951, Painter gives mechanism of resistance under three main categories, viz nonpreference, antibiosis and tolerance. In nonpreference insects found plant undesirable. The plant become bad host to the insect for feeding, oviposition.or shelter. Antibiosis has adverse effect on the insect after it feed upon host plant. The main reason of antibiosis is lack of nutrients and due to presence of toxicant in host plant. The insect feeding on resistant plant may develop symptoms like larval death in early instars, abnormal growth rate, failed to pupae, development of abnormal adult, restlessness, reduction in longevity, reduction in fecundity and death. Tolerance is heritable character of host plant to withstand the insect population without reduction in yield. These types of resistance do not develop selection pressure and prevent biotype development. The resistant plants have ability to regrow, prevent lodging and produce additional branches to compensation yield loss. Plant breeder should know sources of resistance for development of resistance plants.

Sources of resistance

The main sources of resistance found in nature are cultivated varieties, wild species, germplasm collections, mutations and introduction of microorganism. The use of cultivated varieties is one of the easiest as well as quickest methods of developing resistance plants. Cultivated varieties which are grown by farmers are to be evaluated extensively for resistance. The cultivar varieties response differently to different insect species. If
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some cultivars are resistance to one insect may not be resistance to other insect species. The genes responsible for insect resistance may be found in some cultivated varieties. In case of cotton crop varieties like SRT1, Khandwa2, DHY286, and B 1007 are source of resistance against Jassid. The desirable genes responsible for resistance can be selected from the cultivated varieties and introduced in susceptible varieties. The second source of resistance is wild species. The wild spices of plants have potential to tolerate/ resist many insect species because of adaptations and morphological characteristics. The wild species of cotton crop like Gossypium lomentosum, G. anomalum and G. armuoriuman are found resistance to Jassid. If the desirable genes of resistance not found in cultivar varieties and wild species, it should have to search in germplasm collection. In India, National Bureau of Plant Genetic Resource, New Delhi is major agency which is responsible for introduction and exchange of new genotype. The screening of germplasm for specific insect is method use to develop resistnt varieties. For example when 2000 apples germplasm lines are screened for rosy aphid resistant out of which only 14 lines found resistant. The utilization of germplasm is difficult when compared with cultivar and wild species for development of resistant cultivar. For screening test lines extensive labours are required.

Some time insect resistance also developed due to natural mutation but the matation is realer and don’t take place spontaneously. In mutants genes sequences changes or deleted which make plants resistance to various insect species. In modern technology, microorganisms are also used as source of resistance to insects. For example, transfer of gene from Bacillus thuringiensis (Bt) into the system of cotton plant through genetic engineering. They are major source of resistance against bollworms.

Genetic type of resistance

The various type of resistance found on the basis of genes viz., Monogenic resistance Oligogenic resistance and polygenic resistance. In monogenic only one major gene is responsible for resistance so can be easily incorporated in host plants. The major disadvantage of that it can be break early by insect species. The oligogenic resistance is regulated by few genes where as in polygenic many minor genes are responsible of resistance. The cumulative effect of minor genes is difficult to break by insect species. The monogenic and oligogenic resistance is regulated by few genes whereas in polygenic many minor genes are responsible of incorporated in host plants. The major disadvantage of that it can be break early by insect species. The oligogenic resistance is regulated by few genes whereas in polygenic many minor genes are responsible of resistance. The cumulative effect of minor genes is difficult to break by insect species. The monogenic and oligogenic resistance is regulated by few genes whereas in polygenic many minor genes are responsible of resistance. The cumulative effect of minor genes is difficult to break by insect species.

Table 1. Genetics of Insect resistance in some crops

<table>
<thead>
<tr>
<th>Crop</th>
<th>Insect pest</th>
<th>Genetics of resistance</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cowpea</td>
<td>Cowpea aphid (Aphis craccivora)</td>
<td>2D (Rac1, Rac2)</td>
</tr>
<tr>
<td>Alfalfa</td>
<td>Pea aphid (Acyrthosiphon pisum)</td>
<td>1D +1d</td>
</tr>
<tr>
<td>Apple</td>
<td>Rosy apple aphid (Dysaphis plantaginea)</td>
<td>1D (smh)</td>
</tr>
<tr>
<td>Barley</td>
<td>Cereal leaf beetle (Oulema melanopus)</td>
<td>1 d</td>
</tr>
<tr>
<td></td>
<td>Corn leaf aphid (Rhopalosiphum maidis)</td>
<td>2 d (s1, s2)</td>
</tr>
<tr>
<td></td>
<td>Green bug (S. graminum)</td>
<td>2 D (Rsg-1a, Rsg-2b)</td>
</tr>
<tr>
<td></td>
<td>Hessian fly (M. destructor)</td>
<td>3 D (Hf, Hf, Hf)</td>
</tr>
<tr>
<td>Cotton</td>
<td>Boll weevil (Anthonomus grandis)</td>
<td>3D (R-1, red colour, H1, H2, hairiness) + 1d (fg, frego bract)</td>
</tr>
<tr>
<td></td>
<td>Jassids (Empoasca spp.)</td>
<td>3 D (H1, H2, H3, hairiness)</td>
</tr>
<tr>
<td></td>
<td>Thrips (Thrips spp.)</td>
<td>3 D (H1, H2, hairiness; SM, glabrous)</td>
</tr>
<tr>
<td></td>
<td>Tobacco budworm (Heliotis virescens)</td>
<td>2 D (Gl2, Gl3, glands)</td>
</tr>
<tr>
<td>Cucumber</td>
<td>Two-spotted spider mite</td>
<td>1 D (B1)</td>
</tr>
<tr>
<td>Maize</td>
<td>European corn borer (Ostrinia nubilalis)</td>
<td>3 gene</td>
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<tr>
<td></td>
<td>Western corn root worm (Diabrotica virgifera)</td>
<td>1 d</td>
</tr>
<tr>
<td>Pumpkin</td>
<td>Fruit fly (Dacus cucurbitae)</td>
<td>1 D (Fr)</td>
</tr>
<tr>
<td>Rice</td>
<td>Brown plant hopper (Nilaparvata lugens)</td>
<td>4D(Bph1, 3, 6, 9)+ 5d (bph2,4,5,7,8)</td>
</tr>
<tr>
<td></td>
<td>Gallmidge (Orseolia orzya)</td>
<td>3 d (gm1 to gm3)</td>
</tr>
<tr>
<td></td>
<td>Green leaf hopper (Nephotettix virescens)</td>
<td>6 D (Glh1,3,4,5,6,7)+ 2d (glh2, glh8)</td>
</tr>
<tr>
<td>Sorghum</td>
<td>Greenbug (S. graminum)</td>
<td>Single gene</td>
</tr>
<tr>
<td>Soyabean</td>
<td>Cyst nematode</td>
<td>3 d</td>
</tr>
<tr>
<td>Wheat</td>
<td>Greenbug (Schizaphis graminum)</td>
<td>5 D (Gb1 to Gb5) + 1 d (gb)</td>
</tr>
<tr>
<td></td>
<td>Hessian fly (Mayetiola destructor)</td>
<td>25 D (HI etc.) + 1 d (h4)</td>
</tr>
<tr>
<td></td>
<td>Wheat stem sawfly (Cephus cintus)</td>
<td>3 genes for stem solidness</td>
</tr>
</tbody>
</table>

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### Polygenic inheritance

<table>
<thead>
<tr>
<th>Crop</th>
<th>Pest or Pathogen</th>
<th>Type</th>
</tr>
</thead>
<tbody>
<tr>
<td>Alfalfa</td>
<td>Spotted aphid ((\text{Therioaphis maculata}))</td>
<td>Polygenic</td>
</tr>
<tr>
<td>Brassica spp.</td>
<td>Pink boll worm ((\text{Aonidiella aurantii}))</td>
<td>Polygenic</td>
</tr>
<tr>
<td>Cucumber</td>
<td>Two-spotted spider mite ((\text{Tetranychus urticae}))</td>
<td>Polygenic (additive)</td>
</tr>
<tr>
<td>Maize</td>
<td>Corn earworm ((\text{Heliothis zea}))</td>
<td>Polygenic</td>
</tr>
<tr>
<td></td>
<td>European corn borer ((\text{O. nubilalis}))</td>
<td>Polygenic</td>
</tr>
<tr>
<td></td>
<td>Corn leaf aphid ((\text{Rhopalosiphum maidis}))</td>
<td>Polygenic</td>
</tr>
<tr>
<td></td>
<td>Fall armyworm ((\text{Spodoptera frugiperda}))</td>
<td>Polygenic</td>
</tr>
<tr>
<td>Rice</td>
<td>Stem borer</td>
<td>Polygenic</td>
</tr>
<tr>
<td>Sorghum</td>
<td>Shoot fly ((\text{Atherigona socata}))</td>
<td>Polygenic</td>
</tr>
<tr>
<td>Soybean</td>
<td>Mexican bean beetle ((\text{Epilachna varivestis}))</td>
<td>Polygenic</td>
</tr>
<tr>
<td>Wheat</td>
<td>Cereal leaf borer ((\text{Oulema melanopus}))</td>
<td>Polygenic</td>
</tr>
</tbody>
</table>

### Cytoplasmic inheritance

<table>
<thead>
<tr>
<th>Crop</th>
<th>Pest or Pathogen</th>
<th>Type</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cotton</td>
<td>Boll weevil ((\text{A. grandis}))</td>
<td>Cytoplasmic</td>
</tr>
<tr>
<td></td>
<td>Tobacco budworm ((\text{H. virescens}))</td>
<td>Cytoplasmic</td>
</tr>
<tr>
<td>Lettuce</td>
<td>Root aphid ((\text{Pemphigus bursarius}))</td>
<td>Cytoplasmic</td>
</tr>
<tr>
<td>Maize</td>
<td>European corn borer ((\text{O. nubilalis}))</td>
<td>Cytoplasmic effects</td>
</tr>
<tr>
<td>Tomato</td>
<td>Potato aphid ((\text{Macrosiphum euphorbiae})) and whitefly ((\text{Bemisia tabaci}))</td>
<td>1 D ((\text{Mi-I}))</td>
</tr>
</tbody>
</table>

**Breeding method to develop resistance varieties:** Breeding methods for development of resistance plant varieties are introduction, selection and hybridization. The introduction is quicker and easier method which plant having resistance genes are introduced in new environment where they were not grown before. The major disadvantage is that introduced plant may not perform well in new environment and it may act as host of other insect species. A breeder can introduce new varieties of already grown crop in that area, wild relatives and totally new crop plants. The process of introduction may be at domestic level and international level. In domestic introduction can be within states or between different states within the country whereas for international introduction it is between different countries. The introduction of plant required to following quarantine rules and regulations to ensure that no weed or insect associated to that plant could not entry new territory. It can be classified into two categories viz Primary and secondary introduction. In primary introduction is directly introduction of plant without any change in original genetic composition whereas in secondary plant varieties not directly introduced. They are subjected to selection, isolation and transfer one or few desirable characters before introduction in new area.

**Selection:** The resistance gene can already exist in grown varieties. On the basis of it performer (insect resistance) in field conditions can selection for grown in next season. This practice is done by farmer in ancient times also. The process of selection can be natural and artificial. The selected plant can be isolation from plant of same or from mixed population. The natural selection play litter value in plant breeding because current breeding programmes are basis on artificial selection. The natural method of selection is more time consuming as compared to artificial method of selection. The basic principal of selection is that it utilized the variation of plants which already exist in plants. Pure Line Selection and Mass Selection are commonly used breeding method of selection.

**Hybridization:** Hybridization is one of the methods for developing new variety by crossing two lines which have unlike genes in other word it also called as crossing two plants having dissimilar genotype. The hybridization is responsible for genetic variation in plant. Segregation and recombination produce many new gene combinations in F2 and the subsequent generation. The degree of variation produced by hybridization in the segregating generation depends upon the number of heterozygous genes in the F1, and this depends upon the number of gene for which two parents differ. The aim of hybridization may be transfer of one or few qualitative characters, the improvement in one or more quantitative character or the use of F1 as a hybrid variety. These objectives are grouped into two classes combination breeding and transgressive breeding. In combination breeding method transfer of one or more characters into single plant. These may be oligogenes or polygenes. In transgressive breeding characters are segregation which plays important role in yield improvement of plants. The aims at improving yield or its contributing character through transgressive segregation. The F2 generation produce by segregation are superior to both parents.
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Evaluation of developed plants: After development of resistant plant varieties they should evaluated under field condition. The screening method is only used to test it performance. It is most difficult for breeder to identification of insect resistant during segregation generation. The screening can be done under field as well as in green house. Under field condition breeder should ensure that unformed infection should take place as well as test insect should not exposes to insecticide spray and bio control agents. The experiment should conducted in condition which is favourable to insect growth and development. The screening of individual insect become difficult because performance of one insect may be effect by other insect species present in filed. The glasshouse screening is more reliable than filed screening. Under glasshouse insect infestation is uniform. The test insect is not effect by biotic as well as abiotic stress. In glass houses screening of test insect is not effect by other insect. After evaluation by breeder the promising material is to be commercialized for general cultivation. The development of resistance plants varieties like all other techniques also have both advantages and disadvantages. The major advantage is that it is seed based technology. If farmer grow resistant seed he not need to use extra skill for it application. It required no extra cash investment is required for it application. The resistance varieties are cheaper as compared to insecticides application and also reduce the cost of production. They are highly comparable to all the management techniques. The resistance can be monogenic to polygenic so can be use against wider range of insects. The following demerits found to be associated with this technology are: It is a long term process which takes 10-15 years to develop agronomically acceptable variety even when the source of resistance are readily available. In some cases, breeding for resistance to one pest leads to the susceptibility to another pest. This is because the host plant feature associated with resistance to one insect is associated with susceptibility to another insect. For example, hairiness, in cotton is associated with Jassid resistance but confers susceptibility to whitefly and bollworms. In many cases, genes for disease and insect resistance are available only in the related wild species. Interspecific gene transfer poses many problems. Moreover, resistant genes are associated with some undesirable characters. It takes a long time to discard undesirable genes in breeding programmes.

References
ENVIRONMENTAL IMPACT OF NEONICOTINOIDS USE IN AGRICULTURE

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ABSTRACT

Pesticides are chemical preparations used to kill fungal or animal pests. Over 98% of sprayed insecticides and 95% of herbicides reach a destination other than their target species, because they are sprayed or spread across entire agricultural fields. Runoff can carry pesticides into aquatic environments while wind can carry them to other fields, grazing areas, human settlements and undeveloped areas, potentially affecting other species. Other problems emerge from poor production, transport and storage practices. Over time, repeated application increases pest resistance, while its effects on other species can facilitate the pest's resurgence. Neonicotinoid pesticides were first introduced in the mid-1990s, and since then, their use has grown rapidly. They are now the most widely used class of insecticides in the world, with the majority of applications coming from seed dressings. Neonicotinoids are water-soluble, and so can be taken up by a developing plant and can be found inside vascular tissues and foliage, providing protection against herbivorous insects. However, only approximately 5% of the neonicotinoid active ingredient is taken up by crop plants and most instead disperses into the wider environment. Since the mid-2000s, several studies raised concerns that neonicotinoids may be having a negative effect on non-target organisms, in particular on honeybees and bumblebees. In response to these studies, the European Food Safety Authority (EFSA) was commissioned to produce risk assessments for the use of clothianidin, imidacloprid and thiamethoxam and their impact on bees. These risk assessments concluded that the use of these compounds on certain flowering crops poses a high risk to bees.

In this modern era of agriculture, in order to feed the fast growing global population, there should be tremendous increase in the crop productivity from the limited land available. Hence, modern agriculture relies on chemical insecticides to reduce the pest attack, thereby, increasing the crop production. Within the different classes of insecticides, neonicotinoids which include imidacloprid, acetamiprid, clothianidin, thiamethoxam, thiacloprid, dinotefuran and nitenpyram, are neurotoxic specifically acting as agonists of the insect nicotinic acetylcholine receptors (nAChR) (Matsuda et al., 2001. Neonicotinoids are broadly divided into two groups which include nitro-substituted group and cyano-substituted group. Among them, nitro-substituted compounds are found to be more toxic to insects (Iwasa et al., 2004. Unlike nicotines, these chemicals have a much higher affinity to insect receptors than for the mammalian receptor, resulting in a much more favourable toxicological profile than nicotine. They have both contact and ingestion action in insects. Among the various neonicotinoids, imidacloprid was widely used. Imidacloprid was introduced since 1990’s and are effectively used for the control of important agricultural crop pests by spraying, seed dressings and soil application. Large-scale use of neonicotinoids was started in 2004 and has rapidly increased to make neonicotinoids the most widely used class of insecticides world-wide. They are registered in 120 countries and made up 80% of all seed treatment sales in 2008 (Jeschke et al., 2011).

Neonicotinoids are widely used insecticide due to their systemic in action, can be applied by different means viz., seed coatings, soil drenches or granules, foliar sprays, by injection into tree trunks or by chemigation and less toxic to birds and other mammals as compared to old insecticides. It has recently emerged that neonicotinoids can persist and accumulate in soils. They are water soluble and prone to leaching into waterways. Being systemic, they are found in nectar and pollen of treated crops. On the basis of these findings, the European Union adopted a partial ban on these substances in May 2013. Much of the recent work has focused on the impact of neonicotinoids on bees, a growing body of evidence demonstrates that persistent, low levels of neonicotinoids can have negative impacts on a wide range of free-living organisms. Neonicotinoids affect the beneficial insects, impact the efficiency of natural biological control. Also affect a soil health, depend on soil invertebrates that play a major role in decomposition and nutrient cycling. Imidacloprid and clothianidin used as seed treatment may pose risks to small birds and cause mortality or reproductive impairments. Imidacloprid and clothianidin exert sublethal effects ranging from genotoxic and cytotoxic, impaired immune function, reduced growth and reproductive success in fishes, mammals and birds. Due to these negative environmental impacts, several countries restricted the use of imidacloprid, thiamethoxam and clothianidin.
CURRENTLY AVAILABLE NEONICOTINOIDS, MANUFACTURER AND THEIR TRADE NAMES

<table>
<thead>
<tr>
<th>Name</th>
<th>Company</th>
<th>Products</th>
<th>Formulation registered</th>
</tr>
</thead>
<tbody>
<tr>
<td>Imidacloprid</td>
<td>Bayer CropScience</td>
<td>Confidor, Admire, Gaucho, Advocate</td>
<td>17.8% SL, 70% WS, 48% FS, 30.5% SC, 70%WG, 0.3% GR</td>
</tr>
<tr>
<td>Thiamethoxam</td>
<td>Syngenta</td>
<td>Actara, Platinum, Cruiser</td>
<td>25% WG, 70% WS, 30%FS</td>
</tr>
<tr>
<td>Clothianidin</td>
<td>Bayer CropScience</td>
<td>Poncho, Dantosu, Dantop</td>
<td>50% WDG</td>
</tr>
<tr>
<td>Acetamiprid</td>
<td>Nippon Soda</td>
<td>Mospilan, Assail, Chipco</td>
<td>20% SP</td>
</tr>
<tr>
<td>Thiacloprid</td>
<td>Bayer CropScience</td>
<td>Calypso</td>
<td>21.7% SC</td>
</tr>
<tr>
<td>Dinotefuran</td>
<td>Mitsui Chemicals</td>
<td>Starkle, Safari, Venom</td>
<td>20% SG</td>
</tr>
<tr>
<td>Nitenpyram</td>
<td>Sumitomo Chemical</td>
<td>Capstar, Bestguard</td>
<td></td>
</tr>
</tbody>
</table>

Environmental impact of pesticides

The synthetic organic insecticides used widely in agriculture are general biocides having innate ability to cause injury to all living organisms as well as to quality of our environment. The presence of residues of these pesticides in food commodities and other components of environment has proved toxic to humans, domestic animals, birds, fishes and non target fauna of agroecosystems. There are various environmental categories on which pesticides may pose risks

Human toxicity and health effects
1. Insecticide residues
2. Insecticide resistance
3. Insect resurgence
4. Toxicity to non target organisms (Natural enemies, bee toxicity, soil organisms and fishes)

Impact of neonicotinoids on abiotic agent abiotic agent- air

Seed coating/dressing is the leading delivery method for neonicotinoids in agriculture throughout the world. Contamination of the air and surrounding environment was the result of the abrasion and separation of the insecticide coating away from seed kernels during planting and the subsequent expulsion of insecticide particles into the environment via the exhaust fan system of the sowing machine. Concerns regarding pesticide-contaminated dust from neonicotinoid treated seeds originated from reports of honeybee losses in several countries following the planting of treated maize in spring. These incidents have been reported in Italy, France, Slovenia, Germany, USA, and Canada dating as far back as 1999 and as recently as 2013 (Krupke et al., 2012 and Van der Geest 2012. In all cases, a great number of dead and dying bees were found near the hive entrance and neonicotinoids used in seed treatments were consistently found in pollen stored in affected hives (Krupke et al., 2012. Given that bee deaths have occurred in conjunction with the sowing of treated seeds, much attention has focused on possible routes of exposure for honeybees, both during and shortly after the planting period. Neonictotinoid-contaminated dust also poses a risk to nontarget organisms through a variety of mechanisms whether they are exposed to insecticides by contact (dust cloud or deposition on vegetation) or through the ingestion of contaminated plant products (pollen, nectar, etc.).

Abiotic agent- soil

The primary method for application of the systemic neonicotinoids for agricultural pest control is the planting of seeds that are coated with the insecticide. For other pest control uses, insecticides can be applied directly to soils for uptake by plants or to the plants themselves by stem injections (Kreutzweiser et al., 2009. The subsequent breakdown of plant material containing insecticide residues can release concentrations back into the soils, thereby providing a further route of soil contamination (Horwood 2007. Neonictotinoid have been shown to pose a risk of harm to earthworms and other soil invertebrates (Pisa et al., 2014. In doing so, they have the potential to adversely affect soil ecosystem services (Chagnon et al., 2014. Neonictotinoids can remain present in measurable concentrations for long periods (months to years) in the soil. Bonmatin et al., (2005) analyzed the concentration of imidacloprid in 74 soils covering a broad range of climates, soil type and agricultural practices in France. Imidacloprid was detected in 91 % of the samples (>0.1 μg/kg), although only 15 % of the sites had been planted with treated seeds during the same year.

Abiotic agent- water

Systemic pesticides used on agricultural fields, grass, turf, hard surfaces such as lawns or concrete may contaminate surface or groundwater through runoff, leaching, drains, spillage, greenhouse wastewater and spray or dust drift (Gerecke et al., 2002).
Contamination by Spray or dust drift

Spray application may lead to direct contamination of surface water. This may be caused by unintentional overspray, careless application, or wind dispersal. In addition, dust emission from treated seeds during planting has the potential to drift to adjacent areas.

Contamination by Leaching

Neonicotinoids are mobile in the soil and thus represent a potential contamination threat to surface water and groundwater. Leaching of pesticides is one of the main mechanisms responsible for the contamination of groundwater and surface water. The leaching process is highly variable across different soil types, pesticide formulations, and application methods (Huseth and Groves 2014. Dinotefuran and clothianidin have a very high leaching potential, imidacloprid and thiamethoxam have a high leaching potential, while nitenpyram are classified as possible leachers (PPDB 2012. Contrary to the other systemic pesticides, acetamiprid and thiacloprid break down readily in soil, thereby decreasing the risk of leaching.

Contamination by Runoff

Neonicotinoids often used to control insect pests in urban or residential areas. Use of these insecticides on ornamental plants or near impervious surfaces creates a potential mode of contamination for aquatic ecosystems through runoff during rainfall or irrigation (Thuyet et al., 2012. Runoff of these pesticides can also occur in agricultural settings. Residues can occur on plant surfaces after foliar applications or accumulation of pesticide-contaminated dust and these residues can be washed off during rain events leading to contamination of surface waters. Climate change is expected to play a role in altering pesticide environmental fate in the future. The likelihood of runoff increases with precipitation levels, with increased frequency and intensity of storm events and with increasing pest pressure under climate change effects. As a consequence, the risk of pesticide runoff is likely to be elevated (Kattwinkel et al., 2011. Bloomfield et al., (2006) examined the impacts of this for pesticide behavior in groundwater and surface water in the UK. Pesticide mobility is expected to increase through more frequent heavy rainfall events, increased soil erosion, and cracking of soils leading to faster by-pass flows in winter. In the drier periods, lower flow in rivers also has the potential to increase pesticide concentration and accumulation in sediments (Masiá et al., 2013. On the other hand, higher soil and surface water temperatures due to climate change will decrease some pesticide half-life times.

Contamination by Drainage

Systemic pesticides are also used in greenhouses, where application techniques include drenching of flower bulbs or chemigation. The wastewater drainage from these greenhouses is often released into surface water and contains high levels of neonicotinoids.

Impact of neonicotinoids on biotic agent:

Impact of neonicotinoids on pollinators- honeybees

Being systemic insecticide, they can easily translocate through the plant system and thereby leaves significant amount of residue on various plant parts, such as pollen and nectar. So potentially bees could be exposed at a large scale to insecticide residues and which could be one of the probable reasons for the decline of honey bee population. This decline in the pollinating species will lead to a parallel decrease of the plant species (Goulson et al., 2008).

Relative toxicity of neonicotinoids in comparison with other groups

Among the various systemic insecticides used, imidacloprid causes the acute toxicity with the LD$_{50}$ value of 0.005μg/ bee, whereas, the LD$_{50}$ values for dimethoate and cypermethrine has 0.152 and 0.160 μg/ bee, respectively. This data showed that imidacloprid was found to be highly toxic to bees in comparison with other commonly used systemic insecticides (Suchail et al., 2001).

LD$_{50}$ Values of different neonicotinoids (Hopwood et al., 2012)

<table>
<thead>
<tr>
<th>Neonicotinoid</th>
<th>Oral LD$_{50}$ (ng /bee)</th>
<th>Contact LD$_{50}$ (ng /bee)</th>
<th>Soil half life (Days)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Clothianidin</td>
<td>2.8-3.79</td>
<td>22-44</td>
<td>148-1155</td>
</tr>
<tr>
<td>Imidacloprid</td>
<td>3.3-81</td>
<td>17.9-243</td>
<td>40-997</td>
</tr>
<tr>
<td>Thiamethoxam</td>
<td>5</td>
<td>24-29</td>
<td>25-100</td>
</tr>
<tr>
<td>Dinofuran</td>
<td>7.6-23</td>
<td>24-61</td>
<td>138</td>
</tr>
<tr>
<td>Thiacloprid</td>
<td>8.510-17,300</td>
<td>14,600-38,830</td>
<td>1-27</td>
</tr>
<tr>
<td>Acetamiprid</td>
<td>8,850-14,520</td>
<td>7,100-8,091</td>
<td>1-8</td>
</tr>
</tbody>
</table>
Reason for higher toxicity of nitro derivatives as compared to cyano derivatives

Neonicotinoids (e.g. Imidacloprid, Thiacloprid, Acetamiprid, Clothianidin, Dinotefuran, Nitenpyram and Thiamethoxam) as a group are extremely toxic to insects while remaining safe for mammals. Imidacloprid is possibly the most widely used insecticide, both within the mode of action group and in the worldwide market. ‘Systemic’ means it is absorbed by the roots of plants and then moves to the above-ground parts (leaves, twigs, and branches), where it is toxic to any sucking or chewing insects. In the case of Imidacloprid it translocates in the plant and its insecticidal proprieties are enhanced by some of the products of its breakdown (metabolites), particularly it’s 5-hydroxy and olefin metabolites that are also in themselves insecticides. Scientifically, it has been proved that in younger, 7 day old bees Imidacloprid and its metabolite only have one receptor target (Receptor 1), whereas in older, 8 days old bees, a new Imidacloprid sensitive receptor appears (Receptor 2). Additionally, this new nicotinic receptor appears to have a high affinity for Imidacloprid and its 5-hydroxy metabolite, meaning that these molecules are able to strongly interact with the honeybee’s nicotinic receptor. Young honeybee workers are typically involved in ‘simple’ in-hive duties, but as they grow older, they perform more complex tasks until they reach the forager status that is associated with more demanding duties outside the hive and their by increasing susceptibility of older workers to pesticide poisoning (Guez et al., 2003. Cyano-substituted neonicotinoids like acetamiprid were less toxic to honey bees by more than two orders of magnitude as compared to the nitro substitution in laboratory studies where the insecticides were topically applied. This reduced toxicity appears to be the result of increased metabolism by P450s and the fact that the metabolites have low bee toxicity. However, in cage studies where acetamiprid and triflumizole were applied in combination to alfalfa at the maximum recommended rate, no bee mortality was detected, suggesting that certainly acetamiprid alone and even acetamiprid in combination with a potent P450 inhibitor, is safe to honey bees. More research is needed to further validate this conclusion (Iwasa et al. 2004).

Sub-lethal effects of neonicotinoids

Honeybees exposed to sublethal doses below the LD$_{50}$ value will have various effects on bees like impair learning behaviour, short and long-term memory loss, reduced fecundity (fertility and reproduction), and altered foraging behaviour and motor activity of the bees, depressing bees’ immune systems or increasing their susceptibility to biological infections- alone or in combination with fungicides (Wu et al., 2011 and Pettis et al., 2013).

Route of pesticide exposure

Since the introduction of imidacloprid in early 1990’s, the use of different neonicotinoids has grown considerably. They are used extensively for the control of important agricultural crop pests by spraying and also widely used in seed dressings and soil additions. In the latter two cases residues of these systemic insecticides can be present at ‘trace’ levels in the plant pollen and nectar. These residues get easily translocated through the plant systems and remain in trace amount in the floral parts like the pollen and nectar, to which the worker bees more frequently visit. These foragers in turn carry this contaminated pollen and honey back to their hives, where the other members are exposed either through trophallaxis. As the result the colony strength is affected considerably. Either through the direct contact with the insecticidal spray or by the residual impact, honey bees are potentially under the risk of population decline.

There are different thoughts related to the honey bee population decline. Some argue for the use of neonicotinoids whereas, some defend it. At first we deal with the argument that neonicotinoids are the cause of bee decline. Honey bees could be exposed to neonicotinoids in two general ways:

1.Direct contact with insecticides 2. Indirect contact by drifts and residues present on the floral parts

Direct impact of the insecticidal spray:

Losses of bees have been reported in Italy mainly in the fields where maize coated with neonicotinoids was sown using pneumatic drilling machine used. Due to this the insecticidal drifts fall on the nearby vegetables. Foragers were exposed to these drifts may have direct acute toxicity. An experiment was conducted to study the acute toxicity, wherein, bees fed with guttation drops and dew collected from the surrounding vegetation. Results revealed that there was no acute toxicity. Hence another experiment of direct aerial spraying of pesticides was undertaken, where the bees were exposed in the sown area under caged condition. The data showed that bees were killed by powder, only if held in high humidity, indicating synergistic effect of neonicotinoid and humidity (Marzaro et al., 2011). About 2-20% of a seed treatment is absorbed by the plant. The rest is either blown into the air by seed drills, or remains in soil. The toxic airborne dust from seed drills can kill flying bees and ultimately pollutes non-target plants and soil. Seed treatments and soil drenches are finally washed away into ground and surface water (Krupke et al., 2012. Amounts used on ornamentals lead to residues 12-16x greater than found on crop plants (Hopwood et al., 2012).
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Indirect impact of the insecticidal spray

More than the direct impact, bees are subjected to the indirect impact on the insecticidal spray via, residues. An experiment was conducted by Stoner et al., 2012 to study the translocation of Imidacloprid and Thiamethoxam into Nectar and Pollen of Squash (Cucurbita pepo). It was found that imidacloprid concentration in the nectar and pollen was 10±3 ppb and 14±8 ppb respectively, whereas, the thiamethoxam concentration was 11±6 ppb and 12±9 ppb respectively.

Sub-lethal effect of imidacloprid doses on foraging activity

An experiment conducted by Bortolotti et al., in 2003 to study the sub lethal effect of imidacloprid doses on the foraging activity. Bees were trained to forage on an artificial feeder filled with a 50% sucrose solution. Later on, these feeders were moved gradually to 500m away from the foraging sites. Randomly about thirty bees were captured, individually colour marked and transferred into a flying cage and maintained as control. Feeder was then replaced with new bees and fed with imidacloprid supplemented sucrose solution. Again randomly about thirty bees were captured, individually colour marked and transferred into a flying cage. Three concentrations of imidacloprid were tested 100 ppb, 500 ppb and 1000 ppb respectively. Bees fed with 100 ppb returned to the hive 24 hours after the release. Those fed with 500 ppb &1000 ppb completely disappeared after the release and were seen neither at the hive nor at the feeding site. Results revealed that neonicotinoids interfered with the foraging capacity of the bees. Here the main drawback being that some bees were not effectively tracked during foraging and their disappearance was proved mysterious. Hence some modern techniques was used to refine overcome the draw backs of the above earlier experiments.

Rfid tracking of foragers to study the sub-lethal effect of neonicotinoid doses

Schneider et al., 2012 conducted an experiment to study the sub-lethal effect of imidacloprid and Clothianidin doses on foragers using Imidacloprid treated bees

<table>
<thead>
<tr>
<th>Parameters</th>
<th>1.5 ng</th>
<th>3ng</th>
</tr>
</thead>
<tbody>
<tr>
<td>Return to the hive</td>
<td>100 %</td>
<td>95  %</td>
</tr>
<tr>
<td>Duration of a single foraging trip</td>
<td>50 %</td>
<td>130%</td>
</tr>
<tr>
<td>Feeder Visit</td>
<td>47%</td>
<td>98%</td>
</tr>
<tr>
<td>Flight time from hive to the feeder</td>
<td>64.7%</td>
<td>241.1%</td>
</tr>
<tr>
<td>Time spent at the feeder</td>
<td>27.5%</td>
<td>45.6%</td>
</tr>
<tr>
<td>Flight time from feeder to the hive</td>
<td>20%</td>
<td>210%</td>
</tr>
<tr>
<td>Time spent within hive</td>
<td>33%</td>
<td>993%</td>
</tr>
</tbody>
</table>

Bees treated with imidacloprid showed peculiar symptom like, foragers can’t fly directly to the hive, reduced mobility, followed by a phase of motionlessness with occasional trembling and cleaning movements.

Clothianidin treated bees

<table>
<thead>
<tr>
<th>Parameters</th>
<th>0.5ng</th>
<th>1ng</th>
<th>2ng</th>
</tr>
</thead>
<tbody>
<tr>
<td>Return to the hive</td>
<td>94.4 %</td>
<td>73.8 %</td>
<td>20.6%</td>
</tr>
<tr>
<td>Duration of a single foraging trip</td>
<td>20 %</td>
<td>32.2%</td>
<td>109.3%</td>
</tr>
<tr>
<td>Feeder Visit</td>
<td>31.1%</td>
<td>71.1%</td>
<td>74%</td>
</tr>
<tr>
<td>Flight time from hive to the feeder</td>
<td>14.1%</td>
<td>39.6%</td>
<td>101.8%</td>
</tr>
<tr>
<td>Time spent at the feeder</td>
<td>14.1%</td>
<td>39.6%</td>
<td>101.8%</td>
</tr>
<tr>
<td>Flight time from feeder to the hive</td>
<td>30%</td>
<td>40%</td>
<td>90%</td>
</tr>
<tr>
<td>Time spent within hive</td>
<td>15.8%</td>
<td>36.7%</td>
<td>95.9%</td>
</tr>
</tbody>
</table>

Bees treated with Clothianidin showed peculiar symptom like, bees move around with an awkwardly arched abdomen, sometimes followed by a phase of turning upside down and lying on the back with paddling leg movements. Results revealed that there was significant reduction of foraging activity and longer foraging flights was observed at lower doses of clothianidin and imidacloprid. RFID-method is an effective way to record short-term alterations in foraging activity after insecticides have been administered once, orally, to individual bees.

Sub-lethal effect of noenicotinoid doses on growth and development

A study conducted by Wu et al., 2011 revealed that delayed development was observed in bees reared in combs containing high levels of pesticides particularly in the early stages (day 4 and 8) of worker bee development. Adult longevity was reduced by 4 days in bees exposed to pesticide residues in contaminated brood comb during development. Survivability increased in bees reared in treatment comb after multiple brood cycles when
pesticide residues had been reduced. There was no larval mortality but demonstrated delayed worker development when brood was reared in highly contaminated brood combs. Sub-lethal effects, including delayed larval development and adult emergence or shortened adult longevity, can have indirect effects on the colony such as premature shifts in hive roles and foraging activity. In addition, longer development time for bees may provide a reproductive advantage for parasitic Varroa destructor mites.

**Sub-lethal effect of noenicotinoid doses on memory and learning**

**Imidacloprid-induced facilitation of the Proboscis Extension Reflex habituation in the honeybee**

Proboscis extension reflex (PER) it is part of a bee's feeding behavior, used to study bee learning and memory in the context of foraging. The effects of imidacloprid at a dose that does not affect sensory or motor functions are used to study the associative learning abilities in honeybee. Imidacloprid was topicaly applied on the thorax (1 μl)- at higher concentration of 5, 10 and 20 ng/ bee, gustatory threshold increased. At low dose of 1.25 ng/bee, there was no effect on the gustatory function but increased the locomotor activity. Slight activation of the cholinergic system with a low dose of imidacloprid can facilitate a simple form of learning in the honeybee which relates to how adult workers respond to and remember tastes and odours (Lambin et al., 2001).

**Sub-lethal effect of neonicotinoid doses on behaviour**

**Imidacloprid-Induced Impairment of Mushroom Bodies and Behavior of Stingless Bee**

Hudson et al., in 2012 conducted an experiment to study the effects on larval survival, development, neuro-morphology and adult walking behavior in stingless bee. More than 50% survival rates were observed at doses lower than 0.0056 mg active ingredient (a.i.)/bee. No sub-lethal effect on body mass or developmental time was observed in the surviving insects, but negatively affected the development of mushroom bodies in the brain and impaired the walking behavior of newly emerged adult workers. It was found that walking impairment increased with the increase in the concentration and it was subsequently reduced as the days increased.

**Effect on sucrose responsiveness and waggle dancing**

Eiri and Neih, 2012 conducted an experiment to study the effect of imidacloprid on two specific behavioural aspects namely Sucrose responsiveness and waggle dancing. Proboscis extension response assay was used to study the Sucrose responsiveness. When imidacloprid was used at a concentration of 0.21 and 2.16 ng per bee, the sucrose response thresholds were significantly higher and bees performed fewer waggle dance circuits. Therefore imidacloprid temporarily increased the minimum sucrose concentration that foragers would accept and reduced waggle dancing also it altered colony food intake, based on this they concluded that a sublethal dose of 0.21 ng per bee of commonly used pesticide may impair colony fitness.

**Interaction effect of neonicotinoids and other pathogens**

**Pesticide exposure in honey bees results in increased levels of the gut pathogen**

Recently Pettis et al., 2013 reported that the effect sub-lethal doses of imidacloprid to the newly emerged bee followed by the treatment of gut parasite, Nosema spp. Pesticide dosages used at low levels had no effects on longevity or foraging in adult honey bees. Nosema infections increased significantly in the bees from pesticide-treated hives when compared to bees from control hives. Interactions between pesticides and pathogens could be a major contributor to increased mortality of honey bee colonies, including colony collapse disorder, and other pollinator declines worldwide.

**Colony collapse disorder – a complex phenomenon**

The sudden disappearance of honey bees from hives has been reported by beekeepers and researched by scientists for decades and called “Disappearing Disease” (Wilson and Menapace 1979. In 2006 the widespread appearance of this phenomenon in the United States was noted and referred to as Colony Collapse Disorder by researchers, beekeepers, and the media. This increase in colony losses also corresponded to increased use of neonicotinoid pesticides (Cresswell et al., 2012. This has led to speculation that there is a causative relationship between the increased use of neonicotinoids and widespread decline in bee populations (Suryanarayanan 2013). However, it is important to look at all the variables associated with CCD. The definitive cause for the declines in the United States and Europe has yet to be fully understood. More than sixty-one variables have been associated with CCD, although none have been clearly identified as the definitive cause of the phenomenon (VanEngelsdorp D 2009.

**Impact of neonicotinoids on predator and parasitoid insects**

Insects can be exposed to neonicotinoids in various direct and indirect ways. Direct contact occurs when foliar sprays are applied to plants and spray contacts the insect or when the insect comes in contact with spray residues on the surface of vegetation or residues in the soil. Beneficial insects may also be indirectly exposed when they consume prey or plant materials that are contaminated with an insecticide.
A number of studies have looked into the impacts of direct contact due to spray applications or residues on vegetation. Dinofeturan sprays at label rates were highly toxic to a parasitoid wasp *Leptomastix dactylopiii* and spray applications of acetamiprid, clothianidin and dinofeturan were toxic to mealybug destroyer beetles *Cryptolaemus montrouzieri* (Cloyd and Dickenson 2006). Imidacloprid is toxic to the predatory *Rodolia cardinalis* when exposed to foliar treatment of citrus and by feeding poisoned cottony cushion scale. Adult survival, progeny and larval development were significantly reduced exposed to treated citrus leaves. When beetle stages were fed insecticide-treated cottony cushion scale toxic effects were more severe than contact toxicity alone (Cardwell and Gu 2003). Per cent survival of adult green lacewing was reduced after feeding on flowers from plants treated with a soil application of imidacloprid. Per cent survival was only 6%, 14 % at twice of label rate (12g) and label rate (6 g), respectively while 79% in control. Trembling of predators was observed in imidacloprid treatments (Rogers et al., 2007). Encyrtid parasitoid wasps (*Anagyrus pseudococci*) showed reduced mobility and lower survivorship after chronic exposure to flowers from plants treated with label rates of soil applied imidacloprid (Krischik et al., 2007). Lady beetles are common in agricultural crops, where they feed on aphids and other crop pests. The multicoloured Asian lady beetle (*Harmonia axyridis*), will also feed directly on corn seedlings to obtain plant specific nutrients. Larvae that fed briefly on seedlings grown from seeds treated with clothianidin or thiamethoxam experienced significantly higher mortality, but also sublethal effects like trembling, paralysis or loss of coordination (Moser and obrycki, 2009).

**Impact of neonicotinoids on mites**

Studies on mites have found a positive effect on population numbers. Sublethal doses of imidacloprid, which is used for the control of green peach aphid (*Myzus persicae*) significantly increased the hatching rate of eggs and pre-adult survivorship of the carmine spider mite (*Tetranychus cinnabarinus*) (Zeng and Wang 2010).

Application of neonicotinoids supressed expression of cotton and tomato plant defence genes. These genes alter the level of phytohormones and decrease the plant’s resistance to spider mites (*Tetranychus urticae*). When mites added to the crops, population growth increased from 30 to 100% on neonicotinoid-treated plants in the greenhouse and up to 200% in the field experiment (Szczepaniec et al., 2013).

**Impact of neonicotinoids on earthworms**

Earthworms are vitally important members of the soil fauna, especially in agricultural soils where they can constitute up to 80% of total soil animal biomass (Luo et al., 1999). Soil fertility is enhanced by earthworm effects on biogeochemical cycling (Bartlett et al., 2010), breakdown of plant litter (Knollengberg et al., 1985) and the mixing of litter with soil (Wang et al., 2012a). Neonicotinoid and other systemic insecticides can pose a risk of harm to earthworm survival and behaviour, potentially disrupting soil development and maintenance processes. The same neural pathways that allow neonicotinoids to act against invertebrate pests are also present in earthworms (Volkov et al., 2007). Thus, when neonicotinoids are applied for the protection of agricultural and horticultural crops, earthworms can be exposed by direct contact with the applied granules or seeds, or with contaminated soil or water. Moreover, their feeding activities may result in ingestion of contaminated soil and organic particles (Wang et al., 2012b). Foliar residues in plant litter after systemic uptake from soils or from direct plant injections also pose a risk to litter-feeding earthworms that consume the contaminated plant litter (Kreutzweiser et al., 2009).

**Effect of neonicotinoids on survival**

When compared to other common insecticides, neonicotinoids tend to be among the most toxic to earthworms. Wang et al., (2012a) tested the acute toxicities of 24 insecticides to *E. fetida* and found that the neonicotinoids were the most toxic in soil bioassays and that acetamiprid and imidacloprid in particular were the two most toxic insecticides overall. They also reported that a contact toxicity bioassay demonstrated that the neonicotinoids were extremely toxic by a contact route of exposure (LC$_{50}$ of 0.0088 to 0.45 μg cm$^{-2}$), although the units of contact toxicity concentration were difficult to compare to standard lethal concentrations. Across a broader range of 45 pesticides, Wang et al., (2012b) found that in soil bioassays, the neonicotinoid insecticide, clothianidin, was the most toxic pesticide to *E. fetida*.

**Effect of neonicotinoids on reproduction**

Only a few studies tested sublethal effects of neonicotinoids on earthworm reproduction, but it is apparent that reductions in fecundity can occur at low concentrations. Baylay et al., (2012) reported EC$_{50}$ for imidacloprid and thiacloprid against cocoon production by *Lumbricus rubellus* of 1.5 and 1.3 ppm, respectively, while Gomez-Eyles et al., (2009) found similar EC$_{50}$ for the same two insecticides at 1.4 and 0.9 ppm for *E. fetida*. The latter study also reported measurable reductions in cocoon production at 0.3 ppm of thiacloprid.

**Effect of neonicotinoids on behaviour**

The behavioural attributes considered here are avoidance behaviour, burrowing, cast production and weight change. Among the 31 reported values for behavioural effects, weight change was the most common, followed by burrowing, avoidance behaviour and cast production.
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<table>
<thead>
<tr>
<th>Species</th>
<th>Insecticide</th>
<th>Effect</th>
<th>Lowest effective concentration</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Lumbricus terrestris</em></td>
<td>Imidacloprid</td>
<td>Burrowing</td>
<td>2 ppm</td>
<td>Dittbrenner et al., 2011</td>
</tr>
<tr>
<td><em>L. terrestris</em></td>
<td>Imidacloprid</td>
<td>Body mass change, cast production</td>
<td>0.6 ppm 0.6 ppm</td>
<td>Dittbrenner et al., 2010</td>
</tr>
<tr>
<td><em>L. terrestris</em></td>
<td>Imidacloprid</td>
<td>Cast production, body mass change</td>
<td>1.89, 0.189 ppm</td>
<td>Capowiez et al., 2010</td>
</tr>
<tr>
<td><em>Aporrectodea nocturna</em></td>
<td>Imidacloprid</td>
<td>Weight loss, avoidance, burrowing</td>
<td>0.5, 0.1, 0.05 ppm</td>
<td>Capowiez and Berard 2006</td>
</tr>
<tr>
<td><em>A. nocturna</em></td>
<td>Imidacloprid</td>
<td>Burrowing</td>
<td>0.01 ppm</td>
<td>Capowiez et al., 2003</td>
</tr>
<tr>
<td><em>A. icterica</em></td>
<td>Imidacloprid</td>
<td>Weight loss, avoidance</td>
<td>0.5, 0.01 0.05 ppm</td>
<td>Capowiez and Berard 2006</td>
</tr>
<tr>
<td><em>A. icterica</em></td>
<td>Imidacloprid</td>
<td>Burrowing</td>
<td>0.1 ppm</td>
<td>Capowiez et al., 2006</td>
</tr>
<tr>
<td><em>Dendrobaena octaedra</em></td>
<td>Imidacloprid</td>
<td>Weight loss</td>
<td>3 ppm</td>
<td>Kreutzweiser et al., 2008b</td>
</tr>
<tr>
<td><em>Eisenia andrei</em></td>
<td>Imidacloprid</td>
<td>Avoidance</td>
<td>0.13 ppm</td>
<td>Alves et al., 2013</td>
</tr>
</tbody>
</table>

Impact of neonicotinoids on microbes

Soil microbial communities have also been affected by imidacloprid, which play major role in leaf decomposition and nutrient cycling. Imidacloprid (3-11 mg kg\(^{-1}\)) systemically applied to sugar maple tree for control of the Asian longhorned beetle. Senescent leaves falling from systemically treated trees with residue levels, sufficient to reduce natural decomposition process through adverse effects on non-target decomposer organisms (Kreutzweiser et al., 2008a. Yao et al., 2006 reported acetamiprid at field concentrations significantly inhibited soil respiration. Acetamiprid applied at normal field concentration (0.5 mg kg\(^{-1}\) dried soil) and at high concentration (5 and 50 mg kg\(^{-1}\) dried soil) showed strong negative influence on soil respiration at these concentrations.

Impact of neonicotinoids on aquatic species

Fish may be affected indirectly by reductions in food resources, particularly aquatic invertebrates (Hayasaka et al., 2012a, b. Imidacloprid was shown to cause a stress syndrome in juvenile Japanese rice fish (medaka). As often happens with stressed fish, a massive by a parasite, *Trichodina* ectoparasite, was observed in medaka fish in imidacloprid-treated fields. Hayasaka et al., 2012 performed experiment by using two successive annual treatments of imidacloprid on aquatic communities of paddy mesocosms. Body size of adult medaka fish and their juveniles were periodically smaller in the imidacloprid treated mesocosms than in control over two years. Imidacloprid reduced the benthic arthropod prey, led to reductions in growth of medaka fish.

<table>
<thead>
<tr>
<th>Species</th>
<th>Effect on</th>
<th>Neonicotinoid</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fathead minnow</td>
<td>Growth and development</td>
<td>Clothianidin 20 mg/L</td>
<td>DeCant and Barrette 2010</td>
</tr>
<tr>
<td><em>Pimephales promelas</em></td>
<td>(reduced weight and length)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Nile tilapia, <em>Oreochromis niloticus</em></td>
<td>Growth and development</td>
<td>Imidacloprid 0.134 mg/L</td>
<td>Lauan and Ocampo 2013</td>
</tr>
<tr>
<td></td>
<td>(extensive disintegration of</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>testicular tissue</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Nile tilapia, <em>Oreochromis niloticus</em></td>
<td>Growth and development</td>
<td>Imidacloprid &lt; 0.134 mg/L</td>
<td>Ocampo and Sagun 2007</td>
</tr>
<tr>
<td></td>
<td>(changes to gonads)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Medaka, <em>Oryzias latipes</em></td>
<td>Immunotoxic</td>
<td>Imidacloprid 0.03-0.24 mg/L</td>
<td>Sanchez-Bayo and Goka 2005</td>
</tr>
<tr>
<td></td>
<td>(Juveniles stressed, led to ectoparasites infestation)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Impact of neonicotinoids on birds

Pesticides can exert their impact on vertebrates either directly through their toxicity or indirectly by reducing their food supply. Direct effects may be the result of several different exposure pathways through ingestion of the formulated product (e.g. birds eating seeds coated with insecticide), through uptake via the skin following a spray event or by eating contaminated prey and indirect effects which are typically mediated through loss in...
quantity or quality of prey associated with pesticide use or through habitat modification. Birds are mainly affected by seed treatment with neonicotinoids. A single corn kernel coated with a neonicotinoid can kill a songbird. Birds depend heavily on the aquatic invertebrates as a prey but neonicotinoid contamination levels in surface and groundwater are strikingly high, sufficient to kill many aquatic invertebrates. About 1/10th of a lethal dose can cause chronic and reproductive effects.

<table>
<thead>
<tr>
<th>Species</th>
<th>Effect on</th>
<th>Insecticide &amp; dose</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mallard, <em>Anas platyrhynchos</em></td>
<td>Reproduction</td>
<td>Imidacloprid mg/kg/day</td>
<td>Mineau and Palmer 2013</td>
</tr>
<tr>
<td>Red-legged partridge, <em>Alectoris rufa</em></td>
<td>Survival (reduced chick survival &amp; adult survival)</td>
<td>Imidacloprid mg/kg/day</td>
<td>Lopez-Antia et al., 2013</td>
</tr>
<tr>
<td>Red-legged partridge, <em>Alectoris rufa</em></td>
<td>Reproduction (reduced fertilisation rate &amp; chick survival)</td>
<td>Imidacloprid mg/kg/day</td>
<td>Lopez-Antia et al., 2013</td>
</tr>
<tr>
<td>Red-legged partridge, <em>Alectoris rufa</em></td>
<td>Immunotoxic (reduced immune response)</td>
<td>Imidacloprid mg/kg/day</td>
<td>Lopez-Antia et al., 2013</td>
</tr>
<tr>
<td>Northern bobwhite quail, <em>Colinus virginianus</em></td>
<td>Reproduction</td>
<td>Clothianidin mg/kg/day</td>
<td>Mineau and Palmer 2013</td>
</tr>
<tr>
<td>Northern bobwhite quail, <em>Colinus virginianus</em></td>
<td>Growth and development</td>
<td>Imidacloprid mg/kg/day</td>
<td>Mineau and Palmer 2013</td>
</tr>
<tr>
<td>Japanese quail, <em>Coturnix japonica</em></td>
<td>Reproduction (testicular anomalies, reductions in embryo length)</td>
<td>Imidacloprid mg/kg/day</td>
<td>Tokumoto et al., 2013</td>
</tr>
<tr>
<td>Japanese quail, <em>Coturnix japonica</em></td>
<td>Genotoxic (increased breakage of DNA in males)</td>
<td>Imidacloprid mg/kg/day</td>
<td>Tokumoto et al., 2013</td>
</tr>
<tr>
<td>House sparrow, <em>Passer domesticus</em></td>
<td>Neurobehavioural (Incoordination, inability to fly)</td>
<td>Imidacloprid mg/kg/day</td>
<td>Cox 2001</td>
</tr>
</tbody>
</table>

**Impact of neonicotinoids on mammals**

Mammals have various routes of exposure *viz.*, From ingestion of treated seed, by residues in or on the crop, drinking water, nearby vegetation or invertebrates, by dermal exposure and by inhalation.

<table>
<thead>
<tr>
<th>Species</th>
<th>Effect on</th>
<th>Insecticide &amp; dose</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rat, <em>Rattus norvegicus</em></td>
<td>Reproduction (reduced sperm production)</td>
<td>Imidacloprid mg/kg/day</td>
<td>Bal et al., 2012</td>
</tr>
<tr>
<td>Rat, <em>Rattus norvegicus</em></td>
<td>Growth and development (reduced weight gain)</td>
<td>Imidacloprid mg/kg/day</td>
<td>Cox, 2001</td>
</tr>
<tr>
<td>Rat, <em>Rattus norvegicus</em></td>
<td>Neurobehavioural</td>
<td>Imidacloprid 337 mg/kg</td>
<td>Abou-Donia 2008</td>
</tr>
<tr>
<td>Rat, <em>Rattus norvegicus</em></td>
<td>Immunotoxic (Significant effect on leukocyte count, immunoglobulins and phagocytic activity)</td>
<td>Imidacloprid 0.21 mg/kg/day</td>
<td>Mohany et al., 2011</td>
</tr>
<tr>
<td>Rabbit, <em>Sylvilagus sp.</em></td>
<td>Reproduction (Increased frequency of miscarriage)</td>
<td>Imidacloprid 72 mg/kg/day</td>
<td>Cox, 2001</td>
</tr>
<tr>
<td>Rabbit, <em>Sylvilagus sp.</em></td>
<td>Reproduction (Increase in premature births)</td>
<td>Imidacloprid &gt; 25 mg/kg/day</td>
<td>DeCant and Barrett 2010</td>
</tr>
</tbody>
</table>

**REGULATION**

In 2008, Germany revoked the registration of clothianidin for use on seed corn after an incident resulted in death of millions of honey bees (Alison B 2008. Neonicotinoid seed treatment is banned in Italy, but foliar use is allowed. This action was taken based on preliminary monitoring studies showing that bee losses were correlated with the application of seeds treated with these compounds (Keim B 2010. In France, sunflower and corn seed treatment with imidacloprid are suspended, imidacloprid seed treatment for sugarbeets and cereals are allowed, as is foliar use (US EPA, 2011. In 2012, European Commission asked the European Food Safety Authority to study the safety of imidacloprid, thiamethoxam and clothianidin. European Commission recommended a restriction of three neonicotinoids for two years from 1 December 2013 across the European Union. The Law restricts the use of three neonicotinoids for seed treatment, soil application and foliar treatment in crops attractive to bees (Gibson 2013.)
CONCLUSION

Neonicotinoids are widely used insecticides. They are systemic, persistence and water soluble in nature. Available in the environment at that levels that are known to cause lethal and sublethal effects on non target organisms. They may cause direct mortality or sublethal effects viz., impaired learning behaviour, short and long-term memory loss, reduced fecundity, altered foraging behaviour and motor activity of the bees and depressing immune system in bees. Neonicotinoids affect the beneficial insects, impact the efficiency of natural biological control. Also affect a soil health, depend on soil invertebrates that play a major role in decomposition and nutrient cycling. Imidacloprid and clothianidin used as seed treatment may pose risks to small birds and cause mortality or reproductive impairments. Imidacloprid and clothianidin exert sublethal effects ranging from genotoxic and cytotoxic, impaired immune function, reduced growth and reproductive success in fishes, mammals and birds. Due to these negative environmental impacts, several countries restricted the use of imidacloprid, thiamethoxam and clothianidin.

References


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Mineau P and Palmer C . 2013. The impact of the nation’s most widely used insecticides on birds. *American Bird Conservancy*, USA.


Recent Trends in Integrated Pest and Disease Management


**Recent Trends in Integrated Pest and Disease Management**

**GLOBAL SCENARIO OF TOMATO PIN WORM, TUTA ABSOLUTA (MEYRICK) AND THEIR MANAGEMENT**

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²ICAR-Indian Institute of Sugarcane Research, Lucknow- 226 002 (U.P.), India.

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**Abstract**

*Tuta absoluta* is a damaging pest for tomato plant. Attack of this pest causes heavy losses in crop yield. It feeds on every part of the plant either stem or calyces or fruits. Controlling this pest is important. Application of integrated control method is an effective method against *Tuta absoluta* of tomato. There is a need for using such combination products as a pest management strategy for achieving expected results. New insect management strategies should be based on treatments that have been already established not on occurrence (presence/absence) of pest. This will help in reducing the costs. Effective use of these management strategies in combination with intensive exploration of population quantity of pests will enhance sustainability of the tomato.

Alien species are species of plants, insect pests that inter a country or region accidentally outside their natural habitat. Invasive alien species is an alien species which becomes established in natural or semi-natural ecosystems an agent of change and threats native biological diversity. This pest does not belong to that area where the occurrence of damage has been reported and so these are considered to be the invasive ones. *Tuta absoluta* is a neotropical, oligophagous pest which infests more several solanecous crops and others too. Since 1960s, this moth has attained the position amongst the key pests of tomato in South America. An invasive species can be any kind of living organism that is not native to an ecosystem which causes harm. They can harm the economy, environment, human health. South American tomato pin worm, *Tuta absoluta* (Meyrick, 1917) (Lepidoptera: Gelechiidae) commonly called as tomato leaf miner is amongst one of the critical invasive pest seen for the first time infesting tomato crop in Maharashtra, India. On basis of classification it is the most severe danger for tomato all through world. This pest has covered South America to many parts of Europe, entire Africa and is now being spreading at a rapid pace in India. The infestation of this pest on plants is either direct or through sites of infection of other pest. Reports have shown that 90 per cent of yield losses and poor quality of fruit due to this pest attack on tomato. Its occurrence in India was reported for the first time by different workers in different parts of India. Its occurrence in and around Bangalore was reported by Sridhar et al. (2014); around Pune by Shashank et al. (2015); in Malnad and Hyderabad-Karnataka region by Kalleshwaraswamy et al. (2015) and in Telangana by Kumari et al. (2015. EPPO (2010) had showed that propagation of *T. absoulta* could be due to transportation of packaging materials from country to country. Occurrence of this pest in any country had been reported to be the most damaging pest in respect to tomato plant (Germain et al., 2009. In India, recent reports stated the attack of this pest in tomato but due to Oligophagous nature of this pest, it can cause damage to other solanaceous plants which makes this pest of utmost importance. In this chapter, focuses have been made on its life cycle, symptoms, damage per cent and management strategies which will be helpful for awaring the farmers from this problem.

**Global Spread**

*Tuta absoluta* was only present in South America at the end of 2006. It was first detected in the province of Castello (Spain) where significant damage is reported (Urbaneja et al., 2007. The incidence of *T. absoluta*, a new pest in Integrated

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Fig. 1: Incidence of *T. absoulta* in Tomato plant in the world
(Source: EPPO and Office of international research, education and development, Virginia Tech)
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pest management tomato fields in the northeast of Spain and also its natural enemies were observed (Arno et al., 2009). The occurrence of *T. absoluta* in Bulgaria, for the first time was noticed in a glasshouse with tomato plants and severe infestation by the pest was observed which had rapidly spread after the last treatment with pesticides. The leaves were the most heavily damaged plant parts with an average of 9.42 and 8.75 mines per leaflet on the middle and upper layer of the canopy, respectively (Harizanova et al., 2009). The occurrence of *T. absoluta* (Meyrick) (Lepidoptera: Gelechiidae) in the Bosnia and Herzegovina was noticed based on the moths trapped from pheromones placed in the greenhouses and open field. From Spain it had spread to European and Mediterranean countries (Urbaneja et al., 2009), later it was noticed in Turkey and then in Iraq later it was reported in east of Iran (Desneux et al., 2010). Due to its exceptional speed of spread, Desneux et al. (2011) predicted it might reach India. Following its introduction to Afro-Eurasia in 2010 it was in the quarantine watch-list of India. In 2014, *T. absoluta* was first time noticed in Pune, India (Shashank et al., 2015).

In India, this pest was first recorded from states like Karnataka and Maharastra in October 2014. Later, it spread to neighbouring states like Andhra Pradesh, Telangana and Tamil Nadu. Recently its incidence is also reported from Delhi and mid-hills of Himachal Pradesh. Now this new invasive pest has crossed all the borders and spread to Varanasi and Mirzapur areas of Uttar Pradesh. So, it is important to keep a vigil on incidence, extent of damage by this new pest in tomato and its spread to other vegetable crops in the regions.

Reason for Spread and Distribution

Fruit import & commercialization of the crop. Long distance dissemination – packaging boxes through infested countries Wind currents seem to be especially favourable for its dispersal in Spain, International trade & movement of materials – Increased risk of alien species invasion. Import of Tomato to fulfill domestic needs of Kathmandu valley has led to introduction of *T. absoluta* to Nepal from India during 2014.

Host Range

Tomato (*Solanum lycopersicon*) Potato (*Solanum tuberosum*), Eggplant, Tobacco, Poplar, Sweet cucumber, Beans etc., Other alternate hosts : *Solanum nigrum*, *S. eleagnifolium*, *S. bonariense*, *S. sisymbriifolium*, *S. saponaceum*, *S. lyratum*, *Lycopersicum puberulum*, *Datura stramonium*, *D. ferox*, *Nicotiana glauca*. The larvae of *T. absoluta* preferred tomato leaves at the most followed by brinjal leaf and tomato fruit and Chilli was least preferred.

Identification

The moth has a grey-brown colour, is approximately 6 mm in size and has a wingspan of about 10 mm Newly-hatched caterpillars are approximately 0.5 mm in size and have a yellowish colour. When maturing, caterpillars turn yellow-green and a black band develops behind the head. Fully grown caterpillars are approximately 9mm in size with a pinkish colour on the back. The pupa is light brown and approximately 6mm in size.

Nature of Damage and Symptoms:

This pest loves to feed on Solanaceous vegetables but it feeds on other vegetables too. The young stage of this pest nourishes on stems, leaves, buds, calyces and even on fruits (young ones/ripened ones. When larva feeds on stems of plants they act as miners but when they do on fruits, they act as borer. The infestation of these pests causes damage of 50-100 per cent loss in yield and quality of fruit. ETL level 2-5 larvae per plant. Symptoms
revealed that white patches were observed on leaves and later these patches get dried giving an appearance of burnt leaves. In fruits, pin holes are being observed at the point of entrance and exit (Fig. 5. These sites cause secondary infections and rotting in fruits.

![Symptoms of Tuta absoluta](image)

**Fig. 5. Symptoms of Tuta absoluta on tomato plant a. Infected leaves b. Infected fruits c. Larva infesting on tomato**

**Management Strategies:**

Integrated pest management (IPM) strategies are the effective ones for controlling of *Tuta absoluta* insect. The integrated control method recommended employs the following (1) Use light traps and pheromone traps for monitoring (2) Deep summer plough in to expose the resting stage of pests (3) Removing of crop residues and alternate hosts (4) Crop rotation with non- solanaceous crops (5) Destroy the affected plants and damaged fruits (6) Selection of pest (whitefly/spider) resistant wild tomato varieties for resistance of this pest (7) Biological control agent such as *Egg parasitoid Trichogramma pretiosum* has been widely used to control *Tuta absoluta* (8) The usage of chemicals for controlling this pest such as imidacloropid in the irrigation water after 8-10 days of planting, chlorantraniliprole 20SC (Rynaxpyr) @ 0.35 ml/l or cyantraniliprole 10 OD (Cyzapyr) @ 1.8 ml/l or indoxacarb 14 SC @ 75 (g a.i./ha) or dimethoate 30 EC @ 275 (g a.i./ha) sprayed at 10 days interval for control of this pest.

**References**


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WEED MANAGEMENT IN BLACKGRAM

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Abstract

Now days, various weed control methods were found to be effective in controlling weeds in Blackgram and also its each other methods have their own merits and demerits based on resource available or environmental condition. However, efficient and cost-effective weed control can be achieved by using either combination of herbicides or combining herbicide alone or any one of the weed control method may not control the weeds effectively. In such condition, an integrated weed management (IWM) practice involving both chemical and other agronomic manipulation may be an efficient tool, as increasing crop density seems to be an alternative to shift crop weed competition in favour of crop. An integrated weed management practice involving both chemical and other agronomic manipulation may be an efficient tool, as increasing crop density seems to be an alternative to shift crop weed competition in favour of crop.

Black gram [Vigna mungo L] is one of the most important pulse crops of India, it belongs to family Leguminoseae. India is the largest producer and consumer of black gram in the world. It is rich in protein, which contains about 26 per cent protein, and supplies a major share of protein requirement of vegetarian population of the country. It is consumed in the form of split pulse as well as whole pulse, which is an essential protein supplement of cereal based diet. The combination of pulse-rice or dal-roti is an important ingredient in the average Indian diet. The biological value improves greatly, when wheat or rice is combined with it because of the complementary relationship of the essential amino acids such as arginine, leucine, lysine, isoleucine and valine etc. Uncontrolled Weeds at critical period of crop-weed competition caused a reduction of 80-90 per cent in Blackgram yield depending upon type and intensity of weed infestation (Kumar et al., 2001). To control weed the traditional method of weed control i.e. hand weeding although effective but it is expensive, tedious and time consuming (Yadav et al., 2009). Moreover, hand weeding and mechanical weeding are difficult due to continuous rainfall and less availability of labours at the critical stage of crop-weed competition. Use of herbicide not only improve crop yield but also makes available significant labour for other productive activities (Kurchania et al., 2001).

Weed flora in black gram

Weeds are often called as plants out of place. They are unwanted, useless, prolific, competitive and often harmful to the total environment. They interfere with agricultural operation and reduce the production potential of crop plants. Weeds are competitive and adaptable to all adverse environments. Weeds are a major problem for successful cultivation in Blackgram as their initial growth is relatively slow. Early emerging weeds are thus more competition. The weed flora in Blackgram differs widely with environment and soil conditions. Generally, weeds are found in larger numbers with more aggressive nature, because of their wider adaptability even under extremities of climate, edaphic and biotic stresses. The Blackgram crop with wide range of weeds species of grasses, sedges and broad leaf weeds while various weeds flora, grassy weeds are dominating after broad leaved weeds has offer a competition for crop weed condition of growth factors. The information on the weed spectrum of Blackgram is essential for the effective weed control strategies. Here under, dominant weed flora associated with Blackgram field in various environmental conditions is shared. Satish Singh et al. (2002) reported that during kharif season Echinochloa spp, Cyperus spp, Trianthema portulacastrum were major weeds in Blackgram field. Vaishya et al. (2003) observed that during kharif season at Cynodon dactylon, Echinochloa colona and Cyperus rotundus were major weed species in Blackgram field. Chand et al. (2003) noted in Pantnagar (Uttarakhand) during kharif season Cyperus rotundus, Echinochloa crusgalli, Cynodon dactylon, Eleusine indica, Trianaemia monogyna, Celosia argentia and Physalis minima were dominant weeds in urdbean. Rathi et al. (2004) reported that during kharif season and found that Cyperus rotundus, Parthenium hysterophorus, Trianthema monogyna, Phyllanthus niruri were major weeds in Blackgram. Bhandari et al. (2004) reported that Amaranthus viridis, Medicago denticulata,Trihananthe monogyna, Cynodon dactylon and Cyperus rotundus were major weed species in Blackgram field. Raman et al. (2005) found that the prominent weed species in the experiment conducted at Annamalainagar (Tamilnadu) were Trihananthe portulacastrum, Cyperus rotundus, Euphorbia hirta, Phyllanthus niruri, Commelina bengalensis and Digitaria sanguinalis. They also observed the maximum dry weight of weeds (104 g m-2) in weedy check plot. Sweta and Singh V. K. (2005) reported that the weed species in the experimental field of Blackgram were; Echinochloa colona,
Cyperus rotundus (sedge), Eleusine indica, Trianthema monogyna, Commelina benghalensis (broad leaf weeds) and Cynodon dactylon, Eleusine indica, Trianthema monogyna, Commelina benghalensis. The studies of Rao (2008) also revealed that the field was dominated by Echinocloa colona, E. crusgalli, Leersia hexandra, Panicum repens, Cyperus rotundus, C. kyllinga, Eclipta Alba, Grangea maderaspatana, Cardenthera uliginosa, Xanthium strumarium, Ammannia baccifera and Commelina benghalensis. The studies of Rao (2008) also revealed that the field was dominated by Echinocloa colona link, which constituted (80%) of the total weed weed population. Other weeds like Dinebra retroflexa (5%) Cyperus rotundus (3%) and broad leaved weeds Xanthium strumarium (2%), Cleome cheledoni (3%), Euphorbia virgata (2%), Nasturtium indicum (1%) where also present in less numbers. Nandan et al. (2011) reported Echinocloa colona (80%), Cynodon dactylon (15%), and Cyperus rotundus (5%), in monocots whereas among dicots weeds, Commelania bengalensis (75%) and Agerratum conozoides (15%) were predominant. Khot et al (2013) found that the weed flora was comprised of Cynodon dactylon, Cenechras biflorus, Dactylolotanium aegyptium, Boerhavia diffusa, Corchorus olitorius, Portulaca oleracea, Tribulus Terrestris, Spargula arvensis and Cyperus rotundus. Aggarwal et al. (2014) found that the major weed flora in the experimental field included Dactylocterium aegyptianum (Crowfoot grass), Cyperus rotundus (Purple nut grass), Cyodon dactylon (bermuda grass), Commelina benghalesis (benghal day flower), Eragrostis pilosa (soft love grass), Trienthema partulacastrum (horse purslane), Digitaria arvensis (wild crab grass), etc. during the two year. Patel et al. (2014) revealed that the predominant of monocots weeds like Eleusis indica, Digitaria sanguinalis, Cyperus rotundus, Cyperus iria and Cyodon dactylon, in blackgram. However, some dicots weeds like Euphorbia hirta, Amaranthus spinosus and Phyllanthus niruri were also marked their presence in few numbers. It could be concluded that several weeds were found competitive with crop depending on the agro-ecosystem of the growing region. Therefore, the critical study for each region is necessary to identify the suitable weed control methods according to existing weed flora.

**Crop weeds competition**

Crops and weeds compete for light, water and nutrients. Weeds benefit from crop management practices such as irrigation, fertilization and pest control that are intended to benefit the crops. Competition begins when crops and weeds grow in close proximity and the supply of any necessary growth factor falls below the demand of both. Competitiveness in both crops and weeds is related to their ability to exploit and sequester the environmental resource upon which plant growth depends. Competition is important only during the crop-growing season. Therefore relative crop / weed competitiveness generally depends on plant performances during only a part of each year.

Crop weed competition has been established as a major deterrent for its low productivity causing yield reductions to the extent of 40 to 80 per cent depending upon type and density of weed species present in the field. Crop type and soil properties had the greatest influence on the occurrence of weed species. The type of irrigation, cropping pattern, weed control measures and environmental factors also had a significant influence on the intensity and infestation of weeds Punia et al. (2009). Weeds, being naturally hardy and emerge faster, cause severe competition at an early stage of crop in respect of light, nutrients, water and space reflecting in considerable reduction in crop yield. Thus, it becomes essential to study crop weed competition scientifically and how it can be reduced to maximum Phajage S.K. (2014). Weed emergence in this crop during the first week is quite high. The initial 4 to 5 weeks are considered to be crucial for weed crop competition in Blackgram. Competition between plants is maximum when available resources for crop growth become limiting Rao A.S. (2008). In general, competition between crops and weeds was more severe when the competing plants have similar vegetative habits and demands upon resources.

**Critical period of crop weed competition**

The association of weeds occurs naturally with crop growth period, still need to catch out the exact time when the weeds are reducing the maximum crop productivity which as period or stage as ‘critical period of crop weed competition. In this, situation or condition is the best for effectively manage or control the weed species with real weed control techniques. The adverse effect of weeds on black gram would be severe in the early growth stages as in other short duration crops Rao A.S. (2008a). The critical period of weed competition in pulses crops is generally during the first 30 DAS. According to Jagraj et al. (2002) concluded that the reduction in the yield due to weed competition was throughout the cropping period (46.8 per cent. When weedy conditions were maintained for first 20, 30 and 40 DAS reduction in Blackgram grain yield was 4.1, 22.1 and 44.7 per cent respectively. The maximum crop weed competition in Blackgram was observed during the period from 10 to 30 DAS Sumachandrika D. et al. (2002). In summer Blackgram, maximum crop weed competition occurred during the period up to 30 DAS. An initial period of 20 to 40 days is very critical and season long weed competition has been found to reduce Blackgram yield to the extent of 87 per cent depending on the type and intensity of weed flora Bhandari et al. (2004). When in fact, Vivek et al. (2008), weed free situation was kept for 30 to 45
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DAS to prevent the potential loss in Blackgram grain yield. Therefore, it can be revealed that crop-weed competition period in Blackgram from 15-45 DAS.

Effect on yield Weeds are majorly compete with crops by moisture, nutrients, sun light and space at critical growth period it leads to reducing the yield of Blackgram. Hence, more effect of weeds on crops will be discussed hereunder. Yield Blackgram is one of the crops sensitive to weed competition. Among all the crop pest and diseases, weeds alone are responsible for about one third yield loss in crop production. Nevertheless, Jagraj et al. (2002) the reduction in the yield due to weed competition throughout the cropping period was 46.8 per cent. When weedy conditions were maintained for first 20, 30 and 40 DAS, which was reduced the grain yield (4.1, 22.1 and 44.7 per cent, respectively) of summer Blackgram. Most likely, Mishra et al. (2004), the reduction in yield due to the infestation of Cuscuta in Blackgram cultivars varied from 12.7 to 39.3 per cent. Parvender et al. (2008), the weeds infestation if not checked after 20 DAS, severe yield reduction to the extent of 38 per cent was recorded in contrast to 20 per cent yield reduction with unchecked weed infestation till 20 DAS. Almost certainly, Echinichloa was reported to be a dominant weed and yield reduction up to 53 per cent was reported due to uncontrolled weed growth in rice fallow Blackgram Venkateshwarlu et al. (2011).

Effect on nutrient availability

Nutrients, Nitrogen (N), phosphorus (P) and potassium (K) are the primary plant nutrients required for plant growth. When the crop growth is interfered by weed growth, it reduced the nutrient utilization of crop plant. In general, weeds have a larger nutrient requirement and will absorb as much or more than the crop. In the same way, Choubey et al. (1999) adoption of weed management practices significantly enhanced NPK uptake by Blackgram and reduced removal of nutrients by weeds as compared to that of unweeded check with saving of 29.1 to 52.3 per cent N, 26.8 to 56.6 per cent P, and 16.9 to 54.3 per cent K2O. Weeds removed 33.53, 15.78 and 72.19 kg/ha of N, P, and K2O kg/ha respectively in weedy plots Gaikwad and Pawar (2002) On the other hand, Rao A.S. (2010 -11) weed growth particularly Echinichloa spp. is severe and effectively competitive with the crop for residual moisture, nutrients and reduces the Blackgram yield up to 75 per cent. Quality of grain a heavy infestation of weeds hampers not only the growth and yield as well as infest the quality of pod or seed. Protein content of Blackgram significantly influenced by weed management practices. Significantly the highest (22.76 per cent) and the lowest (21.90 per cent) protein content were observed with Pendimethalin @ 1.0 kg/ha along with one hand weeding and inter culturing at 20 DAS, respectively Patil N.M. (1999., Singh et al. (1999., However, the experiment laid out on summer Greengram at Pantnagar (Utaranchal) and noted that protein content was significantly higher in weed free plots and the lowest in weedy check plot. Harmoniously, the weed species are affecting the quality of pod size and seed due to long time presence of weed growth and also reducing the market value of produces Davi D. (2004. Thus weed flora as well as weed population in unweeded control plot affected quality adversely.

Weed management strategies in Blackgram

Weed free crop situation has creating stable place to crop for getting effective growth environmental circumstance. Wherever, select the weed control techniques based on the economic threshold levels of weed growth for providing weed free competition and also reduce the environmental biodiversity Adpawar et al. (2011). The popular or effective weed management strategies to find out the weed species, weed control methods, time of scheduling to be practiced. In this context, decrease or minimize weed growth may be use of cultural, physical or mechanical and herbicides application have been improved in growth and spread of weeds.

Manual method

Hand weeding is the oldest method of weed control. It is time – consuming, labour – intensive, back – breaking and often costlier than chemical method of weed control. It effectively controls annual weeds, but not perennial weeds. Manual laborers use small implements e.g. Khurpi, which is useful to remove those weeds along with roots and thus it could be manual – cum – mechanical in nature, this loosen soil surrounding the rhizosphere of crop plant and thereby enhance crop growth and yield.

In India, weeds are controlled mostly either manually or mechanically in Blackgram. Manual weed control techniques manage weed populations through physical methods that remove, injure, kill, or make the growing conditions unfavorable. Hand weeding at 20-25 DAS and followed by another weeding at 12-15 DAS interval up to 50-55 days of the crop. One of the important method of hand weeding by hoe is effectively controlling the weed species in the inter row spaces of a line sown crop. This method might be provides good physical and environmental condition to the crop growth by way of soil aeration through stirring of the soil. Still now, this method could be effective for eliminating weeds particularly annual and biennial weeds in cropped and non-cropped situations. Respectively, Manish B. and Kewat M.L. (2002) the minimum seed yield was recorded when weeds were allowed to grow throughout the crop season and yield was highest in weed free plots received hand weeding twice at 20 and 40 DAS. Similarly, Rajput R.L. and Kushwah S.S. (2004) the first hand weeding

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at 20 DAS and followed by another weeding at 40 DAS received more seed yield of 1860 kg/ha. Hand weeding recorded significantly the highest yield (1120 kg/ha) of Blackgram due to effectively reduced the density of weed species and also its dry weight at critical crop growth period Gousia B. and Rao A.S. (2006. In the same way, Kalita et al. (2008) two hand weeding tremendously increased the seed yield and yield parameters of blackgram. For all that, lower weed biomass, lesser weed density, weed index and weed control efficiency were observed with hand weeding followed by mechanical weeding in both Blackgram and Greengram Veeraputhira R. (2009). Two hand weeding at 15 and 40 DAS was found to be more effective in controlling weed species in Blackgram and it led to higher seed yield and yield attributes, weed control efficiency, net profit, benefit cost ratio and lowest weed index were observed by Naidu et al. (2011). Application of 20 kg N/ha as basal plus 20 kg N/ha as split at 30 DAS plus two hand weeding (30 and 45 DAS) recorded the more number of pods/plant (24.96), number of seeds/pods(7.47) and 1000 seed weight (38.34 g) in Blackgram Ahmad et al. (2011).

Mechanical method
In the recent past, weed control is affected more by chemical means supplemented by mechanical weeding. Increasing demand for labour and escalating cost of agro-chemicals together with Phytotoxicity effects pose the farming community to think of mechanical measures, which will help the crop production to free itself from the scourge of weed menace with limited labour Kathiresan R.M. (2002). Mechanical weeding can be done by unskilled labour and is generally economical, nonpolluting without residual problems and is relatively safe to the operator. In the past, there were no mechanical weeders to fight this enemy and farmer had to use his hands to pull them out. Manual weeding is laborious, back breaking and time consuming and hence efficient mechanical weeders are being developed for weeding operation and help to obtain expected yields from the farm. Although it has undergone a spectacular advancement, to use of simple weeders with hand weeding and it would be easily operating, economically more effective in controlling the weed flora and led to increase the productivity of crops Sumachandrika et al. (2002). Rotary weeder was effective in controlling weeds present in inter-row space, but failed to control the weeds in intra-row space or those in vicinity of the crop Choubey et al. (1998). Similarly [44], use of improved weeders increased yield from 169.5 per cent to 329.6 per cent over control. Mechanical control of weed controls because physical changes in the immediate environment that may cause positive or negative effects. The suppression of the targeted weeds will open niches in the environment and may also stimulate the growth of other weeds by decreasing their competition and making their environment more favorable. If a desirable plant does not fill the niches, they will eventually be taken over by another weed.

Cultural method/ecological method
Cultural/ecological method of weed control exploits the crops competition behaviour growing environmental and management practices towards smothering of weeds. The farmers should adopt a good crop husbandry. Every practice head to be adopted with due care and should result in or aim at boosting up of initial growth of crop. (Walia). Weed control is one of the most important objectives of cultural operations. Following proper cultural operations is more than half the weed control envisaged on a farm. While directly it includes a healthy growth of crops, indirectly it maintains a crop environment that is detrimental to weeds. Blackgram is highly sensitive to abiotic stresses and thus, its yield levels are usually low. Among the production factors known to determine the crop yield, date of sowing has been recognized as the most important non-monetary input affecting the growth and yield in view of the change in the environmental conditions. The optimum time of sowing ensures the complete harmony between the vegetative and reproductive phases on one hand, and the climatic rhythm on the other and helps in realizing the potential yield Singh and Dhangra (1993). Weed population and weed dry weight were 16 and 12 per cent lower, respectively, in line sowing than broadcast sowing of blackgram Singh and Singh (2010). While, Ramakrishna et al. (2006) application of mulches reduce the weed infestation, increase the soil temperature and conserve the soil moisture in the field. Planting the crop at optimum time therefore, plays a key role in obtaining high seed yields Rathore et al. (2010). The reduction in weed population and less dry matter production of weeds may be due to an appreciable smoothing effect on weed as broad bed method leaving very little space weed to grow offered better crop weed competition in favour of crop resulting higher grain yield of Blackgram Darvin et al. (2015). Besides various methods of weed control. A good crop cover by adopting right inter-row and intra-row spacing will smother the growth of the weeds. Crop rotation also affects weed population in the preceding crops like maize or sorghum Ramakrishna et al. (2006).

Chemical method
In reality, crop fields are seldom adequately weeded by hand; weeding is tedious and time consuming. Laborers are not always available when needed. Weeding is often done late, causing drastic losses in yield. Due to scarcity of labour at peak times of agricultural operations, different herbicides based weed management technologies have been developed and as an alternative and test verified Rashid et al. (2012. Chemical weed
control by pre-sowing, pre-emergence and post-emergence application of herbicide and combinations of them are all effective way to control weeds for first few weeks after sowing of crop. Manda P. (2011) The use of herbicides has gained impetus from the general rise in farm wages for consistently increase the economic levels of farms as well as provide the non-farm employment opportunities, and drastically use of herbicide as a result of rising opportunity costs of labour across the developing world Hossain M.M. (2015. Based on income and labour use per hectare, herbicide technology was found superior to various weed control strategies. To create an awareness or knowledge to farmers about the proper use of pre and post emergence herbicide techniques to controlling weed flora in Blackgram.

**Pre - emergence herbicides**

Pre-emergence herbicides are applied one or two days after sowing of a crop but before the emergence of crop. Although the emergence of crop is taken into consideration, the emergence of weeds is equally important for designating many herbicides pre-emergent. Several pre-emergence herbicides viz., Pendimethalin, Oxyfluorfen, Nitrofen, Alachlor, Clethodim, Terbutryne, Fluchloralin, etc to control the germination of weeds in Blackgram at early stages. Pre emergence herbicide is preferred because of its better efficiency along with time involvement. Also, it causes no mechanical damage to the crop that happens during manual weeding Ram et al. (2004. Moreover, the control is more effective as the weeds even within the rows are killed, which invariably escape, because of morphological similarity to crop, during mechanical control. Effective weed control depends on the proper selection of herbicides, type of weed flora infesting the crop, time of application and further use of optimum dose of herbicide Chum et al. (2010. Application of Pendimethalin as pre emergence @1.5 kg/ha along with hand weeding at 30 DAS observed maximum weed control efficiency it lead to increase the productivity of Blackgram Ramanathan and Chandrashekarhan (1998. In the same way, pre emergence application of Pendimethalin at 1.50 kg/ha in combination with raised seed bed and ridge planting was effective to control Polygonum alatum and Ageratum conyzoides Suresh and Angiras (2005) and improving the physiological parameters (dry matter production, leaf area index and chlorophyll content) and further develop the nodules in Blackgram were significantly influenced by Fluchloralin @ 1.0 kg/ha followed by Pendimethalin @ 0.75 kg/ha Ram et al. (2004. Almost certainly, application of Pendimathali (0.75 kg/ha) plus hand weeding at 30 DAS drastically reduced density and dry weight of Triamethoxy monoxygena Sharma and Yadav (2006. Congruently, Pendimethalin @ 0.75 kg/ha in integration with one hand weeding at 45 DAS resulted in highest seed yield of Blackgram and minimum weed number and dry matter accumulation as observed by Suresh and Angiras (2008. However, the highest seed and haulm yield as influenced by Pendimethalin at 0.75 kg/ha as pre emergence along with one hand weeding at 40 DAS in summer Blackgram Patel et al. (2011. Harmoniously, effective suppression of newly emerging grasses and broad-leaved weeds by the application of Pendimethalin after dibbling of black gram seeds Sasikala et al. (2014).

**Post emergence herbicides**

The use of post-emergence herbicides alone or in combination may broaden the window of weed management by broad-spectrum weed control Henglata et al. (2016. Recently, some new post emergence herbicides viz. Imazethapyr, Acifluorfen sodium and Clodinafop propargyl, Quizalofop-p-ethyl, Fenoxaprop-p-ethyl, Cyhalofop-butyl etc. are being marketed with the assurance of selective control of weeds in blackgram. The imazethapyr allows much flexibility in timing of the applications. Imazethapyr may be applied as pre-plant initiation, pre-emergence or as post-emergences York et al. (1995. Although, Reddy et al. (2000) application of fenoxaprop-p-ethyl @ 60 g/ha effectively controlled the predominant weeds like Echinochloa colonum and Paspalum distichum and recorded significantly lower weed dry matter and higher grain yield. Similarly, postemergence application of tralkoxydin @ 0.4 kg/ha and fenoxaprop-p-ethyl @ 80 g/ha at 30 DAS recorded significantly lower weed dry weight, weed density and recorded higher weed control efficiency and grain yield of nurms on clay loam soil Singh and Tripathi (2001. In rice fallow Blackgram, thiobenthiocarb at 2.0 kg/ha as sand mix application at 9 DAS was more effective with 70 per cent weed control efficiency and recorded the highest yield of 385 kg/ha and was on par with imazethapyr at 63.5 g/ha applied as post emergence on 20 DAS Rao et al. (2001. Weed control efficiency of fenoxaprop-p-ethyl applied @ 75 g/ha was found to be higher than that of fenoxaprop-p-ethyl applied @ 45, 60 g/ha and provide effective control of Echinocloa colonum and Echinocloa crus-galli on clay loam soils of Pantnagar Singh et al. (2003. However, Kushwah and Vyas (2005) reported that imazethapyr at 75 g/ha were effective against both monocot and dicot weeds and was at par with one hand weeding at 20 DAS, however it was more effective against grassy weeds. If enhanced the grain yield by 45.3 per cent over weedy check. Application of imazethpyr @ 63 g/ha resulted in minimum dry weight of sedges and broad leaved weeds and also registered highest grain yield (930 kg/ha) in Blackgram Rao A.S. (2008. In the same way, Veeraputhiran et al. (2008) observed that the effect of imazethapyr on weed density, weed dry weight and weed control efficiency was at par when applied either on 21 or 28 DAS. The post-emergence herbicides like fenoxaprop-p-ethyl, clodinafop-propargyl and cyhalofop-butyl significantly reduced Echinochloa colona growth
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and increased Blackgram yield by 27 to 42 per cent over weedy check without any crop injury Rao A.S. (2008a. The weed control efficiency using imazethapyr @ 150 g/ha and increase the seed yield Naidu et al. (2011) of rice-fallow blackgram. Respectively, Harithavardhani et al. (2016) post emergence as imazethapyr at 25 g/ha had no adverse effects on rain-fed blackgram. Among the herbicidal treatments Mundra and Maliwal (2012), application of quizalofopethyl 50 g/ha increase in growth and yield attributes might be due to the reduction in weed competitiveness with the crop, which ultimately favored better environment for growth and development of crop. Application of fenoxaprop-p-ethyl @ 75 g/ha or cyhalofop butyl @ 100 g/ha drastically reduced the density of grassy weeds in rice fallow Blackgram. Post emergence application of acifluorfen sodium + clodinafop propargyl at 300 and 240 g/ha sprayed at 15 DAS registered higher weed control efficiency (70.6 and 68.0 per cent, respectively) due to greater reduction in weed biomass in Blackgram Harithavardhani et al. (2016.

Integrated weed management strategies

Shweta and Singh (2005) observed Initial weed control through application of herbicide (Pendimethalin @ 0.75 kg/ha) and further weed growth was drastically reduced by hand weeding at 40 DAS which situation to crop creating best growth condition Rathi et al. (2004), Kumar et al. (2006. In the same way, combined effect of cultural (seed rate), mechanical (hand weeding at 40 DAS) and chemical methods (Pendimethalin @ 0.75 kg/ha) markedly reduce the weed density and weed dry weight of Blackgram which led to increase the productivity and ultimately providing higher benefit cost ratio Velayudham K. (2007). In general, sequence application of weed control methods like pre emergence herbicide prevent or kill the germinated weed seeds and further vigour weed growth was controlled by hand weeding for superior methods than individual application of other control methods of weeds Rao A.S. (2010. Application of pre emergence herbicides as pendimethalin (1.00 kg/ha) or oxyfluorfen (0.18 kg/ha) followed by mechanical weeding (hand weeding + inter-cultivation or two hand weeding at 20 and 40 DAS respectively) creating a better weed free situation and also provides economically safe to farmers Balyan et al. (2016. Post-emergence herbicide as quizalofop-ethyl 50 g/ha at 30 DAS was significantly superior in reducing weed density both at 30 and 60 DAS while remained at par with the treatments of inter-culture 15 DAS fb imazethapyr 100 g/ha 30 DAS, inter-culture 15 DAS fb quizalofop-ethyl 50 g/ha at 30 DAS, and imazethapyr 100 g/ha 20 DAS Pratap et al. (2016. Crop grown under line sowing with the application of quizalofop ethyl @ 50 g/ha recorded lowest weed dry weight followed by broad bed method and ridge method Darvin et al. (2015. However, pre-mix application of imezathapyr + pendimethalin (1000 g/ha) or imazethapyr + imazamox (pre-mix) 70 g/ha reduced total weed population by 63.2 and 62.3 per cent, respectively so given as better performance of combination of herbicides might be due to synergistic effect between the two herbicides reducing the population as well as dry matter accumulation of different weed species Rao et al. (2010a. Regulation of various weed control methods should be such that they give the competitive edge to crop over weeds. The integration of these methods with chemical measure is advisable to avoid the ill effects caused by the sole dependence on the herbicides. Some of the negative impacts of sole dependence on herbicides are evolution of herbicide resistance weed flora shift and soil and environmental pollution. Also, the continuous dependence on single method of weed control leads to shift of weed flora in favour of more tolerant and difficult to control species and to tackle this problem, there is need to adopt integrated weed management practices. The rising cost of labour and input will wipe out the profits of farmers unless an integrated approach with focused attention of ecology and herbicides is adopted.

Reference

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INSECT PESTS OF RICE AND THEIR MANAGEMENT

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Abstract

Rice is an important cereal crop and serves as a staple food for more than half of the world’s population. It is grown on over 145 million ha in more than 110 countries, and occupies almost one-fifth of the total world cropland under cereals. In India, rice is grown on about an area of 36.95 million hectares with a production of 80.41 million tons. There are over 100 insect species damaging rice in one way or other but about 20 species are of major importance and of regular occurrence. The insect pests cause huge economic loss to the rice-growing farmers in the developing world. Average rice yield loss due to various insect pests was estimated to be 31.5% in Asia (excluding mainland China) and 21% in North and Central America in the year 1967. Rice is attacked by a large number of insects, among these insect pests, plant hoppers, leaf hoppers, stem borers, and gall midges are the most serious pests of rice.

Host plant resistance assumes central role in pest management in order to increase production and productivity of the crops. Conventional breeding in conjunction with molecular techniques and transgenic approaches have a great promise to reduce pest associated crop losses, and accelerate the progress in developing cultivars with resistance to insects. Although, considerable progress has been made over the past two decades in manipulating genes from diverse sources to develop plants with resistance to insect pests, deployment of molecular techniques for insect resistance, understand nature of gene action and metabolic pathways, but rapid and cost effective development and adoption of biotechnology-derived products will depend on developing a full understanding on the interaction of genes within their genomic environment, and with the environment in which their conferred phenotype interact. A good beginning has been made in developing genetic linkage maps of many crops, but the accuracy and precision of phenotyping for resistance to insect pests remains a critical constraint in many crops. Improved phenotyping systems will have substantial impact on both conventional and biotechnological approaches to breed for resistance to insect pests, in addition to the more strategic research that feeds into these endeavours.

Insect-pests of rice - Rice is attacked by a number of pests at different stages of its growth.

**Lepidopteran pests**
- Yellow stem borer, *Scirpophaga incertulas* Walker (Pyralidae)
- White stem borer, *Scirpophaga innotata* Walker (Pyralidae)
- Pink stem borer, *Sesamia inferens* Walker (Noctuidae)
- Pale headed stripes borer, *Chilo suppressalis* Walker (Pyralidae)
- Dark headed stripes borer, *Chilo polychrysa* Meyrick (Pyralidae)
- Rice caseworm, *Nymphula depunctalis* Guenee (Pyralidae)
- Rice leaf folder, *Cnaphalocrocis medinalis* Guenee (Pyralidae)
- Swarming caterpillar or armyworms, *Spodoptera mauritia* Boisduval (Noctuidae)
- Rice ear cutting caterpillar, *Mythimna separata* Walker (Noctuidae)

**Dipteran pests**
- Rice gall midge, *Orseolia oryzae* Wood-Mason (Cecidomyiidae)
- Rice whorl maggots, *Hydrellia philipendulina* Ferini (Ephydridae), Orthopteran pests
- Rice grasshopper, *Hieroglyphus banian* Fabricius (Acrididae)

**Hemipteran pests**
- Brown plant hopper, *Nelaparvata lugens* Stål (Delphacidae)
- White backed plant hopper, *Sogatella furcifera* Horvath (Delphacidae)
- Green leaf hopper, *Nephotettix virescens* Distant and *N. nigropictus* Stål (Cicadellidae)
- Ear head bug or Gandhi bug, *Leptocorisa acuta* Thunberg (Coreidae)
- Mealy bugs, *Ripersia oryzae* Green (Pseudococcidae), Coleopteran pests
- Rice hispa, *Dicladispa armigera* Olivier (Chrysomelidae), Root weevil, *Echinochernes oryzae* Marshall (Curculionidae)
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Damages caused by rice pests

Several damage symptoms are caused by rice pests which lead to the severe yield loss. Most important among them are dead heart and white ear head by yellow stem borer, hopper burn by brown plant hopper, onion shoot by gall midge, chaffy grains by gundhi bug. Due to these severe damages, a great barrier is created for healthy rice production and yield. In addition, hoppers transmit a number of diseases. Diseases like grassy stunt and ragged stunt are transmitted by brown plant hopper. Tungro, rice yellow dwarf, transitory yellowing are transmitted by different species of green leaf hoppers. Orange leaf disease is transmitted by zigzag leafhopper.

Management of rice pests

Cultural practices like Summer ploughing, selection of healthy seeds, timely planting, removal of weed, balanced use of fertilizers as per recommendations are the important cultural practices that are followed for pest management in paddy. Mechanical practices like removal and destruction of pest infested plant parts, clipping of rice seedling tips and collection of egg masses. Biological control agents like coccinellids, spiders, damsel flies, dragonflies should be conserved. Behavioural control like pheromone traps are installed at the rate of 20 traps/ha to trap yellow stem borer at 10 days after transplanting. Chemical control measures are used under IPM as a last resort. Application of pesticides has to be need based and proper crop health monitoring, observing ETL and conservation of natural biocontrol agents has to be ensured before deciding in favour or use of chemical pesticides. As hoppers are very destructive pest of rice, so to combat this problem resistance breeding technique, being a very handy tool can be efficiently used along with other management programme.

Plant resistance

In every plant species there exists a great deal of diversity with respect to the extent of damage done by an insect. Individual plants which show lesser damage are called resistant and those showing more damage are called susceptible, thus, these terms are relative. Host plant resistance is the result of interaction between two biological entities, the plant and the insect under the influence of various environmental factors. Painter (1951) described plant resistance as the “relative amount of heritable qualities that influence the ultimate degree of damage done by the insect. In practical agriculture, resistance represents the ability of a certain variety to produce a large crop of good quality than do ordinary varieties at the same level of insect population.”

Paddy germplasm containing resistance genes for hoppers

The number of rice accessions in germplasm collections is now very large. At IRRI, more than 80,000 accessions are conserved. More than 40,000 accessions each have been screened for resistance to blast (BI), bacterial blight (BB), green leafhopper (GLH), and brown plant hopper (BPH). Wild races are a rich source of insect resistant germplasm and may provide new genes which can be used in the various gene development strategies. The embryo rescue strategies have been utilized in the crossing of *O. sativa* and wild rice species in breeding for BPH resistance.

<table>
<thead>
<tr>
<th>Species</th>
<th>2n</th>
<th>Genome</th>
<th>Characteristics</th>
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</thead>
<tbody>
<tr>
<td><em>O. nivara</em></td>
<td>24</td>
<td>AA</td>
<td>Grassy stunt virus resistance</td>
</tr>
<tr>
<td><em>O. rufipogon</em></td>
<td>24</td>
<td>AA</td>
<td>Source of CMS</td>
</tr>
<tr>
<td><em>O. glaberrima</em></td>
<td>24</td>
<td>AA</td>
<td>GLH resistance, early vegetative vigour</td>
</tr>
<tr>
<td><em>O. punctata</em></td>
<td>24, 48</td>
<td>BB, BBCC</td>
<td>BPH, WBPH, GLH resistance</td>
</tr>
<tr>
<td><em>O. officinalis</em></td>
<td>24</td>
<td>CC</td>
<td>BPH, WBPH, GLH resistance</td>
</tr>
<tr>
<td><em>O. eichingeri</em></td>
<td>24</td>
<td>CC</td>
<td>BPH, WBPH, GLH resistance</td>
</tr>
<tr>
<td><em>O. minuta</em></td>
<td>48</td>
<td>BBCC</td>
<td>BPH, WBPH, GLH resistance</td>
</tr>
<tr>
<td><em>O. australiensis</em></td>
<td>24</td>
<td>EE</td>
<td>BPH resistance, drought resistance</td>
</tr>
</tbody>
</table>

Screening techniques for resistance to plant hoppers and leaf hoppers

With the advent of insect-infestation devices and simple agronomic procedures for growing healthy plants, it is now possible to adopt techniques for mass screening large populations of segregating plant materials. This is an initial step in the screening technology needed to eliminate the majority of susceptible segregants and select the resistant ones. Such large-scale evaluation where insects are offered a free choice of plant materials can be accomplished in the greenhouse, screen house, or small field plots. The approach for screening and evaluating resistance will depend on the insect and the crop under study, the required insect numbers, as well as the availability of research facilities. If insect damage occurs at more than one stage of plant growth, it is important to evaluate resistance at each of those stages.
The conventional seedbox screening test is a rapid method for screening large numbers of rice germplasm accessions for qualitative resistance to hoppers. Seed sowing and infestation are timed according to the hopper-rearing schedule. Seeds are sown in rows in a standard seedbox (60 x 40 x 40 cm. The number of insects per seedling can be determined easily. Seven days after sowing (DAS), when the seedlings are at the two- to three-leaf stage, the seedboxes are placed in a water pan inside a screened room. The seedboxes are kept in the pan containing 5 cm of water. Plant hopper nymphs (2nd instar) cultured on a susceptible variety are uniformly distributed on the test seedlings by holding the base of the feed plant and lightly tapping the plants and blowing on them. In this way, approximately 10 hopper nymphs are deposited on each seedling. Grading of the entries in each seedbox is done when about 90% of the susceptible check seedlings in that box are dead. The Standard Evaluation System (SES) scale (0–9) for rice (IRRI 1988) is used to score seedling damage: 0 = no damage; 1 = very slight damage; 3 = first and second leaves of most plants are partially yellow; 5 = pronounced yellowing and stunting or about half of the plants wilting or dead; 7 = more than half of the plants wilting or dead; 9 = all plants dead.

Genes showing resistance to rice hoppers

**Brown plant hopper**

Sources of resistance to *N. lugens* were first identified in 1967. The gene designated, Bph1, bph2, Bph3, bph4, bph5, Bph6, and bph7 have been identified from different sources through genetic segregation analysis and bph8 and Bph9 through allelism tests. Since then, many donors for resistance to *N. lugens* have been identified and used in breeding *N. lugens*-resistant varieties and exploring more resistance source of rice continues. Some of the donors are Mudgo, ASD7, ADR52, Rathu Heenati, Babawee, ARC10550, Swarnalata, T27A, IR64, GX2183, PTB18, PTB33, T12, Chin Saba, Balamawee, *O. officinalis, O. australiensis, and Ovalipes minuta* from cultivated and wild species of rice. So far, 27 genes for *N. lugens* resistance have been identified from the gene pool of cultivated and wild rice species. A total of 126 rice varieties possess *N. lugens* resistance genes.

**Green leaf hopper**

A large number of varieties have been screened for GLH and many resistant donors (Pankhari 203, ASD7, Sigadis, Pt8, DV5, Asmaita, ARC10313, ARC11554, *O. rufipogon*) have been identified. Genetic analysis has revealed 11 dominant and three recessive genes [Glh1, Glh2, Glh3, glh4, Glh5, Glh6, Glh7, glh8, Glh9, glh10, Glh11, Glh12, Glh13, and Glh14].

**White backed Plant hopper**

WBPH occurs in all the rice-growing countries of Asia and does moderate damage to the crop. With the mass screening technique, germplasm collections have been evaluated and donors for resistance have been identified. More than 300 cultivars resistant to the WBPH have been identified and 80 of them have been analyzed genetically. Eight genes for resistance (*Wbph1, Wbph2, Wbph3, wbph4, Wbph5, wbph6, Wbph7 (t) and Wbph8*) have been identified. QTLs are mapped for resistance to white backed plant hopper. QTLs for ovicidal response have also been reported.

**Zigzag leaf hopper**

The zigzag leafhopper (ZLH) occurs in the tropics and subtropics of Asia. However, it is a minor pest of rice. Some donors (Rathu Heenati, Pt821, Pt833) for resistance have been identified. Single dominant genes that segregate independently of each other and that conveyed resistance to ZLH were designated Zlh1 (Rathu Heenati), Zlh2, (Pt821), and Zlh3 (Pt833). Tests for the independence of the various genes for resistance to leafhoppers and plant hoppers revealed that Zlh1, Zlh2, and Zlh3 are independent of Wbph3, Zlh2, and Zlh3 and also segregated independently of bph2 and Bph3.

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WEED MANAGEMENT THROUGH INSECTS

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Relationships between insect pests and weeds

Weeds are an important plant resource for insects, although feeding by insects on weeds can have both positive and negative effects on crop productivity. Weeds also indirectly affect crops via their influence on beneficial insects, and by harboring plant and insect diseases. Weeds may affect the ability of dispersing insects to locate crop plants. The host relationship between insects and plants is highly variable, ranging from very specialized to generalized feeding behaviors. Most insects, including crop pests, are specialists, and preadapted to feed only on some plants, often within a single plant family. Even polyphagous insects often have a distinct preference hierarchy, feeding more widely only when preferred hosts are unavailable. Use of plants by insects is a dynamic interaction, with characteristics of the insect (e.g., mandible structure) and the plant (e.g., allelochemicals) affecting feeding behavior. Thus, weeds that are closely related to crops are particularly important in harboring insects that attack those crops. Crop production practices should seek to sever the taxonomic association between the crop and the weeds found within the crop, and nearby, by eliminating weeds related to the crop. This will make it less likely that insects will move easily from weed to crop plants, that damaging population densities of insects will develop in the field, and that insect vectors that harbor plant diseases will be harbored in the field. (John L. Capinera, 2005)

Weeds are a food resource for insects

Weeds are a primary resource for many phytophagous insects. From the perspective of crop protection, this has both positive and negative aspects. In a positive sense, insect feeding on weeds makes water, soil nutrients, and sunlight more available to crop plants, thereby reducing weed competition with crops. Many insects feed exclusively, or nearly so, on weeds. For example, the sesiid moth, Carmentahaematica (Ureta) attacks only snakeweeds, Gutierrezia and Grindelia spp., in the family Asteraceae (Cordo et al. 1995). Other insects prefer weeds, but may damage crops readily in the absence of attractive weeds. For example, the Colorado potato beetle (Leptinotarsa decemlineata Say) prefers to oviposit on hairy nightshade (Solanum sarrachoides Sendtner) rather than on potato (Solanum tuberosum L.), and eggs are less abundant on potato in the presence of nightshade (Horton and Capinera, 1990).

Other example is the army cutworm, Euxoa auxiliaris (Grote), will serve to demonstrate some of the complexities of the insect-weed-crop plant interaction. Army cutworm is a common lepidopteran pest of wheat (Triticum aestivum L.). It is known predominantly as a pest of small grains, it feeds on a large number of plants, including many weeds in preference to grain crops. According to John L. Capinera army cutworm, Euxoa auxiliaries clustered on pinnate tansy mustard [Descurainia pinnata (Walt) Britt.]. Also, the effect of army cutworm feeding on this weed is clearly evident. The foliage is completely consumed by the army cutworm and only the base of the plant remains. Note that the wheat plants on both sides of the weeds are free of cutworms and cutworm feeding. Indeed, up to this point the army cutworms are beneficial insects, serving to reduce competition by the weeds with the young wheat by killing or severely inhibiting the growth of the tansy mustard plants.

Weeds Affect Host-Finding by Herbivores

Weeds can modify the attractiveness of crops to the insect herbivore, thereby affecting the rate of colonization. Both vision and odor play an important role in host location by most insects (Stanton 1983. In the case of vision-based host finding, it is the spectral profile (non visible to humans as well as visible) to which the insect responds. Weeds can also modify the attractiveness of crops to insects by affecting the colour of the foliage; as first demonstrated conclusively by V. Moericke (Kennedy 1976), many herbivorous insects are attracted to
yellow or yellowish green during the host-seeking phase, relative to dark green or other colors (Kostal and Finch 1996; Moericke 1969. Thus, light green weeds interspersed among darker green crops could be relatively more attractive to alighting insects. Weeds also affect chemical-based host finding. Many insects do not depend on vision and then use odor to identify a suitable host. On the other hand, in weedy fields, the chemical stimuli may be less concentrated or confusing to the potential herbivore.

Why It's Important to Remove Weeds
Man's struggle against weeds is endless. Even today there are millions of acres of valuable lands lost to weeds. Some weeds are just hard to kill. Some, although easily killed, grow on lands too low in value or too inaccessible for control by conventional means (C. B. Huffaker, 1959). “Weeds are notorious yield reducers that are, in many situations, economically more harmful than insects, fungi or other crop pests,” said a study, published in the journal, Crop Protection.

Weeds lead to India losing an average of $11 billion each year in 10 major crops, shows data from 1,581 farm trials in 18 states. Insects, diseases and weeds are the three main biological factors for losing crop yield and causing economic loss to farmers. Unlike the visible impact of diseases and insects, the impact of weeds goes unnoticed, said Dr. Yogita Gharde, lead author of the paper and scientist at the directorate of weed research at Indian Council of Agriculture Research. According to Dr. Yogita Gharde, if weed growth is not stopped at a critical time, it results in massive crop loss, sometimes as high as 70%.

Choice of phytophagous insects for weed control:

Generally, alien weeds are especially amenable to biological control because of the lack of effective and host-specific natural enemies to keep them under check in the new area(s) which they have colonised. In India Parthenium hysterophorus L. (Compositae) is one of the best examples of such an alien weed. In what is commonly known as classical biological control the first step is to trace the native range of the alien weed, investigate the biotic agents that regulate its population. Select the most promising natural enemies, subject them to a strict protocol of host-specificity screening tests, and then to introduce the successful candidate species into the new area for field evaluation. Many of the insects attacking a potential weed in its native home may be expected to be polyphagous, which will automatically disqualify them for use in biological control in other areas in view of the risk of their attacking useful plants so, host-specificity is the most essential requirement for a bioccontrol agent used in weed control.

Some native insects that attack indigenous weeds and exhibit monophagy or oligophagy have been studied and used in weed control by breeding them in the laboratory and releasing them to augment the otherwise low natural populations at critical stages in the life-cycle of the weed. Bactra verutana Zeller (Lepidoptera, Tortricidae) has been used against Cyperus rotundus L. (Cyperaceae) in the USA (Frick and Chandler 1978) and periodical releases of the noctuid Episamea pectinicornis Hmps. are now a standard control method against Pistia stratiotes L. (Araceae) in Thailand (Napompeth 1982. The use of native biotic agents may be of value against weeds for the control of which there is little or 110 scope for introduction of additional, more effective, biocontrol agents from other geographical areas. Genetic diversity in the introduced insect population will enhance and improve the chances of success.

Insect damage potential and its impact on weeds

Insects that damage different parts of the same weed may kill a weed or contribute to an overall reduction in its growth, vigour and reproductive potential. Therefore, if one species fails to control a weed adequately others may be used to enhance the level of control. Synergistic action by two species may also inflict more damage on the weed than that possible by either species working alone, as with N. eichhorniae and the water hyacinth mite, Orthogalumana terebrantis Wallwork (Del Fosse 1978.Species that severely damage the root system or the main stem may kill the plant within a short time while defoliators and flower feeders are less drastic in their effect. Most of the phytophagous insects themselves are subject to parasitism, predation and pathogenic diseases, and asa result many species occur at low populations, particularly at the most vulnerable stages in the life-cycle of the weed. With annual weeds that only propagate by seed, fruit- and seed-feeding insects are likely to be more effective control agents than they are with weeds that propagate vegetatively as well as by seed. Larvae of the noctuid, Eulocastra argentisparsa cause complete loss of seed in individual Striga plants but their natural populations are always very low. A microsporidian disease is known to infect them. Moreover Striga is an annual weed and its seed-maturing phase is short. Eulocastra is unable to build up its population early enough to reduce the total seed output in an infested area significantly.
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A gall-forming weevil, *Smicronyx albovariegatus* Fst. attacks *Striga* spp. in India but most of the galls are produced on the root, stem and branches and yet do not impair the vigour or reproductive capacity of the host-plant. The host-plant is able to withstand, if not 'resist, the weevil attack. Some other *Smicronyx* occurring in Africa produce fruit galls and thereby arrest seed production. However, parasitism limits their value as control agents. Larvae of the tortricids *Bactra (Nannobactra) minima minima* Meyr and *Bactra (Chiloides) venosana* Zeller bore into the stem and occasionally into the rhizome of *Cyperus rotundus* L. in India but, since even a partially damaged rhizome can produce a fresh shoot, the insect is not able to kill the plant. The new shoot may also be infested by the borer. Such plant regeneration and insect counter-attack have the final effect of dwarfing the plant by keeping it under constant stress, which gives cultivated plants a competitive advantage over the weed in crop fields. Here again various parasites reduce the effect of these borers (Sankaran and Rao 1972). The gall-fly *Proccecidocharesutilis* is widely established on *Eupatorium adenophorum* in many parts of India but its control action is weakened by native parasites.

Biological Control of Weeds with Insects

*Lantana camara* Linnaeus- This plant is a serious pest of grazing lands in Queensland and New South Wales, Australia, but, to the banana grower it is useful because it immediately occupies land being "rested" from banana cropping (121) and can be easily removed when planting is resumed. It may also be of value in water and soil conservation. Lastly, there is fear that control of Lantana may foster the spread of *Eupatorium adenophorum* Sprengel, which is considered a more serious threat to pastures in parts of eastern Australia (121). Nonetheless, Parham et al. (79) consider it a fallacy to view Lantana as beneficial on plantations in Fiji.

*Opuntia* spp.-The urgency for controlling prickly pears in Australia in the 1920's was unchallengeable, and the subsequent success of that work has stood as a landmark not likely to be equalled. However, *Opuntia* spp. are not always considered pests. Use of these plants in Hawaii, South Africa, Madagascar, the United States, and Mexico, as human food, fodder, and a source of water for stock on dry ranges has been reported [Fullaway(36); Huffaker (49); Imms (56); Pettey (85)]

Importation of *Cactoblastis cactorum* (Berg) into the United States has long been denied largely on such grounds, although it has been stated that, in West Texas alone, an area of some 60,000,000 acres of range lands are infested with *Opuntia* and consequently suffer greatly lowered values (21). On spineless cactus plantations in some areas of South Africa it has been necessary to control the cochineal previously introduced against the related pest cacti [Pettey (85)].

*Echium plantagineum* Linnaeus-In Australia this plant, although generally considered a weed, has some fodder value in certain areas and during droughts [Wilson (123)].

*Ulex europaeus* Linnaeus-In New Zealand gorse was early recognized as a serious pest. Yet, in some sections of that country it is used as hedgesand, in its early growth, as a fodder crop. Hence, the insects introducedfor its control were arbitrarily restricted to those which might curtail seedproduction and spread without destroying existing plants [Miller (65)].

*Centaurea solstitialis* Linnaeus-In California (U.S.A.) *yellow star thistle* is a complicated example. It is harmful to range lands and in grain and seed crops. But it is reportedly essential to the maintenance of bees in sufficient numbers to pollinate the nut and fruit crops. The claims in this respect may have been exaggerated, and loss to the bee industry could probably be replaced.

Examples of weed management through insects

1. *Lantana, Lant ana camara* Linnaeus: Hawaii.-The first attempt at biological weed control was made in Hawaii in 1902, where *L. camara* threatened ranching interests. Destruction of this weed by the scale insect *Orthezia insignis* Douglas. There is actually an earlier example: "the ability of insects to destroy prickly pear had been known and applied in India and Ceylon since cochineal insects had been introduced" in 1795 to develop a dye industry, wrote Wilson. Which had arrived accidentally, had been not iced, and ranchers had engaged in establishing it on their ranches. Their efforts gave impetus to the plan of sending Koebele to collect Lantana insects in Mexico and Central America for introduction in Hawaii.

Of the insects introduced, eight became established: a seed fly, *Agromyza lantanae* Froggatt, a lace bug, *Teleonemia scrupulosa* (Stal); a tortricid, *Epinotia lantana* (Busck); a moth, *Platyptilia pusillidactyla* (Walker);

G. Wilson Fernandes & Jean C. Santos, 2008 revealed that the three species of longhorn beetles associated with *L. camara* in November 2004: *Trachyderes succinctus duponti* Aurivillius, 1912; *Andraegodius rufipes zonatus* (Dalman, 1823); and *Dorcacerus barbatus* (Olivier, 1790. *Dorcacerus barbatus* represented more than 95% of all individuals found. To their knowledge, this is the first record of these three cerambycid species on *L. camara*.

![Fig.1. Dorcacerus barbatus (Cerambycidae) on Lantana camara (Verbenaceae. (Left) Feeding on inflorescences. (Right) Feeding on fruits. (Source: G. Wilson Fernandes & Jean C. Santos,2008)](image)

According to Dodd and Davis, the following insects were released in Hawaii to control *Lantana camara* weeds: a cerambycid, *Aerenicopsis championi* Bates, the noctuids, *Catabenaesula* (Druce) and *Diastematigris* Guenee; the pyraustids, *Syngamia haemorrhoidalis* Guenee and *Blepharomastix acutangulalis* (Snellen); and an agrodid, *Hypana jussalis* Guentle. Favorable recoveries of the cerambycid have been made on Hawaii. *C. esula* is established on Hawaii, Maui and Oahu. *B. acutangulalis* and *D. tigris* have not been recovered. *S. haemorrhoidalis* is also well established.

2. Prickly pears, *Opuntia* spp.: This classic example of biological control is so well known that no effort will be made here to give it the coverage its importance merits. The problem in Australia and the ultimate solution were well documented by Dodd in 1940. *Dactylopiusindicus* Green (≡*D. ceylonicus* n.n.) was introduced in 1903. Cactus moth, *Cactoblastis cactorum* is also introduced.

Insects introduced prior to *C. cactorum*, including *Chelinidea tabulata* (Burmeister), *Dactylopius tomentosus* Lamarck, Oyee Uajune to Uneela (Hulst), and a mite, *Tetanychus desertorum* Banks (≡*T. apuntiae* Banks), made rapid progress from 1925 to 1927. From 1930 on, *C. cactorum* mastered the prickly pears and eclipsed all other species.

The other species of the 12 established against *Opuntia* are: a phycitid, *Tucumania tapiocola* Dyar; three cerambycids, *M oneilemaulkei* Horn, *M oneilemavariolare* Thomson, and *Lagocheirus funestus* Thomson; a coreid, *Chelinidea vitliger* Uhler; and a cochineal, *Daetylopius eonfusus* Cockerell (≡*D. newsteadi*Cockerell. India andCeylan.- According to David & Muthukrishnan (22), *Opuntia vulgaris* Miller and *O. elatior* Miller (≡*O. nigricans*
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Haworth) were introduced in India about 1787 to culture the commercial cochineal, valued for its dye, while Opuntia dillenii (Ker-Gawler) was probably introduced much earlier. *O. vulgaris* had long been under effective control by the cochineal, *Dactylapius indicus* Green, which apparently arrived along with the introduced *Opuntias.*

3. Purple nut sedge, *Cyperus rotundus*: *Bactra verutana*, the javelin moth, is a species of moth of the Tortricidae family, was introduced. The effectiveness of *Bactra verutana* Zeller, that is ineffectual in suppressing purple nut sedge, *Cyperus rotundus* L., (Kenneth E. Frick and Clemente Garcia, Jr., 1975). (Source: Figen Efil et al, 2012)

![Figure 3. The damage of Bactra venosana](image1)

![Figure 4. The pupa of Bactra venosana on nutsedge.](image2)

4. Congress, *Parthenium hysterophorus* L.: Different control approaches have been used for the management of *Parthenium*. Although manual and chemical methods are effective strategies to control the weed in agricultural fields, but these are not economical in pastures and large natural areas or wastelands (Krishnamurthy *et al.* 1977. Biological control of *Parthenium* weed is considered to be the most cost effective, environmentally safe and ecologically viable method (Dhileepan *et al.* 2000. It was documented to control *Parthenium* worth of Rs10 million in terms of herbicide cost after initial release of bioagent *Zygogramma bicolorata* Pallisterat Jabalpur, India (Sushil kumar, 2006) and it was estimated that this bioagent has checked the spread of *Parthenium* in about eight million hectares of land since its release in India. *Zygogramma bicolorata* is an effective biocontrol agent that can significantly reduce the vegetative and reproductive growth of *Parthenium* weed. However, the effectiveness of the biological control *Z. bicolarata* can be further enhanced if it is applied at the early growth stages (young or pre-flowering) of *Parthenium* weed.

![Figure 5. Zygogramma bicolorata beetle feeding on Parthenium weed](image3)

5. Water hyacinth, *Eichhornia crassipes* was first introduced from South America into China as a good fodder plant in 1901, and had become a serious environmental problem in China by the early 21st century (Ding *et al.* 2001. The weevils *Neochetina eichhorniae* and *N. bruchi* were released in the USA for water hyacinth control during the early 1970s and have since been used in many other countries (Ted D. Center *et al.* 1999.

![Figure 6. The waterhyacinth weevils. Left: Neochetina eichhorniae Hustache. Right: N. bruchi Warner.](image4)

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6. Alligator weed, *Alternanthera philoxeroides* (Mart.) Griseb. (Amaranathaceae), is an invasive aquatic weed native to South America that began threatening Florida's waterways in the early 1900s. The alligator weed flea beetle, *Agasicles hygrophila* Selman and Vogt, was the first insect ever studied for biological control of an aquatic weed. The introduction of this insect into the United States was approved in 1963, but it was not successfully established on the invasive alligator weed until 1965. The insect was first released in 1964 in California, and subsequently, in Alabama, Florida, Georgia, Louisiana, Mississippi, South Carolina, and Texas. Alligator weed flea beetles kill the alligator weed by destroying its stored food and interfering with photosynthesis by removing leaf tissue. Both adults and larvae feed on the leaves of alligator weed, often defoliating the stems. (Ted D. Center, 2015)

Figure 7. Larvae and adult of the alligator weed flea beetle, *Agasicles hygrophila* feeding on alligator weed (Gary Buckingham, USDA-ARS; Bugwood.org)

7. *Striga hermonthica*: Commonly known as purple witch weed or giant witch weed, is a hemi parasitic plant that belongs to the family Orobanchaceae. It is devastating to major crops such as sorghum (*Sorghum bicolor*) and rice (*Oryza sativa*). The reduction in seed production from gall-forming *Smicronyx* spp. is often substantial, but there has been no successful development of a biological control programme based on these weevils. Attempts to introduce *Smicronyx alb ovariegatus* and the moth, *Eulocastra argentisparsa* from India into Ethiopia apparently failed. Meanwhile, conclusions from a mathematical modelling project have suggested that *Simicronyx* spp. would in any case be unlikely to have a significant impact on *Striga* population dynamics. Other potentially useful organisms for *Striga* management include the following: the butterfly *Precis* (=*Junonia*) species whose larvae feed on leaves, buds and capsules of many *Striga* species. (ICIPE Biennial Scientific Report, 2004-2005)

Figure 8. *Striga* weed (left) and weevil, *Smicronyx* sp. (Right)

Natural enemies associated with *Striga*.

1. *Smicronyx alb ovariegatus* Fst., Larvae make galls on roots, stems and seed pods of *Striga*. and it was reported in India, East Africa, Nigeria, Uganda.
2. *Eurytoma* spp., Adults make galls on stem and it was reported in Kenya.
3. *Ophiomya stri galis* Spencer, Larvae mine the leaves and stems of *Striga* and it was also reported in Kenya.
4. *Diacr iria investigatorurn*, Larvae eat leaves and flowers and it was reported in Africa.
5. *Eriqymnrn unipunctata*, Larvae eat leaves and flowers of *Striga* weed and it was reported in Tanzania.
6. *Spodoptera literalis*, Larvae eat leaves and flowers and it was reported in India and Tanzania.
7. *Eulocastra argentisparsa* Hmps., Larvae devour the seed and it was reported India (AP &Karnataka).
8. Gunonia orithya, Defoliate *Striga* plant and it was also reported in India.
9. Platypilia spp., Defoliate *Striga* plant and it was reported in India, Tanzania & Uganda.
10. Antestia cincticollis, Suck plant sap. It was reported in Uganda (Inderjit, 2004).
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8. *Eupatorium adenophorum*: Commonly known as crofton weed or sticky snakeroot, is a species of flowering plant in the daisy family native to Mexico and Central America. Originally grown as an ornamental plant, it has become invasive into farmland and bush land worldwide. *Procecidochares utilis* is a tephrtitid gall fly, which is known to be an effective biological agent that can be used to control the notoriously weed *Eupatorium adenophorum* (Gao et al. 2014).

Figure 9. Gall formation in crofton weed (left) Crofton weed gall fly (*Procecidochares utilis*), a biological control agent (right) (Source: Forest and Kim Starr, USGS)

9. Scentless chamomile, *Matricaria perforate* (merat):- Also known as mayweed, scentless mayweed or daisy, has white daisy-like flowers and its finely divided fern-like leaves, in the early stages this weed is often confused with pineapple weed, stinking mayweed or yarrow as all of these weeds have very finely divided leaves. This plant is native to Eurasia and North Africa. *Omphalapion hookeri*(Kirby). Seed head feeding beetle, which is known to be an effective biological agent that can be used to control the *Matricaria perforata*(ISC, 2014).

Figure 10. Scentless chamomile, *Matricaria perforate* weed (left) and Seed head feeding beetle, *Omphalapion hookeri*(Kirby) (right) (Credit, Gov. of Alberta, 2000.

10. Perennial Sow Thistle, *Sonchus arvensis*: Its native to Europe and occurs in all European countries. It has been spread by humans to large parts of the temperate world, and also to subtropical areas in all continents. It has become naturalized in such areas in 59 countries including India. (CABI, 2017. Several insects attack *S. arvensis*, e.g. a gall former, *Tephritis dilacerata*, which causes galls in flower heads, thereby reducing seed production (Schroeder, 1973 ., Shorthouse, 1980.

Figure 10.a) Perennial Sow Thistle, *Sonchus arvensis* (left) (Credit, Vijay Choudhary,2016).b) a gall former, *Tephritis dilacerata* (Right) (Credit, Raimo Peltonen)

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NEW RECORD OF INVASIVE MOTH ASOTA CARICAE CARICAE (FABRICIUS, 1775) (EREBIDAE :AGANAINAE) FROM WESTERN DOON VALLEY (DEHRA DUN, UTTARAKHAND) WITH SYSTEMATIC ACCOUNT, DISTRIBUTION AND NATURAL HISTORY

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ABSTRACT

The present communication deals with the occurrence of invasive moth Asota caricae caricae (Fabricius, 1775), the Tropical Tiger Moth, in Western Doon Valley, Dehra Dun (Uttarakhand) and its systematic account, distribution and natural history.

Recently, the first author came across a moth inside a mosque building at Muradnagar, Ballupur road, Western Doon Valley, Dehra Dun (Uttarakhand) during the day time which looked interesting and on examination was found belonging to Asota caricae caricae (Fabricius, 1775), the Tropical Tiger Moth, under family Erebidae. The host plants are likely to be Broussonetia papyrifera, the Paper Mulberry and Ficus religiosa, the Peepal tree as present in the compound of the mosque. Earlier, it was recorded from some localities in North-Eastern Doon Valley and Rajaji Tiger Reserve by Sondhi & Sondhi (2016).

SYSTEMATIC ACCOUNT, DISTRIBUTION AND NATURAL HISTORY OF ASOTA CARICAE CARICAE (FABRICIUS, 1775)

Genus: Asota Hubner, 1891
Asota caricae caricae (Fabricius, 1775)
Tropical Tiger Moth
Hypsa (Damalis) plaginota Butler, 1875. Bull. Trans. Ent. Soc.: 320 (type-locality: India also N. E. India, larger and paler, expense of wings 2 inches 8 lines.
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Classification


Material Examined

1 example from Rehmania Mosque, Muradnagar, Chor khala (near Vijay Park), Ballupur Road, Dehra Dun (at 648 m altitude); 30.ix, 2018; Coll. Akhlaq Husain (the first author.

Flora around

Peepal tree (Ficus religiosa), Mango tree (Mangifera indica), Paper Mulberry plants (Broussonetia papyrifera) on Peepal tree trunk and ‘pushta’ walls of mosque compound and other seasonal vegetation.

Diagnostic Features

Palpi with black spots on 1\textsuperscript{st} and 2\textsuperscript{nd} joints, head yellowish-brown, with basally and apically a black spot; thorax also yellowish-brown with three black dots; abdomen also yellowish-brown with a black mark on top of last few segments; fore-wings buff brown with a whitish medial spot in centre at lower angle of cell, a basal orange patch with two sub-basal black spots and a series of three smaller spots on its outer edge, veins streaked with white; hind-wing deeper yellowish-orange with a black spot at end of cell, one beyond, one below vein 2 and a sub-marginal irregular series smaller spots which may become nearly confluent at margin and veins crossing it yellowish, black spots on middle much larger.

Male: Antenna weakly bristled. Genitilia with uncus long; tegument long and narrow; vinculum and saccus shorter; valvae simple, elongate, extending to rounded apex and usually with a single, relatively basal harpe at distal end of sacculus; saccular process prominent; aedeagus normally short and broad; vesica large and bearing a small group of cornuti or a single cornutus; with a prominent cornutus; coremata of 8\textsuperscript{th} segment small, especially the lateral pair.

Female: Genitilia as of other species but ductus bursae not sclerotised at base, signa (if present in bursa) not in scobinate bands, more circular in shape.

Wing-span: 62-67mm- male, 72-76 mm-female (Hampson, 1892; Chandra & Nema, 2006); 50-55 mm-male, 60-63 mm- female (van Eecke, 1928); 40-56 mm- male, 47-63 mm- female (Moriuti, 1996); 68 mm (Chandra & Nema, 2008); 51-56 mm- male, 58-70 mm- female (Gurule & Nikam, 2013); 52-65 mm (Ulziijargal et al., 2016); 51-58 (Wikipedia; WikiVisually.

Altitudinal Range Records

Present (648 m); Gagar (2,400 m. Maheshkhan (2,200 m), Jones Estate, Bhimtal (1,500 m), Ranibagh (450-00 m) (Peter, 2008); Dhoran Khas (794 m), Malsi (824 m), Danda Lokhand (802 m), Whelam Girls’ College (673 m) and Rajaji Tiger Reserve (328-760 m) (Sondhi & Sondhi, 2016.

Distribution

Dehra Dun: Muradnagar, Chor khala (near Vijay Park), Ballupur Road, 648 m altitude (present); Dhoran Khas, 794 m altitude (Rajpur area); Malsi, 824 m altitude; Danda Lokhand, 802 m altitude (Sahatradhara road); Whelam Girls’ College, 673 m altitude (Dalanwala) and Rajaji Tiger Reserve 328-760 m altitude (Sondhi & Sondhi, 2016.

Rest of Uttarakhand: Nainital, Rajaji Tiger Reserve (Haridwar part).

Rest of India: Andaman & Nicobar Islands (Port Blair), Assam (Relief Yard Colony Road, Lumding), Chhattisgarh (Kanger Valley National Park, Bastar), Himachal Pradesh, Jharkhand (Dalma Wildlife Sanctuary, East Singhbhum district), Kerala (Thodupuzha-Udumbanoor Road, Neyyassery), Madhya(Seoni and Umaria;
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Bandhavgarh National Park, Satpura hill ranges, Umaria district), Maharashtra (Amrawati, Dhule, Nandurbar and Nashik districts), Manipur, Meghalaya, Nagaland, Sikkim, Tamil Nadu (Thoothukudi district), West Bengal (Durgapur; Neora Valley National Park, Darjeeling) and Western Ghats.

Outside India: NE Australia (Northern Territory and Queensland), Bangladesh (Dacca), Bhutan, Borneo (Brunei, Indonesia and Malaysia) Greater Sunda Islands), Cambodia, SW China, Hong Kong, Indonesia (Ambon, Biak, Java, Malaku, New Guinea, Numfor, Papua, Salawati, Seram/Ceram, Sulawesi, Sumatra, Sumbawa and Yapen Islands), Japan, Malaysia (Penang Island), Myanmar, Nepal, New Hebrides, New Ireland, Papua New Guinea, Philippines, Solomon Islands, Sri Lanka, Sunda Islands (Brunei, East Timor, Indonesia and Malaysia), Taiwan, Thailand and Vanuatu.

**Habitat:** Low lands forests and agricultural areas.

**Breeding**

Eggs laid in batches on host plant leaves and often covered by female with hair scales from her body; larvae blackish, gregarious when young or until last instar; caterpillar black above and pale red or rusty-brown below, with a series of black specks on sides and sparse black hair, dorsally a narrow central black stripe flanked by broad white/yellow ones and an undulant sub-spiracular white band, a sub-dorsal black spot on each somite; thorax black; head reddish; pupation in a thin cocoon, fixed on a curled leaf of host plant.

Larva black above, brown below, two dorsal white bands, a sub-dorsal black spot on each somite; a series of lateral black specks; sparse black hair; head reddish.

**Host Plants**

*Broussonetia papyrifera*, the Paper Mulberry, *Ficus religiosa*, the Peepal and other spp. (Moraceae), *Camellia sinensis*, the Tea plant (Theaceae), *Crotalaria juncea*, the Sun-hemp (Fabaceae), *Mesua ferrea*, the Indian Rose Chestnut (Calophyllaceae), *Shorea robusta*, the Sal tree (Dipterocarpaceae) and *Tectona grandis*, the Teak (Lamiaceae), (Mathur et al., 1958; Browne, 1968.

*Ficus racemosa*, the Wood-fig and *F. hispida* (Moraceae), *Lansium domesticum* (Meliaceae), *Lycopersicon esculentum* - Tomato (Solanaceae) and *Psidium guajava* (Myrtaceae), (Kononeko & Pinratana, 2005.

*Broussonetia* and *Ficus* (Moraceae), *Mesua* (Calophyllaceae), *Shorea* (Dipterocarpaceae) and *Tectona* (Lamiaceae) (Holloway, 1988.

*Carica papaya*, the Paw-paw (Caricaceae), *Ficus oppositifolia*, the Opposite-leaved Fig and *F. racemosa*, the Wood-fig (Moraceae), (vide Butterfly House.


**Remarks:** *Asota caricae melanesiensis* Viette, 1951 (from New Caledonia) and *A. caricae euroa* Rothschild, 1897 (distribution not available) are other recognised subspecies.

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Photo 2: Asto- underside  
Photo: Asto- Upper side