

Soilborne Diseases of Mulberry and their Management

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Soilborne diseases pose a serious problem for mulberry cultivation during nursery plantation and established gardens, which cause severe loss in revenue generation of mulberry growers as compared to foliar diseases. Various soilborne diseases affect mulberry. Among them, root knot and root rot affect the established plantation resulting in severe loss in leaf yield apart from deterioration in leaf quality, which is a prerequisite in successful sericulture to get the good quality of cocoons. Besides, stem-canker, cutting rot, collar rot and die-back, affect the initial establishment and survivability of mulberry plantation in nursery. The problem is difficult to handle, due to the complex nature of the diseases and also involvement of various biotic and abiotic factors. This is compounded by the occurrence of disease complex (especially nematode + soilborne pathogenic microbes) in established mulberry gardens, which facilitates quick spread of the disease and enhance the plant mortality, resulting substantial loss in leaf yield. Therefore, prevention and timely control measures need to be taken up to protect the mulberry plants from different soilborne plant pathogens. In this review article, symptomatology, epidemiology, disease cycle and control measures of soilborne diseases of mulberry are discussed.

Key words: Soilborne diseases, Symptomatology, Management, Mulberry

Introduction

Mulberry (*Morus* spp.), the sole food plant of silkworm is a perennial crop and usually cultivated as a monocrop.

Leaf production and utilization is once in 70 days and within a year 5–6 crops are taken. Due to the repeated harvesting of leaf, soil gradually becomes poor in nutrients making the crop susceptible to various soilborne pathogens. Once the causative pathogens established in the mulberry gardens it is very difficult to eradicate due to the involvement of various biotic and abiotic factors and due to the complex nature of diseases. As the production of quality cocoons depends upon the quality and quantity of mulberry leaves, timely control measures for protection of mulberry plants from different soilborne diseases are essential. It is expected that this review may help researchers, students, educated farmers in concerned with mulberry production in general and diseases in particular.

Nematode/Root Knot Disease

More than 42 species of nematode belonging to 24 genera (Table 1) are reported to cause the different diseases in mulberry all over the world. Among them, *Meloidogyne incognita* (Kofoid & White) Chitwood, a non-segmented worm belongs to the Family Heteroderoidea, Order Tylenchida of Class Secerentia under Phylum Nematoda causing root knot disease is very serious and chronic to the mulberry. The disease is widespread and more prevalent in sandy soils under irrigated farming system. *M. incognita* has a wide range of host plants, which infects more than 2000 species of plants including almost all agricultural, horticultural, oilseeds, ornamentals, plantation crops, etc. *M. incognita* is more dangerous to mulberry not only because of the direct damage to the crop but also it predisposes the plant to various soilborne plant pathogens (Todia and Keereewan, 1991; Bhagyathy *et al.*, 2000; Sukumar *et al.*, 2000; Nishitha Naik *et al.*, 2003).

Distribution of root knot nematode

The root knot nematode was reported from almost all the countries where mulberry is cultivated. Bessey (1911)

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Table 1. Nematode species reported on mulberry and their geographical distribution

Nation	Species
Brazil	<i>Helicotylenchus dihystra</i> ; <i>Rotylenchus reniformis</i> ; <i>Xiphinema vulgaris</i> (Sharma and Loof, 1972); <i>Meloidogyne incognita</i> (Lordello and Lordello, 1996).
China	<i>Meloidogyne incognita</i> ; <i>M. arenaria</i> ; <i>M. hapla</i> ; <i>M. mali</i> ; <i>M. javanica</i> ; <i>M. arenarithmetici</i> (Ertian, 2003).
Egypt	<i>Pratylenchus</i> spp.; <i>Rotylenchus reniformis</i> (Youssef, 1998).
Europe	<i>Meloidogyne arenaria</i> (Castillo <i>et al.</i> , 2001).
France	<i>Xiphinema meovuitenezi</i> (Dalmasso, 1969).
India	<i>Atlantodorus</i> spp. (Khan <i>et al.</i> , 1993); <i>Carcholaimus mujtabain</i> (Jairajpuri, 1968); <i>Hoplolaimus indicus</i> (Saha <i>et al.</i> , 1983); <i>Meloidogyne incognita</i> ; <i>M. Javanica</i> (Swamy and Govindu, 1965); <i>Hemicriconemoides</i> spp.; <i>Pratylenchus</i> spp. (Edward <i>et al.</i> , 1963); <i>Rotylenchus reniformis</i> (Swarup <i>et al.</i> , 1964); <i>Xiphinema basiri</i> (Thangavelu <i>et al.</i> , 1989); <i>Helicotylenchus</i> spp.; <i>Longidorus</i> spp.; <i>Tylenchorhynchus</i> spp. (Vadivelu, 1996); <i>Helicotylenchus wazihi</i> (Sultan, 1981).
Israel	<i>Meloidogyne</i> spp. (Minz, 1956).
Italy	<i>Xiphinema index</i> ; <i>X. italiae</i> (Martelli and Raski, 1963; Lamberti and DiErrico, 1980).
Japan	<i>Crossonema multisquamatum</i> ; <i>Longidorus martini</i> ; <i>Macrophostonia sphaerocephala</i> ; <i>M. xenoplax</i> ; <i>Nothocriconema mutabile</i> ; <i>Neolobocriconema serratum</i> ; <i>Variasquamata querci</i> (Toida and Momatu, 1981); <i>Gracilacus yokooi</i> ; <i>Meloidogyne arenaria</i> ; <i>M. Javanica</i> ; <i>Ogma coffeae</i> (Toida, 1984); <i>Helicotylenchus erythrinae</i> ; <i>H. dihystra</i> ; <i>Variasquamata gracile</i> (Toida <i>et al.</i> , 1978); <i>Meloidogyne mali</i> (Toida and Yaegashi, 1991); <i>M. suginamensis</i> (Toida and Yaegashi, 1984); <i>Pratylenchus elachistris</i> ; <i>Trichodorus porosus</i> ; <i>Xiphinema americanum</i> ; <i>X. arcum</i> ; <i>X. insigne</i> ; <i>X. basiri</i> (Yokoo, 1970).
Korea	<i>Xiphinema basiri</i> (Lee and Han, 1976).
Pakistan	<i>Neopsilenchus bilineatus</i> ; <i>Trichodorus pakistanensis</i> ; <i>Tylenchus skarduensis</i> (Siddiqui, 1962; Maqbool and Shabina, 1987)
Thailand	<i>Criconemella</i> spp; <i>Ecphyadophora quadralata</i> ; <i>Hoplolaimus seinhorsti</i> ; <i>Helicotylenchus dihystra</i> ; <i>Rotylenchus reniformis</i> ; <i>Tylenchorhynchus</i> spp. (Keereewan and Leeprasert, 1975; Toida and Keereewan, 1991)
Turkey	<i>Pratylenchus allenii</i> ; <i>P. enzurumensis</i> (Yuksel, 1977).
USA	<i>Pratylenchus</i> spp.; <i>Trichodorus porosus</i> ; <i>Xiphinema</i> spp. (Siddiqui <i>et al.</i> , 1973)
USSR	<i>Aphenlenchoides</i> spp.; <i>Helicotylenchus digitiformis</i> ; <i>H. retuses</i> ; <i>Paratylenchus</i> spp.; <i>Longidorus</i> spp. (Adylov, 1976; Kiryanova and Shagalina, 1976).

reported the root knot nematode in mulberry for the first time from USA and subsequently it was reported from other countries *viz.*, Japan (Nozu, 1940), Israel (Minz, 1956), Australia (Colbran, 1958), Brazil (Campos *et al.*, 1974), USSR (Adylov, 1976), China (Zhou, 1982), etc. In India, it has been reported first by Swamy and Govindu (1965) from Mysore and few years later by Mathur *et al.* (1969) from Uttar Pradesh and Haque (1976) from West Bengal. All the six major species of *Meloidogyne viz.*, *M. incognita*, *M. javanica*, *M. hapla*, *M. arenaria*, *M. arenarithmetici* and *M. mali* have been reported to cause disease in mulberry (Todia, 1984; Ertian, 2003). However, *M. incognita* and *M. javanica* have been reported on mulberry from India (Swamy and Govindu, 1965; Narayanan *et al.*, 1966; Sharma and Sarkar, 1998). Besides *Meloidogyne* species, occurrence of various other nematodes such as *Xiphinema basiri*, *Paratylenchus clanchistus*, *Longidorus martini* and *Rotylenchulus reniformis*, etc. has also been reported in mulberry (Table 1) from all over the world.

Survey has revealed that the root knot disease incidence and intensity were found to be very high in red sandy soils followed by red loamy soils while in black cotton soils, the incidence was found very less. Among the different farming systems, the incidence and intensity found to be very high under irrigated gardens, while under rainfed gardens it was very poor (Hirata, 1971; Keerwan and Leeprasert, 1975; Todia, 1984; Teotia *et al.*, 1992; Govindaiah *et al.*, 1994; Philip *et al.*, 1997; Sharma and Sarkar, 1998).

The dynamics of established nematode population (*M. incognita*) at various depths of rhizosphere soil *viz.*, 0 – 15; 15 – 30; 30 – 45; 45 – 60; 60 – 75 and 75 – 90 cm indicate that there is significant increase in density of nematode population up to the depth of 15 – 30 cm, and gradually decreases up to 75 – 90 cm depth. Further, the distribution of nematode population was very high at rhizosphere region as compared to inter-stump (space between plants) and inter-ridge regions *i.e.*, space between rows (Sharma, 1998). Such information may help to formulate the appropriate strategies for disease management.

Symptoms

The severely affected mulberry plants show stunted growth with marginal chlorosis and necrosis of leaves. The underground symptoms include the formation of knots/galls on the roots. The root galls are spherical and vary in size; young galls are too small and yellowish-white while old galls are big and blackish brown in colour. Histopathological observations revealed that due to nematode infection, the vascular tissues and cortex of roots are disorganized hampering the uptake of water and minerals from the soil to aerial parts of the plant (Shetty and Rudramuniyappa, 1992; Ertian, 2003). The deaths of grown up plants are very rare until or unless there is an involvement of some other soilborne pathogens viz., *Fusarium solani*, *F. oxysporum*, *Botryodiplodia theobromae*, etc.

Source of infection

The disease spreads primarily through contaminated soil, farm implements and run-off irrigation water from infected plot to the other plot. Plantation of infected saplings, cultivation of other susceptible crops along with mulberry, and growth of some susceptible weeds in and around the mulberry gardens acts as the secondary sources of infection (Govindaiah *et al.*, 1989; Teotia *et al.*, 1992; Sharma, 1999a).

Economic threshold level and crop loss

M. incognita is found to be highly pathogenic to mulberry causing significant reduction in plant growth and leaf yield at 1000 larvae/plant or 150 larvae/250 cc soil under field conditions. The avoidable leaf yield loss is estimated up to 15% (Govindaiah *et al.*, 1991a; Sharma *et al.*, 1998; Sharma, 1999a). Besides reducing the leaf production, it also affects the nutritive value by reducing the protein content, which is an important nutrient for silkworm growth. Feeding of the leaves harvested from infested plots affects normal growth of silkworms and the quality of the silk produced (Saha *et al.*, 1983; Paul *et al.*, 1995).

Races of *M. incognita*

International *Meloidogyne* Project has identified four races of *M. incognita* viz., Race-1, Race-2, Race-3 and Race-4 and more interestingly all the four races are found in India and other countries (Hartman and Sasser, 1985). Govindaiah *et al.* (1993a) reported that only Race-2 of *M. incognita* is found to infect the mulberry in India.

Epidemiology and disease cycle

During its life cycle, the nematode passes through three stages such as egg, larva and adult. Eggs and larvae are the main sources to spread the disease. The eggs are ellipsoidal with a diameter of 40 – 45 µm and length of 90 – 100 µm while second stage juvenile larvae are vermiform with

15 – 20 µm diameter and 400 – 450 µm length. The second stage juvenile larvae invade the roots through root tips by making the hole with the help of their stylets. After entry, they start feeding on the parenchymatous cells. While feeding on the roots of parenchymatous cells, the larvae seem to inject the secretion in to the roots. The secretion is called saliva, which contains the proteolytic enzyme and flows forward in to the oesophagus and ejected through the stylet leading to enlargement of the cells. Due to the process of hypertrophy and hyperplasia tumors or cancerous galls are formed on the roots. Usually, four moults occur to complete the life cycle, first moult occurs within the egg, which hatches to form the second stage larva. The larvae inside the roots undergo three moults and develop into mature adults. The male is cylindrical worm-like, transparent, colourless with stripes across the body having length of 1400 – 1450 µm and diameter of 30 – 35 µm. The female is pear shaped, elongated and pyriform with a length of 700 – 750 µm and diameter of 400 – 450 µm (Minamizawa, 1997; Sharma, 1999a). The reproduction takes place mostly by parthenogenesis and very rarely by sexual means, as occurrence of males is very rare. Each female lays 300 – 500 eggs covered with gelatinous substance called as egg mass (Ertian, 2003). Electron microscopic study has revealed that two ways of infestation of *M. incognita* occurs viz., primary infestation by second stage juveniles from soil and secondary infestation by juveniles developed within the root tissues (Babu *et al.*, 1996). The nematode produces six generations within a year and life span is completed within 30 – 40 days. Temperature from 15 – 35°C (optimum being 20 – 30°C), soil humidity of more than 60% and soil pH of 4 – 8 are favourable for the development of the nematodes (Murthy, 1986).

Management

Because of the perennial nature of mulberry it is not an easy task to eradicate/control the nematode by any single method. Therefore, an integrated approach involving cultural, chemical, biological and host resistant methods is required to minimize the nematode population below the economic threshold level.

Physical/cultural control

The nematode population in the infected gardens can be reduced by deep digging/ploughing to a depth of 30 – 40 cm during summer. The higher temperature and lower humidity developed by this practice kill the nematode eggs and larvae present in the soil because they are sensitive to heat (Minamizawa, 1997; Sharma, 1999a).

Hot water treatment of roots of mulberry saplings may also be followed for elimination of nematode population.

The root system of mulberry saplings (three-month-old) should be dipped in hot water at a temperature of 48°C for 20 min (Sharma and Govindaiah, 1996). Ertian (2003) has suggested soaking of infected saplings up to the root in hot water at 50–53°C for 20 min before planting to kill the nematodes inside the root.

Application of neem oil cake (obtained from seeds of *Melia azadirachta* L. after oil extraction) at the rate of 2 MT/ha/year in four equal split doses around the basin of the plants during intercultural operations/fertilizers application is recommended, which reduces root galls and egg masses of nematode to an extent of 65–70 % resulting in 16–18% increase in leaf yield. The neem oil cake should thoroughly be incorporated to the soil by ploughing/digging followed by irrigation (Govindaiah *et al.*, 1994, 1997; Sharma *et al.*, 1998).

Intercropping of plants like marigold (*Tagetes patula*), sesame (*Sesamum indicum*) or sun hemp (*Crotalaria spectabilis*) at 30 cm distance in between mulberry rows reduces 60–65% root galls and egg masses of nematode resulting in marginal increase of 8–10% in leaf yield. Mulching of these plants after full growth enriches the soil fertility, in addition to controlling the disease (Govindaiah *et al.*, 1991b, 1997).

Chemical control

Application of Organophosphate and Carbamate nematicides like Furadan 3G (Carbofuran) at the rate of 40 kg/ha/year or Rugby 10G (Sebufos) at the rate of 30 kg/ha/year in four equal split doses at an intervals of 3 months can effectively reduce 70–75% root galls and egg masses of the nematode resulting in 12–16% improvement in leaf yield. The nematicide should be applied during intercultural operations/fertilizers application particularly around the basin of the plants by broadcasting. After broadcasting, it should be well mixed in the soil by ploughing/digging followed by irrigation. After 40–45 days of nematicide application, the leaves can be used for silkworm rearing (Govindaiah *et al.*, 1993b, 1997). In China and Japan, the application of 3–4 liters of dibromochloropropene (DBCP, trade name: Neamgone) emulsion after diluting with 25–50 liters of water at a depth of 15 cm by making the furrows or by injection method is recommended. Soil disinfection with chloropicrin or carbon disulphide or lime nitrogen is reported to be controlling the disease effectively (Minamizawa, 1997; Ertian, 2003).

Soil fumigant like Durofume (Ethylene dibromide + methyl bromide, 1:1) is more effective compared to the nematicides and it reduces the number of root galls and egg masses to an extent of 86–97%. As Durofume is not economical, therefore, it can only be used during nursery plantation to get the disease free saplings (Govindaiah *et al.*, 1993c; Minamizawa, 1997).

Biological control

In this method, talc-based bionematicide (Bionema) produced by a biocontrol agent, *Verticillium chlamydosporium* Goddard, is used along with neem oil cake (Sharma, 1999a). *V. chlamydosporium* parasitises the eggs of *M. incognita* and stops further hatching (De Leij and Kerry, 1991) whereas the compounds present in the neem oil cake (nimbodin, thionemone and phenolics) kill the nematode larvae present in soil (Alam, 1993).

Method of application

One kg Bionema is mixed with 24 kg neem oil cake and 200 kg farmyard manure (sufficient for 1000 plants) and added 30–32 liters of water to maintain moisture up to 15–20%. The mixture is stored under the shade for about one week by covering with gunny cloth. Roots are exposed by digging to a depth of 15 cm, bunches of exposed knots from roots are cut off and destroyed by burning. The mixture is applied at the rate of 200 g/plant around the exposed roots. The mixture of Bionema and neem oil cake is recommended 3 times in a year at an interval of 4 months during cultural operations/fertilizer application followed by irrigation.

Bionema and neem oil cake application reduces the disease severity by 85–90% and increases the leaf yield up to 23%. *V. chlamydosporium* is compatible with neem oil cake, Furadan 3G, Rugby 10G and chemical fertilizers, VA-mycorrhiza, bacterial biofertilizer (*Azotobacter*) and vermicompost. Application of Bionema along with neem oil cake does not show any residual toxicity on silkworms even after 2 days of application (Sharma *et al.*, 2002).

On farm-trials in hot spot areas for the control of root knot disease using application of Furadan, neem oil cake and Bionema along with neem oil cake have confirmed the laboratory results, which recorded reduction of disease by 79–90% with an improvement of leaf yield by 20–22% (Sharma *et al.*, 1998; Rao *et al.*, 2002).

Host resistance

In Brazil, Campos *et al.* (1974) have screened several mulberry varieties against *Meloidogyne* spp. and variety Calabresa was found to be fairly resistant. In India, 250 mulberry varieties were screened for resistance against root knot (*M. incognita*) and none of them were found to be immune or resistant to the disease. However, varieties like S-13, S-30, S-1096, RFS-135, V-1 and Assambola were found to be moderately resistant to the disease (Govindaiah *et al.*, 1996; Sharma *et al.*, 2001). Vadivelu (1996) screened the mulberry varieties against *X. basiri* and found Kanva-2, MR-2, S-13 and S-36 to be resistant. These results could be utilized in disease resistance breeding programmes for evolving nematode-resistant varieties.

Integrated management

Integration of different methods could be utilized for managing the disease depending up on the severity. Integration of moderately resistant variety (V-1/S-13/S-34) and soil application of Bionema along with neem oil cake in disease prone areas reduces the disease severity to an extent of 95 – 98% with an improvement of 28 – 30% leaf yield (Sharma *et al.*, 2001). In moderately resistant varieties, the penetration of juveniles into the roots is lesser resulting in lesser number of root knots, egg masses and nematode population (Govindaiah *et al.*, 1996). Therefore, varieties moderately resistant to nematode also play an important role in Integrated Disease Management programme to keep the nematode population below economical threshold level (Sharma *et al.*, 2001). The following control measures are found to be more appropriate to avoid the spread of nematode disease (Sharma, unpublished).

If the soil is heavily infested with nematodes, fumigation of soil with Durofume is found to be economical and effective during nursery plantation to raise disease free saplings.

When the infection is very severe in mulberry gardens, Bionema along with neem oil cake treatment should be given for 2 – 3 consecutive years. Application of Bionema/ neem oil cake in combination with intercropping of marigold has to be taken in gardens with moderate infection.

Soil solarization during summer season followed by application of Bionema along with neem oil cake and plantation of moderately resistant variety (V-1/S-13/S-34) are more appropriate in disease prone areas.

Destroy all the weeds around the mulberry gardens so that the freshly hatched II stage juveniles die due to starvation because weeds are considered as the carrier of many nematodes and also help them in multiplication. Govindaiah *et al.* (1989) listed the resistant and susceptible weeds against *M. incognita*, which are generally found in mulberry gardens (Table 2). If the saplings have

root knot symptoms, it should be treated with hot water before plantation. Farm implements needs to be disinfected either with 5% formalin solution or dipping in boiling water for 5 – 10 min before use. By using appropriate control measures depending on the severity of infection, root knot disease can be mitigated.

Root rot diseases

Root rot is the most dangerous disease due to its epidemic nature and potentiality to kill the plants completely and poses a serious problem during mulberry cultivation in almost all the sericultural countries. Various types of the mulberry root rot diseases have been reported from all over the world. These are dry root rot, black root rot, charcoal root rot, violet root rot, white root rot, *Armilaria* root rot and bacterial root rot. Among them, the dry (*Fusarium*), black and charcoal root rots are reported in India (Philip *et al.*, 1995, 1997; Sukumar and Padma, 1999). However, violet, white, *Armilaria* and bacterial root rots are observed in other countries like China, Japan, Russia, Sri Lanka and Thailand (Aoki and Matsu, 1959; Kakuliya and Gogeliya, 1970; Aoki, 1971; Takahashi, 1975; Zhing *et al.*, 1988; Ertian, 2003). The disease was reported from almost all types of soils, under varied agro climatic conditions throughout the year. It initially appears in an isolated patch in few plants, which act as an infective centers for the spread of the disease leading to the death of plants within a short period. Survey has revealed that the mortality of plants with leaf yield loss up to 14% due to these root rots.

Dry (*Fusarium*) root rot

In India, earlier it has been reported that violet and white root rots damage the mulberry (Ragaswami *et al.*, 1976) but various surveys indicate that the disease is dry root rot caused by *Fusarium solani* (Mart.) Sacc. and *F.*

Table 2. List of root knot nematode *Meloidogyne incognita* resistant and susceptible weeds of mulberry garden

Resistant weeds	Susceptible weeds
Anagallis arvensis, Argemone maxicana, Acanthospermum hispidum, Amaranthus viridis, Cassia occidentalis, Chrozophora rottleri, Commelina tenghalensis, Crotalaria spectabilis, Cyperus rotundus, Datura metel, Dianthes chinensis, Digitaria adscendens, Desmodium triflorum, Euphorbia geniculata, E. hirta, Evolvulus alsinoides, Flaveria australasica, Glinus oppositifolium, Jatropha gossypifolia, Leucas aspara, Ocimum americanum, Pavonia zeylamica, Phyllanthus urinaria, Tephrosia pumila, Tridax procumbens, Trichodesma indicus, Tribulus terrestris.	Achyranthus aspera, Alternanthera pungens, Borreria ocy-modies, Calotropis procera, Cannabans sativa, Chenopodium album, C. muranticolar, C. murale, Cleome viscosa, Croton bonplandianum, Cyanodon dactylon, Digerea muricata, Eclipta alba, Euphorbia sp., Mimosa pudica, Oxalis corniculata, Parthenium hysterophorus, Solanum nigrum, Trianthema portulacastrum.

oxysporum Schlecht., (Philip *et al.*, 1995, 1997). Both these pathogens belong to the Family Tuberculariaceae, Order Moniliales of Class Deuteromycetes under Division Mycota. There are reports on *Fusarium* spp. causing root rot disease in China and Philippines which is restricted to certain areas and occurring occasionally (Luo and Zhaoxuan, 1989; Telan and Gonzales, 1999).

Symptoms and nature of damage

The first above ground symptoms of the disease appear as sudden withering and defoliation of leaves followed by death of the affected plant. Withering and defoliation start from bottom of the branch and progress upwards. The underground symptoms include decaying of root and decayed roots become black due to black powdery mass below the bark. Later, the tissues below the bark *i.e.*, cortex of primary and secondary roots and conducting tissues (xylem and phloem) turn gummy and paste-like. The bark in such case can be peeled-off easily. Sometimes, white mycelial strands on the root surface on rotten bark can also be seen. The leaves show withering symptom when most of the roots are decayed. On severity, the entire root system gets decayed and the plants die. Affected plants after pruning either failed to sprout or if the plant sprouted bears small and pale yellow leaves with rough surface. The severely affected plants loose the hold in the soil and can easily be uprooted.

Source of infection

The disease spreads fast primarily through contaminated soil, farm implements, and irrigation. The secondary source of infestation is through diseased saplings (Philip *et al.*, 1996).

Epidemiology and disease cycle

The root rot pathogens invade the plant mainly through the wounds inflicted in roots. *F. solani* and *F. oxysporum* produce numerous micro and macro conidia and chlamydospores. Micro conidia of *F. solani* are small, oval, non-septate measuring $4 - 6 \times 10 - 19 \mu\text{m}$ and macro conidia are linear, slightly curved, 2 - 3 septate measuring $5 - 9 \times 27 - 51 \mu\text{m}$. In case of *F. oxysporum*, the micro conidia are small, ovate, non-septate measuring $2 - 3 \times 5 - 7 \mu\text{m}$, and macro conidia are sickle shaped $3 - 4$ septate measuring $4 - 5 \times 20 - 35 \mu\text{m}$. The chlamydospores of both the pathogens are oval to spherical and thick walled. The chlamydospores can survive for longer period in plant debris in the soil even in the absence of the host plants. The disease is favored by soil temperature in the range of $26 - 35^\circ\text{C}$, soil moisture below 40% and pH of 5 - 10. Incidence of nematodes enhances the intensity of root rot as nematodes cause wounds on mulberry roots

while feeding or penetration, which acts as entry points for soilborne fungi like *Fusarium* species.

Management

Various cultural, chemical and integrated methods are suggested for control of the root rot in mulberry.

Cultural method

Sufficient quantity of organic manure should be applied in affected areas. The infested land should be deep ploughed and the soil should be exposed to sunlight during summer months to kill the pathogens. The disease may spread to disease free areas through saplings raised in infected plots. Therefore, sapling raising should be done in disease free plots. Dead plants should be uprooted and burnt immediately. The uprooted area should be heated by burning dry leaves and grasses.

Chemical method

Philip *et al.* (1995) and Sharma (1999b) suggested the chemical method for the control of dry root rot, which involves the root dipping of saplings in 0.1% Bavistin (Carbendazim 50% WP) solution for half an hour and planting in pits dusted with 10 g of Dithane M-45 (Mancozeb 75% WP). As soon as the initial symptoms like wilting/ withering of leaves appear on plants, Dithane M-45 should be dusted at the rate of 10 g/plant after removing the soil around the infected plants to a depth of 15 cm. If the disease is very severe, affected plants should be uprooted and destroyed. Later, 10 g of Dithane M-45/pit is applied in the diseased patches and plant the saplings treated with Bavistin. Dithane M-45 is also applied to surrounding plants near the diseased patch. Four doses of Dithane M-45 should be applied at an interval of 3 months with in a year.

Integrated method

This method involves the combined application of Dithane M-45 and talc-based biofungicide named as Raksha produced by *Trichoderma harzianum* Rifai (Philip *et al.*, 1996; Sharma, 1999b). In integrated method, Dithane M-45 reduces the pathogen load in soil whereas *T. harzianum* is inhibiting the growth of pathogens by the various types of phenomena such as hyperparasitism, competition and antibiosis. During antibiosis, the antagonist is also known to produce hydrolytic enzymes and antifungal compounds like chitinases, trichodermin and gluconase, which dissolves the cell wall of the pathogenic fungi or arrest the growth and leads to inactivate (Tronsmo, 1996).

Method of application

The diseased plants are uprooted and burnt. Ten gram of

Dithane M-45 per pit should be applied and plant the new saplings. After 15 – 20 days, the Raksha mixture at the rate of 500 g/plant in the root zone has to be applied and irrigated. The Raksha mixture can be prepared by mixing of 1 kg Raksha with 50 kg farmyard manure (sufficient for 100 plants) and added 8 – 10 liters of water to maintain the 15 – 20% moisture. The mixture should be stored under shade for about one week by covering with gunny cloth for enhancing the multiplication of *Trichoderma* colonies. Application of Raksha should be continued for one year at an interval of 3 months. *T. harzianum* does not show any residual toxicity on mulberry and silkworms.

Black root rot

Botryodiplodia theobromae Pat. [syn. *Lasioidiplodia theobromae* (Pat.) Griffon & Maubl.] causes black root rot disease in mulberry. The disease is prevalent in Karnataka and Tamil Nadu, India (Radhakrishnan *et al.*, 1995; Sukumar and Padma, 1999). The fungus belongs to the family Sphaeropsidaceae; order Sphaeropsidales of class Deuteromycetes. The above ground level symptoms appear as sudden wilting and defoliation of leaves while below ground symptoms include decaying of roots. Only few branches are affected initially and as disease become severe complete drying of all branches occurs. Under advanced stage, the grayish black mycelium of pathogen is observed on infected roots resulting the death of the conducting tissues leading to death of plant. Rotting and peeling of bark some times extends up to the stem region near the soil line.

The pathogen enters the host through cut ends of the stem after pruning. The fungus prefers 25 – 30°C for good growth and does not survive after 40°C. The pathogen survives on all types of substrates such as soil, weed debris and stacked mulberry twigs. Once the plants are vulnerable to infection the fungus dominates inside the roots multiplying the hyphae rapidly in the cortical tissues and extending up to the pith. It enters the xylem vessels and causes death of the plants. The disease can easily be controlled through spray of 0.2% Bavistin or Captan 50% WP on the stumps immediately after pruning followed by soil application at the rate of 8 kg/acre. The soil application should be given after 10 days of aerial spray (Sukumar *et al.*, 2000).

Charcoal rot

It is caused by *Macrophomina phaseolina* (Tassi.) Goid belonging to the family Sphaeropsidaceae; order Sphaeropsidales of class Deuteromycetes. In India the disease is observed various parts of Tamil Nadu. Occurrence of this disease at present is marginal but it may become severe in future. The disease symptoms start from yellowing of leaves followed by drooping of branches. The affected roots are brown, and then turning black like charcoal and tissues

become weak breaking off easily. In severe cases, the sclerotial bodies are seen scattered on the affected root tissues. The infected plants fail to sprout after pruning and dry up completely (Sridhar *et al.*, 2000; Chowdary *et al.*, 2003).

Since the above disease is of minor importance much attention has not been paid on control measures. However, antagonistic microbes *Bacillus subtilis* have shown inhibitory effect against *M. phaseolina* under laboratory condition (Sridhar *et al.*, 2000). In addition, general field sanitation practices and protective chemical spray/soil application may help to reduce the disease intensity.

Violet root rot

The disease is caused by *Helicobasidium mompa* Tanaka belongs to the order Tremellales of class Basidiomycetes. It is more prevalent in China, Japan, Korea, USSR and Sri Lanka and observed in areas where the water logging condition is more throughout the year. In China, it is called mould root disease while in Japan, it is known as murasaki mompa. The symptoms are similar to mineral deficiency, the leaves become yellow and small in size and buds fail to sprout. On the surface of roots, violet red growth of fungal mycelium is observed. The mycelium forms ball like structure called as sclerotium. In severe conditions roots decay and plant dies. The affected plants or stumps lose hold on the soil and can be uprooted easily without using much force.

The fungus overwinters on the diseased roots or root stumps in the soil in the form of mycelium, sclerotium and rhizomorph. The violet colour mycelial strands produce basidia at their terminal ends. Each basidium bears four basidiospores at the top. Under favourable conditions mycelia form sclerotia and rhizomorph in the soil and also on root system and infect the plants. The pathogen lives in the soil up to 1 – 1.5 m deep. The fungus can survive saprophytically in the soil up to 3 – 5 years. Soil temperature around 26 – 28°C with relative humidity of 70% and pH of 4.2 – 7.2 are favourable for disease development (Minamizawa, 1997; Ertian, 2003).

For the management of the disease, strict quarantine and use of disease free saplings is a must. The diseased plants should be uprooted and soil should be treated with 2% formalin solution and plant the new saplings. Before planting, the saplings should be disinfected with 0.2% Bavistin for 30 min at a temperature of 45°C or 0.3% bleaching powder for 30 min. Intercropping of susceptible crops in mulberry should be avoided. Crop rotation with rice or corn is recommended. 0.2% DBCP (80%) is also advised as soil application at the rate of 5-ml/pit before plantation. Disinfection of the soil with 150 g of chloropicrin per m² or Penta chloro nitro benzene (PCNB) at the rate of 2 kg per 108 sq. ft is also recommended for the control of vio-

let root rot. A mixture of PCNB and DBCP can also be used at the rate of 2 g after mixing with one kg air-dried soil (Minamizawa, 1997; Ertian, 2003). Varieties such as Negumigaesi, Russkaya, Adrenelli-02 and 03 are reported to be resistant to this disease in Russia (Kakuliya and Gogeliya, 1970).

White root rot

White root rot is caused by *Rosellinia necatrix* Berl. ex. Prill. belongs to the order Sphaeriales of class Ascomycetes. It is also prevalent in China, Japan, Korea, USSR and Sri Lanka in water stagnated gardens throughout the year. Affected plants show stunted growth and yellowing of leaves. On the root, white mycelial growth is seen due to which the affected roots become rotted. In severe conditions, plants become weak with feeble leaf bud growth and leaves wither off leading to death of the plants.

The fungus over winters in the form of hyphal strands, sclerotia and rhizomorph on the diseased roots along with root stumps in the soil. It reproduces asexually by chlamydospores, sclerotia and rarely through conidia while sexual reproduction takes place by asci and ascospores in closed fruiting body called perithecium. Each ascus is having 8 ascospores, which are dark brown and spindle shaped. Ascospore germinates under favourable conditions and infects the root system. The fungus can survive in the soil up to a depth of 100 cm for 4 – 5 years. Soil temperature 25 – 30°C with more than 70% humidity is favourable for disease development (Minamizawa, 1997; Ertian, 2003).

Diseased plants have to be uprooted and burnt. Before plantation, the saplings should be dipped in 20% lime suspension or 1% CuSO₄ for one hour and later soil should be disinfected with 1% CuSO₄ or dusting of sulphur powder at 6 kg/acre. The saplings are then planted in disinfected field followed by irrigation. The other preventive methods followed are same as that of the violet root rot disease.

Armilaria root rot

A Basidiomyceteous fungus, *Armilaria mellea* (Vahl. ex. Fr.) Karsten is the casual agent of the root rot disease. The disease is sporadic in nature and reported from Japan and China. The fungus produces black masses of mycelial strands on the roots. The affected roots start decaying and rotting due to which the aerial parts of plant become weak and droop. The leaves of diseased plants fall off prematurely and ultimately the plant dies. The fungus produces turnip shaped black coloured hyphae, which develop clustered or single fruiting bodies near the main trunk. The fruiting body has radial gills, which produce numerous basidia and basidiospores. These spores germinate and produce hyphae, which infect the plant. The preventive

measures followed are same as that of the violet root rot (Minamizawa, 1997; Ertian, 2003).

Bacterial root rot

The disease is caused by a bacterium, *Pseudomonas (Ralstonia) solanacearum* (Smith) Dowson and is more prevalent in China during rainy season (Zhing *et al.*, 1988) while the same disease is known to occur sporadically in India (Mathew *et al.*, 1994). It is also known as bacterial wilt. The bacterium belongs to family: Pseudomonadaceae; order Pseudomonadales of class Schizomycetes. The pathogenic bacterium infects the under ground portion of plant like root system via the root tissues. The symptoms of the disease are wilting of plants and rotting of the roots and xylem tissues become black and rotten due to formation of brown strips in the xylem. Finally, roots become brown or black in colour and the root bark also become rotten and can be easily peeled off. While cutting the mulberry shoot or root some dirty white bacterial ooze is observed.

The pathogen is a soil inhabitant and can remain in a particular soil for several years. The bacterium is Gram-negative, without spore and capsule having 1 – 3 polar flagella at each end, measuring 1.90 × 0.80 µm. Soil and diseased plants are the primary sources of inoculum. The Pathogen overwinters in the soil debris. Secondary infection takes place by irrigation, spore dissemination by rain, and through appliances used for cultivation. For management of the disease, water logging condition should be avoided. Contaminated soil should be disinfected with fomaline solution after diluted with water (1:100) or bleaching powder at the rate of 10 – 20 kg/m². Variety like Qing no. 10, no. 4 and Dong 9 are reported to be resistant against this bacterium in China (Zhing *et al.*, 1988; Minamizawa, 1997; Ertian, 2003).

Besides these, various other pathogens/factors, which cause root rot disease, are also reported on mulberry. These pathogens occur occasionally and restricted to certain areas only. In Thailand, Miura *et al.* (1992) reported a non-infectious root rot. The disease is caused by topographical factors, physical and chemical properties of the soil. The disease is closely related to pedological status. Further, the occurrence of root rot caused by *Phytophthora* spp. was also observed in the same country. *Diplodia morina* Syd. (Roy, 1943) and *Helicobasidium compactum* Boedijn (Munshi *et al.*, 1992) were also reported to cause root rot in mulberry from India.

Diseases of nursery

In any crop either annual or biennial or perennial, initial

establishment plays an important role in growth and crop yield. In India, mulberry is propagated through stem cuttings either raise saplings or direct plantation of cuttings in the main field whereas in other countries, the crop is propagated through seeds. Further, the breeders use the seedlings as stock for grafting in all sericultural countries including India for the conventional breeding programme. Both of the propagated materials are attacked by several soilborne pathogens, which reduce the survival percentage of saplings/seedlings considerably in addition to spreading the diseases in the disease free areas.

Seedling diseases

When seeds are sown in either green house or in field, seedlings are affected by a number of seedborne/soilborne pathogens. Chandrashekar and Shetty (1997) reported that various pathogenic fungi such as *Alternaria alternata* (Fr.) Keissler, *B. theobromae*, etc. affect mulberry seeds and some of them cause the root rot in mulberry. Siddaramaiah and Hegde (1988) reported that *F. solani* and *B. theobromae* is caused root rot in mulberry seedlings. Yoshida and Shirata (1998) have observed that a sudden death or delay of growth caused by *Colletotrichum dematium* (Pers. Ex Fr.) was reported on mulberry seedlings. The fungus belongs to order Melanconiales of class Duetromycetes. The disease is transmitted through infected soil. In china, woolsorters (Anthrax) caused by *C. morifolium* Hara is a major disease often found in seedlings during summer and autumn seasons. The symptoms appeared on the lower parts of the stem of seedlings as bright red spots. Later, many spots merge to form larger spots due to which high mortality occurs during nursery stage. The disease is transmitted through soil. The fungus in the soil is the primary and main source of infection (Ertian, 2003). Recently, the damping-off of mulberry seedlings caused by *F. solani* and *A. alternata* is noticed, which drastically hampers the seedling vigour. Both the pathogens cause pre and post emergence damping-off. The symptoms appear any time from the emergence of young seedlings up to transplanting. In case of pre-emergence stage, the young seedlings are killed before they reach the surface of the soil whereas post emergence of seedlings is characterized by rotting of emerged seedlings near the soil line leading to fall on the ground (Nishitha Naik, unpublished).

Management

The damping-off can be managed by soil drenching with combined application of 0.1% solution of Bavistin and Dithane M-45 (Nishitha Naik, unpublished). Further, in order to ensure good emergence and crop stands, the mulberry seeds before sowing in soil should be dipped in 0.1% Bavistin and Dithane M-45 solution for 30 min, so

that the seeds are free from contamination. (Sharma, unpublished). Spraying of 0.2% Bavistin solution on sprouted seedlings can prevent withering and death of seedlings (Ertian, 2003).

Cutting/sapling diseases

While preparation of the stem cuttings, various wounds are inflicted. These wounds form the main portal for entry of many soilborne pathogens causing various diseases during plantation of the cuttings. These diseases affect the sprouting of cuttings and saplings affecting the survivability to an extent of 30–35%. The mortality is still severe (more than 50%) in case of high-yielding mulberry varieties, which are poor rooters (Luke and Paul, 1982; Singh and Chohan, 1984; Shree and Boraiah, 1987; Sukumar *et al.*, 1991; Gupta *et al.*, 1997).

Stem canker, cutting rot, collar rot and dieback are the major diseases, which affect mulberry during nursery stage and are usually called nursery diseases (Singh and Chohan, 1984; Gupta *et al.*, 1997; Gupta, 1998). These diseases are caused by deuteromycetous fungus. Stem canker caused by *Botryodiplodia theobromae* is manifested as presence of greenish-black eruptions on stem-cuttings. Due to this, the bark becomes dead and decayed resulting in the failure in sprouting of stem-cuttings. Cutting rot is caused by *Fusarium solani* (Mart.), which appears as rotting of whole cuttings and decaying of bark resulting in death of sprouted cuttings while collar rot caused by *Phoma mororum* and *P. sorghina* manifests as brown to black discolouration of bark and rotting of cuttings near the soil line *i.e.*, collar region of cutting (Sukumar and Padma, 1999). *B. theobromae* causes die-back, which is characterized by the wilting of saplings at the apex and progressing downward resulting in death of saplings. It has been observed that cutting rot and stem canker are affecting the sprouted cuttings while collar rot and die back attack during sapling stage (Gupta *et al.*, 1997; Gupta and Sarkar, 1999).

Source of infection

The nursery diseases spread with rain and irrigation water. The primary infection occurs through contaminated soil and farm implements. The secondary infestation takes place by plantation of infected stem-cuttings to the field.

Epidemiology and disease cycle

The fungus, *B. theobromae* produces numerous dark coloured conidia in fruiting bodies known as pycnidia. These conidia are bi-celled and ovoid at maturity while *F. solani* produces numerous small, oval, non-septate micro conidia; and linear, slightly curved, 2–3 septate macro conidia as well as oval to spherical, thick walled character-

istic chlamydospores. However, *Phoma* spp. produces pycnidial type of fructification consisting of numerous rounded, minute and un-septate conidia. *Fusarium* and *Phoma* spp. invade mulberry stem-cuttings/saplings through cut-ends/wounds and cause cutting rot and collar rot diseases, respectively. Die-back disease caused by *B. theobromae* kills the mulberry saplings, as the fungal mycelium seems to reach the phloem of young mulberry plants by invading cortex and cambium cells. The cell contents of affected tissues are exhausted resulting in death of the saplings.

Management

There are various ways such as cultural, biological, chemical and integrated methods to control the nursery diseases in mulberry.

Cultural method

During summer months, the land should be deeply ploughed and exposed to the sunlight after covering the plots with 30 µm thickness transparent polythene sheets for about a month, which helps in destruction of various soilborne pathogens. This method provides better establishment of saplings in the soil (Gupta *et al.*, 1999).

Biological method: It involves the use of lignite-based biofungicide named as “Nursery-Guard” developed by a biocontrol agent, *Trichoderma pseudokoningii* Rifai, which can be used as soil application before plantation of stem cuttings (Gupta and Sarkar, 1999). *T. pseudokoningii* colonizes in the rhizosphere of soil within a short span. Application of *T. pseudokoningii* makes the soil disease-suppressive by the process of competition (colonization), hyper parasitism and antibiosis (production of hydrolytic enzymes and antifungal compounds) and inhibits the growth and proliferation of the pathogens. In addition, *T. pseudokoningii* stimulates the growth of saplings, as it is also known to produce the plant growth promoting substances (Tronsmo, 1996).

Method of application

One kg Nursery-Guard has to be mixed with 60 kg farm-yard manure (sufficient for 2000 stem cuttings) and added 10 – 12 liters of water to maintain the 15 – 20% moisture. The mixture is stored under the shade for one week by covering with gunny cloth to enhance the multiplication of *T. pseudokoningii* colony. The mixture is then broadcasted in nursery beds at the rate of 2 kg/m² and incorporated in to the soil by light digging. The stem-cuttings are then planted in treated beds, followed by irrigation. For direct plantation of stem-cuttings in main fields, the Nursery-Guard mixture is applied in planting pits at the rate of 50 g/pit before plantation followed by irrigation.

Chemical method: Stem cuttings should be soaked in

0.2% Dithane M-45 for half an hour before plantation. Later, the treated cuttings have to be planted in nursery beds. Further, 25 – 30 days old saplings should be sprayed with 0.2% Bavistin solution. This method enhances the saplings production by 15 – 20%, apart from improving the saplings health and vigour (Gupta and Sarkar, 1999).

Integrated method: Integration of soil application of biofungicide, Nursery-Guard and soaking of stem-cuttings in 0.1% solution of Dithane M-45 for half an hour before plantation controls the nursery diseases resulting in better establishment, survivability and growth of mulberry plants (Gupta 1998; Gupta and Sarkar, 1999).

Disease complex

It has been observed that root knot nematodes predispose the mulberry plants making them vulnerable to various other soilborne pathogens causing the disease complex. In Thailand Somkuan *et al.* (1989) and Toida and Keerewan (1991) reported that nematodes like *Ecphyadophora quadralata* and *Hoplolaimus seinhorsti* are found to be associated with root rot pathogens. Yagita and Komura (1992) reported that in Japan mulberry ring spot virus (MRV) is transmitted by the nematode, *Longidorus martini*. They observed that nematode acted as vector to spread the mulberry ring spot virus disease. In India the interaction between *M. incognita* with *F. solani*/Rhizoctonia *bataticola* cause disease complex in mulberry and *M. incognita* provides a good site for easy entry of developing hyphae of these pathogens in to the roots of mulberry (Bhagyathy *et al.*, 2000; Sukumar *et al.*, 2000; Nishitha Naik *et al.*, 2003). Chowdary *et al.* (2003) have observed that root rot disease in mulberry is occurred due to the microbial complexity and microbes were identified as *F. solani* (Fs), *F. oxysporum* (Fo), *B. theobromae* (Bt), *M. phaseolina* (Mp), and black sterile mycelia (Bsm). Pathogenicity trials revealed that all the 5 fungi are pathogenic to mulberry and Fs, Fo along with Bt was found to be frequently associated causing the root rot disease in different parts of Tamil Nadu. In Andhra Pradesh the association of Fs, Fo along with Mp was observed while in Karnataka Fs, Fo, Bt along with Bsm were noticed. The severity of Bt was more in mulberry gardens adjacent to coconut (*Cocos nucifera* L.) plantation whereas Mp severity was observed in the gardens adjacent to ground nut (*Arachis hypogaea* L.) fields (Chowdary, unpublished).

Other soilborne diseases

Besides the diseases mentioned above, there are a few

other soilborne diseases reported on mulberry in India and other countries. The fungus, *Sclerotium rolfsii* Sacc. belongs to the order *Mycelia sterilia* of class Deuteromycetes causes various types of the diseases viz., wilt of sprouted cuttings, stem rot, white thin silk and branch sclerotium. The first two diseases have been reported from India (Siddaramaiah and Patil, 1984; Singh, 1992) while other two diseases are from China (Ertian, 2003). The wilt of sprouted cuttings is characterized by the appearance of black water soaked spots on the collar region of the stem, which rapidly encircle the whole stem and white mycelial mat with a large number of chocolate brown sclerotial bodies are visible over infected tissues. Stem rot is manifested as light brown water soaked patches around the infected portion of the stem, which was in contact with infested soil. However, in case of white thin silk disease, the fungus affects the root and trunk portions near the soil line with the appearance of white silky matter. Branch sclerotium appears on the basal portion of new branch in which the cortical tissues and stem are rotted and peeled off easily. The fungus produces the white mycelial mat with a large number of chocolate brown sclerotial bodies over infected tissues. The above diseases can be controlled by soil drenching with Bavistin (0.05%) or PCNB, Brassicol (0.2%) or 0.2% Captan (Siddaramaiah and Patil, 1984; Ertian, 2003).

It has been observed that *B. theobromae* and *Phoma* spp. also cause the diseases in established mulberry gardens occasionally. *B. theobromae* causes death of stump, which appears as blackish brown necrotic zone on the bark or blackening of central portion (pith region) from the cut-end of stems after pruning. *Phoma* spp. causes sudden wilting and death of affected branches or shoots. Spraying of Bavistin solution (0.2%) on affected mulberry stumps or plants can control these diseases effectively (Siddaramaiah and Patil, 1984; Dayakar Yadav and Sukumar, 1987).

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