

Over Summering of *Phytophthora* Causing Abnormal Leaf Fall Disease of Rubber*

M K George and Thomson T Edathil

Rubber Research Institute of India Kottayam 686 009

Phytophthora is a facultative parasite causing diseases in several crops and observed to be active in nature only during the rainy season. Almost all parts of the rubber plant, are being infected by this pathogen. This fungal infection causes severe damage to the rubber plants resulting in considerable loss of crop (5). In India three *Phytophthora* spp have been recorded causing abnormal leaf fall disease in rubber. McRae in 1918 (2) studied the abnormal leaf fall disease of *Hevea* and observed oospores of *Phytophthora* in infected pods. The pathogen causing the disease was reported as *P. meadii* by McRae. In 1961, Ramakrishnan and Radhakrishna Pillai reported the causal organism as *P. palmivora* (Butl.) Butl. causing leaf fall disease of rubber (4). Subsequently in 1968 Thankamma *et al* recorded the occurrence of *P. meadii* (McRae) also causing abnormal leaf fall disease of rubber in India (5). Recently *P. nicotianae* var *paravitica* was also found to be causing this disease of *H. vea* (6).

The organism produces sexual spores (oospores) inside the host tissue and sporangia on infected plant parts. Sexual spores are produced as an aid for over summering while the asexual spores facilitate quick multiplication of inoculum and rapid spread of the disease. The different types of spores produced by the pathogen may remain on the tree or ultimately reach the soil.

Materials & Methods

Phytophthora infected and dried up pods, fruit stalks, shoots, petioles, bark and seeds were collected after the disease incidence during several seasons from different locations and examined for oospores.

Pericarp of infected and dried up fruits and affected bark having oospores were powdered and suspended in 1% glucose solution in a beaker and incubated at low temperature, $22 \pm 2^\circ\text{C}$, as well as in room temperature and examined after 24 hrs. Healthy pods were inoculated with the powdered pericarp and bark containing oospores after puncturing and were incubated at low temperature and high humidity. These pods were kept under observation for ten days.

Soil samples were collected from rubber growing areas during the off season to isolate the fungus, using a selective medium used by Flowers and Hendrix (1) with slight modification. The medium used was not chilled before seeding with diluted soil suspension. The soil samples collected from the ground surface were air dried and dispersed in sterile water to a dilution of 1 in 50. One millilitre of the diluted soil suspension was added to the surface of the specific medium contained in 9 cm. petridishes. The plates were incubated at low temperature, $22 \pm 2^\circ\text{C}$ in darkness. *Phytophthora* colonies were observed to develop in the seeded plates after 48 hrs. These colonies were transferred to leaf extract agar medium in petridishes.

By the advent of the south west monsoon and just before the disease incidence, green healthy pods were buried in soil and also left exposed on the ground, in mature rubber areas. These pods were frequently examined. Similar studies were conducted under laboratory conditions with soil samples collected from different rubber growing areas. Green healthy pods after puncturing were also inoculated with soil samples in the laboratory.

Healthy rubber fruits were inoculated in the laboratory with *P. meadii* and *P. palmivora*, alone and in combination. These pods were incubated under high humidity and at low temperature for 12 days and frequent observations were made.

Artificial inoculation studies were also carried out with the isolates obtained from germination of oospores from infected and dried up materials and from soil, as per the method described by Radhakrishna Pillai and Chee, (3) to ascertain the pathogenicity of these isolates on rubber.

Results

Oospores of *Phytophthora* measuring 16 to 23 μ were observed in the pericarp of infected and dried up fruits, seeds, fruit stalks, petioles, shoots and bark. The specimens collected during different years from several locations were found to contain *Phytophthora* oospores.

* Paper presented at the Rubber Planters' Conference, India 1974 held at the Rubber Board on 4th and 5th, January 1974.

Oospores from powdered pericarp and bark suspended in 1% glucose solution and incubated at low temperature as well as at room temperature germinated, after 24 hrs. Pure isolate of *Phytophthora* was obtained from germinated oospore found in nature which was identified as *P. meadii*. This isolate when inoculated on rubber twigs produced typical symptoms of abnormal leaf fall disease. Fruit rot was also observed when healthy green fruits were inoculated with powdered pericarp and bark, after puncturing. Rotting pods during the disease incidence season showed different stages of oospore formation, when examined *Phytophthora* infected and callused shoot lesions had oospores after the disease incidence season. *Phytophthora* colonies were observed after 48 hrs. in selective medium in plates seeded with diluted soil suspension, incubated in darkness. The colonies were transferred to leaf extract agar. The isolates thus obtained produced sporangia after 72 hrs. in leaf extract agar and lima bean agar. A number of isolates were made from different samples of soils collected from estates during the off season and identified as *P. meadii*. Artificial inoculations were carried out with those isolates on green pods and shoots of rubber and typical symptoms of infection as observed in nature were produced by the pathogen. The pathogen was reisolated and found to be similar to the original isolate.

Phytophthora was isolated from the fruits buried in the soil and left exposed on ground just before the disease incidence season. Similar studies conducted with fruits buried in soil and inoculated with soil samples in the laboratory also showed symptoms of *Phytophthora* infection and production of sporangia.

The green healthy fruits inoculated with *P. meadii* alone and *P. meadii* and *P. palmivora* together, produced plenty of oospores, but no oospore was observed in the case of pods inoculated with *P. palmivora* alone.

During the beginning of disease incidence season observations were made on the development of primary infection of *Phytophthora* on rubber. Primary infection was noticed on pods at the top of the tree as well as on pods on lower branches, from where the disease was observed to spread.

Discussion

Oospores of *Phytophthora* are resting spores enabling the pathogen to tide over the unfavourable season. The infected and dried up plant parts with oospores remain on the plant or drop on the ground carrying the resting spores with them. In the soil the pathogen can survive till the advent of favourable climatic conditions during the subsequent disease incidence season when the fungus becomes active again and produces the inoculum required for the primary infection.

Phytophthora oospores collected from nature germinated under laboratory conditions and produced new colonies. These isolates are found to be pathogenic to rubber. Hence it is evident that the oospores observed in the infected plant parts germinate during rainy season and cause primary infection by the advent of favourable climatic conditions.

The isolates from soil and dried up fruits when artificially inoculated on twigs and green healthy pods of rubber were found to be pathogenic. The pods inoculated with soil sample showed symptoms of *Phytophthora* infection and produced sporangia. The pods left in the field prior to disease incidence also showed *Phytophthora* infection. Thus, isolation of the pathogen from soil using a special medium, recovery of the fungus from soil baiting with rubber fruits and isolation of *Phytophthora* from fruits showing symptoms of infection after inoculation with soil, show that the primary inoculum for the disease infection develops from the soil. The observation made regarding the incidence of primary infection on the pods on lower branches close to the ground confirms this finding.

Successful pod infection obtained by inoculation of healthy pods with powdered pericarp of infected and dried up fruits and germination of oospores present in the dried up plant parts resulting in production of *Phytophthora* colonies, pathogenic to rubber also indicate that these materials act as sources of production of primary inoculum on the plant by the advent of favourable climatic conditions during the subsequent season.

Summary

Studies on over summering of *Phytophthora* infecting rubber were carried out by collecting infected specimens and soil samples from different estates and isolating the pathogen from such materials, during the off season. The fungus was isolated from oospores from infected dried up plant materials. Similarly the pathogen was isolated from soil samples using a selective medium and by baiting with healthy green fruits. The isolates obtained from soil and dried up plant parts were identified as *P. meadii*. The sources of inoculum for primary infection of abnormal leaf fall disease on rubber were discussed.

Acknowledgement

The authors are grateful to Dr C K N Nair, Director of Research, RRII, for evincing keen interest in this study and to Shri P N Radhakrishna Pillai, Deputy Director, Plant Pathology for all the valuable suggestions and guidance. The authors also wish to express their sincere thanks to CMI, Kew, England for the help rendered in identification of the isolates.

Bibliographical References

- 1 FLOWERS (RA) and HENDRIX (JW) Gallic acid in a procedure for isolation of *Phytophthora parasitica* var *nicotianae* and *Pythium* spp from soil. *Phytopathology* 59; 1969; 725-731.
- 2 MCRAE (W). *Phytophthora meadii* n sp. on *Hevea brasiliensis*. *Mem Dep Agri; India Bot Ser* 9; 1918; 219-273.
- 3 RADHAKRISHNA PILLAI (PN) and CHEE (KH). Susceptibility of *Hevea* Rubber clones to leaf disease caused by two species of *Phytophthora* F A O plant prot bull 16; 1968; 49-51.
- 4 RAMAKRISHNAN (TS) and RADHAKRISHNA PILLAI (PN). Abnormal leaf fall of rubber (*Hevea brasiliensis*) caused by *Phytophthora palmivora* (Butl) Butl in South India I. *Rubb Board Bull* 5; 1961; 11-20.
- 5 THANKAMMA (L), GEORGE (MK) and GEORGE (KV). Occurrence of two spp of *Phytophthora* on *Hevea brasiliensis* in India. *Rubb Board Bull* 10, 1; 1968; 43.
- 6 RUBBER RESEARCH NEWS LETTER (RRN). 4; 1973; 7.