Plant Pathology Journal

ISSN 1812-5387

science alert

ANSInet
an open access publisher
http://ansinet.com

Plant Pathology Journal 10 (2): 85-88, 2011
ISSN 1812-5387 / DOI: 10.3923/ppj.2011.85.88

Etiology of *Trichosporium vesiculosum* Butl. Causing Blister Bark Disease in *Casuarina equisetifolia* Forst

[1]A. Karthikeyan [2]T. Muthukumar and [1]I. Sureka
[1]Institute of Forest Genetics and Tree Breeding, R.S. Puram, Coimbatore-641 002, Tamil Nadu, India
[2]Root and Soil Biology Laboratory, Department of Botany, Bharathiar University,
Coimbatore-641 046, Tamil Nadu, India

Abstract: Blister bark caused by *Trichosporium vesiculosum* Butl. is a lethal fungal disease of *Casuarina equisetifolia* Forst. In plantations, blister bark is more prevalent in trees that are more than two and a half years old. So in this present study, the influence of climatic and edaphic factors on blister bark disease development in *C. equisetifolia* under controlled conditions were investigated. The pathogen isolated from the disease plant tissues was maintained on Potato Dextrose Agar medium. Conidial suspension of the pathogen was inoculated onto 3 to 6 months old. *C. equisetifolia* seedlings raised on different soil types (Alfisol, Vertisol and Ultisol). The pathogenecity study indicated that *T. vesiculosum* attacked *C. equisetifolia* seedlings as early as 5 months old. A temperature range of 25 to 30°C and a relative humidity (%) range of 45 to 60 favoured the conidial development and fungal spread in the seedlings. Similarly soil pH of 5.5 to 6 favoured the pathogen and disease development whereas soil pH<7.5 deters disease development. It was also found that regular watering prevented disease development. Soil type seems to play a major role in disease development as disease incidence and severity was more in seedlings raised in Vertisol than in other soil types. From this experiment it was understood that the favourable temperature, soil factors and soil type for this pathogen to be taken care at the time of planting of *C. equisetifolia* seedlings so as to avoid the blister bark disease.

Key words: Edapho climatic factors, soil pathogen, environment conditions, *Casuarina equisetifolia*, *Trichosporium vesiculosum*

INTRODUCTION

Casuarina equisetifolia Forst. is an important multipurpose tree species mainly grown for its fuel wood. It plays a major role as windbreak and shelterbelt along coasts in several tropical countries including India (Nicodemus, 2009). It is also a nitrogen fixing tree and grows up to 50 m in height and 50 cm dbh. It is widely planted in Andhra Pradesh, Orissa, Tamilnadu and Pondicherry with an area of 50,000 ha (Murthy *et al*., 2006). The average yield of *C. equisetifolia* in India is about 50 to 60 tonnes per ha, under rainfed and 90 to 100 tonnes under irrigated conditions (Karthikeyan *et al*., 2009). It is also used as agroforestry crop and the uses are scaffolding for building construction, ornamental and soil improvement (Nicodemus, 2009). After the devastation of coastal areas by a Tsunami in the year 2004, *C. equisetifolia* gained much importance as the matured trees up to 20 m tall successfully reduced the force of the Tsunami and saved the coastal areas in few parts of Tamilnadu, India (Karthikeyan *et al*., 2009). Though this tree is very strong it is highly susceptible to a destructive fungal pathogen *Trichosporium vesiculosum* Butl. This disease was first reported in India during 1905 and in recent years this disease has been known to cause large-scale mortalities of *C. equisetifolia* in India, China, Kenya and Vietnam (Narayanan *et al*., 1996). This fungus is able to multiply rapidly and spread over the main stem of the tree that causing death of the tree. This disease because of its rapid spread affects many places in Tamil Nadu and Pondicherry. Though the researchers on management of this have carried out considerable research, no concrete control measure is available for want of information on etiology of the disease. Therefore, in this study the mode and spread of infection of this destructive pathogen on the specific host of *C. equisetifolia* were studied under controlled conditions through etiology.

MATERIALS AND METHODS

This study was conducted in Institute of Forest Genetics and Tree Breeding IFGTB), Coimbatore, India from April to October 2008.

Corresponding Author: A. Karthikeyan, Institute of Forest Genetics and Tree Breeding, R.S.Puram, Coimbatore-641 002, Tamil Nadu, India

Isolation of pathogen: Diseased plantations were identified in Panampally (Kerala) situated at 11° 7'N and 77° 7' E has typical warm-humid climate with average annual rainfall of 3,020 mm and mean monthly temperature ranging from 17.5 to 29.5°C and the mean monthly relative humidity range from 75 to 92%. The total area of the affected *C. equisetifolia* is 1 ha (Narayanan *et al*., 1996).

Culture of pathogen: Culture of *T. vesiculosum* was developed by plating the spores collected from the disease trees on to Potato Dextrose Agar (PDA) Medium (potato extract 200 g, Dextrose 20 g and Agar 20 g in 1000 mL of sterile distilled water). Fungal colonies developed after seven days under normal room temperature conditions (21 to 24°C). To identify the influence of temperature on the mycelial growth of *T. vesiculosum* the pathogen was placed under seven temperature conditions (10, 15, 20, 25, 30, 35 and 40°C) in the laboratory using growth gradient chamber. Each treatment was replicated at five times. The colony development was measured after 72 h. The growth incidence was calculated according to the methods of Karthikeyan *et al*. (2003).

Inoculation of pathogen in *C. equisetifolia* seedlings under different environmental conditions: Five millilitre of *T. vesiculosum* spores (10^6 spores mL^{-1}) was inoculated to three months old *C. equisetifolia* according to Davison (1994) and kept under different temperatures (10-40°C) in an environmental chamber (NK system, Nippon chemicals and Laboratory Co. Ltd. Japan). Seedlings inoculated with *T. vesiculosum* were regularly observed for the presence of disease symptoms. Similarly, a second set of inoculated seedlings were kept under different humidity levels (30 to 60°C) in a growth chamber. These seedlings were also periodically observed for the development of blister bark disease.

Influence of irrigation frequency: Seedlings inoculated with *T. vesiculosum* were watered at different intervals (Daily, once in two days, once in 4 days and once in a week) and monitored up to 6 months. The influence of water in development of disease symptoms were also noticed and recorded.

Influence of soil type, moisture and pH: *C. equisetifolia* seedlings were planted and inoculated with *T. vesiculosum* in three different soil types. Vertisol soil was collected from Panampally (Kerala), Alfisol was collected from Sadivayal (Coimbatore, T.N) and Ultisol was collected from Coimbatore (T.N). The plants were monitored for 6 months and the soil moisture and soil pH of each soil type was calculated according to Jackson (1973).

Statistical analysis: All the data were statistically analysed by using the software SPSS (ver.10). The data were presented as means and their standard errors. The data were subjected to Critical difference and Pearson's coefficient correlation was used to determine the degree of association between soil characteristics, growth and disease incidence of *T. vesiculosum* (Zar, 1984).

RESULTS

Growth incidence (%): The growth incidence (%) was higher under temperature of ranged from 25 to 40°C. A significant positive correlation ($p<0.01$) was also occurred between Temperature and growth of *T. vesiculosum* (Table 1).

Disease symptoms under different environmental conditions: Under temperature above 25°C the conidial formation was occurred in the seedlings. Similarly the relative humidity between 40 to 60% was influenced the disease as the diseases symptoms occurred (Table 2). Influence of water was also noticed that caused disease in *T. vesiculosum* inoculated *C. equisetifolia* seedlings daily watered seedlings did not exhibit any disease symptoms. Significant positive ($p<0.01$) correlations were

Table 1: Growth of *T. vesiculosum* under different temperature conditions (mean of 5 replicates)

Growth rate	10°C	15°C	20°C	25°C	30°C	35°C	40°C
Colony diameter (cm^2)	0	1.8	1.8	2.6	2.6	2.6	2.8
Growth incidence (%)	0	11	14.4	42.0*	42.5*	43.3*	43.5*
C.D at 5% level = 5.6							
Pearson's correlation coefficient (r) n = 5							
Temperature vs Colony diameter (cm^2) = 0.848**							
Temperature vs Growth incidence (%) = 0.912**							

*Values significant at 5% of C.D; ** Correlations are significant at p = 0.01

Table 2: Influence of temperature and humidity on *T. vesiculosum* inoculated seedlings of *C. equisetifolia* (mean of 5 replicates)

Temperature (°C)	Humidity (%)	Disease severity score (DSI) and symptoms	% of infection
10	30	0 (No symptoms)	0
15	35	0 (No Symptoms)	0
20	40	1 (Yellowing of needles)	40.5±2.20*
25	45	2 (Wilting and withering of	80.8±2.08
30	50	2 (Wilting and withering of needles)	85.4±2.28
35	55	3 (Drying and dead of seedlings)	86.2±2.13
40	60	3(Drying and dead of seedlings)	88.5±2.42

DSI = 0: No symptoms; 1: Yellowing of needles; 2: Wilting and withering of needles, 3: Drying and dead of seedlings, *±SE of the mean

Table 3: Correlation co-efficient (r) for Temperature, RH (%), Percentage of infection and disease incidence (n = 5)

Factor	% of infection	Disease incidence
Temperature	r = 0.915*	r = 9.70*
RH (%)	r = 0.918*	r = 975*

Correlations are significant at p = 0.01

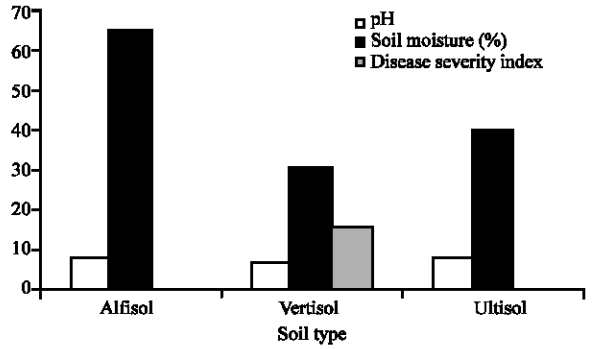

Fig. 1: Influence of soil pH and soil moisture of different soil types on disease severity of *T. vesiculosum* inoculated seeding of *C. equisetifolia*

established among Temperature, RH (%), percentage of infection and disease incidence (Table 3).

Influence of soil moisture and pH: Soil moisture and pH are greatly influence the disease as showed disease symptoms in seedlings of *C. equisetifolia*. Soil pH ranges between 5.5 to 6 highly favours the disease and soil moisture <40% showed the symptoms (Fig. 1).

DISCUSSION

The disease blister bark affects *C. equisetifolia* trees of different ages causing large scale mortalities as high as 90% (Sharma, 1994). Few chemical control measures were developed to control the disease (Narayanan *et al*., 1996; Narayanan, 2000). However; these were not successful in curing the disease in the field (Narayanan and Sharma, 1996). So, to control this disease in field it is essential to understand the edapho climatic factors which may help to prevent the disease in field. Generally the symptoms of the disease include yellowing of the needles (cladophylls) and wilting of the needles. In advanced stage of the disease, small blister appear on the stem and roots. The black spores are exposed when the bark split bursting open blisters. The symptoms of yellowing and wilting of needles were evident at the temperature range of 25°C and above which indicates that the optimum temperature for the pathogen is 25°C and above. It also reflects the water stress which showed chlorotic appearance (Dell and Malajczuk, 1989). Jabnoun-Khiareddine *et al.* (2006) found similar results with Verticillium wilt of tomato under 30 °C temperature conditions. Pensuk *et al.* (2010) supported these results from their experiment stating that peanut bud necrosis virus inoculated pea nut plant to low temperature did not increase the infection. Further, in an earlier study it was showed that the incubation condition of 25°C induced the optimal growth of *Fusarium solani*, *F. oxysporum, Verticillium thalia* and *Rhizoctonia solani* (Fayzalla *et al*., 2008). These all fungi are soil pathogens and belong to fungi imperfecti category. *T. vesiculosum* also falls under the fungi imperfecti category hence the similar results were found in this study. The positive correlation between edapho climatic factors and disease severity showed that favorable environmental conditions activate the growth and disease severity of pathogen on seedlings of *C. equisetifolia*. Similar results were found by Chang *et al.* (2006) in root rot severity at 5-15°C in Cone flowers. Hiber *et al.* (2006) also stated that optimum temperature was more favorable to disease development by fungal pathogens. However, seedlings watered daily failed to exhibit any symptoms in spite of *T. vesiculosum* inoculation. Routine watering may help to prevent the disease development through control of plant temperature. This effect was also observed in plantations of Tamilnadu and Pondicherry where there is a good irrigation. Less soil moisture of vertisol soils are also indicated the results of severe yellowing, drying of needles and formation of black conidial spores. It showed the impact of environment and soil influence the disease (Shearer and Tippett, 1989). The acidic pH causes the disease whereas the pH<7.5 did not show any symptoms. These results supported by Fayzalla *et al.* (2008) as *F. solani*, *F. oxysporum*, *V. thalia* and *R. solani* found grown well under pH 9. Similar results also found in earlier study on impact of soil pH on *Phytophthora cinnamomi* in *Cedrus deodara* (Karthikeyan *et al*., 2000). In this present study, vertisol type soils favours suitable for the pathogen to proliferate and survive. Similarly, Nerey *et al.* (2010) found calcisol soil type is the most suitable soil for *Rhizoctonia solani* causing hypocotyl rot in *Phaseolous vulgaris* which proved that certain soil types are more favourable to particular pathogen than any other soil types. The Temperature, humidity, soil factors and soil types definitely influences the pathogen development which may also cause sever damage in field conditions according to Aigbe and Remison (2010). Hence, it was understood that to prevent this blister bark disease in C. *equiestifolia* seedlings necessary steps have to be undertaken while planting viz., (1) adjusting soil pH to an alkaline range, (2) providing good irrigation system and (3) avoidance of planting sites at vertisol type soils.

CONCLUSION

T. vesiculosum is a severe pathogen which causing blister bark disease in *C. equisetifolia* trees. To control this disease understanding the etiology of the pathogen is very much essential. From this experiment it was understood that the favourable climatic conditions soil factors, soil type for this pathogen to be avoided while planting so as to control the blister bark disease in *C. equisetifolia* trees.

ACKNOWLEDGMENT

The author (A.K) thanked Indian Council of Forestry Research and Education, Govt. of India for financial support in form of a research project.

REFERENCES

Aigbe, S.O. and S.U. Remison, 2010. The influence of root rot on dry matter partition of three cassava cultivars planted in different agro-ecological environments. Asian J. Plant Pathol., 4: 82-89.

Chang, K.F., S.F. Hwang, H. Wang, G. Turnbull and R. Howard, 2006. Etiology and biological control of sclerotinia blight of coneflower using *Trichoderma* species. Plant Pathol. J., 5: 15-19.

Davison, E.M., 1994. Role of environment in die back of Jarrah: Effect of waterlogging in Jarrah and *Phytophthora cinnamomi*. J. R. Soc. W. Aus., 77: 123-126.

Dell, B. and N. Malajczuk, 1989. Jarrah Die Back: A Disease Caused by *Phytophthora cinnamomi*. In: The Jarrah Forest: A Complex Mediterranean Ecosystem, Dell, B., J.J. Hard and N. Malajczuk (Eds.)., Kluwar Academic, Dordrecht, pp: 67-87.

Fayzalla, E.S.A., Y.M. Shabana and N.S. Mahmoud, 2008. Effect of environmental conditions on wilting and root rot fungi pathogenic to solanaceous plants. Plant Pathol. J., 7: 27-33.

Hiber, K., M.D. Remadi, H.J. Khiareddine and M.E. Mahjoub, 2006. Temperature effect on mycelian growth and disease incidence of *Fusarium oxysporum* f.s.p. radicis–lycopersici. Plant Pathol. J., 5: 233-238.

Jabnoun-Khiareddine, H., M. Daami-Remadi, K. Hibar, J. Robb and M. El-Mahjoub, 2006. Effects of temperature on verticillium wilts of tomato in Tunisia. Pl. Pathol. J., 5: 1-6.

Jackson, M.L., 1973. Soil Chemical Analysis. 1st Edn., Prentice Hall of India Pvt. Ltd., New Delhi, India.

Karthikeyan, A., G.S. Goraya, S. Kumar and S. Kalia, 2000. Studies on the mortality of <>(Roxb.) L. Don, in Chail forest (HP) and its causative factors. Ind. For., 126: 1326-1332.

Karthikeyan, A., S. Kumar and S. Kumar, 2003. *Trichoderma viride* a mycoparasite for the control of *Phytopthora cinnamomi*. Ind. For., 129: 631-634.

Karthikeyan, A., B. Deeparaj and P. Nepolean, 2009. Reforestation in Bauxite mine spoils with *Casuarina equisetifolia* Frost. and beneficial microbes. For. Trees Livelihood, 19: 153-165.

Murthy, S.R.V., A. Kumar, A. Chandrasekaran and S. Nakkeeran, 2006. Survey of stem wilt disease of *Casuarina equisetifolia* forst. incited by *Trichosporium vesiculosum* Butler. in Tamil Nadu. Ind. For., 132: 1207-1210.

Narayanan, C. and J.K. Sharma, 1996. Epidemiological studies on blister bark disease of *Casuarina equisetifolia* caused by *Trichosporium vesiculosum*. Proceedings of 3rd International Casuarina Workshop, March 4-7, Vietnam, pp: 80-85.

Narayanan, C., M.J. Dudzinzki, J.K. Sharma and C. Mohanan, 1996. The extent, recognition and management of *Casuarina blister* bark disease. Proceedings of the 3rd International Casuarina Workshop, March 4-7, Vietnam, pp: 74-79.

Narayanan, C., 2000. Studies on wilt disease of *Casuarina equisetifolia* forst. Caused by *Trichosporium vesiculosum*. Ph.D Thesis, Deemed University, Dehra Dun, India.

Nerey, Y., S. Van Beneden, S.C. Franca, A. Jimenez, R. Cupull, L. Herrera and M. Hofte, 2010. Influence of soil type and indigenous pathogenic fungi on bean hypocotyl rot caused by *Rhizoctonia solani* AG4 HGI in Cuba. Soil Biol. Biochem., 42: 797-803.

Nicodemus, A., 2009. Casuarina: A guide for cultivation. Money Spinning Trees 1. IFGTB Publications, India.

Pensuk, U., S. Jogloy and A. Patanothai, 2010. Effect of temperature and relative humidity on the effectiveness of peanut bud necrosis virus inoculation on peanut. Plant Pathol. J., 9: 188-193.

Sharma, J.K., 1994. Pathological investigations in forest nurseries and plantations in Vietnam. Consultant's report. UNDP/FAO Project Vie/92/022. Hanoi, Vietnam, pp: 44.

Shearer, B.L. and J.T. Tippett, 1989. Jarrah Die-back. The dynamics and management of *Phytophthora cinnamomi* in the Jarrah (*Eucalyptus marginata*). Forest of South Western Australia. Research Bulletin No. 3. CALM, pp: 76.

Zar, J.H., 1984. Bio Statistical Analysis. 2nd Edn., Prentice Hall, Englewood Cliffs, New Jersey.