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Effect of Weather Parameters on Blue Pine (*Pinus wallichiana* J.) Needle Blight and Ascospore Release of *Lophodermium pinastri* in India

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Abstract The needle blight disease of Blue pine appeared in February and reached maximum by the end of October. During the study period maximum logrithemic infection rate of 0.0618 and 0.0285 unit/day was recorded during the first fortnight of April in 2008 and 2009, respectively. Weather factors *viz.* temperature and relative humidity were positively correlated with disease intensity (62.1 per cent contribution). However, except temperature all other weather factors were positively correlated with the disease development in terms of infection rate (units/day) (25.7 per cent contribution).The correlation studies of various meteorological factors with disease development revealed significant positive correlation of disease intensity with mean temperature, wind speed and relative humidity. The correlation of disease intensity with rainfall and wind speed was negative. The apparent infection rate was negatively correlated with temperature, but positively correlated with relative humidity, rainfall and wind speed. The multiple linear regression fitted to data revealed that weather factors influenced the disease intensity to the extent of 62.1 per cent in 2008-2009 and for infection rate to the extent of 25.7 per cent. Correlation of various meteorological factors with spore release revealed that spore release and temperature were negatively correlated during whole season (2009) but positively correlated for August-September period. The correlation of spore release with relative humidity, rainfall and wind speed was significantly positive during whole season. The disease incidence depicted negative correlation with temperature, rainfall and wind speed but was positively correlated with RH. Multiple regression analysis of spores released per cm^2 and disease incidence with weather parameters revealed the coefficient of determination (R^2) of 43.9, 72.6 and 21.0 per cent for the spores released was during June-July, August-September periods and in whole season, respectively. However, it was 35.0, 90.9 and 90.3 per cent for disease incidence during June-July, August-September and in whole season on Blue pine plantations.

Keywords: Blue pine, *Pinus wallichiana*, needle blight, weather parameters

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Introduction

Blue pine or Kail pine (*Pinus wallichiana* Jackson) grows widely in moist and dry temperate forests of Western and Central Himalaya from Jammu & Kashmir to Bhutan at an altitude of 1700 to 3700 m amsl and is one of the main commercial timber tree species grown in the Western Himalaya. Blue pine, mainly propagated through nursery-raised seedlings, faces severe regeneration problem under existing agro-climatic conditions. Depending upon the weather conditions, pine trees are often exposed to persistent pathogenic attacks, particularly those causing leaf or needle diseases (Douce *et al*., 2002). Weather parameters viz., temperature, rainfall, relative humidity, sunshine hours and wind speed plays an important role in population dynamics and pathogenic distribution so influence the disease incidence. Losses may be severe if weather and field conditions are favorable for disease spread. Since weather influences plant disease epidemics, therefore, understanding of weather data and climatic conditions in relation to disease development is required to have baseline information for developing simple and reliable disease prediction system. Scanty information is available on the influence of environmental conditions conducive for needlecast disease development. Although weather-based disease forewarning models have been developed in various field and horticulture crops, a functionally viable model for disease forecast in forestry crops is needed for effective integrated disease management (Singh *et al*., 1990, Jayanthi *et al*., 1993 and Prasad *et al.*, 2008). Generally, high humidity (>90%), optimum leaf wetness and conducive temperature (27$\pm$3°C) favour disease development (Picco and Rodolfi, 2002) while wind and rainfall help in spore dispersal to other organs and plants. The impact of environmental conditions and their fluctuations in relation to inoculum build up and disease spread has not been studied quantitatively in needlecast disease. Prediction of environmental factors that have vital role in disease spread are regarded as an economical method for controlling plant diseases.

World-wide the needle blights are caused by several pathogens such as *Scirrhia pini* syn. *Mycosphaerella pini*/*Dothistroma pini* (red band needle blight); *Scirrhia acicola* syn. *Mycosphaerella dearnessii*/*Septoria ascicola* (brown spot needle blight)*; Lophodermium seditiosum* and *L. pinastri* (Lophodermium needlecast); *Ploioderma* spp. (Ploioderma needlecast); *Cercospora pini-densiflora* (Cercospora needle blight); *Cercoseptoria pini* (Cercoseptoria needle blight); *Davisomycellia ampla* (needle blight); *Meloderma desmazierii* (needle blight); *Elytroderma* sp. (Elytroderma needle disease); *Phoma eupyrena* (Phoma blight), etc. (Reddy and Pandey, 1973; Gadgil, 1984; Ivory, 1994; Brown *et al*., 2003; Jerry and Richard, 2008). Among these, Lophodermium needlecast has posed serious threat to pine both in nurseries and field (Miller and Watson, 1971), thereby reducing the plant vigour and health by inducing needle discoloration and defoliation (Johansen and Wade, 1987; Ortiz-Garcia *et al.*, 2003; Hanso and Drenkhan, 2008; Kirk *et al.,* 2008). Severe out-breaks of needle blight (*L. pinastri*) have been reported in 1966-67 in several nurseries at Minnesota, Wisconsin and Michigan (USA) which resulted in the destruction of millions of red pine (*Pinus resinosa* Air.) and Scotch pine (*P. sylvestri*s L.) seedlings (Skilling and Nicholls, 1974). In Europe, needlecast disease has threatened the nurseries for the last 100 years (Millar and Watson, 1971; Staley, 1975; Minter, 1981). In Himalayan mountainous range the needlecast disease incited by several *Lophodermium* species has severely affected the pine nursery seedlings and plantations in forests, parks and gardens during last few years (Ahanger *et al.,* 2012). Severe infestation in saplings has adversely affected Blue pine regeneration programmes and warrants immediate attention. The information on the meteorological factors affecting the needlecast disease is lacking, especially with respect to Indian subcontinent. The information on mechanism of disease development is also lacking for proper diagnosis and to develop effective management strategies.

The present investigations were aimed at to assess the relationship between various weather factors and needlecast disease in Blue pine and generate baseline information on conidial dissemination which may help in developing appropriate strategies for disease management

Materials and methods

Effect of meteorological factors on disease

The role of abiotic factors on the progress of needle cast disease in Blue pine in forest plantations at Forest Nursery, Faculty of Forestry, SKUAST-Kashmir, Shalimar (Srinagar) during the years 2008 and 2009. Six 10 year old Blue pine plants were randomly selected and tagged for scoring of diseased needles under natural epiphytotic conditions. Scoring was done as per 0-4 point scale (Skilling and Nicholls, 1974) wherein 0 = needles/whorls showing no disease symptoms, and 1, 2, 3 and 4 depicted the needles/whorls affected with 1-25, 26-50, 51-75 and $\geq$76% infection, respectively. The scoring was made at fortnightly interval commencing from the 2nd fortnight of February during each year and continued till 1st fortnight of November. The per cent disease intensity was calculated as per the formula:

$$\text{Per cent disease intensity} = \frac{\sum (n \times v)}{N \times 4} \times 100$$

where Σ = Summation, n = number of diseased needles/whorls in each category, v = numerical value of each category, N = number of needles/whorls examined and 4 = maximum grade value. The growth of disease development in terms of apparent infection rate (unit/day) was calculated as per the formulae given by Vanderplank (1963).

$$r_l = \frac{2.3026}{t_2 - t_1} \times \log \frac{x_2}{x_1}$$

where, r is logarithmic infection rate (unit/day) and x_1 and x_2 are disease intensity at time t_1 and t_2, respectively. The data with respect to the weather variables such as temperature, relative humidity, rainfall and wind speed during the study period was procured from the Meteorological Section, Division of Agronomy, SKUAST-Kashmir, Shalimar (Srinagar). The weather data during the study period was correlated with disease development and multiple regressions worked out to determine the influence of individual and combined weather factors on disease development.

Relation between disease incidence, spore discharge and weather parameters

This study was conducted in the year 2009 at Forest Nursery, SKUAST, Shalimar (Srinagar). The study commenced from 2nd June to 30th September, 2009. Twelve 10 year old plantations were randomly selected and on each plantation three vaseline-coated glass slides were hanged on three branches bearing symptomatic needles with hysterothecia. Slides were replaced with new slides at weekly interval and brought to the laboratory to assess the number of spores trapped per square centimeter of glass slide. The spores trapped were observed under a compound microscope (40x). All the spores resembling the Description of Minter *et al* (1982) with respect to *Lophoderemium pinastri* that appeared in the field during four passes across the width of the slide were tallied. The area observed under each microscopic slide at each magnification was determined and ascospore counts converted to the number of spores per mm^2. Further, disease incidence was recorded at weekly interval as per the standard method (Skilling and Nicholls, 1974). The meteorological data during the study period was procured from the Meteorological Section, Division of Agronomy, SKUAST-Kashmir, Shalimar, Srinagar (J&K) and weekly weather data with respect to maximum and minimum temperature, relative humidity, total rainfall and wind speed correlated with the number of spores trapped and percent disease incidence during the corresponding period. Multiple regressions was computed to estimate the effect of combined weather factors on disease development for the whole season as well as at 2 months periodicity taking the number of spores trapped as dependent variable and meteorological factors as independent variables.

Results

Role of weather parameters with disease development

Studies on disease development during 2008 and 2009 and its correlation with weather parameters *viz.,* temperature, relative humidity, rainfall and wind speed revealed that the disease initiated from 2nd fortnight of February and during both the years reached its peak in the month of September (Table 1). In 2008 the disease initiated with intensity of 0.20 per cent in February and attained a peak disease intensity of 25.00 per cent in 1st fortnight of November. No disease progress noticed beyond this period. The disease trend was almost similar during 2009 with initial disease intensity of 0.86 per cent in 2nd fortnight of February and peak disease intensity of 23.33 per cent in 1st fortnight of November. During the study period maximum apparent infection rate of 0.0618unit/day was recorded during the 1st fortnight of April in 2008 and infection rate of 0.0285 unit/day during the same period in 2009.

The mean temperature, relative humidity, wind speed and rainfall varied from 11.68 to 23.90^{o}C, 54.0 to 76.0%, 1.9 to 4.0 and 0 to 4.73, respectively, during infection period in 2008. The temperature, relative humidity, wind speed and rainfall varied from 7.02 to 25.58^{o}C, 56.0 to 71.0%, 1.6 to 3.9 and 0 to 4.94, respectively, during infection period in 2009.

The relationship between weather parameters, apparent infection rate and disease intensity is presented in Table 2. The data revealed that disease intensity was significant and positively correlated with mean temperature (r = 0.42) and relative humidity (r = 0.194). However, disease intensity exhibited negative and signifant correlation with rainfall (r = -0.15) and wind speed (r = -0.79). In case of apparent infection rate, the temperature (-0.315) and relative humidity (-0.036) depicted a negative correlation. However, rainfall (0.339) and wind speed (0.621) were significantly positively correlated with apparent infection rate.

The association between weather parameters and disease intensity vary with season to season and from individual to individual year studies drawing any conclusion is misleading. Hence we have performed the regression analysis with pooled data and conclusion drawn is based only on pooled data analysis. The pooled regression analysis of disease intensity of *Lophodermium pinastri in* relation to weather parameters during 2008-2009 indicated regression coefficients of 0.621.This clearly indicates 62.1% variation is caused by inexpliable reasons or due to errors beyond the control of experiments or due to factors not included in the investigation.

The multiple linear regression equation of pooled data for the years 2008-2009 gave a linear relationship between disease intensity/infection rate (Y) and weather parameters and infection rate (X1=temp, X2= RH, X3=total rainfall, X4=wind speed)

$$Y = 49.9 + 0.158\ X1 - 0.149\ X2 - 0.339\ X3 - 10.3\ X4 \ldots\ldots\ldots\ldots\ldots \text{Eqn.1}$$

($R2=0.621$; $SE =5.12$)

$$Y = 0.0415 - 0.000257\ X_1 + 0.000201\ X2 - +0.00295\ X3 + 0.0156X4 \ldots\ldots\ldots \text{Eqn.2}$$

($R2 = 0.423$; $SE = 0.012$)

This clearly indicates that a unit change in temperature has exerted an influence on the disease intensity up to an extent of 0.158 units in positive direction followed by relative humidity with -0.149 units, cumulative rainfall with -0.339 units and wind speed -10.3 units in the negative direction.

Role of meteorological factors on disease incidence and spores trapped

To ascertain the role of meteorological factors in spore release, weather factors such as temperature, relative humidity, rainfall and wind speed were correlated with the number of spores trapped cm^{-2}. The study revealed that the ascospores were released throughout the season (2nd June to the last week of September, 2009); and the number of spores trapped per cm^2 varied from 11 to 152 (Table 3). Pattern of spore release followed a polynomial curve with total of three peak spore counts, two in June-July period (9-23 June and 7-28 July), and one in August-September period (1-8 September). Maximum spores release occurred in June-July which was approximately 2.5 times more than August-September period. It seemed that optimum temperature, high relative humidity, wind speed and high rainfall (16.39 mm in comparison to 8.98 mm during August-September) during June-July period favoured maximum spore release (787 spores than 305 spores in August-September). The highest peak of spore release may be attributed to favourable wet humid weather and favourable temperature for hysterothecial opening and ascospore release during June-July period.

The correlation between spore release and temperature was significantly negative during June-July period with $r = -0.34$; but was significant and positive in August-September period with $r = 0.17$ (Table 4). However, in whole season the spore release depicted a negative correlation with temperature ($r = -0.30$). The correlation between needle blight incidence and temperature during June-July period was significantly positive ($r = 0.75$) but during August-September period it was significant and negative ($r = -0.79$). However, disease incidence was positively correlated with temperature during the whole season with $r = 0.21$.

The correlation studies between spore release and RH revealed highly significant correlation during August-September period ($r = 0.47$) than during June-July period ($r = 0.23$). The correlation of overall season between spore release and RH was positive with $r = 0.06$. During June-July period the correlation between needle blight incidence and RH was significantly high with correlation coefficient of $r = 0.75$ in comparison to August-September period with $r = 0.24$. The overall correlation between needle blight incidence and RH was significantly positive ($r = 0.46$).

During June-July period the spores release per cm^2 was found positively correlated with rainfall ($r = 0.35$) followed by August-September period ($r = 0.24$). The results indicate that spore release over a season was highly significant with rainfall with a correlation coefficient of ($r = 0.59$). Needle blight incidence showed negative relationship with rainfall throughout the season with correlation coefficients of -0.61, -0.10 and -0.67 during June-July, August-September and overall season, respectively.

The correlation between spore release and wind speed was positive and highly correlated (r = 0.91) during June-July period followed by whole season (r = 0.60) than in August-September period (r = 0.26). However, disease incidence depicted negative correlation with wind speed with correlation coefficients of -0.10, -0.39 and 0.20 in June-July, August-September and overall season, respectively.

Multiple regression analysis of the number of spores released/cm^2 and disease incidence with the prevailing weather parameters was analyzed to know the extent of correlation in June-July, August-September and over the whole season during 2009. The multiple linear regression of spore release per cm^2 in relation to weather parameters during 2009 indicated that regression coefficients for mean temperature, relative humidity, rainfall and wind speed in whole season were 2.61, 3.06, 8.89 and 66.10, respectively. During June-July the regression coefficients for these weather parameters were of 2.39, 1.30, 15.80 and 23.90, respectively. The regression coefficients during August-September period for mean temperature, RH, rainfall and wind speed were 2.64, 3.49, -1.11 and -32.10, respectively (Table 5). The coefficient of determination (R^2) for the number of spores released was 43.9, 72.6 and 21.0 per cent during June-July, August-September and in whole season, respectively.

The multiple linear regression of Lophodermium blight incidence of *Pinus wallichiana* in relation to weather parameters during 2009 indicated regression coefficients of -0.40, 0.15, 0.08 and 8.21 for mean temperature, relative humidity, rainfall and wind speed, respectively, in the whole season. The regression coefficients during June-July for mean temperature, relative humidity, rainfall and wind speed were 1.04, 0.40, -0.07 and 3.75, respectively, and the regression coefficients for these weather parameters during August-September were -0.81, 0.18, -1.48 and 5.45, respectively. The coefficient of determination (R^2) for needle blight incidence on Blue pine plantations during June-July, August-September and in whole season were 35.0, 90.9 and 90.3 per cent, respectively.

Discussion

Role of weather parameters with disease development

Precise studies on climatic requirements of Lophodermium needle blight disease is lacking and the knowledge of climatic requirements may help in effective management of disease through timely adaption of control strategies. The study clearly indicated that the temperature, relative humidity, wind speed and rainfall influence disease expression; and favourable temperature and wind speed are important weather parameters for development of needle blight disease.The maximum disease development during early summer could be attributed to the favourable temperature coupled with higher relative humidity and frequent rains which coincided with the formation of primary needles of Blue pine. Warm moist summers and cool wet falls and abundance of young susceptible hosts reportedly favour disease development (Woods, 2003). The findings are supported by Raspopov (1980); Aminev (1980).,Hanso (1970); Pagony (1971) and Hanso and Hanso, (2001) who found warm wet weather in early spring conducive for disease development on pine plantations. Pagony and Gasko (1977) also observed epidemics of disease due to high seedlings density and moisture. Multiple regression analysis showed that the weather factors accounted for 62.1 per cent variation in disease development, whereas infection rate accounted for 42.3 per cent disease development. The results are in agreement with Kulkerni (2009) who observed similar type of regression equations for anthracnose disease of green gram and reported that weather factors influence the disease to the extent of 57 per cent.

To ascertain the role of meteorological factors in spore release, weather factors such as temperature, relative humidity, rainfall and wind speed were correlated with the number of spores trapped cm^{-2}. The study revealed that the ascospores were released throughout the season showing maximum spores release in June-July than in August-September period. It seemed that optimum temperature, high relative humidity, wind speed and high rainfall during June-July period favoured maximum spore release (787 spores than 305 spores in August-September). The highest peak of spore release may be attributed to favourable wet humid weather and favourable temperature for hysterothecial opening and ascospore release during June-July period. Maximum spore release seemed to be related with the prevailing temperature conditions after the periods of maximum rainfall. However, during August-September period though prevailing temperature were favourable but rainfall was less which delayed ascospore release of *L. pinastri* so resulted in less disease during the period. These findings are in agreement with the findings of Chwalinski (1978), Fedorov and Yakimov (1979) and Raspopov (1980) who observed ascospore release throughout the season (from April to November) and found two spore peaks one in summer (May-June) and another in autumn (August to mid-November). Pagony and Gasko (1977) also observed that the number of spores collected in spring-summer period were 5 times higher than that in summer-autumn. Minter and Miller (1980) reported that hysterothecia open during wet periods and release ascospores. They observed earlier spore dispersal in *L. pinastri* and *L. conigenum* for one to few months than observed in other related species. Raspopov (1980) reported that ascospore dissemination dynamics depends on the number of apothecia with mature ascospores, air and temperature and needle humidity and observed increase in ascospore release at 15-20ºC and decreases at 10-12ºC. He also recorded significant increase in spore dissemination after daily precipitation of 8-10 mm. Linzon (1968) have also reported highest number of ascospore release during the month of June compared to July-August as result of maximum rainfall during June.

The correlation between spore release and temperature was significantly positive in whole season and in August-September period but was negatively correlated in June-July period. Spore release was highly correlated with R.H in August-September period than during June-July period. During June-July period the spores release per cm^2 was found positively correlated with rainfall followed by August-September period. The results indicate that spore release over a season was highly significant with rainfall. Needle blight incidence showed negative relationship with rainfall throughout the season during June-July, August-September and overall season, respectively. The observations are in corroboration with Ostry and Nicholls (1992) who observed positive correlation between rainfall and spore release of *L. pinastri*. Similarly, Chwalinski (1978) reported maximum ascospore production and dispersal as well as highest infection rate in autumn season in case of *L. pinastri*. Pagony and Gasko (1977) observed that maximum spore discharges occurred in mid June to mid July and mid August to ending September. Further, the intensity of spore dissemination was higher in young than in mature pine plantations. Pagony (1971) observed a positive correlation between rainfall and the number of spores trapped during the growing period. Scharpf (1986) reported a maximum ascospore release (1,170 spores) on 10 spore traps in the 1st week of May, 1988 during which a rainfall of 5 cm was recorded.

Multiple regression analysis of the number of spores released/cm^2 and disease incidence with the prevailing weather parameters was analyzed to know the extent of correlation in June-July, August-September and over the whole season during 2009. The multiple linear regression of spore release per cm^2 in relation to weather parameters during 2009 indicated that regression coefficients for mean temperature, relative humidity, rainfall and wind speed in whole season were 2.61, 3.06, 8.89 and 66.10, respectively. During June-July the regression coefficients for these weather parameters were of 2.39, 1.30, 15.80 and 23.90, respectively. The regression coefficients during August-September period for mean temperature, RH, rainfall and wind speed were 2.64, 3.49, -1.11 and -32.10, respectively (Table 5). The coefficient of determination (R^2) for the number of spores released was 43.9, 72.6 and 21.0 per cent during June-July, August-September and in whole season, respectively.

The coefficient of determination (R^2) for needle blight incidence on Blue pine plantations during June-July, August-September and in whole season were 35.0, 90.9 and 90.3 per cent, respectively. The results are in agreement with Kulkerni (2009) who observed similar type of regression equations for anthracnose disease of green gram and reported that weather factors influence the disease to the extent of 57 per cent. Aminev (1980) developed a forecast system for *L. pinastri* on the basis of correlation between the development of needlecast disease in a year and the sum of monthly rainfall, snow depth and available monthly temperature from primary infection period to its renewal in the following year.

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