Int. J. Indust. Entomol. Vol. 22, No. 1, 2011, pp. 11~15

International Journal of
Industrial Entomology

Factors Influencing Development and Severity of Grey Leaf Spot of Mulberry (*Morus* spp.)

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(Received 03 January 2011; Accepted 20 February 2011)

Impact of pruning date, shoot age and weather parameters on the severity and development of grey leaf spot (*Pseudocercospora mori*) of mulberry was studied. The disease severity (%) increased with increase in shoot age irrespective of pruning date. Maximum disease severity was observed in plants pruned during second week of October and minimum in plants pruned during last week of December. Significant (P<0.05) influence of date of pruning, shoot age and their interaction was observed on the severity of the disease. Apparent infection rate (r) was significantly higher during plant growth period from day-48 to day-55. Average apparent rate was higher in plants pruned during first week of September and least in plants pruned during third and fourth week of December. Multiple regression analysis revealed contribution of various combinations of weather parameters on the disease severity. A linear prediction model [$Y=66.05+(-1.39)x_1+(-0.219)x_4$] with significant R^2 was developed for prediction of the disease under natural epiphytotic condition.

Key words: Apparent infection rate, Disease severity, Mulberry, Pruning date, *Pseudocercospora mori*, Shoot age.

Introduction

Mulberry (*Morus* spp.) is cultivated in many parts of India for silkworm rearing. In general, diseases cause deterioration in nutritive value of mulberry leaves by changing biochemical constituents, alter physiological activities of the leaves (Pratheesh Kumar *et al.*, 2003) and reduce quality as well as quantity of the silk produced (Qadri *et al.*, 1999). Leaf spot caused by *Pseudocercospora mori* is an economically important disease of mulberry (Biswas and Pavan Kumar, 1995; Pratheesh Kumar *et al*., 2005). Though some chemicals are reported for control of the disease (Pratheesh Kumar *et al.*, 1999), management using chemical fungicide is not economically feasible, moreover many chemicals are reported be toxic and are non-degradable. Disease evasion is a viable alternative for protection of crop plants from diseases for a better harvest. Since the diseases are interaction of susceptible host, virulent pathogen and favourable weather, age of the plants at a particular condition that favours the pathogen is one of the determining factors of the disease development. Studies on relation between host age and disease development were made in many agricultural crops (Furgo *et al.*, 1994; Sekhon and Sokhi, 1999). Similar possibility for management of mulberry diseases is to be exploited. Therefore an attempt has been made to understand influence of shoot age, date of pruning and weather factors on severity and development of the *P. mori* leaf spot disease of mulberry.

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Materials and Methods

The experiment was conducted in the mulberry farm situated in tropical humid condition with latitude-24º6'N, longitude 88º15' E, and altitude 19 m above msl. A popular mulberry variety S1 was used for the study. Mulberry plants were raised following standard cultural practices (Subba Rao, 1989). Plants with shoot age difference of 7-day were obtained by weekly rotational pruning. Disease severity on 48, 55, 62, 69, 76, 83 and 90-day-old shoots were recorded from plants pruned during first, second, third and fourth week of all the months. Ninety-day-old plants were finally pruned off to repeat he process. Observations on the leaf spot severity were recorded using a 0-5 scale and the disease severity was calculated using standard formula (Govindaiah *et al.*, 1989). This rotational pruning was made for three consecutive years. Data on the epidemiological factors such as temperature (°C), relative humidity (%) and rainfall (mm) were recorded daily from a whether observatory located near the experimental farm. Apparent infection rate (*r*) per unit per day was calculated applying following formula of Van der Plank (1963).

$$\text{Apparent rate of infection } (r) = \frac{2.303}{t_2 - t_1} \log_{10} \frac{x_2 (1- x_1)}{x_1 (1- x_2)}$$

where, r=rate of disease development; t_1=date of first observation; t_2=date of second observation; x_1=disease severity in first observation; x_2=disease severity in second observation. Since whether condition from September to January, favours occurrence and development of the disease, severity and apparent infection rate on shoots of various age groups (plants pruned during different weeks of July to December) during this period was only considered for establishment of influence of plant growth period and pruning dates on severity and apparent infection rate. The data of three consecutive years were pooled and evaluated for two-factor ANOVA and means were compared for significant difference.

Data on severity of disease and meteorological parameters of three years were pooled and multiple regression equations were derived using the meteorological factors such as mean

Table 1. Effect of pruning time on the severity of grey leaf spot disease

Pruning time		Leaf spot severity (%)/ shoot age (days)							Mean
Month	Week	48	55	62	69	72	83	90	
July	I	1.68	3.33	5.15	8.23	9.23	12.71	13.71	7.72
	II	2.13	2.95	4.82	5.96	9.02	14.06	15.53	7.83
	III	2.11	4.84	6.23	5.23	8.60	9.98	10.46	6.78
	IV	1.19	4.08	4.89	6.12	8.53	10.40	12.31	6.80
August	I	1.10	4.77	5.06	4.48	8.39	8.47	12.11	6.34
	II	0.23	1.06	6.99	5.67	8.58	11.36	10.47	6.34
	III	0.89	2.72	5.41	7.23	8.68	18.47	19.41	8.97
	IV	3.52	7.77	6.92	5.61	9.79	17.06	20.17	10.12
September	I	0.83	2.07	8.41	10.17	13.65	20.43	26.31	11.70
	II	1.10	1.72	10.37	10.58	16.78	20.08	22.31	11.85
	III	1.04	1.03	13.92	12.59	16.10	20.74	24.42	12.82
	IV	10.77	12.45	19.91	19.12	19.53	23.68	27.99	19.05
October	I	10.75	15.70	17.57	17.38	20.42	23.63	27.90	19.05
	II	13.72	20.38	22.05	18.25	23.82	20.03	19.33	19.66
	III	9.48	13.69	15.38	16.12	17.76	16.70	15.38	14.93
	IV	0.65	3.65	9.06	12.99	18.24	16.08	14.92	10.80
November	I	0.51	0.71	4.71	9.86	12.48	12.09	11.89	7.47
	II	0.97	9.15	8.64	7.50	4.93	2.35	3.47	5.29
	III	1.00	2.13	3.11	3.27	4.70	5.39	6.15	3.68
	IV	0.49	1.77	3.49	5.42	7.93	6.80	8.26	4.88
December	I	0.83	0.94	2.26	1.81	2.27	4.60	5.05	2.54
	II	1.73	0.49	0.27	0.89	8.02	5.53	0.11	2.44
	III	0.13	0.00	0.23	0.27	0.13	0.7	0.00	0.12
	IV	0.05	0.00	0.04	0.07	0.33	0.04	0.00	0.08
Mean		2.91	5.02	7.83	8.24	10.90	12.66	13.78	
CD (P<0.05)									
Date of Pruning		0.92							
Shoot age		0.94							
Pruning x shoot age		2.42							

maximum (x_1) and minimum (x_2) temperatures, percent relative maximum (x_3), minimum (x_4) humidity, and rainfall (x_5). The predicted mean disease severity index (Y) equation *i.e.*, $a_1+b_1x_1+b_2x_2+...+b_5x_5$ was derived by multiple regression analysis. Where Y denotes the predicted disease severity a denotes the intercept and b_1 to b_5 the regression coefficient for x_1 to x_5 weather factors. Significance of coefficient of multiple determination (R^2) and partial regression coefficient (b) values were followed at $P<0.05$ (Snedecor and Cochran, 1967).

Results

Mean severity of the disease was higher (>15%) on plants

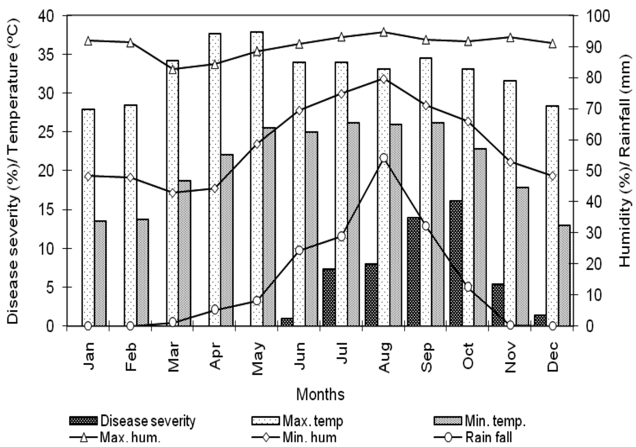

Fig. 1. Grey spot severity and weather conditions during various months, the data is average of three years.

Table 2. Effect of pruning time on the apparent infection rate of grey leaf spot disease

Pruning time		Apparent infection rate (*r*)/ shoot age (days)						Mean
Month	Week	48-55	55-62	62-69	69-76	76-83	83-90	
July	I	0.046	0.039	0.052	0.022	0.033	0.007	0.033
	II	0.056	0.074	0.032	0.057	0.078	0.021	0.053
	III	0.150	0.038	0.027	0.077	0.024	0.007	0.045
	IV	0.175	0.033	0.033	0.051	0.033	0.027	0.059
August	I	0.215	0.009	0.023	0.098	0.004	0.057	0.060
	II	0.138	0.371	0.032	0.062	0.046	0.014	0.095
	III	0.182	0.105	0.045	0.028	0.125	0.009	0.082
	IV	0.120	0.024	0.035	0.094	0.092	0.029	0.046
September	I	0.153	0.222	0.030	0.031	0.087	0.047	0.095
	II	0.068	0.272	0.002	0.075	0.034	0.019	0.078
	III	0.016	0.391	0.017	0.040	0.045	0.031	0.084
	IV	0.020	0.084	0.007	0.003	0.036	0.032	0.028
October	I	0.063	0.019	0.002	0.028	0.027	0.032	0.028
	II	0.070	0.014	0.034	0.048	0.031	0.007	0.010
	III	0.059	0.019	0.008	0.017	0.011	0.013	0.013
	IV	0.274	0.138	0.056	0.059	0.022	0.013	0.082
November	I	0.029	0.301	0.112	0.040	0.006	0.006	0.078
	II	0.357	0.009	0.032	0.060	0.112	0.054	0.034
	III	0.135	0.037	0.024	0.045	0.030	0.024	0.049
	IV	0.260	0.083	0.090	0.053	0.022	0.032	0.083
December	I	0.024	0.173	0.029	0.039	0.105	0.013	0.054
	II	0.166	0.009	0.151	0.344	0.059	0.133	0.021
	III	0.000	0.000	0.032	0.026	0.008	0.00	0.011
	IV	0.000	0.000	0.000	0.159	0.066	0.00	0.015
Mean		0.10	0.099	0.02	0.058	0.019	0.011	
CD (P<0.05)								
Date of Pruning		0.051						
Shoot age		0.026						
Pruning x shoot age		0.125						

Table 3. Correlation of grey leaf spot severity with weather factors

Sl. No.	Multiple regression function	R^2
1	Y=35.645+ (-0.8226**) x_1+(-0.4281*) x_2+(0.1924**) x_3+(-0.1641**) x_4+(0.00289) x_5	0.89
2	Y=35.68+(-0.8215**) x_1+(-0.4296**) x_2+(0.1921**) x_3+(-0.164588) x_4	0.89
3	Y=48.57+(-1.2764**) x_1+(0.1787*) x_3+(-0.2578**) x_4	0.89
4	Y=41.22+(-0.482**) x_1+(-0884**) x_2	0.92
5	Y=66.055+(-1.3917**) x_1+(-0.2195**) x_4	0.88

*Significant at P<0.05, **significant at P<0.01

x_1 = Maximum temperature (ºC); x_2 = Minimum temperature (ºC);

x_3 = Maximum humidity (%); x_4 = Minimum humidity (%); x_5 = Rainfall (mm)

pruned during last week of September to third week of October (Table 1). The disease was however low (<5%) in plants pruned after first fortnight of November. Analysis of variance revealed significant influence of date of pruning (F = 258.32, P < 0.01), shoot age (F = 493.24, P < 0.01) and date of pruning x shoot age interaction (F = 14.21, P < 0.01). The disease severity increased significantly with increase in shoot age. The highest mean disease severity (13.78%) was found in 90-day old plants and least (2.91%) by 48-day old plants. Maximum severity (27.99%) was found in 90-day old shoots which were pruned during last week of September and low (< 1%) in plants pruned during third and fourth week of December irrespective of shoot age. Also there was no significant difference in severity among plants of different shoot ages pruned during third and fourth week of December. Higher mean disease severity was found during the months of September and October (Fig. 1). There was no significant difference in mean disease severity among plants pruned during first fortnight of July, third week of July to second week of August; first and second week of September, fourth week of September to third week of October. The highest mean disease severity (19.66%) was observed on plants pruned during second week of October and least (0.08) in plants pruned during fourth week of December. Infection rate was significantly higher (0.10) during the growth period from 48-55 (Table 2). Infection rate (0.011) was less in 83-90. Analysis of variance revealed significant effect of pruning date (F = 2.62, P < 0.1), growth period (F = 20.91, P < 0.01) and growth period and their interaction (F = 3.42, P < 0.01) on apparent infection rate (r) per unit area per day (Table 2). Infection rate was not significantly differed during growth period from day 48 to day 62. Average apparent infection rate was higher in plants pruned during second week of August to third week of September and minimum in plants pruned after first week of December. No significant difference in mean rate of infection was observed among plants pruned during first week of July to first week of August, among second week of August to third week of September. Stepwise multiple regression analysis revealed that, combination of different weather factors played an important role on the severity of *P. mori* leaf spot. Based on closeness and actual disease severity, five equations were derived (Table 3). The R^2 value of various combinations of weather factors on the severity of disease ranged from 0.878-0.924. Combination of minimum temperature and minimum humidity which has given highest R^2 value (0.924) with minimum combination $Y = 66.05 + (1.39)x_1 - (0.219)x_4$ (where x_1 is maximum temperature and x_4 is minimum humidity) was found as the best-fit equation for prediction of the disease at natural epiphytotic condition.

Discussion

Development of disease depends on the interaction of the susceptible host, a virulent and aggressive pathogen as well as a favorable environment. Escape from disease depends on the lack of coincidence and interaction of these three factors. Whenever a susceptible plant age takes place at a different time, which is out of phase from the appearance of a virulent pathogen, the plant is likely to escape the disease. Escape from disease also may result from early maturing of a variety relating to the arrival and/ or multiplication of the pathogen (Stakman and Harrar, 1957). In the present study, severity increased significantly with increase in shoot age. Highest disease severity noticed in 90-day-old shoots and lowest in 48-day old shoots. Young leaves are reported to be highly resistant to most facultative pathogens and aging leaves show a progressive decrease in resistance (Dowley *et al*., 1975). As leaves age, longer period of leaf wetness is required for infection (Turechek and Stevenson, 1998). Similar results on the higher disease severity in older plants were reported in case of rust infection in mulberry (Pratheesh Kumar *et al*., 2000). The susceptible stage in a plant may appear after the pathogen inoculum has been weakened or exhausted. Maximum grey leaf spot severity was observed in plants pruned during September and October and plants pruned before fourth week of March showed significantly less disease severity. Temperature is an important factor for spread of grey leaf spot, and hence the disease appears with the onset of monsoon.

Generally, leaves of mulberry shoots with age between 60-90 days are utilized for silkworm rearing. In this experiment, plants pruned at various weeks passed through diverse weather conditions, which may be favourable or unfavourable for growth of the pathogen. The interaction between the shoot age and the favourable or unfavourable condition for pathogen elucidates the variation in disease severity observed on plants pruned in different weeks. Significant influence of shoot age and shoot age x pruning date interaction observed in this study indicates influence of both environment and host age on the infection rate (*r*). Infection rate can vary during epidemic of a disease (Gassert, 1976). The rate of infection per unit area per day was noticed higher initially. Many workers made similar observation in a variety of crops (Hegde and Anahosur, 1994). The higher apparent infection rate per unit area per day in young plants rather than in the mature old ones observed may be due to the establishment and colonization of the pathogen in the early growing stage and thereafter the growth of pathogen in a steady rate. The fungi utilize food material available in the leaves for the establishment and colonization which causes a deficit in food materials for subsequent growth of the pathogen reducing in the apparent rate of infection in more mature and older plants (Pratheesh Kumar *et al.,* 2000). Environment influence the epidemics of diseases through its various structural elements. Often the environmental factors have relative rather than absolute effect on epidemics. This favourable level of one or more factor may be compensated for certain deficiency in other factors (Rotem *et al*., 1971). The variation in the infection rate on plant pruned at a certain week during its growth period may be due to various favourable or unfavourable environmental factors along with the host physiological factors that existed during growth season. Since the disease is very prominent and affect the silkworm rearing, early utilization of leaves and change of pruning schedule to minimize the disease severity can be practiced for getting good quality mulberry leaves for silkworm rearing and thereby reducing the loss.

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