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Correlation of disease with meteorological factors and management of *Septoria* leaf spot of chrysanthemum (*Chrysanthemum grandiflorum*)

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ABSTRACT

Leaf spot severity was high at 25°C and lowest at 35°C temperatures. Relative humidity more than 96% influenced the spread of the disease to maximum by causing 57.20% disease severity. In field, the disease usually appears in June and reached to maximum in August with rise in relative humidity and rainfall in both the years (2003–04). The simple, partial and multiple correlations, indicated a positive correlation of disease concerning rainfall and relative humidity while temperature had a negative effect on disease development. The co-efficient of multiple determinations signifies 66.03 and 67.40% variation with the positive weather factors of disease development. In *in vitro* studies, 7 fungicides three from each systemic (contaf, roverol and bavistin) and non-systemic (SAAF, dithane M45 and dithane Z78) category arrested the mycelial growth of the pathogen up to 73% generally at higher concentrations. In field contaf, SAAF, roverol were found equally good in controlling the disease, followed by dithane M45, dithane Z78 and benomyl.

Key words: Biocontrol agents, Chemical control, Chrysanthemum, Epidemiology, Leaf spot, *Septoria obesa*

Septoria leaf spot of chrysanthemum is worldwide in occurrence and known to be one of the destructive diseases of chrysanthemum in India. Two species of *Septoria*, *Septoria chrysanthemella* Sacc. and *S. obesa* Synd. are frequently observed to induce leaf spots. The infected plant develops irregular and circular brownish dead spots that grow progressively upward from stem base of the plant (Huelsma 2001, Kumar *et al*. 2008; Koike and Wilen 2009). The pathogen, *S. obesa* Synd. is more prevalent in Himachal Pradesh.

It is primarily a foliar disease and causes premature defoliation and reduction, but under favourable environmental conditions particularly in rainy season, the severity is more.

Since the magnitude of losses are very high and moreover not much work on epidemiological and control aspects have been reported on this particular disease. Therefore, the present study in relation to correlation between the disease severity and meteorological conditions and chemical control was undertaken.

MATERIALS AND METHODS

Epidemiological studies were conducted to know the pattern of disease development. Six different temperature sets, 10, 15, 20, 25, 30 and 35 °C were adjusted in different incubators. Ten healthy randomly (7–8 times) pin-pricked leaves of chrysanthemum after sterilizing with 0.1% mercuric chloride and subsequently sprayed with 1 ml of spore suspension (60–70 spores/10× microscopic field) were placed gently over the sterilized water agar poured in Petri-plates (6inch dia.) in 3 replications by following leaf detached technique evolved by Lapwood (1968). The Petri-plates were kept at different temperatures under study and disease severity was recorded after 21 days of incubation period in each treatment. The data obtained were analyzed statistically to know the best temperature for development of maximum disease severity (Table1).

Different levels of relative humidity such as 0, 56.8, 75.6, 82.9, 88.5, 92.4, 95.1 and 100% were studied on disease severity according to the method given by McLean and Cook (1941). These levels of relative humidity were maintained using concentrated sulphuric acid in desiccators covered with airtight greased lids. Healthy leaves of almost equal size and age were surface sterilized, pin pricked and inoculated with *S. obesa* in a similar way as mentioned above and placed in Petri-plates lined with 2 layers of blotting sheets. The Petri-plates were kept over wire gauge in desiccators having different humidity levels with 3 replications in each case to record percent disease severity (Table 2).

Correlation of disease severity with meteorological factors

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Table 1 Effect of temperature and relative humidity on disease development in chrysanthemum

Factor	Disease severity (%)
Temperature °C	
10	3.67 (11.02)
15	10.00 (18.42)
20	30.00 (33.42)
25	36.33 (37.07)
30	32.33 (34.65)
35	8.33 (16.77)
P= 0.05	1.56 (1.42)
Relative humidity (%)	
100.0	52.47 (46.72)
98.5	57.20 (49.14)
96.1	53.87 (47.22)
92.9	51.03 (42.74)
88.5	51.00 (45.57)
82.9	46.07 (46.57)
75.6	45.47 (42.00)
56.8	29.43 (32.86)
0.0	0.00 (0.00)
P= 0.05	4.28 (2.52)

Figures in parentheses are arc sine transformed values

under field was worked out. The observations of disease severity were made on 50 randomly selected plants by taking 20 leaves/plant for the two cropping seasons of 2003 and 2004 in research field of Floriculture and Landscaping of Dr Y S Parmar University of Horticulture and Forestry, Solan in randomized block design cultivated every year with a commonly grown variety 'Tata Century' (Table 2). A 0–5 scale as evolved by McKinney (1923) was used for recording disease severity as per the following formula:

Disease severity (%) = sum of all the gradation/no. of leaves × maximum category number × 100.

Standard agronomical operations were carried out in field to raise the crop without application of any fungicides on appearance of symptoms.

The independent variables chosen for correlation were mean air temperature, total rainfall and relative humidity. The values of meteorological factors were obtained from the observatory of Department of Soil Science and Water Management of Dr Y S University of Horticulture and Forestry, Solan. The correlation (simple, partial and multiple) between leaf spot severity and other individual weather factors were drawn and the regression co-efficient of these factors were worked out using multiple least square method by following equation $Y= a+ b_1x_1+ b_2x_2+b_3x_3+ u$, where y is the disease severity and is dependent variables, x_1 is the mean relative humidity (%), x_2 is the mean temperature (°C) and x_3 is the mean total rainfall (mm) –all are weather parameters and independent variables, a is the intercept, b_1 to b_3 are regression co-efficients and u is the error term (Table 3).

Five systemic and seven non-systemic fungicides such as contaf (hexaconazole), bavistin (carbendazim), benomyl (benlate), hilnate (thiophanate methyl), roverol (iprodione), mancozeb (flowable mancozeb), dithane-Z 78 (zineb), dithane-M 45 (mancozeb), antracol (propineb), captan (captan), blitox (copper oxychloride) and SAAF

Table 2 Month-wise disease severity (%) in relation to meteorological factors during 2003-04

Month	Disease severity (%)	Relative humidity (%)	Rainfall (mm)	Temperature (°C)	Disease severity (%)	Relative humidity (%)	Rainfall (mm)	Temperature (°C)
June	17.05	80.0	210.0	23.45	15.92	76.0	148.2	24.21
July	38.15	79.0	155.6	24.60	37.21	77.0	149.1	24.72
August	48.91	80.5	210.0	24.60	47.32	79.6	200.8	25.21
September	40.25	66.0	1.0	22.45	39.85	65.0	1.0	21.05
October	37.21	67.0	0.0	19.60	33.16	66.0	0.0	19.00
November	31.20	57.0	0.0	15.50	27.56	55.0	0.0	15.00

Meteorological data obtained from Department of Soil Sciences and Water Management of Dr YS Parmar University of Horticulture and Forestry, Nauni, Solan

Table 3 Simple and partial correlation co-efficients between weather parameters and disease severity of Septoria leaf spot of chrysanthemum

Parameter	Simple correlation co-efficient with disease severity		Partial correlation coefficient with disease severity	
	2003	2004	2003	2004
Temperature °C	–0.2152	-0.2400	-0.1068	-0.0954
Relative humidity(%)	0.0098	0.2190	0.5473	0.0020
Rainfall (mm)	0.3627*	0.1445*	0.7841*	0.1074*

P= 0.01

Multiple linear regression equation indicating correlation among weather parameters during 2003 and 2004 with disease severity is given here:

Year	Character	Equation	Co-efficient of determination R^2 (%)
2003	Disease severity (%)	X1 =- 65.351+0.17510(RH)-0.565(T)+0.119*(RF) (65.998) (0.18940) (0.37239) (0.667)	66.03
2004	Disease severity (%)	Y1=-64.341+0.0134(RH)-0.12315(T)+0.0304*(RF) (66.851) (0.4646) (0.90847) (0.1992)	67.04

Values in parentheses are standard error, *P= 0.01, T, Temperature; RH, relative humidity; RF,rainfall

(carbendazim–12%+ mancozeb–63%) were evaluated *in vitro* against *S. obesa* by Poisoned Food Technique (Falck 1907). Each treatment was replicated thrice with suitable control (without fungicide). Different fungicides mixed aseptically in equal quantities with double strength sterilized potato dextrose agar medium to give a required concentration of each fungicide. The medium was pre-sterilized in an autoclave at 15 lbs pressure for 20 min. before pouring into Petri-plates. The mycelial discs of 4 mm of an actively growing colony of the test fungus were placed in the centre of the test plates. The per cent mycelial inhibition was observed and was calculated by formula given by Vincent (1947) as follows: Per cent inhibition (I) = C-T/C×100 where, C and T stands for fungal growth rates (mm) in control and treated medium. The results obtained after statistical analysis of the data arresting the fungal radial growth are presented in Table 4.

Field experiments were conducted with cultivar, 'Tata Century' during 2003 and 2004 at experimental farm of Department of Mycology and Plant Pathology of the University. Recommended agronomical practices were followed time to time in growing crop. Ten fungicides selected from the *in vitro* studies, viz contaf (hexaconazole 0.3%), bavistin (carbendazim 0.05%), benomyl (benlate 0.1%), roverol (iprodione 0.4%), mancozeb flowable (mancozeb 0.25%), dithane-Z 78 (zineb 0.25%), antracol (propineb 0.3%), captan (captan 0.2%) and SAAF (carbendazim 12%+ mancozeb 63% 0.2%) were evaluated for their efficacy to check the disease spread (Table 5). The experiment was conducted in randomized block design (RBD) consisting of 11 treatments including one control along with 3 replications. First spray of fungicides was given as soon as the symptoms of the disease become prominent in the field during the second week of June and thereafter 3 subsequent sprays were given at 15 days intervals. The disease severity was recorded

Table 4 In vitro evaluation of systemic and non-systemic fungicides against *Septoria obesa* at different concentrations

Fungicide	Mycelial inhibition(%) at different concentrations (ppm)			Mean
	500	750	1000	
Systemic				
Bavistin	67.29(55.12)	69.90(56.73)	70.59(57.16)	69.26(56.14)
Benomyl	48.00(43.85)	59.27(50.38)	62.42(52.19)	56.56(48.80)
Hilnate	43.67(41.37)	50.26(45.15)	50.89(45.51)	48.47(44.01)
Roverol	66.90954.80)	70.67(57.21)	71.56(57.79)	69.71(56.63)
Contaf	67.92(53.60)	70.98(57.41)	71.51(59.03)	70.80(57.32)
	49.35(43.66)	53.79(45.27)	56.21(46.76)	
	500	750	1000	
Non-systemic				
Captan	46.67(43.08)	53.09(46.78)	56.78(48.90)	52.18(46.25)
Dithane M 45	67.40(55.19)	67.37(53.36)	69.24(56.32)	68.00(55.56)
Blitox	44.26(41.71)	45.87(41.63)	48.24(42.49)	46.12(42.78)
Saaf	69.63(56.56)	72.13(58.14)	75.74(60.53)	72.50(58.91)
Mancozeb (flowable)	55.73(47.14)	56.23(48.58)	59.06(50.22)	56.34(48.65)
Dithane Z 78	60.98(51.35)	64.46(53.41)	70.96(57.40)	65.40(54.05)
Antracol	58.04(49.65)	61.79(51.82)	67.19(55.06)	62.36(52.18)
Mean	49.73(42.86)	52.04(44.23)	55.01(46.02)	

P =0.05 Figures in parentheses are arc sine transformed values
Fungicides (systemic)-1.35; concentration-0.88; fungicide (systemic) × concentration-2.05
Fungicides (non-systemic)-1.52; concentration-0.91; fungicide (non-systemic) x concentration-1.94

15 days after the last spray by following 0–5 scale (Mckinney 1923)

RESULTS AND DISCUSSION

Epidemiological studies

The leaf spot severity increased from 3.67 to 36.33% with increase in temperature up to 25°C and declined to 8.33% at 35°C from maximum intensity of 36.33% recorded at 25°C. Higher levels of relative humidity (96.1 and 98.5%) influenced the disease to maximum causing 57.20 and 53.87% severity, which were found optimum for development of the disease. However, minimum disease severity of 29.43% was obtained with lowest relative humidity level. Relative humidity of 88.5% increased the disease to a maximum (Tok and Kurt 2004, Kurt and Tok 2005).

Correlation studies with meteorological parameters

Disease was reported in the field in June 2003 and 2004 which gradually increased and reached up to maximum in August in both the years when the relative humidity reached to 80.5%, and declined considerably thereafter. The simple and partial and multiple correlation co-efficients (Tables 2, 3) indicate that there was a positive correlation between relative humidity and rainfall while temperature had a negative correlation with the disease severity in both the years under investigation. Correlation co-efficient of disease severity with rainfall was found to be significant at 1% levels of significance, whereas other weather parameter were reported significant at 5% level of significance.

The data set out in above Tables indicated that by keeping one factor constant, the combined effects of relative humidity, total rainfall reported to have a significant correlation, whereas temperature had a negative effect on disease development. The results embodied in this study revealed that the leaf spot severity in chrysanthemum was greatly influenced by two factors simultaneously with increase in relative humidity and total rainfall there was maximum rise in the disease severity level compared to temperature. However, with temperature an inverse relationship was obtained. The co-efficient of multiple determinations was calculated to be 0.6603 and 0.6704, which signifies that 66.03% and 67.40% variation in disease severity in 2003 and 2004 were due to the weather parameters taken into consideration and rest of variation is contributed from the other unknown factors. Hazard *et al*. (2008) also reported relative humidity as one of the prime factors in the development and spread of *Septoria* leaf spot on tomatoes. Relative humidity (100%) and 8 hr dark period at 17°C temperature increased the severity of disease. Nao (2008) reported significant role of rainfall in spread of leaf spot disease in lettuce caused by *S. lactucae*. Leaf wetness for 12 hr or longer increases the disease severity. Lannon and Bertrand (2004) also observed cool weather and high humidity as favourable factors for the rapid spread of *S. tritici* blotch on wheat in field.

Chemical control

All systemic fungicides showed superiority over control in inhibiting mycelial growth of the fungus *(S. obesa*). Contaf, roverol and bavistin of systemic group arrested maximum mycelial growth of 70.80, 69.71, 69.26% and adjudged best fungicides and found significantly same compared to benomyl (56.56%) and hilnate (48.47%). However, all the fungicides at higher concentrations showed maximum inhibition than the lower concentrations (Table 4). SAAF (72.50%), dithane M 45 (68.00%) and dithane Z 78 (65.40%) recorded maximum mycelial inhibition amongst non-systemic fungicides. About concentrations, SAAF and dithane M 45 registered more mycelial inhibition at 500 and 750 ppm. In field trials, the results indicated that contaf, SAAF and roverol were statistically at par in controlling the disease to great extent by giving minimum severity of 11.59,

Table 5 Field evaluation of fungicides against *Septoria* leaf spot of chrysanthemum during 2003-04

Fungicide	Disease severity (%)		Overall mean	Disease control (%)
	2003	2004		
Dithane M 45	17.02 (3.90)	15.24(3.20)	16.13(4.01)	65.65
Benomyl	15.58(3.95)	15.05(3.88)	15.31(3.91)	63.21
Bavistin	21.40(4.62)	17.91(4.25)	15.35(4.49)	62.21
Dithane Z 78	14.85(3.85)	12.82(3.53)	13.83(3.72)	65.45
SAAF	12.32(3.51)	11.41(3.38)	11.86(3.44)	70.32
Captan	20.41(4.72)	18.56(4.30)	19.48(4.06)	51.29
Roverol	13.10(3.62)	11.33(3.36)	12.21(3.49)	69.48
Contaf	11.81(3.43)	11.38(3.37)	11.59(3.40)	70.97
Antracol	15.65(3.45)	15.23(3.90)	15.44(3.93)	61.37
Mancozeb (flowable)	20.37(4.07)	16.57(4.07)	18.37(4.29)	53.98
Control	46.01(43.86)	48.01(42.79)	47.01(43.29)	0.00
(*P*= 0.05)	3.38(0.48)	1.24(0.15)	2.37(0.30)	

Figures in parentheses are arc sine transformed values

11.86 and 12.2%. In order of preference according to the maximum disease control, dithane M 45 (65.65%), dithane Z 78 (65.45%), benomyl (63.21%) were found next best fungicides.

In field, contaf gave lowest disease severity while captan and mancozeb flowable reported higher severity levels and hence less disease control as shown in Table 5.

Ahmad and Ahmad (2000) evaluated carbendazim, thiophante methyl and captan against *Septoria* leaf spot disease and reported their superiority over other fungicides. The evidence of the field efficacy of these fungicides was found by various workers in controlling different species of *Septoria* (Bates 2005 and Lambe 2009). Raudales and McSpedden-Gardena (2008) used microbial biopesticide for the control of *Septoria* diseases in tomato in organic farming. Fungicides like Folcan 460 EC and Opera were used in full and reduced doses of one or split applications against *S. tritici* and *S. nodorum*, leaf blotch complex in wheat. A cover crop mulch and fungicide programme also affected the progress of disease and marketable yield in processing tomato production (Oziambo and Sherm 2005, Wyenandt *et al*. 2008)

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