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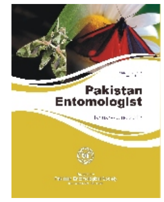

STUDIES ON CITRUS LEAF MINER (CLM) IN RELATION TO ABIOTIC FACTORS ON DIFFERENT HOST PLANTS IN PUNJAB, PAKISTAN

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ABSTRACT

Studies on the relationships between seasonal ambient temperatures (weekly average, maximum and minimum) and the relative abundance and incidence of citrus leaf miner (CLM) on plants of different citrus varieties were conducted in citrus nurseries and orchards at two experimental farms in Pakistan by sampling infestations of CLM at weekly intervals from May through September. Significant relationships were found between CLM abundance and incidence and the temperature variables by correlation and regression analyses. Maximum, minimum and average weekly temperatures had consistent positive relations with CLM abundance and incidence, whereas, rainfall and relative humidity had either a negative or no correlation to insect infestation. Relative humidity at one site had a positive correlation with CLM abundance and incidence (R^2 value ranged from 0.29 to 0.72). Linear regression models were formulated with a CLM abundance as a response variable and stepwise addition of max. temp., min. temp, ave. temp. and RH as concomitant variables. Models explained 10-72% of the variation in the data sets based on the values of the regression coefficient. These results are discussed in view of previous studies of relationship of CLM infestation and its environment.

Keywords: Weather, insect infestation, citrus, environmental entomology

INTRODUCTION

Environmental resistance from abiotic and biotic factors affects the distribution, colonization, survival, abundance, behavior, fitness, and the life history of insects. It is well established that extremes of weather fundamentals limit the geographic range of insect populations, either imparting mortality or depriving them from their food. Temperature, for example, can be the sole factor for initiating dormancy in insects (Danks, 2002).

Models of insect-plant-environment interactions are often based on temperature. Plant phenology is highly dependent upon temperature. Insect-plant-environment interactions are important to understand the population ecology of insect. Horticultural crops are highly sensitive to the extremes of weather factors (Wani *et al*., 2010). Citrus trees produce several flushes in a year and this is directly associated with heavy rainfall and warm temperature. Young trees often appear to flush almost continually during favorable weather conditions (Hall, 2007).

Citrus leaf miners rely on flushing shoots for their reproduction and development. Several studies have stated connection of different weather factors with population dynamics of citrus leaf miner (*Phyllocnistis citrella* Staint. [Lepidoptera: Gracilleriidae). This association has been mostly followed on single citrus species (Bhumanavar and Sing, 1978; Patel *et al*., 1994; Pena *et al*., 1996; Legaspi *et al*., 1999; Ateyyat and Mustafa, 2000; Ujiye, 2000; Rahman *et al*., 2005; Diez *et al*., 2006; Xiao *et al*., 2007; Sinclair and Hughes, 2008; Jesus *et al*., 2008; Malika *et al*., 2010) however, few reports have shown a comparison among different citrus species (You and Wang, 1999; Goane *et al*., 2008).

Citrus leaf miner has also been implicated in the spread of citrus canker (Jesus *et al*., 2006; Atiq *et al*., 2007). There was a significant correlation of maximum temperature and relative humidity with disease development. The correlation of the rainfall and minimum temperature with citrus canker was also significant in a few varieties, while the role of wind was non-significant in all the varieties. It is suggested that farmers should take care of their orchards against citrus canker especially in the month of July, August and September (Khan and Abid, 2007).

Our objective was to measure the seasonal occurrence of

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CLM on several varieties of citrus and correlate the relative abundance of the CLM to weather factors at two geographic locations, in the nursery and in the full grown trees in the orchard. These measurements were analyzed by regression to determine the relationships of the insect infestation to the weather factors.

MATERIALS AND METHODS

Population of citrus leaf miner (CLM) in citrus nurseries

This study was carried out at University of Agriculture, Faisalabad (UAF) and Citrus Research Institute (CRI), Sargodha, the nurseries at UAF and CRI had 7 and 9 citrus varieties, respectively, whereas the groves had 4 and 6 citrus varieties.

Abundance of citrus leaf miner on nursery plant was monitored by taking 20 plants per variety. From each plant a bunch of fresh leaves was selected. From each bunch total fresh leaves and total damaged leaves with citrus leaf miner galleries were counted as number of CLM on fresh leaves/total no. of fresh leaves. The seven citrus varieties at UAF and nine in Sargodha were selected, on which incidence of citrus leaf miner was determined weekly in the years 2010 and 2011.

CLM incidence in citrus groves at PARS and CRI and its relation with abiotic factors

A. Two methods were used to determine CLM infestation level in citrus groves. The first sampling method, measured CLM abundance. Three groves each at University of Agriculture Faisalabad (Square 9), and Citrus Research Institute (Risala 5) at Sargodha were selected. These groves had the same set of citrus varieties, as the nursery, with 7 varieties at UPA and 9 varieties at CRI. In groves, abundance of citrus leaf miner was monitored by randomly selecting 5 plants per variety. 25 leaves per plant were randomly selected from all four sides of a tree to record the relative abundance of CLM mines. Leaf mines were enumerated and classified as with or without live leafminer larvae.

B. For the second sampling method measured CLM incidence. Citrus groves were examined at each site over a wider area than was observed in the first sampling method. The groves at each site were distantly spaced and each grove had one variety, 100 leaves were selected and examined for presence of CLM and data are represented as mean percent CLM..

The measurement of weather factors (maximum temperature, minimum temperature, relative humidity, rainfall) was taken from observatories at each selected area in the years 2010 and 2011. The population dynamics in these cases was statistically analyzed through correlation and regression with abiotic factors in Minitab Version 11.

RESULTS

The CLM population on nursery at UAF and CRI during May to September for the said years showed that Lemon had comparatively high population at UAF (Fig.1) and CRI (Fig. 2) whereas Musambi was at par with Kinnow. In contrast to Sargodha, leaf miner was highest in May and comparably June and July but less than in August and September. Two reasons may be important for this difference; viz., abiotic factors and spray regime. Among Kinnow varieties at CRI, seedless Kinnow had less number of leaf miner than Kinnow and Am Kinnow. Musambi had least number of leaf miner population (Fig. 2).

At Faisalabad, groves under study were maintained at Campus in University of Agriculture. Highest leaf miner population was observed in May in all varieties of citrus, followed by the number in June. Least population was observed in August on all varieties (Fig. 3). Incidence of CLM in citrus groves at Sargodha is given in Fig. 4. There was negligible incidence of CLM on Kinnow seedless (June through September), Sweet orange (June and July) and Feutrell's early (July). The incidence of old galleries on leaves suggest rapid growth of the pest during these times of the season. Variation of galleries in different months in varieties shows the impact of variety on life cycle of the pest.

Temperature and relative humidity had significant, but contrasting relation with CLM incidence. Relation of incidence with rainfall was not uniform in all seven varsities of citrus at UAF. Predictive model of effect abiotic factors on CLM incidence showed higher coefficient of determination value ($p>0.30$) on all varieties except lemon where it was $p<0.30$ at Faisalabad (Table 1).

The relationship between abiotic factors and CLM incidence on nine different varieties of citrus in Sargodha is given in Table 2. Notable in this place is non-significant and positive relation between relative humidity and CLM incidence which was not observed in Faisalabad.

Temperature appears to be playing a major role of change in population. Maximum, minimum and average temperature had positive relationship with CLM incidence. This indicates that CLM infestation level would increase with a concomitant increase in any or all of these factors. Rainfall in CRI appeared to reduce the incidence of CLM with negative r values on all varieties except grapefruit. Coefficient of determination value of effect of abiotic factors on CLM incidence was >0.50 when temperature and relative humidity were included in the predictive model (Tables 3 & 4).

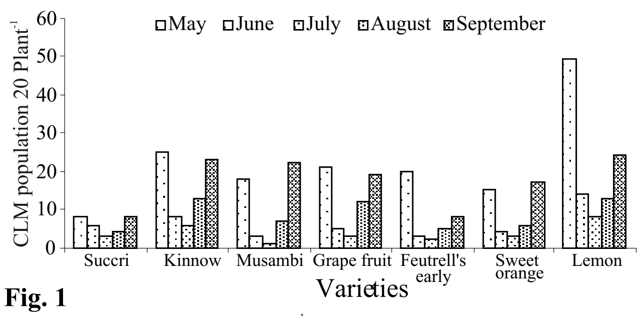

Fig. 1

Leaf miner population (20 Plants^{-1}) in different varieties in citrus nursery at Faisalabad.

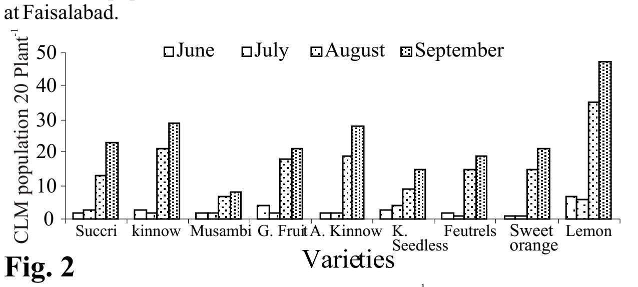

Fig. 2

Citrus Leafminer (CLM) population (20 Plants^{-1}) in different varieities in citrus nursery at Sargodha.

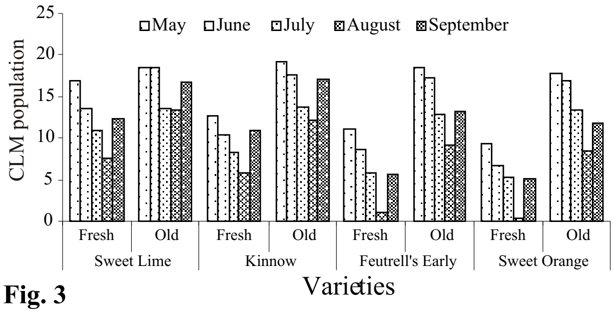

Fig. 3
Incidence of CLM in citrus groves at University of Agriculture, Faisalabad.

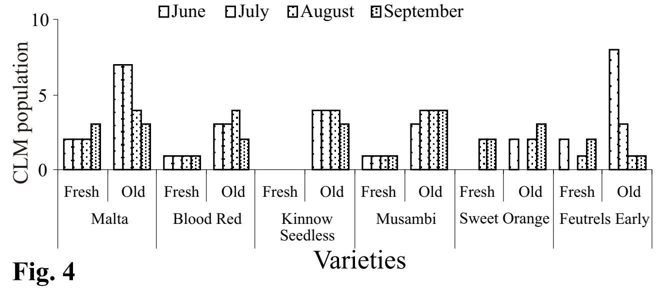

Fig. 4
Incidence of CLM in citurs groves at Sargodha.

Table 1
Correlation coefficient values of relation of abiotic factors with CLM incidence at PARS, University of Agriculture, Faisalabad.

Varieties	Max Temp	Mini. Temp	Aver. Temp.	RH	RF
Succari	0.58*	0.43*	0.50*	-0.58*	-0.33
Kinnow	0.68*	0.63*	0.66*	-0.49*	0.15
Musambi	0.63*	0.64*	0.65*	-0.36	0.47*
Grape Fruit	0.78*	0.72*	0.75*	-0.64*	0.39*
Freutrells Early	0.62*	0.39*	0.48*	-0.79*	-0.12
Sweet orange	0.72*	0.62*	0.66*	-0.65*	0.00
Lemon	0.49*	0.49*	0.50*	-0.37*	0.17

* represents pd0.05

DISCUSSION

The citrus varieties showed pronounced variation of CLM populations in nurseries, however, incidence becomes less pronounced in the groves. Goane *et al.* (2008) found no difference in leaf miner performance among host plants (lemon (*Citrus limon* L. Burm.), orange (*Citrus sinensis* L. Osbeck), and grapefruit (*Citrus paradisi* Macfadyen) in Tucumán province (northwest Argentina). They also concluded that ecological conditions might be more important than physiological adaptation in shaping a probably labile host ranking in this pest species.

The present results have revealed importance of temperature and relative humidity in bringing about change in incidence of CLM in groves which is in line with some of the earlier reported cases. The larval population of leaf-miner was higher during August-September than during the rest of the period. The minimum temperature (r = 0.58) had positive association, whereas sunshine hours (r = -0.59) and vapor-pressure deficit (r=-0.43) hadnegative associationwith leafminer population. The minimum temperature more than 18-degrees-C, vapor pressure more than 20 mm Hg and sunshine less than 6 hr favored the multiplication of leaf-miner. The individual factor showed 35% effect, whereas their combined effect by step-wise regression analysis did not exceed 9% (Patel *et al*., 1994). High population densities of *P. citrella* are usually recorded in spring and summer due to greater availability of leaf flushes and new shoots, as well as higher temperatures (Pena *et al*., 1996; Legaspi *et al*., 1999; You and Wang, 1999; Diez *et al*., 2006). You and Wang (1999) reported that the variations in hatching rates of the eggs, survival rates of the larvae and emergence rates of the pupae were great, but unimodal at different relative humidity. The highest population density of CLM occurred during the dry season. Orange and grapefruit yielded higher numbers of citrus leaf miners and their natural enemies than other citrus species (Bermudez *et al*., 2004). Minimum and medium temperature and relative humidity were the abiotic factors showing the strongest influence in the numbers of *P. citrella* mines and larvae (Jesus *et al*., 2008). Analysis of correlation between population (mixed, nymphs and adults) of citrus psylla and different abiotic factors revealed that minimum, maximum and mean temperatures, relative humidity and vapour pressure had a positive relation with both nymphal and adult population whereas wind velocity had very low impact on adults (Sharma, 2008). However, Ateyyat and Mustafa (2000) reported little role of abiotic factors in reducing incidence of CLM and mortalities were related to parasitism and predation on eureka lemon. Furthermore, populations of CLM were found to be regulated by the action of a combination of different mortality factors including parasitoids, and these, uncontestably, remained the most effective (Malika *et al*., 2010). Biotic factors take considerable toll of leafminer populations, predominance of predation suggest that parasitoid complex on this exotic pest in Florida is depauperate and would likely to improve by additional introductions (Xiao *et al*., 2007). No significant correlations were found between the proportion of mined species at a site and rainfall, latitude or foliar projected cover (Sinclair and Hughes, 2008). In a farmer's grove receiving 78 annual sprays, *P. citri* populations were higher at the end of each season than in nonsprayed trees. *Phyllocnistis citrella* severely attacked the summer flush in sprayed and nonsprayed groves equally, and no parasitism was observed (Catling *et al*., 1977).

The foregoing discussion confirms the earlier reports that environment plays a minor role in the progression of the pest, however, the effect of temperature in the buildup of the pest and its monitoring is advised with rise in temperature throughout the year.

Table 2
Regression analysis of abiotic factors on CLM population at PARS, University of Agriculture, Faisalabad.

Predictors	Regression equation	R^2
Succari		
Max Temp	Y=1.71+0.39X1	0.38
Max, Minim Temp	Y=-3.72+0.85X1-0.48X2	0.42
Max, Minim Temp Avg Temp	Y=-3.72+0.85X1-0.48X2+0.02X3	0.42
Max, Minim Temp Avg Temp RH	Y=-2.31+0.79X1-0.48X2+0.04X3-0.01X4	0.42
Kinnow		
Max Temp	Y=0.16+0.69X1	0.46
Max, Minim Temp	Y=0.50+0.66X1+0.03X2	0.46
Max, Minim Temp Avg Temp	Y=0.61+0.70X1+0.08X2-0.09X3	0.46
Max, Minim Temp Avg Temp RH	Y=-7.93+0.42X1+0.18X2+0.02X3-0.05X4	0.46
Musambi		
Max Temp	Y=-2.88+0.57X1	0.39
Max, Minim Temp	Y=0.84+0.25X1+0.33X2	0.42
Max, Minim Temp Avg Temp	Y=0.55+0.15X1+0.20X2+0.24X3	0.42
Max, Minim Temp Avg Temp RH	Y=1.058+0.13X1+0.21X2+0.24X3-0.004X4	0.42
Max, Minim Temp Avg Temp RH RF	Y=7.12+0.13X1+0.06X2+0.22X3-0.07X4+0.19X5	0.55
Grape fruit		
Max Temp	Y=-6.45+0.68X1	0.60
Max, Minim Temp	Y=-6.28+0.66X1+0.01X2	0.60
Max, Minim Temp Avg Temp	Y=-5.62+0.89X1+0.32X2-0.54X3	0.61
Max, Minim Temp Avg Temp RH	Y=18.07-0.03X1+0.63X2-0.16X3-0.17X4	0.65
Max, Minim Temp Avg Temp RH RF	Y=24.01-0.03X1+0.49X2-0.19X3-0.24X4+0.19X5	0.78
Feutrells early		
Max Temp	Y=-5.04+0.71X1	0.38
Max, Minim Temp	Y=-20.5+2.03X1-1.39X2	0.61
Max, Minim Temp Avg Temp	Y=-19.26+2.46X1-0.81X2-1.009X3	0.62
Max, Minim Temp Avg Temp RH	Y=-15.24+1.11X1-0.36X2-0.46X3-0.26X4	0.82
Sweet orange		
Max Temp	Y=-11.95+0.81X1	0.51
Max, Minim Temp	Y=-15.29+1.09X1-0.30X2	0.52
Max, Minim Temp Avg Temp	Y=-14.36+1.42X1+0.13X2-0.76X3	0.53
Max, Minim Temp Avg Temp RH	Y=15.37+0.26X1+ 0.52X2-0.29X3-0.22X4	0.56
Lemon		
Max Temp	Y=1.67+0.76X1	0.25
Max, Minim Temp	Y=-6.17+0.38X1+0.40X2	0.26
Max, Minim Temp Avg Temp	Y=7.06+0.69X1+0.82X2-0.73X3	0.26
Max, Minim Temp Avg Temp RH	Y=43.35-0.73X1+1.3X2-0.16X3-0.27X4	0.29

Max Temp (X1); Minim Temp (X2); Avg Temp (X3); RH (X4); RF (X5). Critical values for R^2 were $p<0.05$.

Table 3
Correlation coefficient values of relation of abiotic factors with CLM incidence at CRI, Sargodha.

Varieties	Max Temp	Minim. Temp	Avg. Temp.	RH	RF
Succari	0.44*	0.34*	0.38*	0.27	-0.16
Kinnow	0.47*	0.35*	0.40*	0.20	-0.14
Musambi	0.42*	0.33*	0.35*	0.22	-0.22
Grape fruit	0.63*	0.50*	0.58*	0.23	0.15
Am Kinnow	0.51*	0.38*	0.45*	0.13	-0.70
Seedless Kinnow	0.58*	0.48*	0.53*	0.19	-0.10
Feutrells early	0.48*	0.41*	0.44*	0.10	-0.20
Sweet orange	0.17	0.15	0.13	0.37*	-0.30
Lemon	0.58*	0.48*	0.52*	0.16	-0.13

*Represents $p<0.05$. Critical values for R^2 were $p<0.05$.

Table 4
Regression analysis of abiotic factors on CLM population at CRI, Sargodha.

Predictors	Regression	R^2
Succari		
Max Temp	Y=6.83+0.38X1	0.23
Max, Minim Temp	Y=21.85+1.18X1-0.94X2	0.36
Max, Minim Temp Avg Temp	Y=34.27+2.94X1-0.69X2-2.1X3	0.52
Max, Minim Temp Avg Temp RH	Y=-44.11+1.29X1+0.57X2-1.05X3+0.54X4	0.62
Kinnow		
Max Temp	Y=8.49+0.53X1	0.22
Max, Minim Temp	Y=33.21+1.85X1-1.54X2	0.35
Max, Minim Temp Avg Temp	Y=48.69+4.04X1-1.24X2-2.61X3	0.46
Max, Minim Temp Avg Temp RH	Y=-23.04+2.53X1-0.008X2-1.66X3+0.49X4	0.57
Musambi		
Max Temp	Y=3.13+0.40X1	0.17
Max, Minim Temp	Y=17.5+1.16X1-0.90X2	0.24
Max, Minim Temp Avg Temp	Y=33.13+3.38X1-0.59X2-2.63X3	0.39
Max, Minim Temp Avg Temp RH	Y=-37.79+1.88X1+0.55X2-1.69X3+0.49X4	0.55
Grape fruit		
Max Temp	Y=4.44+0.34X1	0.40
Max, Minim Temp	Y=16.65+0.98X1-0.76X2	0.54
Max, Minim Temp Avg Temp	Y=21.09+1.61X1-0.68X2-0.75X3	0.58
Max, Minim Temp Avg Temp RH	Y=-2.02+1.13X1-0.30X2-0.44X3+0.16X4	0.64
Am Kinnow		
Max Temp	Y=4.44+0.34X1	0.40
Max, Minim Temp	Y=16.65+0.98X1-0.76X2	0.54
Max, Minim Temp Avg Temp	Y=21.09+1.61X1-0.68X2-0.75X3	0.58
Max, Minim Temp Avg Temp RH	Y=-2.02+1.13X1-0.30X2-0.44X3+0.16X4	0.64
Kinnow seedless		
Max Temp	Y=1.53+0.44X1	0.34
Max, Minim Temp	Y=13.53+1.08X1-0.75X2	0.41
Max, Minim Temp Avg Temp	Y=19.68+1.95X1-0.63X2-1.04X3	0.45
Max, Minim Temp Avg Temp RH	Y=-59.53+0.28X1-0.65X2-0.001X3+0.55X4	0.77
Feutrells early		
Max Temp	Y=3.11+0.29X1	0.23
Max, Minim Temp	Y=9.63+0.63X1-0.41X2	0.26
Max, Minim Temp Avg Temp	Y=15.17+1.42X1-0.29X2-0.93X3	0.32
Max, Minim Temp Avg Temp RH	Y=-24.21+0.59X1+0.34X2-0.41X3+0.27X4	0.45
Sweet orange		
Max Temp	Y=8.13+0.12X1	0.03
Max, Minim Temp	Y=10.17+0.23X1-0.13X2	0.03
Max, Minim Temp Avg Temp	Y=17.19+1.31X1+0.002X2-1.29X3	0.10
Max, Minim Temp Avg Temp RH	Y=-67.11-0.48X1+1.39X2-0.16X3+0.59X4	0.52
Lemon		
Max Temp	Y=9.23+0.72X1	0.34
Max, Minim Temp	Y=29.14+1.78X1-1.25X2	0.41
Max, Minim Temp Avg Temp	Y=43.33+3.79X1-0.96X2-2.39X3	0.49
Max, Minim Temp AvgTemp RH	Y=-67.99+1.44X1+0.83X2-0.91X3+0.77X4	0.72

Max Temp (X1); Minim Temp (X2); Avg Temp(X3); RH (X4). Critical values for R^2 were $p<0.05$.

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