Journal of Entomology and Zoology Studies

Available online at www.entomoljournal.com

Journal of Entomology and Zoology Studies

ISSN 2320-7078
JEZS 2014; 2 (6): 213-216

Received: 24-10-2014
Accepted: 15-11-2014

Muhammad Mustafa
Department of plant pathology, university of agriculture, Faisalabad, Pakistan.

M. Imran
Directorate of Agriculture, Pest Warning & Quality Control of Pesticides, Punjab, Lahore, Pakistan.

Amer Rasool
Directorate of Agriculture, Pest Warning & Quality Control of Pesticides, Punjab, Lahore, Pakistan.

M. Azeem
Department of Soil Science, Pir Mehr Ali Shah Arid Agriculture University, Rawalpindi, 46300, Pakistan.

Adnan Riaz
Queensland Alliance for Agriculture and Food Innovation. The University of Queensland, Brisbane, QLD 4072, Australia.

M. Afzal
Directorate of Agriculture, Pest Warning & Quality Control of Pesticides, Punjab, Lahore, Pakistan.

Correspondence:
M. Imran
Directorate of Agriculture, Pest Warning & Quality Control of Pesticides, Punjab, Lahore, Pakistan.

Evaluation of commercial citrus cultivars for resistance to citrus leaf miner and its management

Muhammad Mustafa, M. Imran, Amer Rasool, M. Azeem, Adnan Riaz and M. Afzal

Abstract
To combat the problem of citrus leaf miner (CLM) which is the one of the important cause of Canker disease development, a study was conducted in research area of the Department of Plant Pathology, University of Agriculture, Faisalabad 2008-09. Fifteen commercial citrus varieties were screened to find out degree of resistance. Kinnow exhibited the resistant response against CLM attack. Blood red and Mayer developed high CLM populations. Pine apple, mungal singh, jaffa and succari were moderately susceptible to CLM attack, whereas tangerine, malta valentia late, feutral's early, china lemon, grapefruit musambi and sweet lemon showed susceptible response against CLM infestation. Plant extracts *Withania somnifera* (Aksin), *Achyranthes aspera* (Akk) and pesticide (Fon 75%WP used to manage the citrus leaf miner under field conditions on these fifteen citrus varieties. The most effective combination was Fon at .3g/l plus *Achyranthes aspera* at 7.5% against CLM infestation as 17.37 f mean value of CLM incidence as compared to control value 38.74a.

Keywords: infestation, pesticides, plant extracts, screening, susceptible.

1. Introduction
In Pakistan, Citrus has an important value as fruit plant. The present day citrus is delecz, juicy, and seedless and of great nutritional significance as well [10]. Citrus is a member of Rutaceae family and grown in varying densities in countries with tropical or subtropical climates. The most delicious and edible varieties of citrus are Kinnow, Pine apple, Valencia late, Grape fruit, Blood red, Chinese lime, Mayer lime, Sweet lime, Fuetrell's early, Jaffa, Succari, Tangrin, Mungal Singh, and Musambi are included. It is used as best source of Vitamin C, sugars, amino acids and other nutrients [2]. Pakistan has versatile climatic conditions and nearly all types of fruits might be produced successfully. A large area of Pakistan has sub- tropical and tropical climate.

Citrus is one of the most important productive and highly profitable fruit crop but unfortunately its present status is threatened by a number of problems including low production induced by pests. Of all the agricultural pests and diseases that threaten citrus crop, citrus leaf miner (CLM), *Phyllocnistis citrella* is one of the major pests which adversely affects plant health and fruit development and enhances the development of canker disease. The CLM is a pest of citrus, which has undergone a very rapid range expansion during the last ten years [12]. The intensity of the citrus canker disease increased with that of injuries caused by CLM. The symptoms of the citrus canker disease appeared more rapidly, the ratio of plant disease and the incidence were higher, and plants remained susceptible for long period. These factors explained the association between the intensity of the disease symptoms and injuries caused by the insect [1]. Exposure of leaf mesophyll during feeding allowed direct penetration of Xac into leaf tissue. Also CLM larvae became contaminated with bacteria, disseminating them through feeding galleries [8, 5].

The feeding activities of CLM that enhance the bacterial infection. These are tearing of the cuticle opened the mesophyll of the leaf to direct bacterial infection. When splash-dispersed or windblown rain dispersed bacteria came in contact with the leaf surface. CLM wounds healed more slowly than mechanical wounds, allowing a longer periods of exposure for bacterial infection; and CLM larvae were contaminated with bacteria and transported them through feeding galleries [1].

Keeping in view the great economic importance of citrus canker disease and its vector CLM, citrus cultivars are needed to be screened under natural field conditions against CLM.

2. Materials and Methods

2.1 Establishment of disease screening nursery

To evaluate citrus varieties/lines for the relative resistance to citrus leaf miner, a screening nursery was established in the Plant Pathology Research Area of the University of Agriculture, Faisalabad. Varieties encountered for screening were Kinnow(v_1), Pine apple(v_2), Valencia late(v_3), Grape fruit(v_4), Blood red(v_5), Chinese lime(v_6), Mayer lime(v_7), Sweet lime(v_8), Fuetrell's early(v_9), Jaffa (v_{10}), Succari (v_{11}), Tangrin(v_{12}), Mungal Singh (v_{13}), Musambi (v_{14}) and Malta(v_{15}). These varieties were obtained from the fruit plant nursery of Horticulture Department, the University of Agriculture, Faisalabad. The experiment was laid out under Randomized Complete Block Design (RCBD). Each variety consisted of twelve plants, which were planted in two rows having six plants in each row. The recommended agronomic practices were followed to maintain citrus nursery in good condition.

2.2 Varietal screening against citrus leaf miner

Citrus leaf miner population data were collected from the disease screening field on randomly selected plants of each variety used for disease rating. The insect populations from upper, middle and lower 100 leaves per the plants were estimated and the average was calculated on weekly basis by using the Luckshman scale (1998) for resistance/susceptibility of citrus varieties (Table 1). Data regarding insect populations were recorded and subjected to analysis of variance. Difference in CLM populations on fifteen cultivars was assessed using the methodology given [1].

Table 1: Citrus leaf miner rating scale used to evaluate the citrus germplasm

Grade	CLM Severity%	Response
0	01-05	Immune
1	01-09	Resistant
3	10-15	Moderately Resistant
5	16-26	Moderately Susceptible
7	21-30	Susceptible
9	31 and above	Highly Susceptible

2.3 Management of citrus leaf miner under field conditions

Plant extracts *Withania somnifera* (Aksin), *Achyranthes aspera* (Akk) and pesticide (Fon 75%WP were used to manage the citrus leaf miner under field conditions on these fifteen citrus varieties. Sterilized water was used as a control. For this purpose two experiments were conducted under the field conditions to control citrus canker disease and CLM infestation. There were three replications of each treatment including control. Data regarding insect populations were recorded before and after application of treatments and subjected to analysis of variance. The treatments were applied according to following plan;

2.4 Experiment

T_1 = Fon .3g/l

T_2 = *Withania somnifera* (15%

T_3 = Fon .3g/l + *Withania somnifera* (7.5%)

T_4 = *Achyranthes aspera* (15%)

T_5 = Fon .3g/l+ *Achyranthes aspera* (7.5%)

T_6 = Control

3. Results

3.1 Response different citrus cultivars/varieties against citrus leaf miner attack

Citrus leaf miner was also active throughout the year and multiplied on young growth of citrus plants. CLM infected young citrus plants including leaves and young stems were studies. The larvae caused damage by making zigzag shaped mines in the young leaves between the upper and lower epidermal layers, eating the parenchymatic tissue. The injured epidermis took the shape of twisted silvery galleries. On the older leaves, brownish patches formed, which served as foci of infection for citrus canker. Heavily attacked plants could be observed from a distance and young nurseries were most severely affected.

Maximum CLM populations was found on *C. sinensis* cv. blood red and *C. limonia* cv. mayer lemon, showing infestation severities as 30.15 and 31.82 (Means) respectively (Table 2). The minimum CLM population occurred on *C. reticulata* cv. kinnow with severity of 10.49 (Means).

Different citrus varieties expressed varying response towards CLM population. Varieties such as *C. sinensis* cv. pine apple, *C. reticulata* cv. mungal singh and *C. sinensis* cv. succari were moderately susceptible to CLM attack, whereas *C. reticulata* cv. tangerine, *C. reticulata* cv. malta, *C. sinensis* cv. valentia late, *C. reticulata* cv. feutral's early, *C. limonia* cv. china lemon, *C. paradise* cv. grapefruit, *C.. sinensis* cv. succari and *C. limettioides* sweet lemon showed susceptible response against CLM infestation.

Maximum population was found on *C. sinensis* cv. blood red and *C. limonia* cv. mayer lemon and were found to be highly susceptible varieties, In contrast, moderate resistance was observed in *C. reticulata* cv. Kinnow (Table 2).

Table 2: Level of resistance/susceptibility to citrus leaf miner population exhibited by various citrus varieties.

Sr.#	Varieties/cultivars	Population incidence (Mean)	Response
1	Kinnow	10.94 j*	MR
2	Pine apple	17.44 i	MS
3	Jaffa	17.71 hi	MS
4	Mungal singh	17.87 hi	MS
5	Succari	18.13 h	MS
6	Tangerine	20.33 g	S
7	Malta	20.77 f	S
8	Valentia late	20.91 f	S
9	Feutral's early	26.13 e	S
10	China lemon	26.60 d	S
11	Grapefruit	26.89 d	S
12	Musambi	27.49 c	S
13	Sweet lemon	29.83 b	S
14	Blood red	30.15 b	HS
15	Mayer lemon	31.82 a	HS

LSD = 0.42 *Means sharing similar letters do not differ significantly (P>0.05). MR = Moderately resistant
MS = Moderately susceptible
S = Susceptible
HS = Highly susceptibl

3.2 Management of citrus leaf miner under field conditions through pesticide and plat extracts

The treatments Fon at .3g/l, *Withania somnifera* at 15%, *Achyranthes aspera* 15%, Fon at .3g/l plus *Withania somnifera* at 7.5% and Fon at. 3g/l plus *Achyranthes aspera* at 7.5% concentration reduced the infestation of CLM compared to control under field conditions (Table 3). The combination of Fon at .3g/l plus *Achyranthes aspera* at 7.5% and Fon at .3g/l plus *Withania somnifera* at 7.5% concentration controlled CLM population significantly compared to control respectively. Fon and plant extracts applied singly against CLM infestation did not show proper results as compared to the combined tested treatment (Table 3 & 4).

Table 3: ANOVA for evaluation of Fon (75%WP) and plant extracts to control the citrus leaf miner under field conditions.

SOV	DF	SS	MSS	F-value
Replication	2	6.513	3.256	1.10
Treatments (T)	5	2453.653	490.731	165.61**
Days (D)	2	736.045	368.022	124.20**
Interaction	10	124.929	12.493	4.21**
(T x D)				
Error	34	100.747	2.963	
Total	53	3421.886		

** = Highly significant (P<0.01)

Table 4: Effect of Fon (75%WP) and plant extracts to control the citrus leaf miner under field conditions.

Sr. #	Treatments	Mean values of citrus leaf miner incidence
T_1	Fon .3g/l	27.36 b
T_2	*Withania somnifera* (15%)	25.69 c
T_3	Fon .3g/l + *Withania somnifera* (7.5%)	20.70 e
T_4	*Achyranthes aspera* (15%)	23.37 d
T_5	Fon .3g/l + *Achyranthes aspera* (7.5%)	17.37 f
T_6	Control	38.74 a
	LSD	1.642

Means sharing similar letter are statistically non-significant (P>0.05).

4. Discussion

Xiao *et al*. (2007) tested 349 citrus rootstocks under either field or laboratory conditions for resistance to the CLM. Field results confirmed the widespread susceptibility of citrus to the leaf miner. Only 0.9% of the trees checked during one growing season escaped damage by Rushing once early in the season. Atiq (2008) tested the different citrus varieties against CLM attack and found that Kinnow, Jaffa attracted the low numbers of CLM while *C paradise*, China lemon, Musambi, *C limettioides*, Blood red, and Mayer lemon developed high CLM population. Atiq *et al*., 2007) screened fifteen citrus cultivars for the source of resistance against CLM. *C. sinensis* cv. blood red and *C. limonia* cv. mayer lemon exhibited high susceptibility while *C. reticulata* cv. kinnow showed moderately resistant response towards the CLM.

Withania somnifera, *Achyranthes aspera* and the pesticide Fon were also tested against CLM. Fon at .3g/l alone or in combination with plant extract reduced the CLM population significantly. The most effective combination was Fon at .3g/l plus *Achyranthes aspera* at 7.5% against CLM infestation. These results agreeing with Jothi *et al.*, 1993) who have tested Neem and Pongamia seed extract (2%) against CLM in Karanataka, during 1989 and also found that neem seed extract was an effective treatment. A field trial was conducted in Nagpur Maharashtra, India to test the efficacy of some botanicals and synthetic insecticides against infestation of CLM on mandarin during June and September 1990 and it was found that neem oil was effective in the control of CLM [9]. The present studies gave a new approach for the management of citrus canker disease. Extracts of plants especially *Withania somnifera* provide, an opportunity for future biochemical work for the isolation, purification and concentration of active antibacterial compounds. Selection of suitable formulations and methods of application could be future aspects of applied research on plant products. Jayanthi and Verghese (2004) [6] conducted an investigation in an acid lime orchard during 1999, to compare the efficacy of new insecticides with commonly used insecticides along with neem-based formulations, *against P. citrella*. Neem formulations, viz. neem seed kernel extract and Azadirachtin, were the most effective in causing high mortality of CLM larvae.

Plant extracts alone or in combination with Fon reduced CLM populations. Similar results were obtained by YiJing *et al.*, 2003) [14], who tested the deterrent effects of alcoholic extracts from 26 non-host species and a horticultural mineral oil against the CLM. Extracts from *Mikania micrantha*, *Ageratum conyzoides* and *Dicranopteris pedata* exhibited good (greater than 90%) oviposition deterrent effects.

5. References

1. Achor DS, Browning HW, Albrigo LG. Anatomical and histological modification in citrus leaves caused by larval feeding of citrus leaf miner (*Phyllocnistis citrella* Staint). In: Proceedings of the international conference on citrus leaf miner, Orlando, Florida (Hoy, M.A., ed.), Gainesville, FL: University of Florida, 1996, 69.
2. Amad R, Khan HH. Citrus decline problems in the Punjab: A review Pp 20-22. In: 2nd Nat. Conf. PL. Path. Univ. Faisalabad. Pakistan, 1999.
3. Atiq M. Prediction of citrus canker disease and its management. Ph.D. Thesis. Uni. Agri. Faisalabad, 2008, 116-119.
4. Atiq M, Khan MA, Sahi ST. Screening off citrus germplasm for the source of resistance against canker disease caused by *Xanthomonas axonopodis* pv *citri.* Pak J Phytopathol 2007; 19(2):222-226.
5. Christiano RSC, Dalla PM, Jesus JWC, Parra JRP, Amorim L, Bergamin FA. Effect of the citrus leaf-miner damage, mechanical damage and inoculum concentration on severity of symptoms of Asiatic citrus canker in Tahiti lime. Crop Prot 2007; 26:59–65.
6. Jayanthi PDK, Verghese A. Efficacy of new insecticides, and neem formulations in the management of the citrus leaf miner, *Phyllocnistis citrella* Stainton (Phyllocnistidae: Lepidoptera). Div Entom & Nemat Ind Ins Hort Res Bangalore India Ento 2004; 29(1):45-50.
7. Jothi BD, Tandon PL, Verghese A. Evaluation of different plants oils and extract against citrus leaf miner, *Phyllocnistis citrella* (Lepidoptera, Phyllocnistis). Indian Soc Tobacco Science 340-342 (Rev. Agric. Entomol 1993; 81(8):8069.
8. Junior JWC, Junior BL, Amorim L, Christiano RSC, Parra JRP, Filho BA. Injuries caused by citrus leaf miner (*Phyllocnistis citrella*) exacerbate citrus canker (*Xanthomonas axonopodis* pv. *citri*) infection. Fitopatologia Brasileira 2006; 1:277-28.
9. Katole SR, Thakare HS, Mahajan RK. Effect of some plant product and insecticides on the infestation of citrus leaf miner on Nagpur mandarin. J Maharachtra Agric Univ., 18(1): 67-68 (Rev Agric Entomol 1993;

82(8):7368.
10. Khan MA, Khan MM, Haq MI, Javed N. Antibacterial activity of various toxicants against *Xanthomonas campestris* pv. *citri* for the control of citrus canker disease: Proc. 1st Int. Sem. Citriculture in Pakistan, Dec. 2-5, 1992, 311-314.
11. Steel RGD, Torrie JH, Dickey DA. Principles and Procedures of Statistics. A Biometrical Approach. 3rd edit. Mc Graw Hill Book Co., New York, 1997.
12. Urbaneja A, Lacer E, Garrido A, Jacas JA. Interspecific competition between two ectoparasitoids of *Phyllocnistis citrella* (Lepidoptera: Gracillariidae). Cirrospilus brevis and the exotic *Quadrastichus* Sp Biol Con 2003; 28:243-250.
13. Xiao Y, Qureshi JA, Stansly PA. Contribution of predation and parasitism to mortality of citrus leaf miner *Phyllocnistis citrella* Stainton (Lepidoptera: Gracillariidae) Populations in Florida. Bio Con 2007; 40:396-404.
14. Yi JC, Xiong FP, Mao XZ, Qiao SD, Yu YD, Biao W. Oviposition repellent of alcohol extracts of 26 non-preferable plant species against citrus leaf miner. J South China Agric Uni 2003; 24(3):27-29.