

Interactions Between Leafminer Damage and Leaf Necrosis Caused by *Alternaria alternata* on Potato in the Sultanate of Oman

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Four field experiments were carried out from 1998 to 1999 and from 1999 to 2000 growing seasons of potato to investigate the relationship between leafminers and *Alternaria alternata*. The experiments established differential susceptibility among potato varieties to *alternaria* leaf necrosis, relationship between leafminer infestation and the level of necrosis, use of insecticides to reduce leafminer and leaf necrosis damages, and use of polyester fleece to eliminate leafminer and eventually reduce leaf necrosis. Results of the study indicate that control of leafminer is of primary importance because this will automatically lead to control of the damage caused by *A. alternata*.

Keywords : *Alternaria alternata*, chemical control, IPM, *Lyriomyza trifolii*, potato.

Potato is a relatively new crop in Oman. However, the cultivation area and total yield of this crop have gradually increased since 1980, making it the third most important vegetable crop after tomato and onion in the Sultanate. Expansion of potato production began in the early 1980s following the government's support under the seed distribution program. Despite the withdrawal of these subsidies in the early 1990s, potato production has continued to increase due to the interest generated by large commercial farms now yielding up to 35-45 t/ha. The total area devoted to potato production in Oman has increased from approximately 50 ha in 1977 to over 260 ha in 1999, and is expected to continue to expand owing to increasing consumer's demand. The crop is now considered to be one of the nationally strategic important crops by Oman's Ministry of Agriculture and Fisheries.

At present, pests and diseases are among the biggest constraints to the expansion of potato production in Oman. Important pests include the peach potato aphid (*Myzus persicae* Sulzer), whitefly (*Bemisia tabaci* Gennadius),

jassid (*Jacoblasca lybica* Berg. & Zanon), potato tuber moth (*Phthorimea operculella* Zeller), and leafminer (*Lyriomyza trifolii* Burgess). Meanwhile, important diseases of potato in the Sultanate include early blight (*Alternaria solani* [Ell. Mart.] L. R. Jones & Groul), leaf spot (*Stemphylium solani* G. F. Weber and *Alternaria alternata* [Fr.: Fr.] Keissl.), blackleg (*Erwinia carotovora* [Jones] Bergey *et al.*), and a number of different potato viruses that are vectored primarily by whitefly and aphids (Anonymous, 1991; Moghal, 1993).

To control these pests and diseases, current recommendations involve the application of insecticides and fungicides. Products that are currently recommended for pest control include butocarboxim, ethiofencarb and primicarb (systemic carbamates), deltamethrin, alphacypermethrin and fenvalerate (contact pyrethroids), phosphamidon (systemic OP), and phenthoate (contact OP). Products that are recommended for pathogen control include cymoxanil, mancozeb, and copper oxychloride (all protectants). The actual choice of chemical used depends on the price, availability, and crop protection knowledge. Whatever the choice is, chemical is usually applied at high volume using hydraulic application equipment. It is not unusual for farmers to combine an insecticide and a fungicide when spraying, and plants are typically sprayed until run-off (Thacker *et al.*, 2000). There are no action thresholds for pest and disease control, and large commercial farms frequently apply pesticides prophylactically.

The choice of potato variety is largely determined by the availability of imported seeds, usually from Western Europe. The time of availability of exotic seed also determines the planting date of the crop. Most potato crops in Oman are planted in mid-November. Preliminary research has indicated differential sensitivity of potato varieties to leaf spot damage (Khan *et al.*, 2000).

The objective of this study was to examine the significance of a possible interaction between two of the most common pest and disease problems of potato, namely; *L. trifolii* and *A. alternata*. Both the pest and the pathogen frequently cause severe leaf damage under Omani condi-

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tions. Consistent field observations have indicated that these two organisms may interact and enhance their damaging potential on the potato crop. The overall goal of this research was to come up with an integrated management strategy for these two biotic constraints of potato.

Materials and Methods

Four field experiments were carried out for 2 years from 1998 to 1999 and from 1999 to 2000 growing seasons. Evaluations were made of leaf necrosis damage (experiments 1-4) and leafminer damage (experiments 2-4). The assessment procedure for the leafminer and leaf necrosis was the same for all four experiments. Ten plants in the center of a treated plot were sampled from each plot. Leafminer incidence was assessed as the percentage of the leaflets mined on a plant. Disease severity was assessed on a scale of 0-10 (0 = no necrosis, 10 = totally necrotic leaf). The presence of the pathogen was confirmed by culturing field-collected samples on PDA in the laboratory. A summary of the four experiments, treatments, and potato varieties used are presented in Table 1.

Experiment 1. The first experiment was designed to evaluate the presence of differential levels of sensitivity of potato varieties to leaf necrosis caused by *A. alternata*. Twelve rows of potato were established at 60 m long per row, with 1 m distance from adjacent rows. Each row was divided into two and planted with different varieties. Five varieties of potatoes were evaluated, each having four replicate blocks in a randomized block design. Fertilizer applications were made according to minimum local requirements. No pesticide applications were made to the crop, and all weeding procedures were carried out by hand. Natural infection and progression of leaf necrosis symptoms were allowed to develop. Plants were assessed for leaf necrosis damage at weekly intervals between 42 and 77 days after planting.

Experiment 2. Twenty-four rows of potato were divided into six replicate blocks. Each row was 60 m long and was divided into three 20-m sections. Row spacing was 1 m. Within each block, 12 potato varieties were sown, one variety per 20-m section. Varieties

were randomly allocated to sections within a block. Assessments of leafminer and leaf necrosis were carried out at weekly intervals from emergence to crop senescence.

Experiment 3. Sixty potato rows, 60 m long with 1-m spacing, were alternately sown with two varieties. The field was then divided into 12 plots measuring 20 m × 15 m. Each plot was allocated one of the following treatments: no spray, one spray, two sprays. Treatments were allocated randomly. Assessments of leafminer and pathogen levels were carried out weekly from the time of the first spray until crop senescence. Pesticides were sprayed 1 and 2 months after plant emergence. The insecticide lambda-cyhalothrin was applied using a knapsack sprayer fitted with a hollow cone nozzle at a volume application rate of 200 l ha⁻¹.

Experiment 4. Twelve potato rows, 60 m long with 1-m spacing, were sown with a single variety. The plot was then divided into four replicate blocks. Within each block, the three rows were randomly allocated one of the following treatments: no cover, with cover for 34 days, with cover for 41 days. Rows were covered with agryl polyester fleece. Assessments of leafminer and necrosis damage levels were carried out weekly from emergence or from the time of uncovering to crop senescence.

Data analysis. For all experiments, the data were analyzed using correlation and regression techniques. Disease severity was plotted against the percentage of leaves that have been attacked by leafminers. Leafminer and disease severity progress curves over time were also constructed. For the varietal trials, comparisons were made both within and between varieties. The slope and the intercept associated with regression models for all data were then analyzed using analysis of variance. Where necessary, and where mentioned, sigmoidal curves were linearized using the method of Nutter and Parker (1997) prior to analysis of variance on model parameters. Areas under disease progress curves were calculated using the midpoint method of Campbell and Madden (1990).

Results

Experiment 1. The disease progress curves for each of the potato varieties are shown in Fig. 1. There were clear differences between the five varieties. While the overall

Table 1. Potato varieties and treatments used to evaluate the interaction between *Lyriomyza trifolii* and *Alternaria alternata*

Potato varieties	Planting date (Day/Mon/Yr)	Treatment
Experiment 1 Diamant, Estima, Turbo, Spunta, Arinda	16/09/98	Susceptibility to natural infection by <i>A. alternata</i>
Experiment 2 Estima, Fabula, Spunta, Kirrie, Ajiba, Donald, Lady Rosetta, Sterling, Lady Claire, Bydand, Diamant, Mondial	06/09/99	Susceptibility to natural infection by leafminer and <i>A. alternata</i>
Experiment 3 Diamant, Lady Rosetta	27/09/99	Integrated control of leafminer and leaf necrosis by chemical insecticide
Experiment 4 Diamant	15/12/99	Integrated control of leafminer and leaf necrosis through exclusion of leafminer

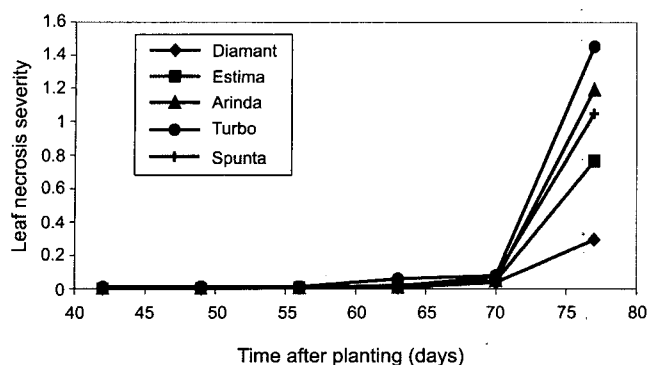


Fig. 1. Disease progress curves for five potato varieties naturally infected by *Alternaria alternata* in Oman.

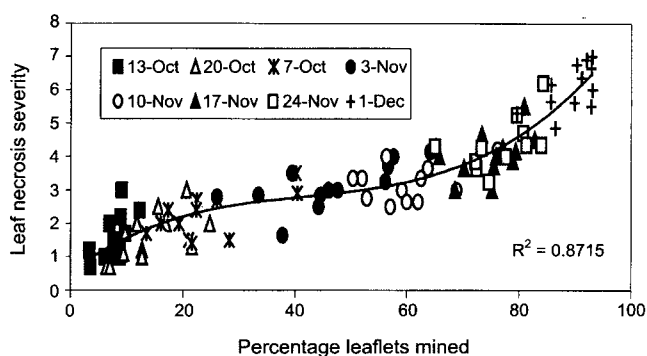


Fig. 2. Regression analysis of leaf necrosis severity against percent leafminer presence. Data are for all potato varieties and all sampling dates.

levels of leaf necrosis were generally low, the level of necrosis in cv. Diamant remained below 0.5 until the final assessment on day 77. For cv. Turbo, the level of leaf necrosis increased rapidly between day 70 and 77, and at the final assessment day the necrosis score was the highest as 1.5.

Experiment 2. During the second experiment, there appeared to be a clear step-wise progression in the extent of leafminer damage and leaf necrosis severity. Fig. 2 shows the leaf necrosis severity plotted against leafminer presence over time for each of the 12 potato varieties. The data were analyzed with a polynomial regression model. The result was significant based on the fit of the model ($y = 0.00002x^3 - 0.0026x^2 + 0.1282x + 0.5273$) to the data set ($R^2 = 0.8715$, $P < 0.001$). The model indicates that necrosis caused by *A. alternata* was highly dependent on leafminer damage. The pattern of damage across all varieties suggests that there may have been two phases of rapid necrosis development. The second phase followed a period of rapid increase in leafminer infestation.

In the second experiment (Table 2), the potato varieties showed clear differences in the extent of potato leaf miner

Table 2. Potato variety response to leafminer and leaf necrosis damages, expressed as the area under the severity progress curve

Potato variety	Leafminer			Leaf necrosis		
	Intercept	Rate	AUDPC ^a	Intercept	Rate	AUDPC
Estima	-6.28	0.100	2384.6	-3.78	0.046	142.3
Fabula	-5.78	0.086	2126.3	-4.26	0.053	148.1
Spunta	-5.08	0.082	2500.8	-2.26	0.027	166.1
Kirrie	-4.91	0.086	2825.4	-2.53	0.035	196.5
Ajiba	-6.31	0.106	2702.0	-3.21	0.047	205.1
Donald	-6.51	0.102	2220.1	-3.71	0.042	125.0
Lady Rosetta	-7.01	0.105	2044.4	-4.47	0.052	124.4
Sterling	-5.57	0.087	2315.6	-2.62	0.029	148.4
Lady Claire	-5.27	0.090	2771.0	-2.82	0.035	177.5
Bydand	-7.08	0.108	2148.3	-3.99	0.051	155.4
Diamant	-6.79	0.109	2378.8	-4.58	0.058	144.0
Mondial	-5.35	0.088	2536.1	-3.64	0.044	141.2

^a Area under disease progress curve.

damage. The time of initiation of leafminer damage was different among the 12 varieties, as indicated by the intercept values of the linear leafminer progress curves. Similarly, the intercept values from the linear necrosis severity progress curves were different among the same varieties. Likewise, the rate of increase in leafminer damage and leaf necrosis severity was clearly different among the varieties. These differences are reflected in the overall measure of damage, expressed as the area under the (non-linearised) leafminer damage progress curve. Similarly, there were clear differences in the extent of leaf necrosis damage, as expressed by the area under the disease progress curve.

There was a significant relationship between the areas under the leafminer and the leaf necrosis severity curves ($y = 0.08x - 36.67$, $R^2 = 0.653$, $P = 0.001$, Fig. 3). Similarly, there was a significant relationship between the leafminer intercept and the leaf necrosis intercept ($y = 0.81x + 1.39$, $R^2 = 0.627$, $P = 0.002$), as well as, between the leafminer rate of increase and the rate of increase in leaf necrosis severity ($y = 0.703 - 0.02x$, $R^2 = 0.515$, $P = 0.009$). This indicates that varieties showing early damage due to leafminer activity are under increased risk of early infection by *A. alternata*. Meanwhile, varieties that are not selected at an early stage for oviposition by female leafminers are subject to delayed infection by *A. alternata*. A limited evaluation was carried out on the density of trichomes on the leaf surface of a selection of potato varieties used in experiment 2. The results show clear differences in trichome density among varieties. Results also show that the levels of leafminer damage (expressed as the area under the leafminer damage progress curve) are related to trichome density (Fig. 4).

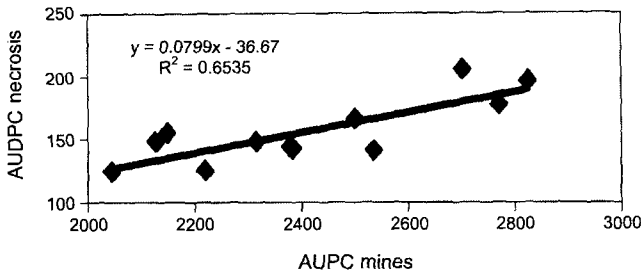


Fig. 3. Relationship between leaf miner damage and leaf necrosis damage on 12 potato varieties.

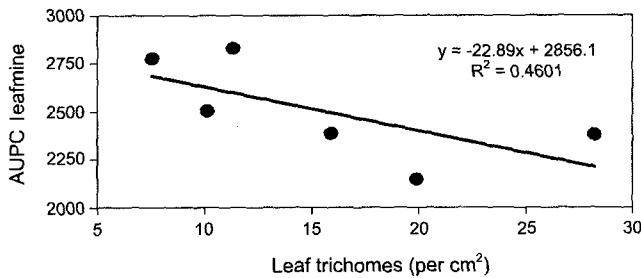


Fig. 4. Relationship between trichome density on the leaves of different potato varieties and leafminer damage.

Experiment 3. The mean percent of leaflets with mines and the mean leaf necrosis severity scores for cvs. Diamant and Lady Rosetta, following treatment with insecticide, are shown in Table 3. In the case of cv. Diamant with no insecticide sprays, there was an increase in percent leaflets mined between the time of the first spray (76.5%) and the time of the final assessment (91.5%). For the same variety receiving one spray, the level of leafminer damage remained constant (approximately 85%). When two sprays were applied to the same cultivar, the overall level of damage declined from 91.2% at the time of the first spray to 72.9% at the final assessment. This was presumably due to

Table 3. Effect of insecticide sprays on the incidence of potato leafminer and the associated severity of leaf necrosis caused by *Alternaria alternata*

Variety	Sprays	0 day after 2 nd treatment		35 days after 2 nd treatment	
		Mines	Necrosis	Mines	Necrosis
Diamant	0	76.5 ^a	2.8 ^b	91.5	5.9
	1	85.5	3.3	85.4	4.7
	2	91.2	4.5	78.4	3.8
Lady Rosetta	0	77.3	2.8	92.2	6.1
	1	82.2	2.4	91.0	5.5
	2	83.2	2.9	72.9	2.7

^aPercentage of leaflets mined by the leafminer.

^bDisease severity was assessed on a scale of 0-10 (0 = no necrosis, 10 = totally necrotic leaf).

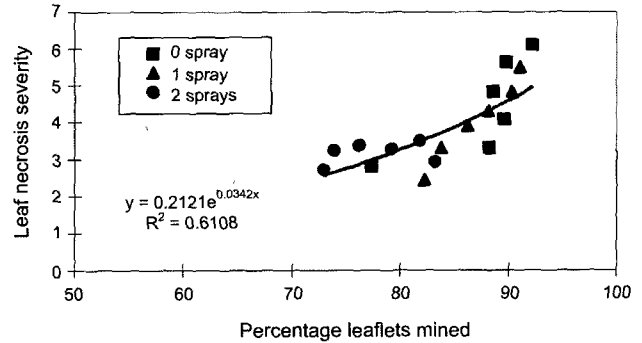


Fig. 5. Regression analysis of leaf necrosis against percent leafminer presence. Data are for all treatments. Sequential symbols within a treatment represent sequential sampling occasions.

the protection given by the insecticide to emerging, undamaged foliage.

A similar pattern was observed for cv. Lady Rosetta. In the untreated plots, the average level of damage increased from 77.3% to 92.2%. Where one spray was applied, there was an increase in the level of damage from 82.2% to 91.0%. When two sprays were applied, the level of damage decreased from 83.2% to 72.9%.

The leafminer damage dynamics are reflected in the severities of leaf necrosis assessed on each of the plots. For both varieties, when no insecticide was applied, there was a large increase in necrosis severity between the time of the first spray and the time of the final assessment (2.8-5.9 for cv. Diamant; 2.8-6.1 for cv. Lady Rosetta). When a single spray was applied to the crops, there was a smaller increase in necrosis severity (3.3-4.7 for cv. Diamant; 2.4-5.5 for cv. Lady Rosetta). With two insecticide sprays, there was a net decrease in the severity of leaf necrosis (4.5-3.8 for cv. Diamant; 2.9-2.7 for cv. Lady Rosetta).

Fig. 5 shows the relationship between leafminer damage and leaf necrosis severity for all crops. The relationship is similar to that observed in experiment 2. It clearly shows the effectiveness of the insecticide treatment in reducing the level of leafminer damage and the associated reduction in

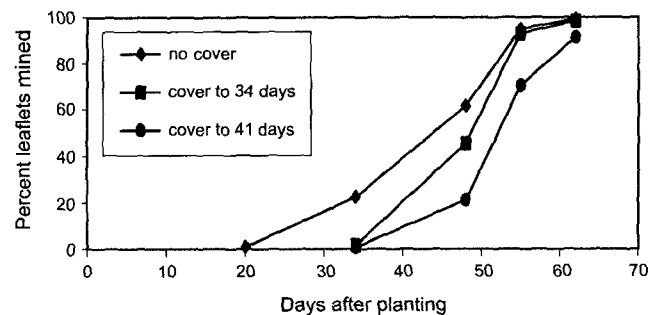


Fig. 6. Progress curves for leafminer damage on potato cv. Diamant following different insect exclusion times using polyester fleece.

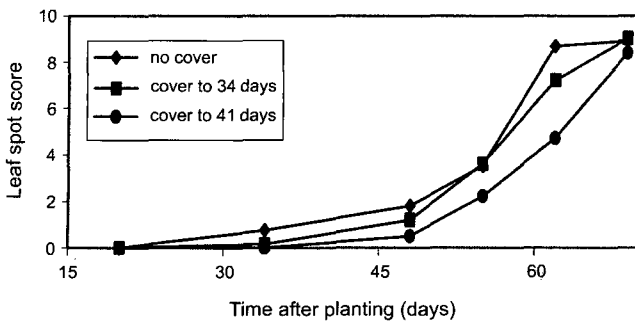


Fig. 7. Leaf necrosis progress curves for potato cv. Diamant on crops from which leafminers have been excluded for different periods of time.

Table 4. Intercept and rate values of leafminer damage and leaf necrosis severity on uncovered and covered potato cv. Diamant with acryl fleece

Treatment	Leafminer		Leaf necrosis	
	Intercept	Rate	Intercept	Rate
Uncovered	-9.21	0.22	-8.30	0.15
Covered for 34 days	-14.46	0.32	-10.74	0.19
Covered for 41 days	-15.04	0.29	-12.16	0.20
LSD (P<0.05)	1.367	0.027	1.197	0.027

the severity of leaf necrosis.

Experiment 4. Figs. 6 and 7 show the progress of leafminer damage and *A. alternata* leaf necrosis on cv. Diamant. Although the acryl cover protected the young foliage from attack by the leafminer, there was a rapid increase in the level of damage on protected crops as soon as the cover was removed. From the results of ANOVA of the linearised progress curves, there was a strong indication that although the polyester cover delayed the initiation of insect damage and leaf necrosis, as soon as the cover was removed the rate of increase of insect damage and necrosis was higher and faster than that in the uncovered ones (Table 4). By day 63, there was no significant difference in the amount of leafminer damage among the three treatments.

Discussion

The data presented indicate a clear relationship between damage to potato crops caused by *Lyriomyza trifolii* and that caused by *Alternaria alternata* in Oman. This is the first report showing this kind of damage association on potato in the Sultanate. Furthermore, this report shows the relationship between weak pathogens, such as *A. alternata*, and damage caused by leafminers and other insects. The puncture holes made by ovipositing female leafminers appear to be creating foci for attack by pathogens on the plants.

Chandler and Thomas (1991) showed a similar relationship between leafminer activity and the incidence of *Alternaria* leaf blight lesions on muskmelon leaves. They also showed a linear relationship between the number of leafminers released under controlled conditions, and the number of lesions developed from artificial inoculations of *Alternaria* spores. There was a similar relationship between the duration of exposure of muskmelon leaves to leafminers and the number of *Alternaria* lesions. Photographic evidence was presented to show the entry of *Alternaria* germ tubes into leaf tissue via the puncture made by the leafminer. Overall, Chandler and Thomas (1991) showed a linear relationship between the number of leafminer punctures per leaf and the number of *Alternaria* lesions per leaf. In their research, they used *A. cucumerina*, a species recognized as an important pathogen of muskmelon in many parts of the world.

In this study, however, *A. alternata*, a weak pathogen, was used. It usually attacks only plant tissue that is already damaged or under some other stresses. For example, *A. alternata* has been observed as a colonizer of necrotic tissue produced by *Urocladium* in cucurbits (Zitter et al., 1996). Chandler and Thomas (1991) noted the strong relationship between the duration of exposure to leafminer and the severity of infection caused by *A. cucumerina*. Results of this study show that there was a clear relationship between the onset of leafminer damage and the severity of leaf necrosis (as measured by the AUDPC) caused by *A. alternata*.

This paper also covered the results of separate evaluations for the integrated management of both an insect pest and a fungal pathogen. The data indicate that the use of polyester fleece may not be an effective strategy. Apart from the cost and logistical implications for the potato producer, the results show that a newly uncovered crop is rapidly attacked by the insect. It appeared that newly damaged hole caused by the insect partly become infection foci for the fungal attack. In the trials reported here, the extent of damage on the newly uncovered crop was more severe, consequently becoming heavily infested by leafminers, than those in adjacent rows which have not been covered. Attack by the pathogen is, therefore, a non-random event but is mediated by damage to the foliage, such as that caused by leafminers in these experiments.

The experiments reported here used different planting dates, therefore, it is possible that some of the differences among trials were due to planting date effects. However, there is no significant trend of increasing or decreasing damage due to leafminer or *A. alternata* as a consequence of planting date. The insecticide tested in this work appeared to partially control the leafminer, as well as, the severity of necrosis caused by *A. alternata*. Further research

is required on chemical control to evaluate the optimum timing of application for leafminer control. Data are also needed on the most effective chemicals for use in an integrated pest and disease management program. What is clear at present, however, is that control of the leafminer may lead to control of leaf necrosis caused by the pathogen *A. alternata*.

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