Zoosymposia 6: 104–110 (2011)
www.mapress.com/zoosymposia/

ISSN 1178-9905 (print edition)
ZOOSYMPOSIA
ISSN 1178-9913 (online edition)

Technical and economic evaluation of different types of control of *Brevipalpus phoenicis* (Acari: Tenuipalpidae) in citrus leprosies management*

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* *In*: Moraes, G.J. de & Proctor, H. (eds) Acarology XIII: Proceedings of the International Congress. Zoosymposia, 6, 1–304.

Abstract

The objective of this work was to evaluate the effect of acaricide applications and pruning of symptomatic branches in citrus leprosis management in Brazil. It was conducted in an orange plantation of the ‘Pera’ variety, grafted onto the ‘Cleopatra’ tangerine, in two seasons (2006–2007 and 2007–2008). The experimental design was randomized blocks in a factorial scheme consisting of the following factors: (A) acaricide, in three levels: spirodiclofen and cyhexatin applied in rotation, lime sulphur; no acaricides; (B) pruning to remove branches that showed symptoms of leprosis, with two levels: with pruning, without pruning. We carried out periodic assessments of *Brevipalpus phoenicis* (Geijskes) populations (vector of the leprosis virus), leprosis incidence and severity, fruit yield, and the economic feasibility of the applied strategies. Based on the results, we concluded that spirodiclofen and cyhexatin were more effective than lime sulphur in *B. phoenicis* control. Control with lime sulphur required more applications than spirodiclofen and cyhexatin in rotation, making it more expensive. Pruning of symptomatic branches used in isolation was not sufficiently effective to control leprosis and significantly increased control costs. Profits were higher when the control involved sprayings of spirodiclofen and cyhexatin in alternation, with or without pruning.

Key words: *Citrus sinensis*, CiLV, Citrus leprosis mite, organic citrus.

Introduction

Leprosis has for several decades been considered one of the most serious citrus disease problems in Brazil. Its causal agent is the non-systemic CiLV (Citrus Leprosis Virus). Particles of CiLV are observed only on tissues with typical leprosis lesions (Kitajima *et al*., 1972). Presence of the only known vector, the mite *Brevipalpus phoenicis* (Geijskes, 1939) (Tenuipalpidae), and the existence of infected plants in the plantation are fundamental conditions for the dissemination of the disease on the plant or between plants of a grove (Kitajima *et al*., 1972). The non-systemic condition of the virus considerably increases the importance of the vector in the epidemiology of the disease (Rodrigues *et al*., 1994).

Control of leprosis is based almost exclusively upon applications of acaricides to control *B. phoenicis*. However, this mite becomes the vector of the causal agent only after feeding on plant tissues infected by the CiLV virus; there is no vertical transmission of the virus in the mite population (Chiavegato & Mischam, 1987).

According to Rodrigues (2002), the epidemiology of such disease in plantations without chemical control indicates that infection levels increase proportionally to the availability of diseased tissues. Thus, it has been suggested that due to particularities of the vector and of the CiLV virus, control measures should not be based only on reducing the vector population, but also on the efficient elimination of virus sources by pruning symptomatic branches (Bitancourt, 1955; Feichtenberger *et al*., 1997). However, the time-consuming nature of pruning and need for specialised labour are the main economic obstacles for the adoption of this practice by growers, as a complement to chemical control of the vector.

Accepted by G. J. de Moraes: 30 Oct. 2011; Published: 20 Dec. 2011

The present study was carried out to evaluate the effect of the use of acaricides with or without pruning in the control of citrus leprosis.

Materials and Methods

The study was conducted at Fazenda São Pedro, municipality of Reginópolis, State of São Paulo, Brazil, in two seasons, 2006–2007 and 2007–2008. The soil of the experimental area is classified as Typic Red-Yellow (LVA), and the region is climatically classified as Cwa type, subtropical with dry winter (Köeppen, 1970).

The 17 year old plants trees used were of the “Pear” variety of orange, grafted onto “Cleopatra” tangerine, spaced at 7 x 4 m and drip irrigated. Initially, 500 citrus plants were inspected to determine the level of leprosis infection, and graded according to the following scale: 1, no leprosis symptoms on any plant part; 2, lesions visible on outer thin branches; 3, lesions visible on outer thin and larger branches as well as on some inner thin branches; 4, lesions visible on many inner, fine branches. Only plants with grade 3 were selected for the experiment, which corresponded to 86% of the evaluated plants. The experimental design used was randomised blocks, in a factorial scheme consisting of the following factors: (A) acaricide, in three levels: spirodiclofen and cyhexatin applied in rotation, lime sulphur; no acaricides; (B) pruning to remove leprosis symptomatic branches, with two levels: with pruning, without pruning. Each combination of factors was replicated four times, with each experimental unit consisting of three plants in a row.

Every fifteen days, the level of infestation by *B. phoencis* was assessed to determine the need for acaricide applications, according to a pre-established control level, as subsequently described. Samples were taken only from the central plant of each experimental unit. At each sampling date, three scabbed (= infected) fruits were taken from the inner part of each of those plants and were examined for mites with a 10 x hand lens (Martinelli *et al*., 1976). In the absence of fruits, three branches were evaluated; in this case, mites were evaluated on a 25 cm–long section taken from the suberized region of the branch (Pattaro, 2003). Each fruit or branch with at least one *B. phoenicis* at any developmental stage was considered infested. Chemical spraying was carried out when at least 8.3% of the fruits/ branches were infested.

The acaricides used, and the respective volumes of commercial product per 100 L of water were: spirodiclofen (Envidor®), 20 mL; cyhexatin (Sipcatin®), 50 mL; and lime sulphur (Fertibom Super S®), 4,000 mL. Other pests and diseases were controlled over the whole experimental area, using products that are not known to affect the mites. Acaricide applications were done with a spray gun, using a volume of acaricide sufficient to completely cover the plants.

Branches and fruits were examined monthly to check for new leprosis symptoms. When they were observed on branches, these were pruned for pruning(+) trees and taken away from the experimental field. When symptoms were detected on fruits, these were removed from the plant and weighed; fruits that dropped due to leprosis were also weighed. During harvesting, healthy and leprosis-infected fruits were weighed separately. Weights of different fruits were then summed to determine total yield (healthy plus infected fruits) and yield losses due to leprosis. Evaluations of leprosis severity were carried out in all plants of each experimental plot, according to the following scale: 0, no symptoms; 1, few lesions on any plant part, restricted to a restrict sector of the plant; 2, lesions on different parts of the plant and/ or distributed to more than one sector; 3, lesions in all plant parts and distributed throughout the plant; 4, lesions throughout the plant, leaf and fruit drop; 5, same as for grade 4 plus branch withering. Each plant was evaluated by two people, and the average of their scores was considered in the analysis. For all evaluations of symptoms, only lesions appearing after the initiation of the study were considered.

Data about symptom severity, yield and yield loss due to leprosis were transformed to ln (x + 5) before submitting to variance analysis, using the computer program Estat of FCAV/UNESP (Estat, 1994); averages were compared using Tukey test, at 5 % level of significance.

At the end of each harvest, Cost of Effective Operation (COE) was determined, taking into account the different costs for each combination of treatments, including depreciation of machinery and social security contributions. The cost of each of the activities developed was registered, considering the land (ha) as a unit, as suggested by González *et al*. (1996). Labor cost was calculated based on the current minimum wage in Brazil, and adding 43% for social security contributions and taxes (Agrianual, 2007). Total revenue was estimated by considering the value of R$10.83 (approximately US$ 6.00) per box of oranges weighing 40.8 kg, and containing fruits considered healthy (IEA, 2008). Operating costs common to all treatments were not considered in the calculations.

Results and Discussion

For both seasons, the acaricide factor was statistically significant in relation to the three parameters measured in the study (Table 1). Conversely, the pruning factor and the interactions of acaricide and pruning were not significant in any season for any of the parameters, indicating that ineffectiveness of pruning symptomatic branches as a leprosis control measure.

TABLE 1. Effect of different treatments for the control of *Brevipalpus phoenicis*. Summary of the variance analysis and significance tests for the acaricide and pruning of symptomatic branches in two seasons (2006–2007 and 2007–2008).

Causes of variation	G.L.	Damage level [1]		Yield [2]		Yield loss [3]	
		Seasons					
		2006–2007	2007–2008	2006–2007	2007–2008	2006–2007	2007–2008
Acaricides (A)	2	0.479*	0.675*	2.277**	8.052*	1.067*	1.401**
Removal pruning (B)	1	0.000^{ns}	0.008^{ns}	0.022^{ns}	0.357^{ns}	0.331^{ns}	0.038^{ns}
A X B	2	0.000^{ns}	0.009^{ns}	0.052^{ns}	0.496^{ns}	0.089^{ns}	0.004^{ns}
Blocks	3	0.097	0.007	1.902	0.497	1.166	0.083
Residue	15	0.031	0.006	0.404	0.310	0.184	0.078
C.V. (%)		9.23	3.71	13.94	14.96	18.31	8.13

[1]Symptomatic damage level (see text for scale); [2]Total fruit yield, considering both damage and healthy fruits; [3]Yield corresponding to fruits that dropped because of damage by *B. phoenicis*. Comparisons of means: ns – not significant; (*) significant $p< 5\%$; (**) significant < 1%.

Incidence of leprosis

At the end of the first season, unsprayed plants showed the highest severity level of leprosis symptoms (Fig. 1). Plants sprayed with spirodiclofen and cyhexatin had the lowest severity level.

Rotational sprays of spirodiclofen and cyhexatin were more efficient in controlling *B. phoenicis* than the application of lime sulphur. In both seasons, four applications of spiridiclofen and four of cyhexatin were necessary, compared to sixteen applications of lime sulphur, to keep the mite population below the control level.

Applications of spirodiclofen resulted in longer periods of control than applications of cyhexatin. A similar result was reported by Ulian (2006). According to that author, the longer period of protection provided by spirodiclofen is due to the fact that only recently this product started to be used in citriculture in the state of São Paulo, and *B. phoenicis* has still not developed resistance to it.

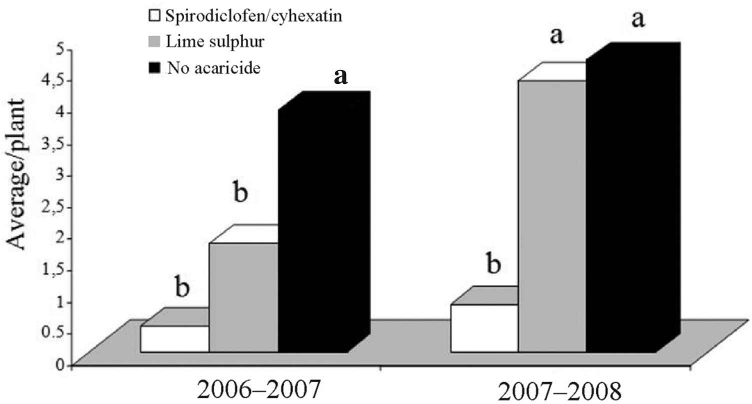

FIGURE 1. Averages levels of leprosis severity on plants treated or untreated with acaricides in two seasons (2006–2007; 2007–2008), independently of pruning to remove branches with symptoms of leprosis.

In the second season, higher leprosis severity symptoms occurred on plants treated with lime sulphur. In this season, the incidence of the disease was statistically the same on plants sprayed with that product and on unsprayed plants. This result can be partially explained by the short residual period and low ovicide effect of lime sulphur against *B. phoenicis* (Pattaro, 2003). The continuous use of lime sulphur could have increased the frequency of resistant individuals. Since long ago (Bitancourt, 1955), applications of lime sulphur were reported to reduce the occurrence of leprosis symptoms, but not to prevent them from occurring. Lime sulphur is the only acaricide allowed for use in organic citriculture in São Paulo. As a consequence, organic citrus producers have faced difficulties in the control of *B. phoenicis* by the unavailability of new options (Turra & Ghisi, 2004).

A comparison of the overall averages showed incidence to be higher in the second than in the first season. Although the elimination of branches infected by leprosis by pruning has been recommended by several authors as an alternative control strategy (Oliveira, 1986; Barreto & Pavan, 1995; Bassanezi, 2004), the observed ineffectiveness of pruning as an auxiliary tactic in leprosis management at our site may be related to the very high levels of incidence of the disease in the experimental area in both seasons.

Yield

In both seasons, citrus yield was significantly higher for trees sprayed alternately with spirodiclofen and cyhexatin than for unsprayed plants (Fig. 2). In the first season, no significant differences were observed between plants sprayed with those acaricides and plants sprayed with lime sulphur, but in the second season plants of the former treatment yielded more than plants of the latter. The lower yield of lime sulphur treated plants in the second season was probably due to the higher levels of leprosis incidence in that season.

Removal of infected branches resulted in substantial reduction of plant parts, especially in plants not receiving any acaricide application. However, this procedure did not affect yield, which was statistically the same on pruned and non-pruned plants in both seasons (Table 1). A comparison of the overall averages showed yield to be higher in the first than in the second season, probably at least in part due to the lower incidence of leprosis in the first season.

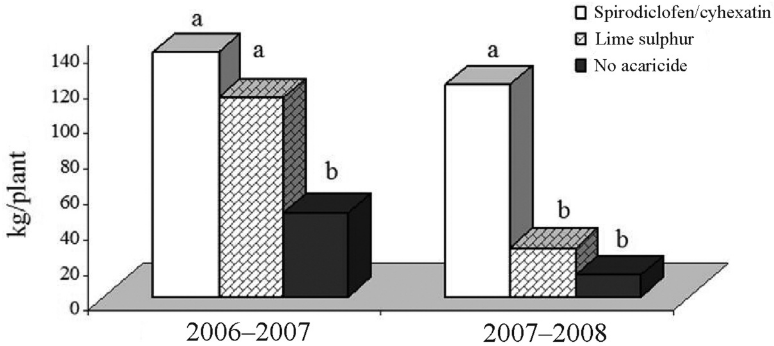

FIGURE 2. Yield (kg/plant) of plants treated or untreated with acaricides in two seasons (2006–2007; 2007–2008), independent of pruning to remove branches with symptoms of leprosis.

Yield loss due to leprosis

In both seasons, yield loss due to leprosis occurred in plants of all treatments (Fig. 3), indicating that none of the treatments evaluated in the study was able to entirely prevent this disease. However loss was lower for plants sprayed with spirodiclofen and cyhexatin than for plants of other treatments, which did not differ between themselves. The similar levels of losses in the lime sulphur and control treatments in both seasons indicate that despite significantly reducing the incidence of leprosis symptoms in the first season, the incidence level achieved was not enough to reduce yield loss due to the disease. This was expected even before weighing the fruits, given the severe defoliation and branch withering of plants of those two treatments.

A comparison of the overall averages showed yield loss to be higher in the second than in the first season, which was related to the higher incidence of the disease in the second season.

Control cost

For all treatments, profits were lower in the second than in the first season (Table 2), most certainly due to the higher levels of incidence of leprosis in that season. Profits were higher in both seasons when the control involved the use of spirodiclofen and cyhexatin in rotation, with or without pruning.

In the second season, profits were negative when lime sulphur was used, with or without pruning. However, in both seasons fruits produced by plants treated with lime sulphur had a better appearance, and could be sold by a higher price for fresh consumption in the organic market. This possibility was not taken into account in the calculation of profits.

The calculated cost of pruning was very high. The high cost, associated with the absence of significant effect on mite control when used by itself did not lend support to its recommendation (Oliveira, 1986; Barreto & Pavan, 1995; Bassanezi, 2004) as the only control measure, at least at the levels of leprosis incidence observed in this study. However, evaluations of the possible efficiency of this practice at lower levels of leprosis incidence should be evaluated. Of course when acaricides were used, the cost of pruning was greatly reduced, but even so profits were lower than when only chemical applications were done; the numbers of required sprays were the same for plants that were or were not pruned.

It became evident that further studies are necessary about the epidemiology of the disease, behaviour of *B. phoenicis*, estimates of damage and control levels, application technology, mite resistance management, causal vector-agent interaction (Bassanezi, 2004; Pattaro, 2003). Those studies could allow the determination of alternative methods to the chemical control of *B. phoenicis*.

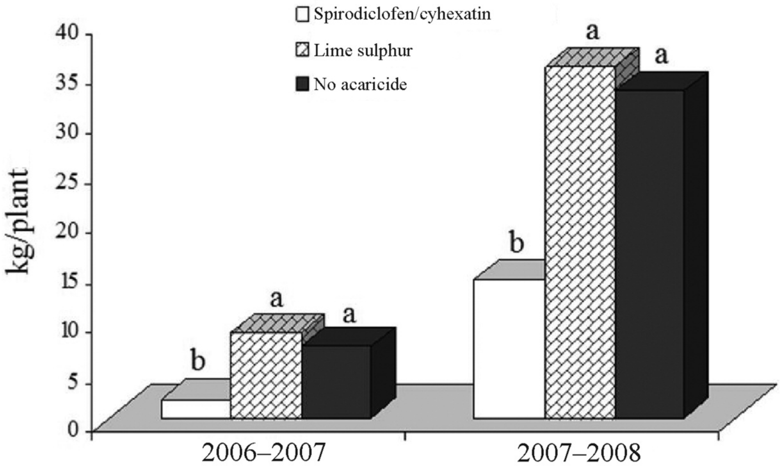

FIGURE 3. Yield loss (kg/plant) due to the incidence of leprosis on plants treated or untreated with acaricides in two seasons (2006–2007; 2007–2008), independent of pruning to remove branches with leprosis symptoms.

TABLE 2. Economic analysis (R$ 1.00= US$ 0.55) of the use of different treatments for the control of *Brevipalpus phoenicis*, vector of the citrus leprosis virus in two seasons (2006–2007 and 2007–2008) in Reginópolis, State of São Paulo.

Treatments	Crop 2006–2007		
	Total		
	Costs (R$/ha)	Revenue (R$/ha)	Profit R$/ha
Spirodiclofen/cyhexatin	4,377.57	13,573.97	9,196.40
Spirodiclofen/cyhexatin + removal pruning	4,704.28	12,784.74	8,080.46
Lime sulphur	7,732.74	11,582.82	3,850.08
Lime sulphur + removal pruning	8,125.15	11,156.92	3,031.77
No acaricide	1,111.80	7,176.36	6,064.56
No acaricide + removal pruning	7,037.02	7,088.24	51.22
Treatments	**Crop 2007–2008**		
	Total		
	Costs (R$/ha)	**Revenue (R$/ha)**	**Profit R$/ha**
Spirodiclofen/cyhexatin	7,988.07	11,456.79	3,468.72
Spirodiclofen/cyhexatin + removal pruning	9,262.64	12,096.43	2,833.79
Lime sulphur	10,049.18	3,182.13	-6,867.05
Lime sulphur + removal pruning	11,958.49	2,641.03	-9,317.46
No acaricide	282.52	1,132.41	849,89
No acaricide + removal pruning	3,239.79	2,549.12	-690.67

Acknowledgements

We are grateful to Fundecitrus, for financial support for the research, and to FAPESP, for the grant awarded to the first author.

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