



GOBIERNO DE CHILE
SERVICIO AGRICOLA Y GANADERO

**MEASURES SUGGESTED FOR QUARANTINE PEST RISK MANAGEMENT IN
CLEMENTINES, MANDARIN ORANGES AND TANGERINES EXPORTED FROM
CHILE TO THE UNITED STATES OF AMERICA**

March 2002

Foreword

International plant protection organizations such as the North American Plant Protection Organization (NAPPO) and the International Plant Protection Convention (IPPC) of the United Nations Food and Agriculture Organization (FAO) provide guidance for conducting pest risk analyses. These guidelines describe three stages of pest risk analysis: Stage 1 (initiation), Stage 2 (risk assessment) and Stage 3 (risk management). The first two of these stages were addressed in the accompanying risk assessment document “Importation of Fresh Commercial Citrus Fruit: Clementine (*Citrus reticulata* Blanco var. ‘Clementine’), Mandarin (*Citrus reticulata* Blanco) and Tangerine (*Citrus reticulata* Blanco) from Chile into the United States— A Pathway Initiated Plant Pest Risk Assessment”. That document identified three pests of concern: *Brevipalpus chilensis*, *Proeulia auraria* and *Proeulia chrysopteris*.

The reduction of phytosanitary risk occurs through the use of mitigation measures that are designed to eliminate, reduce, or prevent the presence of pest populations in shipments of commodities primarily in the country of origin. The appropriate risk management strategy for a particular pest depends on the risk posed by that pest. APHIS risk management programs are risk based and dependent on the availability of appropriate mitigation methods. Details of APHIS risk management programs are published, primarily, in the Federal Register as quarantine notices. Risk assessments and risk management strategies may also be published as a single risk analysis document or, as is the case here, stand alone documents.

As noted in **Section 2.6** of the risk assessment document, in a standard visual inspection, *P. auraria* and *P. chrysopteris* are easy to detect because they are external feeders. Consequently, U.S. import regulations currently permit importation of certain fruits from Chile (*e.g.*, apricots, nectarines, plums, plumcots and peaches) with a preclearance inspection to certify freedom from *Proeulia* species (Title 7 Code of Federal Regulations Part 319 Section 56-2s).

Because *B. chilensis* may be more difficult to detect, USDA has required specific treatment(s) prior to entry for fruit hosts of this pest (*e.g.* cherimoya; Title 7 Code of Federal Regulations Part 319 Section 56-2z). In anticipation of such a requirement for clementines, mandarins and tangerines, the Chilean Servicio Agrícola y Ganadero, in cooperation with the Fundacion para el Desarrollo Frutícola, produced this risk management document entitled “Measures Suggested for Quarantine Pest Risk Management in Clementines, Mandarin Oranges and Tangerines exported from Chile to the Market of the United States, March 2002”. This document describes a risk management program for *B. chilensis* proposed by Chile. The risk management document has not been revised by USDA, APHIS and is presented, along with the draft risk assessment, for public comment as part of the current Federal Register Notice of Availability.

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1 INTRODUCTION

This document proposes a feasible set of risk mitigation measures whose implementation is aimed at mitigating the potential risk of introducing quarantine pests into the United States via the importation of fresh commercial clementine, mandarin orange and tangerine fruits from Chile.

The companion commodity pest risk assessment for these fruit species (SAG / APHIS, 2002) rated the quarantine species *Brevipalpus chilensis* as **Medium** in accordance with the USDA/APHIS PRA Guidelines, Version 5.02 (USDA, 2000). As noted in the Guidelines, pests rated Medium may require specific mitigation.

The risk management measures proposed are based upon the experience gained from the kiwifruit export program to the United States. This program (a Systems Approach) comprises a series of individual measures to mitigate the risk of introducing *Brevipalpus chilensis* into the United States (See Annex 8).

Background information is supplied in Annexes 1 through 7. Technical data are presented on the natural prevalence of *Brevipalpus chilensis* in clementine orchards, the mite's population dynamics, control measures at the orchard level, efficacy of the packing process - considering it to be a post-harvest treatment for the elimination of the mite as well as efficacy of the detection method.

2 RISK MANAGEMENT MEASURES

Risk management measures - or Systems Approach components- for *Brevipalpus chilensis* are given below. These measures, considered feasible and appropriate, can be implemented in an integrated manner from pre-harvest to post-harvest in order to reduce pest risk:

- Orchard Control
- Orchard Registration
- Low prevalence orchard certification
- Harvest timing
- Post-harvest treatments
- Joint SAG/USDA phytosanitary inspection

2.1 Orchard Control

Universidad Católica de Valparaíso (UCV) carried out studies on mite control at the orchard level. These studies show that applications of detergent or oil during the period of highest *B. chilensis* population levels have an efficacy of 92.0 percent and 97.3 percent respectively, in the control of the mite (See Annex 3: Effect of different treatments on *Brevipalpus chilensis* in clementine orchards, Nogales, V Region, Chile; UCV).

A pest survey carried out by Fundación para el Desarrollo Frutícola (FDF) during harvest (May through August, 1999) in different clementine, mandarin orange and tangerine orchards in Regions III through VI, indicated an average *B. chilensis* prevalence level of 0.091 specimen per fruit (See Annex 1: Survey and identification of pests associated to

clementine, mandarin orange and tangerine fruits; FDF).

The above results suggest that **pest control at orchard level** constitutes an effective risk mitigation measure to ensure the phytosanitary condition of the lots to be exported.

2.2 Orchard Registration

For phytosanitary monitoring, each orchard is identified and registered by means of a Registration Card with the following basic data:

- Orchard
- Grower
- Municipality
- Province
- Region
- Area planted to each species
- Number of plants/hectares/species
- Date of Harvest

2.3 Certification of Low Prevalence Orchards

The Servicio Agrícola y Ganadero (SAG) will carry out a program for certification of low prevalence to be applied in each of the registered orchards. The program would be based upon a random fruit sampling carried out in each registered orchard prior to harvest.

Each sample then undergoes the “dragging by washing” procedure – a method that has proved to be effective in the detection of *Brevipalpus* in table grapes and kiwifruit. The efficacy of this mite detection method has been evaluated for clementine fruits (See Annex 7: Efficacy test for detecting *Brevipalpus chilensis* on Clementine fruits).

It is proposed that only orchards certified by SAG shall participate in the annual export program to the United States.

2.4 Harvest Timing

Studies carried out by Universidad Católica de Valparaíso on the population biology of *B. chilensis* in clementine orchards show that the highest population levels of mites present on leaves, twigs and fruits occur from December through March (See Annex 2: Population Monitoring of *Brevipalpus chilensis* in clementine orchards in the IV and V Regions of Chile; UCV).

Harvest starts in April and lasts until September, a period in which the winter population of *B. chilensis* is reduced. Timing of the harvest is therefore considered to be an additional mitigation measure to reduce pest risk.

2.5 Post-Harvest Treatment (washing and waxing)

Washing and waxing fruits are considered normal citrus fruit packaging processes in Chile and are similar to the USDA-APHIS approved “soap water and waxing” quarantine treatment applied to cherimoyas imported into the United States from Chile.

Several studies were undertaken to evaluate the effectiveness of each of the different steps of the packaging process in reducing *B. chilensis* populations. One of the first experiments carried out by Fundación para el Desarrollo Frutícola (FDF) in 2000 evaluated washing the fruits with water and detergent, followed by waxing. The result was an estimated 89.7 percent efficacy in reducing the number of *B. chilensis* mites. (See Annex 4: Exploratory sampling at different stages from harvest to post-harvest of clementine fruits, FDF).

The results of two studies conducted by UCV in June 2000 in the IV and V Regions showed the normal packaging process reduces *B. chilensis* populations 79.9 percent and 81.2 percent, respectively, (See Annex 5: Effect of the Packing Process on the Removal of *Brevipalpus chilensis* in Clementine fruits. UCV).

Similar research conducted by FDF on clementine and mandarin orange fruits during the 2001 season found that the normal packing process reduces *B. chilensis* populations from 85.4 percent to 89.3 percent (See Annex 6)

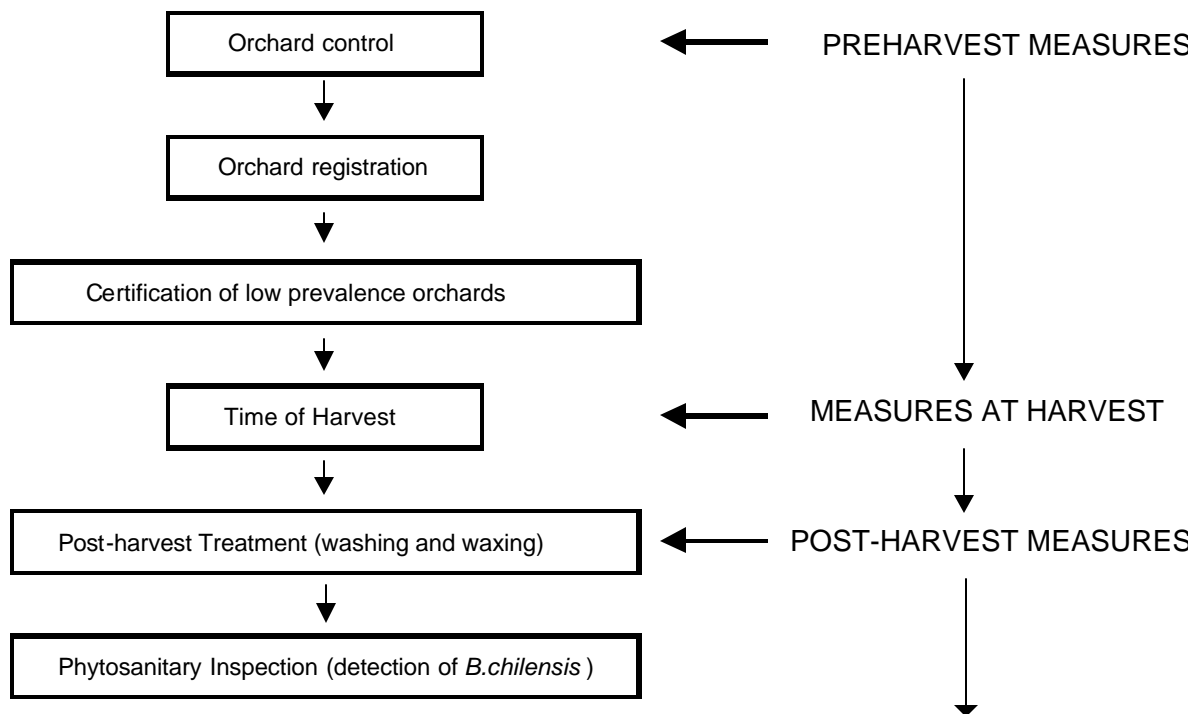
These studies confirm that the washing and waxing treatments clementines, mandarin oranges and tangerines undergo during the packing process produce a significant reduction of *B. chilensis*. These treatments are standard operating procedures in the packing process and constitute an additional pest risk mitigation measure.

2.6 Pre-shipment Phytosanitary Inspection

To verify the presence / absence of *B. chilensis*, it is proposed that a sample of fruits be taken during the phytosanitary packinghouse inspection carried out in Chile jointly by SAG, USDA-APHIS and the Chilean Exporter’s Association (ASOEX) as part of the preclearance program. This sample is to be analyzed by applying the “dragging by washing” procedure similar to the one used for the detection of this mite on kiwifruits exported to the United States.

3 FLOW CHART OF PROPOSED RISK MANAGEMENT MEASURES FOR *Brevipalpus chilensis*.

The flow chart below lists the measures described above. They can be considered a set of integrated measures to mitigate the pest risk posed by *B. chilensis* associated with the export of clementine, mandarin orange and tangerine fruits from Chile to the United States.



4 LITERATURE CITED

SAG / USDA. 2002. Importation of Fresh Commercial Citrus Fruit: Clementine (*Citrus reticulata* Blanco var. 'Clementine') Mandarin (*Citrus reticulata* Blanco) and Tangerine (*Citrus reticulata* Blanco) from Chile into the United States. A Pathway Initiated Plant Pest Risk Assessment. March 2002 (Revised July 2002). Departamento Protección Agrícola, Servicio Agrícola y Ganadero and United States Department of Agriculture. 67 pp.

USDA, 2000. Guidelines for Pathway-Initiated Pest Risk Assessments, Version 5.02. www.aphis.usda.gov/ppq/prc/commodity/cpraguide.pdf. Last accessed 31 July, 2002.

ANNEX 1

SURVEY AND IDENTIFICATION OF PESTS ASSOCIATED TO CLEMENTINE, MANDARINE AND TANGERINE FRUITS

(FUNDACION PARA EL DESARROLLO FRUTICOLA)

FDF

INCLUDING TABLES

- Table 1: Prevalence of *Brevipalpus chilensis* in clementine, mandarine and tangerine orchards.
- Table 2: Registration of clementine, mandarine and tangerine orchards and sampling date aimed at determining the prevalence of *B. chilensis*.
- Table 3: Prevalence in clementine, mandarine and tangerine orchards Results by orchard.
- Table 4: Natural location of *B. chilensis* on fruits and pedicels of clementines, mandarines and tangerines at different periods of sampling.
- Table 5: Other insects and mites detected in clementine, mandarine and tangerine orchards

1. BACKGROUND INFORMATION

The area planted to clementine, mandarin and tangerine fruits is about 1,300 hectares, distributed between Chile's III and VI regions.

Since no concrete information existed stating the presence of *Brevipalpus chilensis* and its prevalence in clementine, mandarin and tangerine fruits, a survey was conducted which encompassed production zones in the country, and included the identification of other pests associated with these fruit species.

2. OBJECTIVE

To identify the presence and level of infestation (prevalence) of *B. chilensis* and other pests associated with clementine, mandarin and tangerine fruits in all of Chile's production zones of these species.

3. METHODOLOGY

3.1 Survey of exporters and register of orchards

Since no up-to-date national land registry existed for the surface area planted to clementines, mandarins and, a survey citrus exporters was conducted. Participants were requested to submit the following information regarding the three species mentioned above: name of orchard, species, region, province, town, number of blocks, age of the plantation, surface area in hectares, number of plants per hectare and harvesting date. The information submitted constituted the basis for surveying for *B. chilensis* as well as other pests associated with clementine, mandarin and tangerine.

3.2 Sampling Plan

Based on the above registry, a sampling plan was defined to carry out a survey for the possible presence of insects and/or mites of quarantine significance on the fruits of these species. This sampling plan called for taking samples in each orchard on three occasions during the harvest.

a) Sampling level and selection of fruits to be sampled

The size of the sample in each orchard was determined depending upon the total number of plants in that orchard, according to the following table

Total N° of plants	% sampling	N° of plants in the sample
Less than 5.000	1,00 %	50
5.001 - 10.000	0,75 %	51 – 75
10.001 - 15.000	0,67 %	76 – 100
15.001 - 30.000	0,56 %	101 – 168
30.001 - 50.000	0,50 %	169 – 250
over 50.000	0,33 %	> 250

From every 10 plants, a **sample of 30 fruits** was taken, with a sub-sample of three fruits per plant; one fruit from the interior of the tree, another from the middle of the tree and a third one from the periphery. For each sub-sample, a different geographic orientation was systematically applied, going clockwise from north to south, east and west of the tree.

b) Selection of plants to be sampled

In every orchard, plants were chosen at random, following a transverse line through the orchard or block. After each chosen plant, two, three or more lines or rows were skipped, in order to cover the whole orchard area.

In those orchards that are divided into several blocks, the blocks were sampled according to their respective plant densities, so that the total sample would truly reflect the whole orchard reality.

c) Handling of the samples

The samples (30 fruits per every 10 plants) were deposited in paper bags. Labels or stickers containing the following data identified these bags:

- Exporter
- Grower
- Orchard
- Region, Province, Commune
- Date of Sampling

The bags containing the samples taken were put in cardboard boxes, with a cardboard cover for sample protection and then transported to the laboratory for analysis.

3.3 Procedure for the detection and identification of insects and mites

For the detection and identification procedure, a method was developed which based upon the current detection system used in the kiwi fruit "systems approach", i.e., by washing the fruits, but introducing some modifications to accomodate certain differences between citrus fruits and kiwi fruits. The procedure consisted of two separate washings: one for the fruits as a whole, and one

for the pedicels (See Annex 6: "Test for the Efficacy of the Method applied"). The washing was done with a solution of soap in water. For the retention of the insects and mites, two sieves of different mesh were used.

The intercepted specimens were kept in an alcohol solution, to which glycerin and acetic acid (AGA solution) had been added. In the case of mites, they were mounted on microscope slides.

a) Materials

- Metal sieve with 200 mesh
- Metal sieve with 20 mesh
- Stereoscopic 40 x magnifying glass
- Washing container with high-pressure water supplied in shower form.
- 300 x to 600 x Microscope
- Tweezers
- Ethanol alcohol
- Petri dishes
- Small glass tubes 3 to 5 ml
- Detergent
- Microscope slides and covers
- Hot plate
- Mounting medium (Hoyer, Euparal)

b) Procedure

The sieves were put one on top of the other, the one with the smaller mesh (mesh 200) below the one with the bigger mesh. Then the fruits and the pedicels were put separately on the sieves; the number of fruits and pedicels washed at one time was appropriate to allow for thorough washing of each of them.

Then the fruits and pedicels were sprayed separately with a solution of water and liquid soap. Every fruit and pedicel was washed thoroughly by means of a pressure water jet using a shower-like nozzle. After completing the first washing, a second one was carried out to assure maximum efficacy.

The 200-mesh sieve was then tilted and washed with a soft water stream so that the material eventually collected would move to one side of the sieve. Then using water and a pair of tweezers, the material was transferred to a Petri plate. This procedure was used for both fruits and pedicels.

Organisms present on the Petri plate were observed with a stereoscopic microscope. Mites and insects present were then transferred to a solution of ethanol, glycerin and acetic acid (AGA solution). Afterwards, the specimens were put on a microscope slide in order to be identified under the microscope. Some insects that could not be put on the slide were left in the above-mentioned solution for subsequent identification.

4. RESULTS

Data on the number of orchards monitored, surface area, number of plants, total samples and total of evaluated fruits, total number of detected *B. chilensis* specimens (living and dead), the level of infestation or prevalence of infested fruits, expressed in terms of average number of specimen per fruit, (live specimens) are summarized in **Table 1**.

As shown in Table 1, the prevalence of *B. chilensis* in Region III was an average of 0.439 specimens per fruit (the result of evaluating two orchards whose areas are the equivalent of 4.17 percent of the total surface area monitored. In contrast, Regions IV and V, whose surface area represented 85.25 percent of the total area surveyed, the prevalence of *B. chilensis* was an average of 0.07 to 0.077 specimens per fruit, based upon 15,150 and 8,910, respectively, fruit evaluated. Further south, in the Metropolitan Region and in Region VI, the levels of infestation declined significantly to an average of 0.049 and 0.004 specimens per fruit, respectively.

In **Table 2**, detailed information is presented on the registered clementine, mandarin and tangerine orchards. Data presented include: Region, Province, Town, area, age of plantation and dates at which the samplings were carried out. Additionally, **Table 3** presents the prevalence results of de *B. chilensis* for each monitored orchard, as well as the exact location (on fruits or on pedicels) where the specimen had been found.

Table 4 summarizes the location and the percentage distribution of *B. chilensis* specimen on fruits and pedicels of clementines, mandarins and tangerines in the different sampling periods.

Table 5 presents a list of the species, both mites and insects, identified during the survey period in clementine, mandarin and tangerine orchards.

TABLE 1

PREVALENCE OF *Brevipalpus chilensis* IN CLEMENTINE, MANDARINE AND TANGERINE ORCHARDS IN CHILE
1999 SURVEY

Region	Surface area		Nº of orchards monitored	Nº of plants		Total Nº of samples	Nº of fruits evaluated	Detection of <i>B. chilensis</i>			Percentage alive specimen	Prevalence Average Nº of specimen per fruit
	total has.	relative %		total	per ha.			Total specimen detected	Total Nº of specimen			
									alive	dead		
III	40.7	4.17	2	36,768	903	67	2,010	1,724	883	841	51.22	0.439
IV	590.1	60.39	15	402,142	681	505	15,150	2,974	1,066	1,908	35.84	0.070
V	242.9	24.86	24	139,189	573	297	8,910	1,839	687	1,152	37.36	0.077
RM	63.3	6.48	10	38,062	601	108	3,240	466	160	306	34.33	0.049
VI	40.1	4.10	3	26,612	664	48	1,440	28	6	22	21.43	0.004
TOTAL	977.1	100.00	54	642,773	658	1,025	30,750	7,031	2,802	4,229	39.85	0.091

Note: Sample=30 fruits in each case

Source: Fundación para el Desarrollo Frutícola.

TABLE 2

**REGISTER OF CLEMENTINE, MANDARINE AND TANGERINE ORCHARDS
AND DATES OF SAMPLING FOR DETERMINING PREVALENCE OF *Brevipalpus chilensis*
1999 SURVEY**

**Third
Region**

	Name of Grower	Name of Orchard	Species	Province	Town	Nº of Lots	Age	Ha.	Nº of Plants	Nº Sam- ples	Harvest Date	Date of Sampling			Comments
												1	2	3	
1	Soc. Agr. Buenaventura Ltda.	Buenaventura	Clementina	Copiapó	Copiapó	2	4	1.0	800	1	5/May	17/M ay	-	-	-
2	Cítricos Uni Agri	Gibraltar	Clementina	Copiapó	Copiapó	7	7	39.7	35,968	66	24/May	18/M ay	18/ Jun	2/ Jul	-
3	Uni Agri	El Canelo	Clementina	Copiapó	Tierra Amarilla	6	1	24.3	15,416						Only plants
Total Region								65.0	52,184	67					

Fourth Region

	Name of Grower	Name of Orchard	Species	Province	Town	Nº of Lots	Age	Ha.	Nº of Plants	Nº Samples	Harvest Date	Date of Sampling			Comments
												1	2	3	
4	Guillermo Peña y Lillo Mora	Fundo Las Rojas	Clementina	Elqui	La Serena	6	4	11.7	6,500	15	30/Jun	23/ Jun	05/ Jul	30/ Jul	-
5	Agrícola Cerrillos de Tamaya S.A.	Fundo La Laguna	Clementina	Limarí	Ovalle	4	6	24.0	15,984	27	1/Jun	25/ May	14/ Jun	01/ Jul	-
6	Agrícola Cerrillos de Tamaya S.A.	El Sauce	Clementina-Mandarina	Limarí	Ovalle	4	1	55.0	33,330						Only plants
7	Raúl Alvarez	Parcela Nº 118	Clementina	Limarí	Ovalle	4	1	8.0	5,328						Only plants
8	Agronova	Camino Socos	Clementina	Ovalle	Ovalle	2		5.0	4,000	17	31/May	31/ May	28/ Jun	9/ Jul	-
9	Soc. Ag. Río Negro Ltda.	Río Negro	Clementina	Ovalle	Ovalle	8	3	18.0	7,000	19	30/Jun	05/ Jul	10/ Jul	12/ Jul	-
10	Soc. Agr. Copequén	Yungay	Clementina	Elqui	Vicuña	6	1	25.0	20,000						Only plants
11	Agrícola Villa Alegre Ltda.	La Granja	Nova-Clementina	Limarí	Punitaqui	7	5	43.2	34,069	52	31/May	31/ May	21/ Jun	08/ Jul	-
12	Agrícola Villa Alegre Ltda.	El Sauce	Clemenule	Elqui	Coquimbo	6	4	147.0	81,585	60	17/May	24/ May	22/ Jun	05/ Jul	-
13	Agrícola Punitaqui Ltda.	Fundo La Palma	Clementina	Limarí	Monte Patria	10	6	39.7	22,153	24	26/Apr	13/ May	25/ May	09/ Jun	-
14	Prodex Guatulame Ltda.	Fundo Antunguayco	Clementina-Satsuma	Limarí	Monte Patria	17	5	26.3	18,791	30	19/Apr	14/May	01/ Jun	16/ Jun	-
15	Mariano Díaz y Cía. Ltda.	Hijuela 5-B Tabalí	Clementina	Limarí	Ovalle	13	3	17.9	10,872	21	30/May	28/ Jun	09/ Jul	13/ Jul	-
16	Agrícola Vespucio	Agrícola Vespucio	Clementina	Limarí	Monte Patria	1	1	30.0	16,680						Only plants
17	Fundo Carretón	San Nicolás	Clementina	Limarí	Monte Patria	3	1	20.0	12,500						Only plants
18	Soc. Agr. El Mirador	Fundo El Mirador	Clementina	Limarí	Monte Patria	6	5	31.1	20,393	5	16/Apr	13/ May	-	-	-
19	Soc. Agr. Uni Agri Ovalle Ltda.	Las Represas	Clementina	Limarí	Monte Patria	14	7	52.5	34,607	54	10/May	19/ May	09/ Jun	24/ Jun	-

20	Soc. Agr. Uni Agri Ovalle Ltda.	Camarico	Clementina	Limarí	Punitaqui	7	6	25.1	15,009	26	1/Jun	10/Jun	30/Jun	06/Jul	-
21	Soc. Agr. Uni Agri Ovalle Ltda.	Nueva Aurora	Clementina	Limarí	Punitaqui	4	5	24.7	14,732	30	1/Jun	10/Jun	30/Jun	06/Jul	-
22	Soc. Agr. Uni Agri Ovalle Ltda.	Santa Rosa	Clementina	Limarí	Monte Patria	2	.	15.1	12,545	27	19/May	03/Jun	11/Jun	26/Jun	-
23	Cítricos Uni Agri S.A.	Unicítricos	Clementina	Limarí	Monte Patria	30	7	108.9	83,902	98	4/May	12/May	27/May	03/Jun	-
Total Region								728.1	469,980	505					

Fifth Region

	Name of Grower	Name of Orchard	Species	Province	Town	Nº of Lots	Age	Ha.	Nº of Plants	Nº Samples	Harvest Date	Date of Sampling			Comments
												1	2	3	
24	Agrícola Paiquén Ltda.	Fundo Colunquen	Clementina	San Felipe	Panquehue	5	6	28.4	15,768	27	14/Jun	15/ Jun	12/ Jul	02/ Aug	-
25	Agrícola Paiquén Ltda.	Fundo Santa Adela	Clementina	San Felipe	Panquehue	1	3	22.5	13,757	27	14/Jun	15/ Jun	12/ Jul	02/ Aug	-
26	La Higuera S.A.	La Higuera	Clementina	San Felipe	Santa María	4	3	22.3	14,822	3	15/Jun	16/ Jun	-	-	-
27	Agrícola San Juan	Fundo Chai-Chai	Clementina	Petorca	Cabildo	1	8	3.9	2,228	6	10/Jun	14/ Jul	03/ Aug	-	-
28	Agrícola El Canelillo	Fundo El Encierro	Clementina	Quillota	La Cruz	8	7	24.2	13,094	27	15/May	01/ Jun	02/ Jul	28 /Jul	-
29	Agrícola El Roble	Fdo. Sta. Marta de Longotoma, Hijueta A	Clementina	Petorca	La Ligua	1	6	4.5	2,497	9	10/Jun	18/ Jun	14/ Jul	03/ Aug	-
30	Agrícola Huerto California	Parcela El Totoral	Clementina	Quillota	Hijuelas	1	6	2.0	1,110	3	15/May	01/ Jun	01/ Jul	27/ Jul	-
31	Agrícola Peppi Ltda.	Parcelas El Romero 1 y 2	Clementina	Quillota	Limache	1	7	8.5	4,747	15	7/Jun	21/ Jun	07/ Jul	27 /Jul	-
32	Agrícola Pucará Ltda.	Parcela N°39-El Carmen	Clementina	Petorca	La Ligua	1	6	4.0	2,220	6	5/May	18/ Jun	14/ Jul	03/ Aug	-
33	Agrícola Pullacón Ltda.	Bien Común N° 7, San Lorenzo, Lote B	Clementina	Petorca	Cabildo	4	6	21.5	11,909	24	28/May	18/ Jun	14/ Jul	03/ Aug	-
34	Agrícola Sta. Cecilia	Parcelas N°17, N°18, N°7-2B	Clementina	Quillota	Nogales	3	7	12.9	8,200	18	8/Jun	31/ May	07/ Jul	28/ Jul	-
35	Agromol Ltda.	Huerto El Maitén	Clementina	Quillota	Hijuelas	1	8	9.8	6,527	15	21/Jun	21/ Jun	07/ Jul	27/ Jul	-
36	Héctor Bozzolo Palma	Bien Común N° 7, San Lorenzo	Clementina	Petorca	Cabildo	1	6	11.4	5,905	15	14/Jun	18/ Jun	14/ Jul	03/ Aug	-
37	Desarrollo Agrario	Fdo. Los Calderones de la Peña	Clementina	San Felipe	Llay Llay	1	5	8.1	5,364	8	12/May	25/ May	14/ Jun	-	-
38	María Paz Greene Concha	Maitenes OCOA	Clementina	Quillota	Hijuelas	2	4	9.0	4,995	17	1/Jun	28/ May	01/ Jul	28/ Jul	-
39	Rodrigo Manubens Smith	Parcela Los Pinos -Lo Rojas	Clementina	Quillota	La Cruz	1	7	5.7	3,184	9	29/May	01/ Jun	02/ Jul	28/ Jul	-

40	Eduardo Moltedo	El Caqui-Parcela 39	Clementina	Quillota	La Calera	1	8	2.5	1,500	6	15/Jun	28/May	02/Jul	27/Jul	-
41	Carlos Moraga	Parcela El Denuncio	Clementina	Quillota	Nogales	5	7	16.6	9,228	21	7/Jun	08/Jun	02/Jul	28/Jul	-
42	Hugo Muñoz Villablanca	Parcela Carén N°38	Clementina	Quillota	Quillota	2	7	7.3	3,833	12	15/Jun	08/Jun	01/Jul	27/Jul	-
43	Miguel Nenadovic	Fundo El Sauce	Clementina	Los Andes	Los Andes	1	1	4.4	2,442	6	5/May	24/May	14/Jun	-	-
44	Eduardo Seidemann Eiserman	Ariztia s/n	Clementina	Quillota	Quillota	1	7	3.2	1,760	6	2/Jul	21/Jun	07/Jul	27/Jul	-
45	Soc. Com. Ag. Los Rosales de Sta. Julia	Chacra Los Rosales 36	Clementina	Quillota	Quillota	3	5	2.6	1,704	6	5/May	01/Jun	01/Jul	27/Jul	-
46	Agrícola Huerto California	Entre Ríos	Clementina	Quillota	San Isidro	1	7	3.0	1,727	9	10/Jun	01/Jun	01/Jul	27/Jul	-
47	Raimundo del Río	Inversiones Tesalía Ltda.	Clementina	San Felipe	San Felipe	1	4	1.2	668	2	1/Jun	25/May	06/Jul	-	-
48	Agrícola El Sobrante	Anfiteatro	Mandarina	Petorca	Petorca	1	1	2.5	1,400						Only plants
49	Agrícola Chalaco Ltda.	Paloma	Mandarina	Petorca	Petorca	1	1	5.8	3,200						Only plants
50	Agrícola Chalaco Ltda.	Algarrobo 2	Mandarina	Petorca	Petorca	1	1	3.6	1,800						Only plants
Total Region								251.2	145,589	297					

Metropolitan Region

	Name of Grower	Name of Orchard	Species	Province	Town	Nº of Lots	Age	Ha.	Nº of Plants	Nº Samples	Harvest Date	Date of Sampling			Comments
												1	2	3	
51	Fernando Cerda	.	Clementina	Melipilla	Mallarauco	1	7	2.0	1,110	6	10/Jun	23/ Jun	07/ Jul	19/ Jul	-
52	Saplums	La Parva	Clementina	Santiago	Colina	1	11	5.9	3,935	12	7/May	14/ May	16/ Jun	6/ Jul	-
53	Luz Montes Bollo	Parcela 14, Carmén Bajo	Clementina	Melipilla	Melipilla	4	7	8.9	6,000	15	1/Jun	14/ Jun	25/ Jun	06/ Jul	-
54	Rafael Palacios	Las Pataguas - La Puntilla	Clementina	Melipilla	Paine	3	5	9.4	5,200	12	15/Jun	16/ Jun	25/ Jun	07/ Jul	-
55	Friederike Volkenborn	Talhuen	Clementina	Melipilla	María Pinto	3	7	10.0	5,551	12	10/Jun	14/ Jun	23/ Jun	07/ Jul	-
56	Enrique Alcalde	.	Clementina	Talagante	Isla de Maipo	2	3	8.6	4,800	15	17/Jun	18/ Jun	06/ Jul	19/ Jul	-
57	Agrícola Las Petras	Fundo Perales	Clementina	Melipilla	Curacaví	1	5	3.5	1,940	6	2/Jun	14/ Jun	23/ Jun	07/ Jul	-
58	Agrícola María Luz Baldrich Ltda.	Reserva Hijueta Siete	Clementina	Melipilla	Mª Pinto	1	6	8.2	4,532	15	24/May	14 Jun	23/ Jun	07/ Jul	-
59	Mallarauco	Patria Vieja	Clementina	Melipilla	Melipilla	1	5	2.2	1,800	-	-	-	-	-	No colaboración
60	Rafael Palacios	Cullipeumo Bajo	Clementina	Maipo	Paine	1	4	3.0	1,946	6	15/Jun	16/ Jun	25/ Jun	07/ Jul	-
61	Rafael Palacios	Santa Inés	Clementina	Maipo	Paine	3	3	7.0	3,892						Only plants
62	Soc. Primavera Agrícola Ltda.	Fundo La Primavera	Clementina	Maipo	Calera Tango	1	6	3.8	3,048	9	8/Jun	11/ Jun	25/ Jun	15/ Jul	-
Total Region								72.5	43,754	108					

Sixth Region

	Name of Grower	Name of Orchard	Species	Province	Town	Nº of Lots	Age of Plantation	Ha.	Nº of Plants	Nº Samples	Harvest Date	Date of Sampling			Comments
												1	2	3	
63	Fernando Tagle	Hijuela Cuarta	Clementina	Cachapoal	Las Cabras	1	5	10.0	5,500	-	-	-	-	-	No colaboration
64	Agrícola Puertallano	Fundo El Parrón	Clementina	Cachapoal	Las Cabras	1	4	14.0	7,770	-	-	-	-	-	No colaboration
65	South Pacific	South Pacific	Clementina	Cachapoal	Las Cabras	2	2	10.0	5,560						Only plants
66	Soc. Agrícola Tejas Verdes	Coliguito	Clementina	Cachapoal	Requinoa	5	7	10.5	7,019	5	25/May	04/ Jun	-	-	-
67	El Tabon	Fundo El Tabon	Clementina	Cachapoal	Las Cabras	5	2	29.3	16,294						Only plants
68	La Rosa Sofruco	La Rosa	Clem - Mandarinas - Tangerinas	Cachapoal	Peumo	10	23	29.0	19,304	41	24/May	04/ Jun	20/ Jul	-	-
69	La Rosa Sofruco	Sofruco	Tangerinas	Cachapoal	Peumo	1	17	0.6	289	2	21/Jul	21/ Jul	02/ Aug	-	-
Total Region								103.4	61,736	48					

TABLE 3

**PREVALENCE OF *Brevipalpus chilensis* IN CLEMENTINE, MANDARINE AND TANGERINE ORCHARDS
RESULTS ORCHARD BY ORCHARD
1999 SURVEY**

**Third
Region**

						Detection of <i>Brevipalpus chilensis</i>										
Name of Grower	Name of Orchard	Province	Town	Ha.	Total		Alive			Dead			Total	% Alive	Nº Specim en per fruit	
					Sam-ples	Fruit	Fruit	Pedi -cel	Tot.	Fruit	Pedi cel	Tot.				
1	Soc. Agr. Buenaventura	Buenaventura	Copiapó	Copiapó	1.0	1	30	18	9	27	7	5	12	39	69.23	0.900
2	Cítricos Uni Agri	Gibraltar	Copiapó	Copiapó	39.7	66	1,980	326	530	856	331	498	829	1,685	50.80	0.432
Total					40.7	67	2,010	344	539	883	338	503	841	1,724	51.22	0.439

Fourth Region

														Detection of <i>Brevipalpus chilensis</i>				
Name of Grower	Name of Orchard	Province	Town	Ha.	Total			Alive		Dead			Total	% Alive	Specimen per fruit			
					Samples	Fruit	Fruit	Pedice	Total	Fruit	Pedice	Total						
4	Guillermo Peñailillo Mora	Fundo Las Rojas	Elqui	La Serena	11.7	15	450	0	0	0	3	5	8	8	0.00	0.000		
5	Tamaya S.A.	Fundo La Laguna	Limarí	Ovalle	24.0	27	810	4	1	5	10	5	15	20	25.00	0.006		
8	Agronova	Camino Socos	Limarí	Ovalle	5.0	17	510	2	0	2	3	2	5	7	28.57	0.004		
9	Soc. Agr. Río Negro Ltda.	Río Negro	Limarí	Ovalle	18.0	19	570	3	1	4	3	0	3	7	57.14	0.007		
11	Villa Alegre	La Granja	Limarí	Punitaqui	43.2	52	1,560	1	4	5	2	0	2	7	71.43	0.003		
12	Villa Alegre	El Sauce	Elqui	Coquimbo	147.0	60	1,800	5	5	10	5	16	21	31	32.26	0.006		
13	Agr. Punitaqui	Fundo La Palma	Limarí	Monte Patria	39.7	24	720	31	27	58	55	50	105	163	35.58	0.081		
14	Prodex	Sector 10 y 1	Limarí	Monte Patria	26.3	30	900	4	4	8	33	25	58	66	12.12	0.009		
15	Mariano Díaz y Cía. Ltda.	Hijuela 5-B Tabalí	Limarí	Ovalle	17.9	21	630	0	0	0	0	0	0	0	-	0.000		
18	Soc. Agr. El Mirador	Fundo El Mirador	Limarí	Monte Patria	31.1	5	150	5	15	20	7	5	12	32	62.50	0.133		
19	Uni Agri	Las Represas	Limarí	Monte Patria	52.5	54	1,620	112	111	223	138	282	420	643	34.68	0.138		
20	Uni Agri	Camarico	Limarí	Punitaqui	25.1	26	780	163	196	359	197	156	353	712	50.42	0.460		
21	Uni Agri	Nueva Aurora	Limarí	Punitaqui	24.7	30	900	16	31	47	22	42	64	111	42.34	0.052		
22	Uni Agri	Santa Rosa	Limarí	Monte Patria	15.1	27	810	62	46	108	86	102	188	296	36.49	0.133		

23	Cítricos Uni Agri	Unicítricos	Limarí	Monte Patria	108.9	98	2,940	116	101	217	291	363	654	871	24.91	0.074
				Total	590.1	505	15,150	524	542	1,066	855	1,053	1,908	2,974	35.84	0.070

Fifth Region

Detection of *Brevipalpus chilensis*

	Name of Grower	Name of Orchard	Province	Town	Ha.	Total		Alive			Dead			Total	% Alive	Specimen per fruit
						Sam- ples	Fruit	Fruit	Pedi- cel	Total	Fruit	Pedi- cel	Total			
24	Agrícola Paiquen Ltda.	Colunquen	San Felipe	Panquehue	28.4	27	810	8	4	12	11	33	44	56	21.43	0.015
25	Agrícola Paiquen Ltda.	Santa Adela	San Felipe	Panquehue	22.5	27	810	2	3	5	5	2	7	12	41.67	0.006
26	La Higuera S.A.	La Higuera	San Felipe	Santa María	22.3	3	90	1	0	1	4	2	6	7	14.29	0.011
27	Agrícola San Juan	Fundo Chai-Chai	Petorca	Cabildo	3.9	6	180	0	1	1	6	2	8	9	11.11	0.006
28	Agrícola El Canelillo	Fundo El Encierro	Quillota	La Cruz	24.2	27	810	29	12	41	52	48	100	141	29.08	0.051
29	Agrícola El Roble	Fdo. Sta. Marta de Longotoma	Petorca	La Ligua	4.5	9	270	7	4	11	11	13	24	35	31.43	0.041
30	Agrícola Huerto California	El Total	Quillota	Hijuelas	2.0	3	90	1	1	2	2	0	2	4	50.00	0.022
31	Agrícola Peppi Ltda.	Parcela El Romero	Quillota	Limache	8.5	15	450	32	13	45	32	27	59	104	43.27	0.100
32	Agrícola Pucará Ltda.	Parcela N° 39	Petorca	La Ligua	4.0	6	180	0	1	1	0	0	0	1	100.00	0.006
33	Agrícola Pullacón	Bien Común N°7, San Lorenzo	Petorca	Cabildo	21.5	24	720	15	4	19	38	26	64	83	22.89	0.026
34	Agrícola Santa Cecilia	Parcela N° 17	Quillota	Nogales	12.9	18	540	6	2	8	14	22	36	44	18.18	0.015
35	Agromol Ltda.	Huerto El Maitén	Quillota	Hijuelas	9.8	15	450	10	11	21	23	31	54	75	28.00	0.047
36	Héctor Bozzolo Palma	Bien Común	Petorca	Cabildo	11.4	15	450	19	10	29	23	20	43	72	40.28	0.064

		Nº7, San Lorenzo														
37	Desarrollo Agrario	Fdo. Los Calderones de la Peña	San Felipe	Llay Llay	8.1	8	240	32	63	95	64	84	148	243	39.09	0.396
38	María Paz Greene	Fundo Maitenes	Quillota	Hijuelas	9.0	17	510	1	2	3	1	3	4	7	42.86	0.006
39	Rodrigo Manubens	Parcela Los Pinos	Quillota	La Cruz	5.7	9	270	25	9	34	59	33	92	126	26.98	0.126
40	Eduardo Moltedo	El Caqui-Parcela 39	Quillota	La Calera	2.5	6	180	35	26	61	59	33	92	153	39.87	0.339
41	Carlos Moraga	Parcela El Denuncio	Quillota	Nogales	16.6	21	630	86	82	168	87	93	180	348	48.28	0.267
42	Hugo Muñoz	Parcela Carén	Quillota	Quillota	7.3	12	360	10	0	10	11	0	11	21	47.62	0.028
43	Miguel Nenadovic	Fundo El Sauce	Los Andes	Los Andes	4.4	6	180	14	29	43	30	53	83	126	34.13	0.239
44	Eduardo Seidemann	Ariztía S/N	Quillota	Quillota	3.2	6	180	3	0	3	3	0	3	6	50.00	0.017
45	S.C.A. Los Rosales de Sta. Julia	Chacra Los Rosales	Quillota	Quillota	2.6	6	180	49	17	66	24	25	49	115	57.39	0.367
46	Agrícola Huerto California	Entre Ríos	Quillota	San Isidro	3.0	9	270	5	0	5	15	0	15	20	25.00	0.019
47	Raimundo del Río	Inversiones Tesalía Ltda.	San Felipe	San Felipe	1.2	2	60	2	1	3	9	19	28	31	9.68	0.050
Total					239.3	297	8,910	392	295	687	583	569	1,152	1,839	37.36	0.077

Metropolitan Region

															Detection of <i>Brevipalpus chilensis</i>				
Name of Grower	Name of Orchard	Province	Town	Ha.	Total		Alive			Dead			Total	% Alive	Specimen per fruit				
					Sam- ples	Fruit	Fruit	Pedi- cel	Total	Fruit	Pedi- cel	Total							
51	Fernando Cerda	Parcela N° 2 Santa Teresa	Melipilla	Mallarauco	2.0	6	180	13	12	25	22	46	68	93	26.88	0.139			
52	Saplums	La Parva	Chacabuco	Colina	5.9	12	360	11	10	21	10	21	31	52	40.38	0.058			
53	Luz Montes Bollo	Parcela N° 14	Melipilla	Melipilla	8.9	15	450	7	0	7	10	3	13	20	35.00	0.016			
54	Rafael Palacios	Las Pataguas - La Puntilla	Maipo	Paine	9.4	12	360	0	0	0	0	2	2	2	0.00	0.000			
55	Friederike Volkenborn	Talhuen	Melipilla	María Pinto	10.0	12	360	1	9	10	15	20	35	45	22.22	0.028			
56	Enrique Alcalde		Talagante	Isla de Maipo	8.6	15	450	40	43	83	50	68	118	201	41.29	0.184			
57	Agrícola Las Petras	Fundo Perales	Melipilla	María Pinto	3.5	6	180	0	0	0	2	4	6	6	0.00	0.000			
58	Agr. M ^a Luz Baldrich Ltda.	Reserva Hijueta Siete	Melipilla	Melipilla	8.2	15	450	6	5	11	9	13	22	33	33.33	0.024			
60	Rafael Palacios	Collipeum o Bajo	Maipo	Paine	3.0	6	180	0	2	2	3	3	6	8	25.00	0.011			
62	Soc. Primavera Agrícola Ltda.	Fundo La Primavera	Maipo	Calera de Tango	3.8	9	270	0	1	1	2	3	5	6	16.67	0.004			
Total					63.3	108	3,240	78	82	160	123	183	306	466	34.33	0.049			

Sixth Region

Detection of <i>Brevipalpus chilensis</i>																
	Name of Grower	Name of Orchard	Province	Town	Ha.	Total		Alive			Dead			Total	% Alive	Specimen per fruit
						Sam-ples	Fruits	Fruit	Ped-icel	Total	Fruit	Pedi-cel	Total			
66	Soc Agr. Tejas Verdes	Coligüito	Cachapoal	Requinoa	10.5	5	150	0	0	0	0	0	0	0	-	0.000
68	La Rosa Sofruco	La Rosa	Cachapoal	Peumo	29.0	41	1,230	3	3	6	14	8	22	28	21.43	0.005
69	La Rosa Sofruco	Sofruco	Cachapoal	Peumo	0.6	2	60	0	0	0	0	0	0	0	-	0.000
Total					40.1	48	1,440	3	3	6	14	8	22	28	21.43	0.004

TABLE 4

**NATURAL LOCATION OF *Brevipalpus chilensis* ON FRUITS AND PEDICELS OF CLEMENTINES, MANDARINES AND TANGERINES
EN DIFERENTES PERÍODOS DE MUESTREO
1999 SURVEY**

Detection of <i>Brevipalpus chilensis</i> (N° of specimen)								
REGION	Monitoring N° 1		Monitoring N° 2		Monitoring N° 3		Total N° of samples evaluated	Total N° of fruits evaluated
	ON FRUIT	ON PEDICEL	ON FRUIT	ON PEDICEL	ON FRUIT	ON PEDICEL		
III	313	387	192	280	177	375	67	2,010
IV	514	575	446	474	439	526	505	15,150
V	370	351	300	238	305	275	297	8,910
RM	64	99	78	113	59	53	108	3,240
VI	15	9	2	2	0	0	48	1,440
Total	1,276	1,421	1,018	1,107	980	1,229	1,025	30,750

PERCENTAGE DISTRIBUTION OF *Brevipalpus chilensis* ON FRUITS AND PEDICELS OF CLEMENTINES, MANDARINES AND TANGERINES

Detection of <i>Brevipalpus chilensis</i> (%)								
REGION	Monitoring N° 1		Monitoring N° 2		Monitoring N° 3		Total N° of samples evaluated	Total N° of fruits evaluated
	ON FRUIT	ON PEDICEL	ON FRUIT	ON PEDICEL	ON FRUIT	ON PEDICEL		
III	44.7	55.3	40.7	59.3	32.1	67.9	67	2,010
IV	47.2	52.8	48.5	51.5	45.5	54.5	505	15,150
V	51.3	48.7	55.8	44.2	52.6	47.4	297	8,910
RM	39.3	60.7	40.8	59.2	52.7	47.3	108	3,240
VI	62.5	37.5	50.0	50.0	-	-	48	1,440
Total	47.3	52.7	47.9	52.1	44.4	55.6	1,025	30,750

TABLE 5

OTHER INSECTS AND MITES DETECTED IN CLEMENTINE, MANDARINE AND TANGERINE ORCHARDS.

1999 SURVEY

Name of Grower	Name of Orchard	Region	Province	Commune	Sampling Date	Identification
Cítricos Uni Agri	Gibraltar	III	Copiapó	Copiapó	18-Jun	<i>Thrips tabaci</i>
Cítricos Uni Agri	Gibraltar	III	Copiapó	Copiapó	2-Jul	<i>Planococcus citri/Thrips tabaci</i>
Cítricos Uni Agri	Gibraltar	III	Copiapó	Copiapó	2-Jul	<i>Pseudococcus sp./Planococcus sp./Thrips tabaci</i>
Guillermo Peña y Lillo Mora	Fundo Las Rojas	IV	Elqui	La Serena	23-Jun	<i>Pseudococcus longispinus</i>
Tamaya S.A.	Fundo La Laguna	IV	Limarí	Ovalle	5-May	<i>Pseudococcus longispinus</i>
Agronova	Camino Socos	IV	Limarí	Ovalle	31-May	<i>Pseudococcus longispinus/Planococcus citri</i>
Agronova	Camino Socos	IV	Limarí	Ovalle	28-Jun	<i>Pseudococcus sp./Pseudococcus longispinus</i>
Agronova	Camino Socos	IV	Limarí	Ovalle	9-Jul	<i>Pseudococcus longispinus</i>
Soc. Agr. Río Negro Ltda.	Río Negro	IV	Limarí	Ovalle	10-Jul	<i>Pseudococcus longispinus</i>
Villa Alegre	La Granja	IV	Limarí	Punitaqui	8-Jul	<i>Coccus hesperidum</i>
Villa Alegre	El Sauce	IV	Elqui	Coquimbo	05-Aug	<i>grupo sin importancia agrícola</i>
Prodex	Sector 1 y 10	IV	Limarí	Monte Patria	1-Jun	<i>Icerya purchasi</i>
Mariano Díaz y Cía. Ltda.	Hijuela 5-B Tabalí	IV	Limarí	Ovalle	9-Jul	<i>Tetranychus sp.</i>
Mariano Díaz y Cía. Ltda.	Hijuela 5-B Tabalí	IV	Limarí	Ovalle	13-Jul	<i>Pseudococcus longispinus/Pseudococcus sp.</i>
Soc. Agr. El Mirador	Viejo	IV	Limarí	Monte Patria	13-May	<i>Icerya purchasi</i>
Uni Agri	Camarico	IV	Limarí	Punitaqui	10-Jun	<i>Pseudococcus viburni</i>
Uni Agri	Camarico	IV	Limarí	Punitaqui	10-Jun	<i>Pseudococcus longispinus/Planococcus citri</i>

Uni Agri	Camarico	IV	Limarí	Punitaqui	6-Jul	<i>Pseudococcus longispinus/Pseudococcus affinis</i>
Saplums	La Parva	RM	Santiago	Colina	14-May	<i>Icerya purchasi</i>
Saplums	La Parva	RM	Santiago	Colina	16-Jun	<i>Icerya purchasi/Thrips tabaci</i>
Luz Montes Bollo	Parcela N° 14	RM	Melipilla	Melipilla	6-Jul	<i>Icerya purchasi</i>
Enrique Alcalde	Enrique Alcalde	RM	Talagante	Isla de Maipo	18-Jun	<i>Post. Ac. Tenuipalpidae</i>
Enrique Alcalde	Enrique Alcalde	RM	Talagante	Isla de Maipo	6-Jul	<i>Pseudococcus longispinus/Icerya purchasi</i>
Enrique Alcalde	Enrique Alcalde	RM	Talagante	Isla de Maipo	20-Jul	<i>Pseudococcus longispinus/Pseudococcus sp.</i>
Soc. Primavera Agr. Ltda.	Fundo La Primavera	RM	Maipo	Calera de Tango	11-Jun	<i>Pseudococcus longispinus</i>
Soc. Primavera Agr. Ltda.	Fundo La Primavera	RM	Maipo	Calera de Tango	25-Jun	<i>Pseudococcus longispinus/Pseudococcus sp.</i>
Soc. Primavera Agr. Ltda.	Fundo La Primavera	RM	Maipo	Calera de Tango	25-Jun	<i>Pseudococcus longispinus/Pseudococcus sp.</i>
Soc. Primavera Agr. Ltda.	Fundo La Primavera	RM	Maipo	Calera de Tango	15-Jul	<i>Pseudococcus longispinus</i>

ANNEX 2

POPULATION MONITORING OF
Brevipalpus chilensis
IN CLEMENTINE ORCHARDS
IN THE IV AND V REGIONS
OF CHILE

(UNIVERSIDAD CATOLICA DE VALPARAISO)

UCV



POPULATION FOLLOW UP ON *Brevipalpus chilensis* BAKER IN
CLEMENTINE ORCHARDS IN THE IV AND V REGIONS

CHILE

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INTRODUCTION

Brevipalpus chilensis is a mite associated with deciduous fruit crops (*e.g.*, grapevines and kiwi plants) as well as fruit trees with evergreen leaves, like citrus and cherimoya. Its population behavior has been mainly studied in deciduous species. There is no information generated from population follow-ups to shed light on its behavior on evergreen fruit species.

The following study was conducted to measure *B. chilensis* population fluctuations during the course of the year, to establish the existence of overwintering stages and to determine the plant structures used by *B. chilensis* as locations for feeding and reproduction.

MATERIALS AND METHODS

The research was carried out between March 2000 and March 2001. Four commercial clementine orchards were selected, two in the IV Region and two in the V Region, all of them with data of previous *B. chilensis* detection. In the IV Region the population follow up was carried out in the El Palqui (UNIAGRI) and Ovalle Bajo (AGRINOVA) sectors; in the V Region the research was conducted at Cabildo and Nogales.

Samples were taken every 15 days. Each sample consisted of a total of 12 fruits, 4 twigs of 20 cm and 80 leaves per tree from 10 preselected trees at every orchard. For the purpose of this study and so as not to interfere with the population development of *B. chilensis*, the preselected trees were excluded from the usual chemical treatments applied in the orchards.

Samples were collected in paper bags, identified with the date of sampling, the orchard sampled as well as the tree from which the sample had been taken. The samples were immediately taken to the Entomology Laboratory of the Catholic University of Valparaiso (UCV); the eggs and mobile stages present on leaves, twigs and fruits were counted using a stereoscopic magnifying glass. Two different tallies were made for fruit, one for the findings on the fruit surface and one for the findings under the pedicel.

The data for each orchard were tabulated and total mite population counts or population counts by plant part sampled were graphed.

RESULTS AND DISCUSSION.

Fig. 1 shows total populations of *B. chilensis* obtained from all samples from every orchard under study. The study finds that the population shows a gradual decline, starting with the samples taken in March and until winter. At the end of winter, a new rise in population is observed, a rise that eventually shows an important increase between December and March. A strong population increase was observed especially at Cabildo and at El Palqui (Uniagri). On the other hand, since the orchard at Ovalle Bajo presented a scarce population throughout the whole sampling year, we found that the natural *B. chilensis* populations, although influenced by climatic conditions, most probably are numerically regulated by management conditions that are characteristic of each orchard.

It must also be stated that *B. chilensis* did not achieve pest status in any of the orchards sampled in this study nor was it associated with any injurious symptoms that could be attributed to its presence.

When studying the presence of *B. chilensis* with regard to its distribution on various plant parts sampled (Fig. 2,3,4,5), we found that, in general, larger numbers of mites were detected on fruits and twigs; mite detection on twigs is largest after finishing the harvest when no fruits are available for *B. chilensis*

development. Once the fruits start developing after fruit set, *B. chilensis* colonies start moving from the twigs to the fruits and prefer to stay on them.

When analyzing the mite's behavior on the fruit, differences can be observed with regard to the mite's location, whether on the fruit surface or under the pedicel. Fig. 6, 7, 8 and 9 show the distribution of *B. chilensis* on fruits from the four monitored orchards.

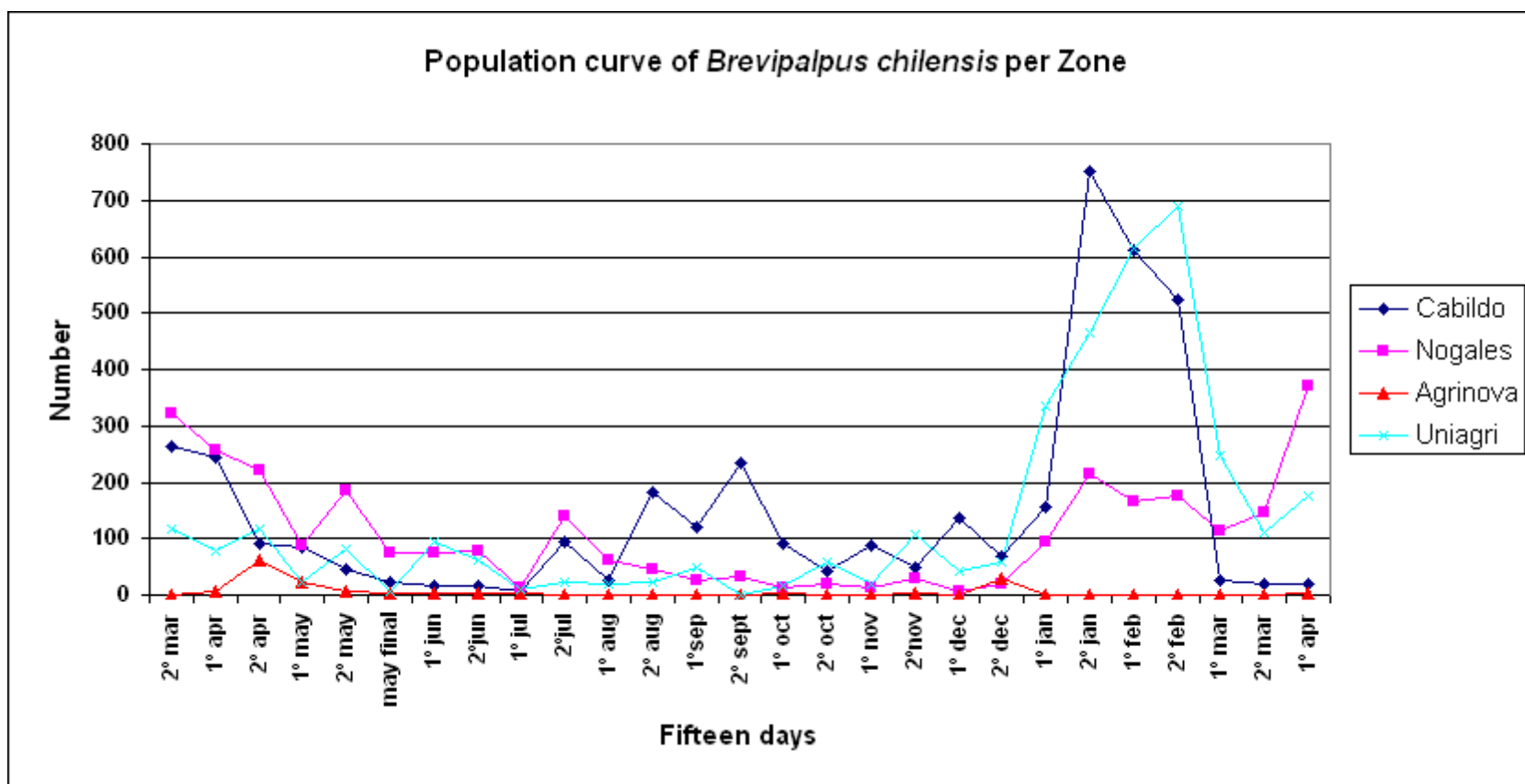
Those Figures show that the mite prefers the pedicel as its favorite place of colonization, since under the pedicel they find a place offering them shelter and food. A more erratic colonization behavior was observed at the Nogales orchard, where the mite's preferences for one location or another strongly fluctuated from one sample to the other.

Regarding *B. chilensis* populations on clementine, mandarin and tangerine plants in winter, they are made up both of mobile stages and of eggs. In this way, their behavior on citrus is different from deciduous fruit trees, where the mite overwinters as a fertilized female.

Population follow up of *Brevipalpus chilensis*

Fig 1. Summary population follow up of *Brevipalpus chilensis* per sample

Fifteen days																												
Orchards	2º mar	1º apr	2º apr	1º may	2º may	may final	1º jun	2ºjun	1º jul	2ºjul	1º aug	2º aug	1ºsep	2º sept	1º oct	2º oct	1º nov	2ºnov	1º dec	2º dec	1º jan	2º jan	1º feb	2º feb	1º mar	2º mar	1º apr	
Cabildo	263	244	91	83	46	24	16	16	7	94	26	181	120	235	92	43	88	50	136	67	156	752	611	523	26	18	19	
Nogales	322	258	222	88	186	75	76	77	12	140	61	46	25	34	14	19	12	28	7	18	95	215	165	174	114	147	372	
Agrinova	0	5	63	22	6	2	4	2	4	1	0	1	0	0	2	0	1	4	0	28	1	0	0	0	0	0	3	
Uniagri	116	78	116	22	80	6	95	62	9	22	18	22	48	1	15	57	20	108	42	59	334	465	616	691	247	109	175	



Population follow up of *Brevipalpus chilensis*

Fig 2. Population follow up of *Brevipalpus chilensis* per structure

	Date																								
	21-Mar	07-Apr	18-Apr	2-May	16-May	30-May	11-Jul	26-Jul	08-Aug	23-Aug	5-Sep	3-Oct	16-Oct	31-Oct	17-Nov	28-Nov	12-Dec	29-Dec	09-Jan	26-Jan	8-Feb	21-Feb	8-Mar	20-Mar	04-Apr
Leaves	0.108	0.094	0.041	0.023	0.014	0	0	0.001	0.003	0.055	0.008	0.006	0	0.005	0.015	0.056	0.091	0.034	0	0.276	0.201	0.23	0	0	0
Twigs	0.65	1.55	0.3	0.83	0.15	0.25	0.1	0.68	0.05	0.73	0.4	0.98	2.3	0.98	1.9	0.13	1.58	1	0.83	4.08	3.03	2.05	0.23	0.05	0.28
Fruits	1.26	0.89	0.39	0.27	0.24	0.12	0.03	0.55	0.18	0.9	0.82	1.6	0	0	0	0	0	0	1.03	3.07	2.74	2.14	0.14	0.13	0.07

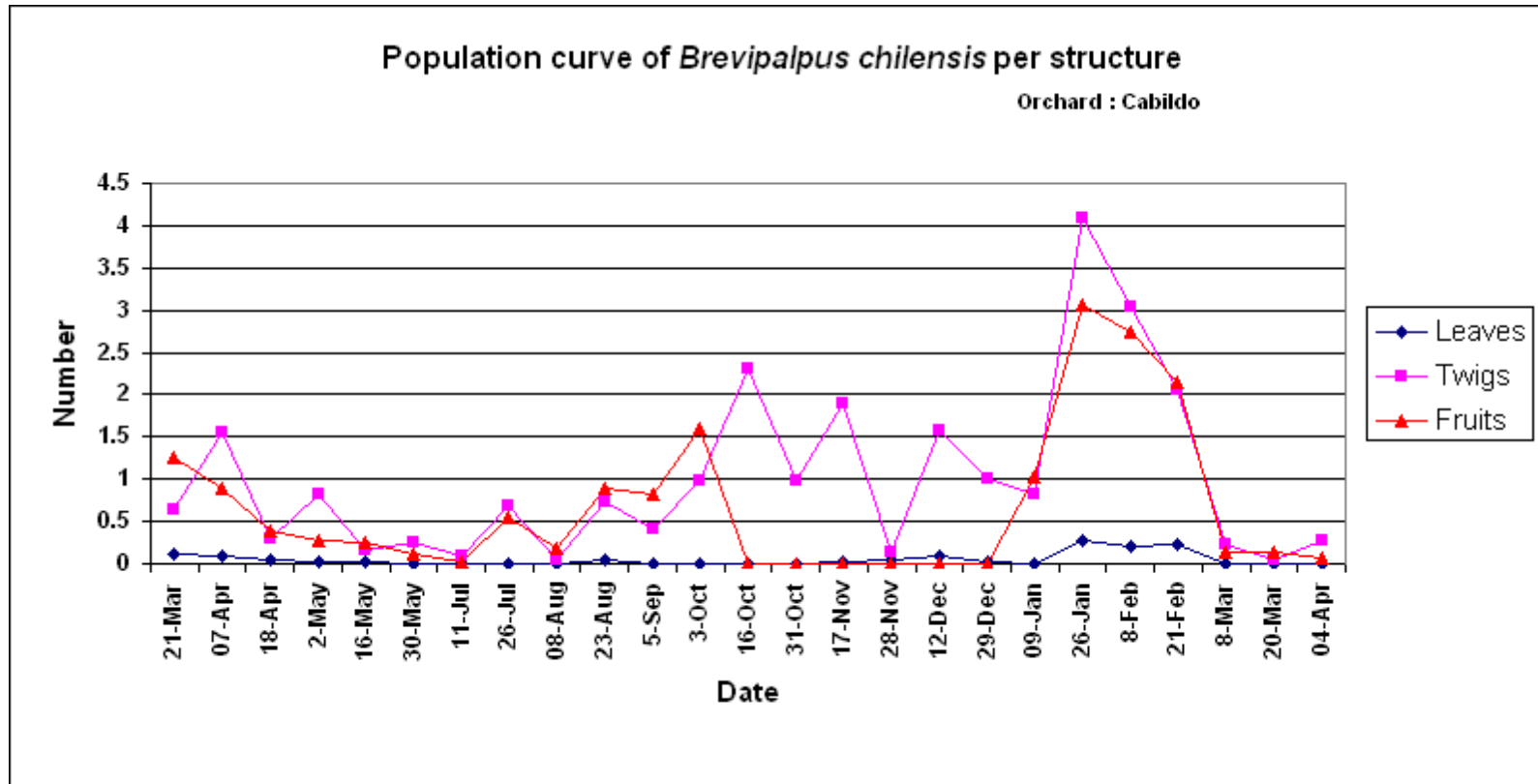


Fig 3. Population follow up of *Brevipalpus chilensis* per structure

	Date																									
	22-Mar	05-Apr	25-Apr	5-May	16-May	30-May	28-Jun	12-Jul	26-Jul	08-Aug	23-Aug	5-Sep	4-Oct	17-Oct	31-Oct	16-Nov	28-Nov	13-Dec	28-Dec	10-Jan	25-Jan	7-Feb	22-Feb	8-Mar	22-Mar	04-Apr
Leaves	0.04	0.01	0.02	0	0.01	0	0	0.01	0	0.01	0	0.01	0.01	0	0	0	0	0	0	0	0	0.01	0.01	0	0	0
Twigs	0.15	0.8	1.33	0.25	0.48	0.3	0.28	0	0.78	0.05	0.13	0.33	0.35	0.35	0.48	0.28	0.7	0.18	0.45	1.6	3.2	0.5	0.73	0.68	0.1	0.65
Fruits	2.36	1.8	1.25	0.63	1.35	0.52	0.55	0.06	0.91	0.42	0.34	0.05	0.14	0	0	0	0	0	0	0.23	0.72	1.16	1.13	0.73	1.19	1.84

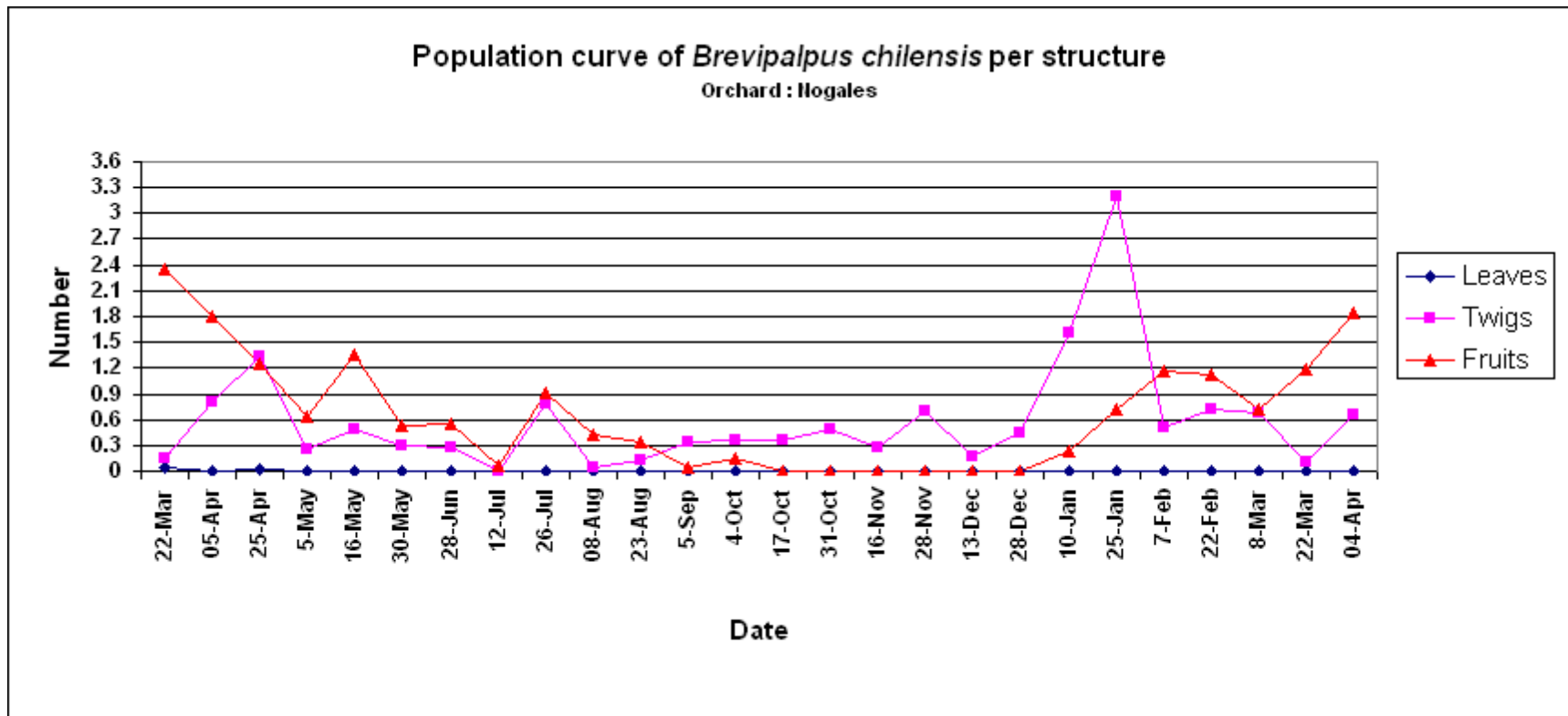


Fig 4. Population follow up of *Brevipalpus chilensis* per structure

	Date																											
	17-Mar	31-Mar	11-Apr	25-Apr	10-May	24-May	7-Jun	20-Jun	4-Jul	18-Jul	02-Aug	16-Aug	29-Aug	27-Sep	12-Oct	26-Oct	8-Nov	21-Nov	05-Dec	21-Dec	04-Jan	17-Jan	31-Jan	15-Feb	28-Feb	15-Mar	29-Mar	
Leaves	0	0	0.045	0.008	0	0.003	0	0	0	0	0	0	0	0	0	0	0	0	0	0.001	0	0	0	0	0	0	0	
Twigs	0	0.075	0.025	0.05	0	0	0.025	0	0.1	0	0	0	0	0	0	0	0.025	0.1	0	0.675	0	0	0	0	0	0	0	
Fruits	0	0.02	0.22	0.12	0.05	0	0.03	0.02	0	0.01	0	0.01	0	0	0.02	0	0	0	0	0	0.01	0	0	0	0	0	0.03	

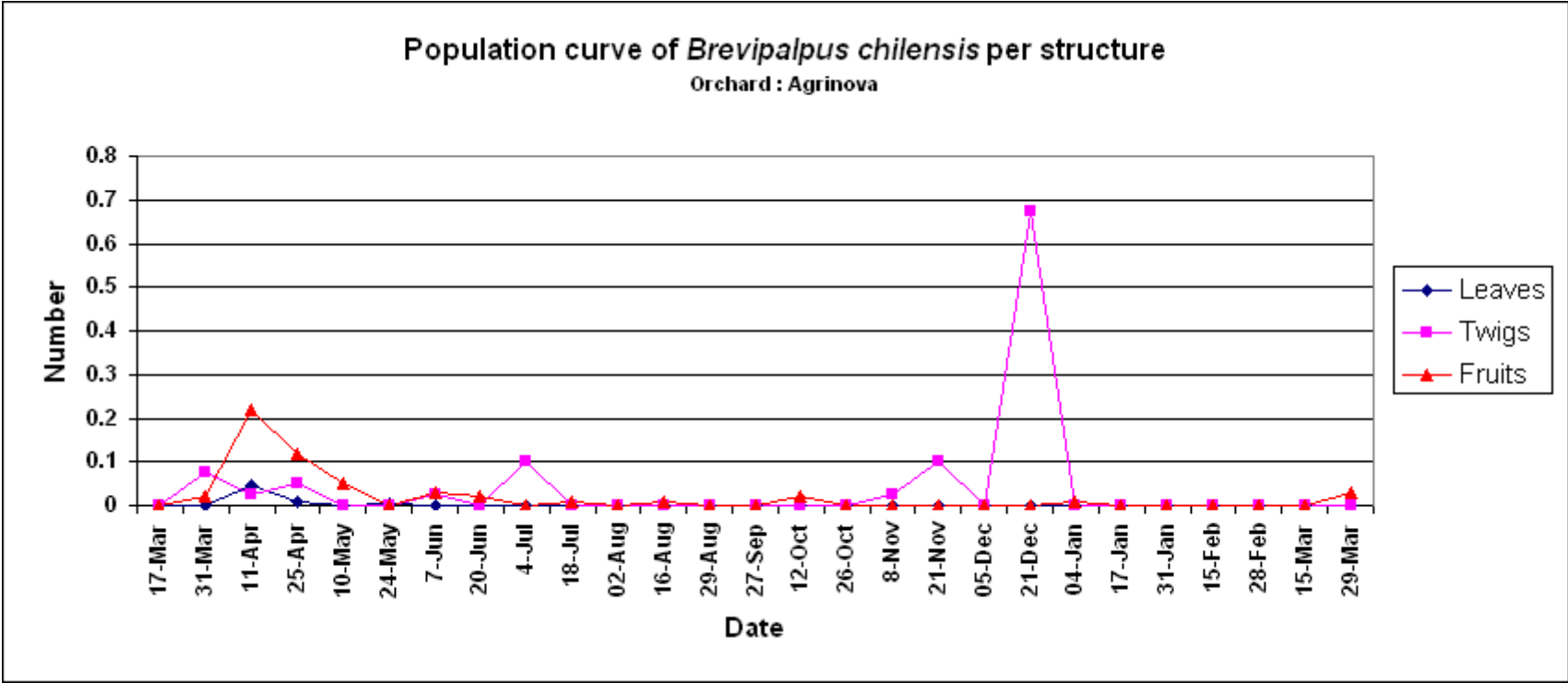


Fig 5. Population follow up of *Brevipalpus chilensis* per structure

	Date																										
	17-Mar	31-Mar	11-Apr	25-Apr	10-May	24-May	7-Jun	20-Jun	4-Jul	18-Jul	02-Aug	16-Aug	28-Aug	27-Sep	12-Oct	27-Oct	8-Nov	22-Nov	06-Dec	21-Dec	04-Jan	17-Jan	31-Jan	15-Feb	28-Feb	14-Mar	
Leaves	3.63	0.13	1.75	0	0	0	0	0	0.25	0	0.13	0	0	0	0	0	0	0	0	0	0	1.38	0.25	0.75	1.63	0	0
Twigs	0.88	0.25	0.9	0.08	0.05	0	0.03	0.18	0.18	0.13	0.1	0	1.2	0.03	0.38	1.43	0.5	2.7	1.05	0.83	5.8	2.28	1.5	6.6	1.95	0.8	
Fruits	0.44	0.56	0.55	0.16	0.65	0.06	0.78	0.46	0	0.15	0.11	0.18	0	0	0	0	0	0	0	0.22	0.76	3.1	4.58	3.45	1.41	0.64	

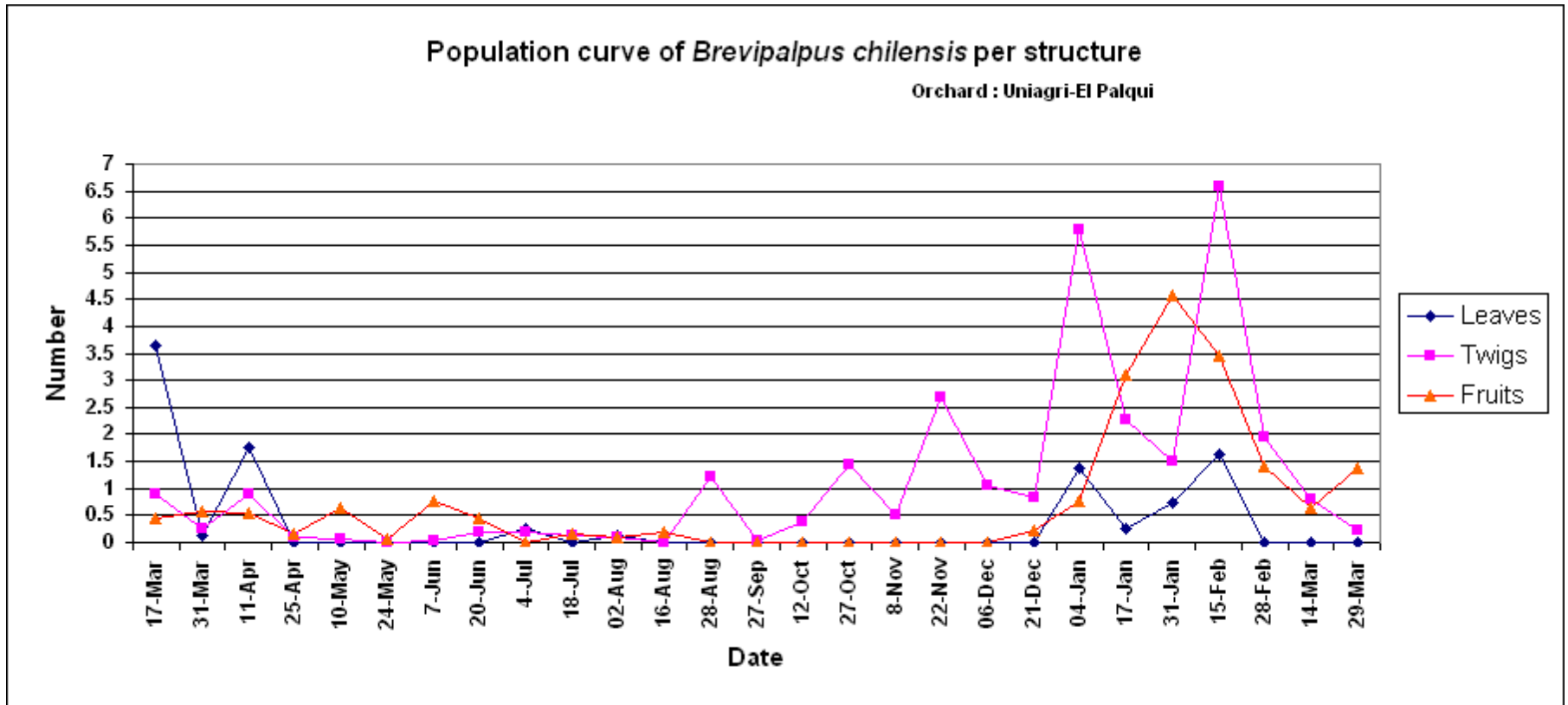


Fig 6. Population follow up of *Brevipalpus chilensis* per fruit

	Date																								
	21-Mar	07-Apr	18-Apr	2-May	16-May	30-May	11-Jul	26-Jul	08-Aug	23-Aug	5-Sep	3-Oct	16-Oct	31-Oct	17-Nov	28-Nov	12-Dec	29-Dec	09-Jan	26-Jan	8-Feb	21-Feb	8-Mar	20-Mar	04-Apr
Fruit surface	0.93	0.33	0.21	0.1	0.11	0.09	0.01	0.27	0.04	0.64	0.62	0.72	0	0	0	0	0	0	0.38	2.1	2.7	1.98	0.09	0.09	0.03
Pedichel	0.33	0.56	0.18	0.17	0.13	0.03	0.02	0.28	0.14	0.26	0.2	0.88	0	0	0	0	0	0	0.65	0.97	0.04	0.16	0.05	0.04	0.04

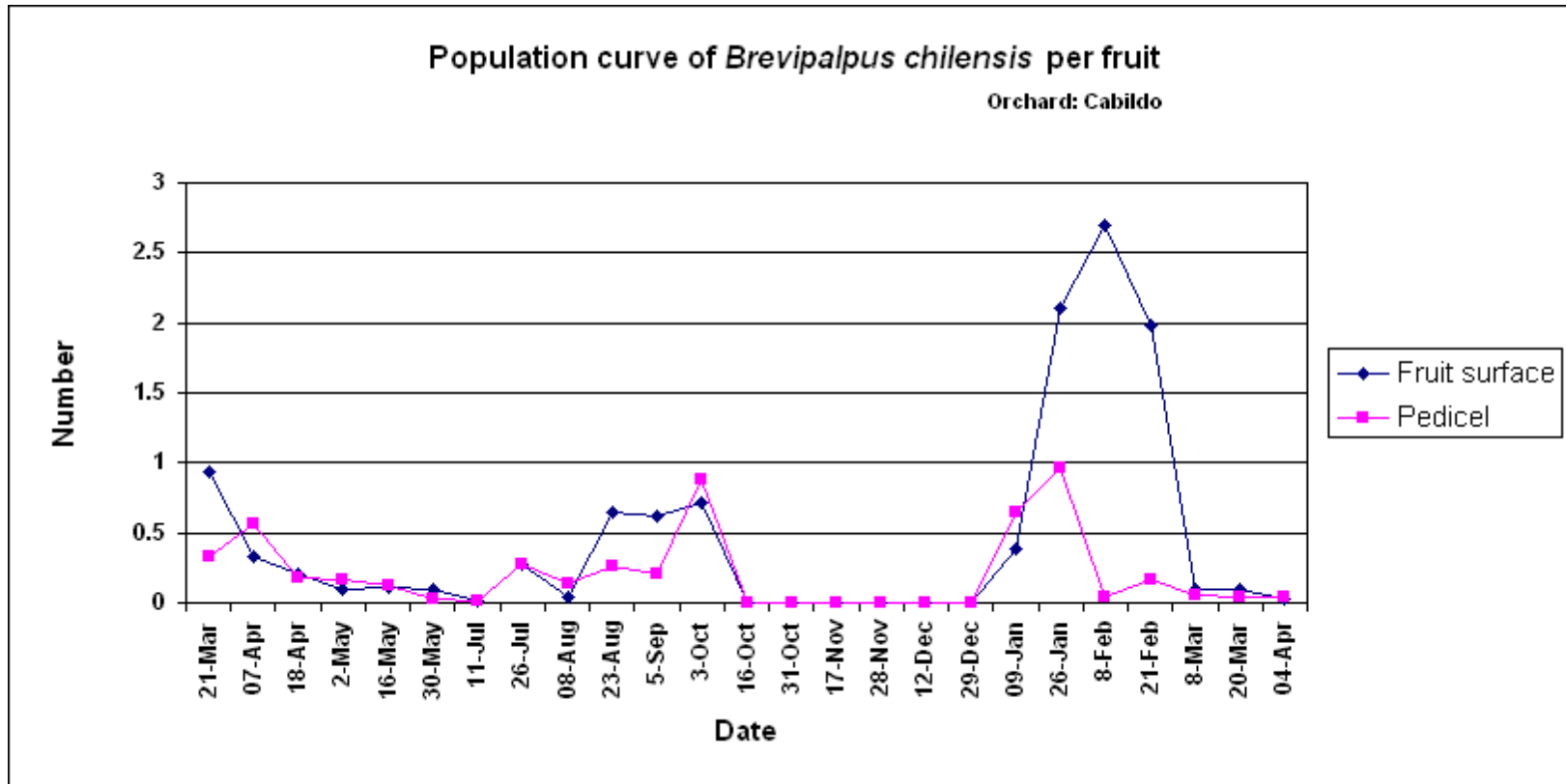


Fig 7. Population follow up of *Brevipalpus chilensis* per fruit

	Date																									
	22-Mar	05-Apr	25-Apr	5-May	16-May	30-May	28-Jun	12-Jul	26-Jul	08-Aug	23-Aug	5-Sep	4-Oct	17-Oct	31-Oct	16-Nov	28-Nov	13-Dec	28-Dec	10-Jan	25-Jan	7-Feb	22-Feb	8-Mar	22-Mar	04-Apr
Fruit surface	1.26	1.2	0.81	0.32	0.68	0.23	0.28	0.03	0.41	0.24	0.1	0.02	0.02	0	0	0	0	0	0	0.08	0.18	0.69	0.57	0.27	0.68	1.84
Pediceal	1.1	0.6	0.45	0.31	0.67	0.29	0.27	0.03	0.5	0.18	0.24	0.03	0.12	0	0	0	0	0	0	0.15	0.54	0.47	0.56	0.46	0.51	1,03

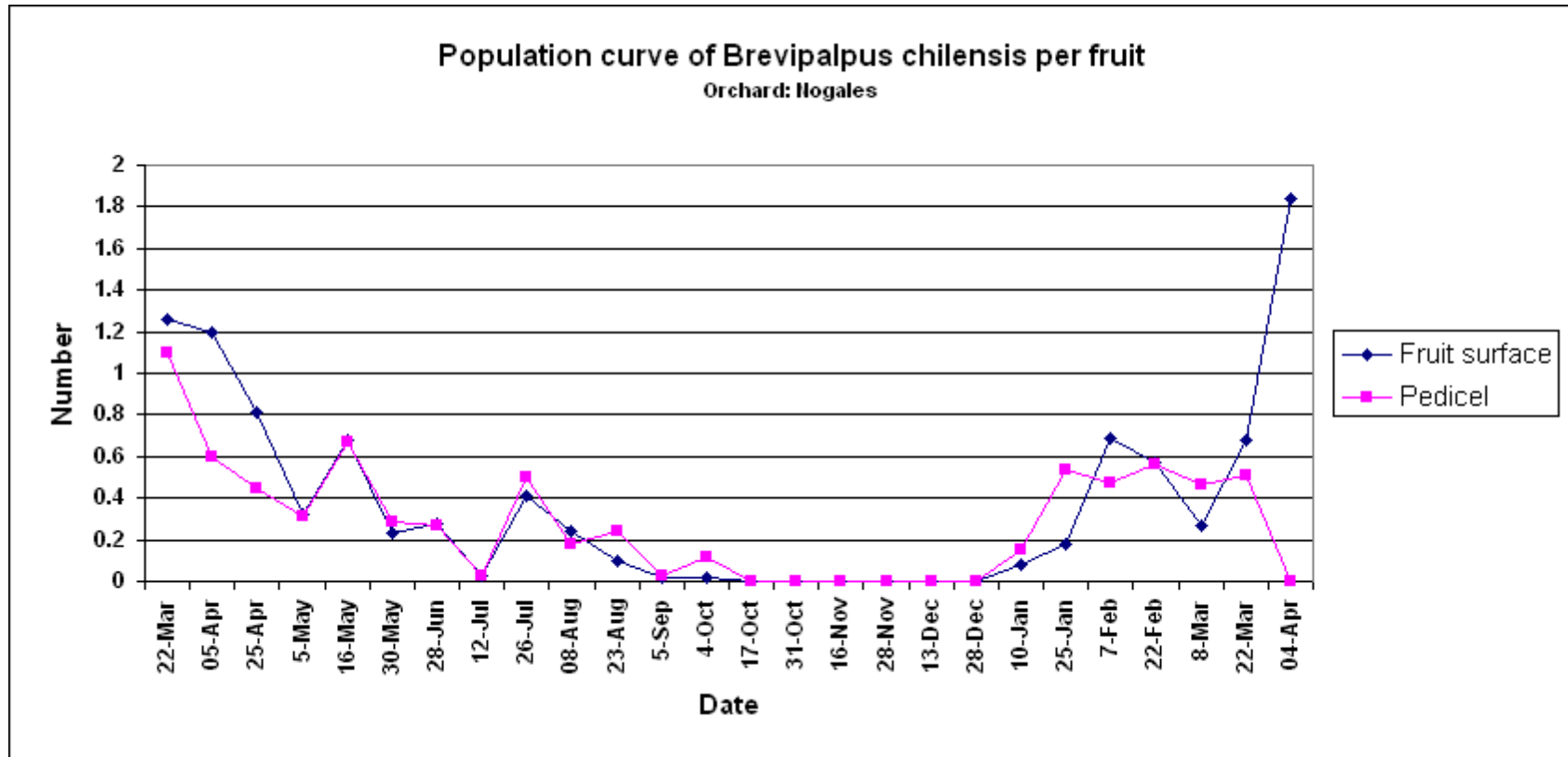


Fig 8. Population follow up of *Brevipalpus chilensis* per fruit

	Date																										
	17-Mar	31-Mar	11-Apr	25-Apr	10-May	24-May	7-Jun	20-Jun	4-Jul	18-Jul	02-Aug	16-Aug	29-Aug	27-Sep	12-Oct	26-Oct	8-Nov	21-Nov	05-Dec	21-Dec	04-Jan	17-Jan	31-Jan	15-Feb	28-Feb	15-Mar	29-Mar
Fruit surface	0	0.01	0.11	0.07	0.01	0	0	0	0	0.01	0	0.01	0	0	0.01	0	0	0	0	0	0	0	0	0	0	0	0
Pedichel	0	0.01	0.11	0.05	0.04	0	0.03	0.02	0	0	0	0	0	0	0.01	0	0	0	0	0	0.01	0	0	0	0	0	0.03

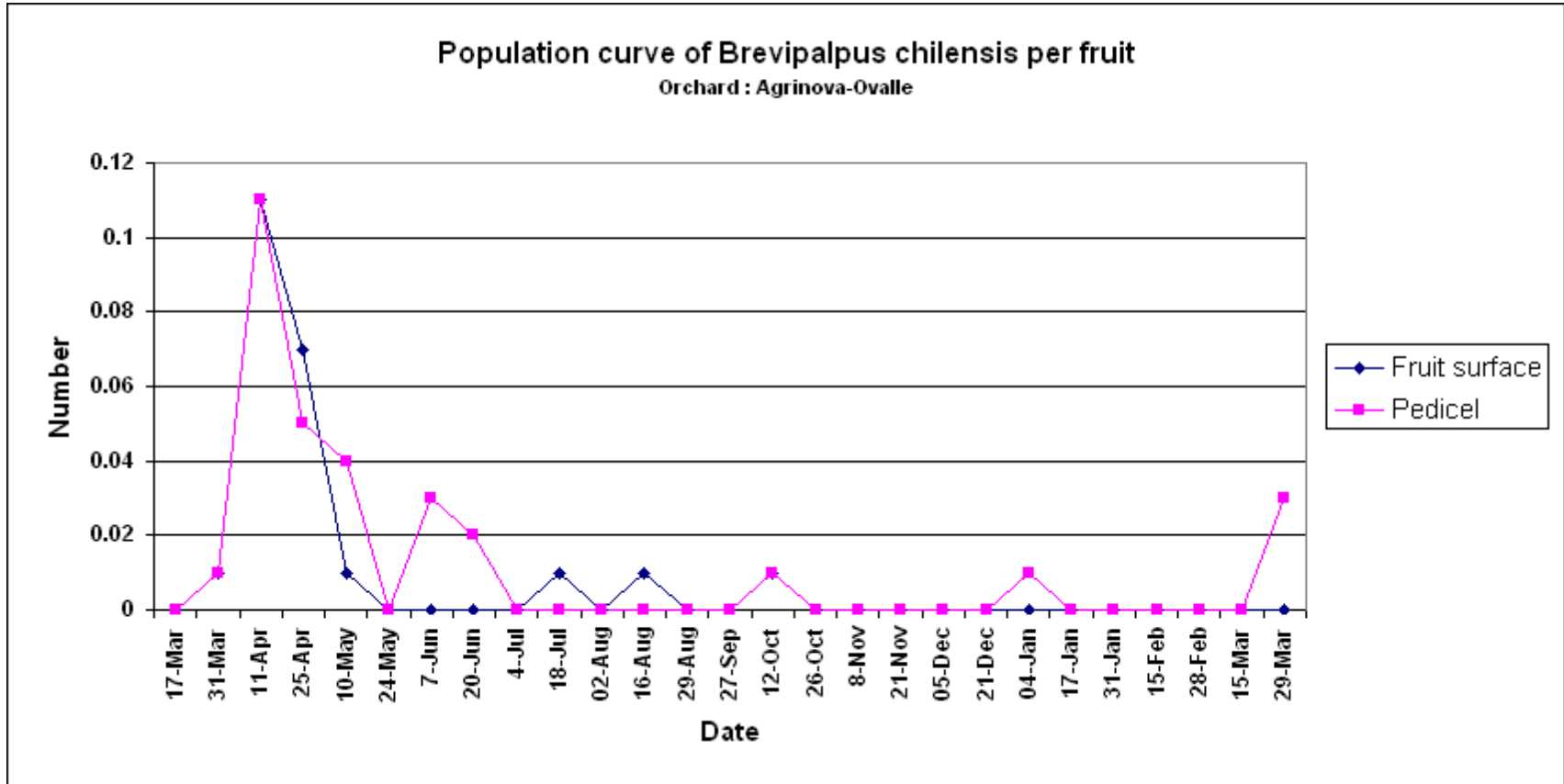
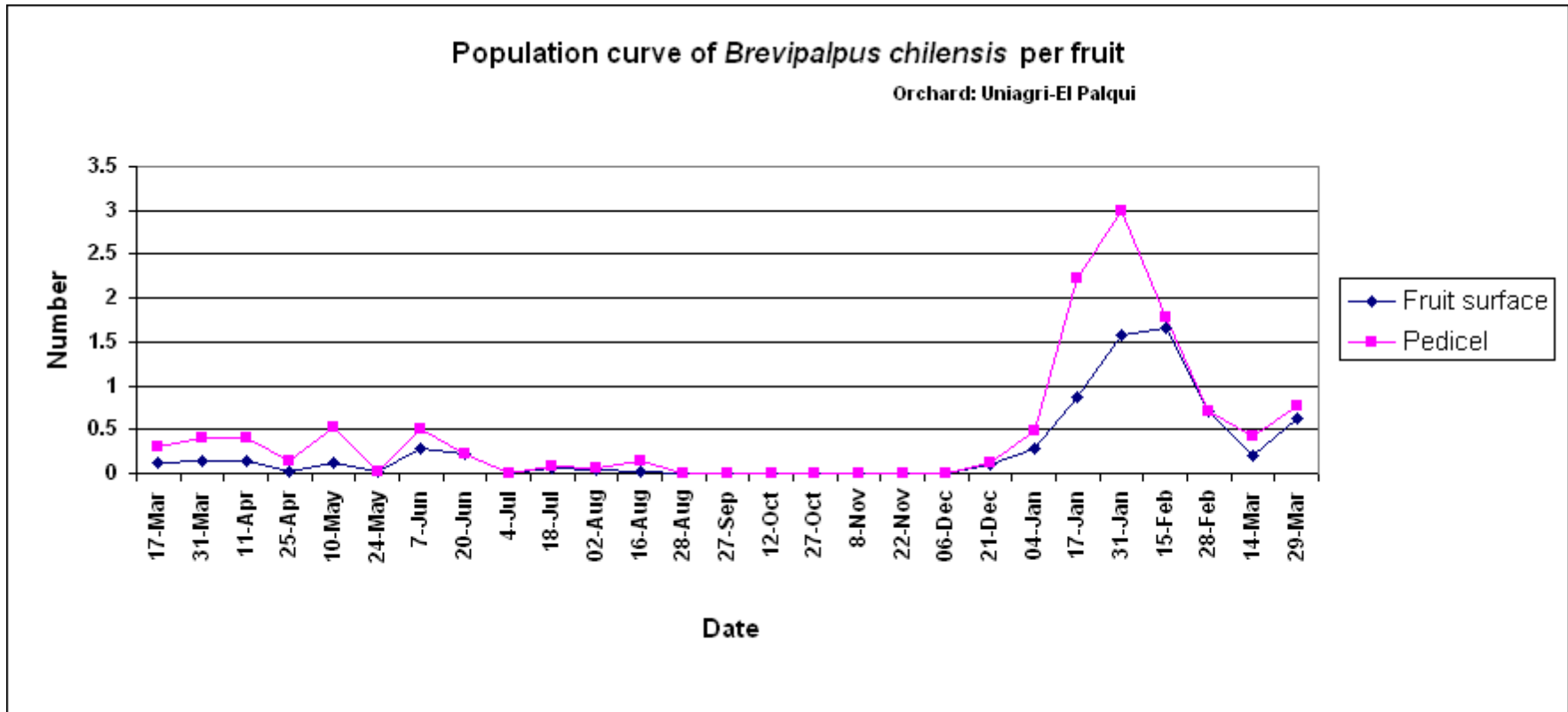


Fig 9. Population follow up of *Brevipalpus chilensis* per fruit

	Date																									
	17-Mar	31-Mar	11-Apr	25-Apr	10-May	24-May	7-Jun	20-Jun	4-Jul	18-Jul	02-Aug	16-Aug	28-Aug	27-Sep	12-Oct	27-Oct	8-Nov	22-Nov	06-Dec	21-Dec	04-Jan	17-Jan	31-Jan	15-Feb	28-Feb	14-Mar
Fruit surface	0.13	0.15	0.15	0.02	0.12	0.03	0.28	0.23	0	0.07	0.05	0.03	0	0	0	0	0	0	0	0.1	0.28	0.88	1.58	1.66	0.7	0.21
Pedicle	0.31	0.41	0.4	0.14	0.53	0.03	0.5	0.23	0	0.08	0.06	0.15	0	0	0	0	0	0	0	0.12	0.48	2.22	3	1.79	0.71	0.43



ANNEX 3

EFFECT OF DIFFERENT CONTROL TREATMENTS ON
Brevipalpus chilensis
IN CLEMENTINE ORCHARDS

NOGALES, V REGION

(UNIVERSIDAD CATOLICA DE VALPARAISO)

UCV

EFFECT OF DIFFERENT TREATMENTS ON
Brevipalpus chilensis BAKER
IN A CLEMENTINE ORCHARD AT NOGALES, V REGION, CHILE

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April, 2001

INTRODUCTION

Brevipalpus chilensis Baker has been detected in Chile in clementine orchards both on leaves and on twigs and fruits.

Since its population levels are very low, this pest is usually not controlled in clementine orchards whether because the mites are not detected or because they are not associated with any damaging symptoms.

In order to even further reduce the mite's natural populations and in this way to mitigate the potential risk of the mite's presence on fruits destined to the U.S. market - where *B. chilensis* is considered to be a quarantine pest - the following trial treatments were designed. For this purpose, several products of known acaricide effect were tested, taking care to use only products that are compatible with the integrated pest management programs that are a feature of the Chilean clementine producing sector.

MATERIALS AND METHOD

The trials were conducted at Nogales, situated in Chile's V Region, on December 28, 2000 and a week later, in January 2001. For this purpose, a clementine orchard in full production was chosen, and 8 trees were selected for the application of each of the following treatments:

T1:	Detergent "Quix"	40cc/100lt
T2:	Sunspray Oil	0,8lt/100lt
T3:	Water	3.000 lt/ha

Owing to the low natural population of *Brevipalpus chilensis* in that orchard, the selected trees had to be artificially inoculated by putting highly mite infested privet (*Ligustrum sinensis*) branches on the selected trees.

In order to establish the initial *B. chilensis* population existing on the trees before starting with the application of the treatments, samples were taken from each of the 8 trees, which constituted the repetitions for each treatment. The samples taken were made up of six fruits per tree, as well as one 20 cm twig and 20 leaves. All samples were kept separate to indicate each repetition and each treatment.

The samples were brought to the Entomology Laboratory of the Catholic University at Valparaiso to be evaluated under a stereoscopic magnifying glass. Here, total *B. chilensis* mites (both mobile stages and eggs) present on the three analyzed plant parts were counted and recorded.

The application of the acaricide products was made by means of a piston pump with a spraying power of 3000 liter/hectare.

After the application of the treatments, a new sample (of the same size and characteristics of the first one) was taken and the first post-application count was conducted in the same way as described above.

A second count was then conducted seven days after application, taking samples of the same characteristics as above described and processing them in the same way.

The data obtained were analyzed statistically by means of a Complete Randomization Design. Duncan's Multiple Comparison Test at 95% confidence was used to compare the three treatments.

Tables 1 and 2 present the pre-application counts of *B. chilensis* mites as well as the counts one day and seven days after application.

Table 1: Effect of different treatment applied in clementine orchard. Observed One day after treatment application (Nogales, V Region, January 2001)

Treatments	Total N° of mites (2)	Total N° of mites	Efficacy (1)	Standard	Confidence interval
	Before treatment	One day	% of elimination	Error	For average at 95%
		After treatment			
Detergent	154	8	92.0 (a)	5.0	(68.8 - 100)
Oil	147	6	97.3 (a)	1.6	(73.9 - 100)
Water	161	24	82.4 (a)	4.5	(59.2 - 100)

N° of replications = 8

(1) Values followed by same letter do not differ statistically, according to Duncan test for 95% CL.

(2) Observations only on twigs and fruits (leaves, mites not detected)

Table 2: Effect of different treatment applied in clementine orchard. Observed one week after treatment application (Nogales, V Region, January 2001)

Treatments	Total N° of mites (2)	Total N° of mites	Efficacy (1)	Standard	Confidence interval
	Before treatment	One day	% of elimination	Error	For average at 95%
		After treatment			
Detergent	154	52	68.3 (a)	6.8	(10.7 - 100)
Oil	147	31	76.3 (a)	8.8	(18.8 - 100)
Water	161	55	48.3 (a)	22.2	(-9.0 - 100)

N° of replications = 8

(1) Values followed by same letter do not differ statistically, according to Duncan test for 95% LC.

(2) Observations only on twigs and fruits (leaves, mites not detected)

In the sampling conducted one day after the application of the treatments, it could be observed that detergent, oil and water removed over 80% of the mites; no statistically relevant differences between the three treatments could be established.

When observing the treatment's efficacy seven days post-application, no significant differences between the three treatments could be found; mite elimination efficacy percentages varied in a range of 48.3 to 76.3 percent.

Consequently, the conclusion can be drawn that all three treatments applied in a clementine orchard contribute to the control of *Brevipalpus chilensis*.

The foregoing conclusion is consistent with field experiences in mite control on other citrus fruits and avocado fruits, where there are *Panonychus citri* and *Oligonychus yothersi* respectively.

ANNEX 4

EXPLORATORY SAMPLING AT DIFFERENT STAGES OF
Brevipalpus chilensis
FROM HARVEST TO POST-HARVEST IN
CLEMENTINE FRUITS
(2000 SEASON)

(FUNDACION PARA EL DESARROLLO FRUTICOLA)

(FDF)

EXPLORATORY SAMPLING AT DIFFERENT STAGES FROM HARVEST TO POST HARVEST OF CLEMENTINE FRUITS

David Castro and Paola Astudillo
FDF, SEASON 2000

OBJECTIVE

The following work plan was carried out for collecting additional information on the presence of *Brevipalpus chilensis* at different stages of the clementine production process from harvest to post-harvest, thus evaluating the pre-and post-harvest risk mitigation effect.

METHODOLOGY

Six orchards were selected at random from fruit producing companies in the III, IV, V Regions and the MR. The following methodology was applied for each orchard:

1. During the harvest in each orchard **30 fruit of similar caliber were collected at random** from the following stages of the process:

- a) 30 recently harvested fruits originating in bins.

For this purpose 3 bins were selected at random and 10 fruits were selected at random from each bin. The bins were marked in order to take a sample in the following stage from the same bins (of the same lot).

- b) 30 fruits collected from bins after fruit curing.

Following the same procedure as above, 10 fruits were taken out of each marked bin until completing the 30-fruit sample.

- c) 30 fruits collected at completion of the packing process (finished product).

Upon completion of the packing process (finished product) 30 fruits were selected at random, making sure they came from the bins of the same fruit lot.

2. Each 30-fruit sample was placed in a 8 kg capacity paper bag bearing an appropriate identification of each of the above described process stages, as well as with the name of the Grower, Orchard, Packing Plant, Region, Province and Commune.

3. The information below was included in order to complete the sampling results.
 - Orchard Registration containing all the required information (Region, Province, Town, Surface Area, Number of Plants, etc.).
 - Phytosanitary Treatment(s) performed in each orchard.
 - Description of the packing process as regards the use of showers or immersion with detergent, wax, etc.
4. Methodology for *Brevipalpus chilensis* detection.

For the detection of *B. chilensis* the methodology described in Annex 1 was applied. The above is based on the fruit samples washing method with detergent and shower over 20 and 200 mesh screens. The pedicels previously removed from the fruits were submitted to this process separately.

RESULTS

Tables 1 and 2, graphically represented in Figures 1 and 2, show the results of the sampling at the different stages of the clementine process from harvest to packing.

As shown in said Tables, the packing process has a significant effect on the reduction of the mite populations, with an average efficacy of 89.79%. This is particularly so in cases with high prevalence levels at harvest time, such as Gibraltar, Santa Rosa, El Encierro and San Francisco orchards. In general, the packing process in these orchards includes the fruits passing through shower systems with detergent and rinsing with shower over rollers, continuing with a waxing process. This would cause an important reduction of the presence of the mite (see attached cards). It is worth noting that in the only two positive cases of *B. chilensis* detection on fruit, once the packing process was completed, the mite was mainly located under the pedicels (Table 2).

Concerning orchards El Sauce and La Granja, there was no *Brevipalpus chilensis* detection on fruits coming from harvest bins (Tables 1 and 2). Subsequently, only one case (El Sauce orchard) with low level of prevalence (an average of 0.03 mite per fruit) was detected in the final stage of the packing process. It should be noted that, according to the 1999 season survey (Ref. Annex 1), these orchards presented low prevalence values ranging between averages of 0.006 and 0.003 mite per fruit, respectively.

In the analysis of the phytosanitary program of these two orchards (2000 season) an application of detergents was applied upon commencing the harvest or during the June-August harvest, which could have caused this significant reduction in mite presence.

CONCLUSIONS

In accordance with these preliminary findings the following can be concluded:

1. The packing process generates an important risk mitigation effect on the *Brevipalpus chilensis* pest, with an estimated average efficacy of 89.79%.
2. The detergent applications at the beginning of the harvest or during the harvest in June-August can cause an important reduction in the mite presence on fruit, thus preventing subsequent re-infestations.

TABLE 1: DATA CARD OF PHYTOSANITARY TREATMENT OF ORCHARD

Name of Exporter: DEL MONTE FRESH PRODUCE (CHILE) S.A.

Name of Exporter or Business Name	Name of Orchard	Date of Application	Phenological State	Product	Formulation	Dose	Water Volume	Pest/Disease
Agrícola Villa Alegre El Sauce	Fundo El Sauce	Oct-99	Flower	SU-120	Liquid detergente	250 cc/100 lt water	8.000 lt/ha	White fly
		Dec-99	Flower + fruit set	Terra-Sorb foliar	Fert. Foliar líquido	150 cc/100 lt water	2.000 lt/ha	Foliar application
		Jan-00	Set Fruit	Nutra Spray Zn-M	25 % WP	200 gr/ 100 lt water	2.000 lt/ha	Foliar application
				SU-120	Liquid detergente	250 cc/100 lt water	8.000 lt/ha	Red mite
				Terra-Sorb foliar	Fert. Foliar líquido	150 cc/100 lt water	2.000 lt/ha	Foliar application
		Feb-00	Growing fruit	Citroliv Miscible	Miscible oil 95%	2%	3.000 lt/ha	Red mite and scale
				Lorsban 4E	48% EC	120 cc/100 lt water	3.000 lt/ha	Grape snout beetle
				Nutra Spray Zn-M	25 % WP	200 gr/ 100 lt water	2.000 lt/ha	Foliar application
		Mar-00	Growing fruit	Citroliv + Lorsban		2 lts + 120 cc/100 lt water	5.000 lt/ha	Scale
				Fert-All Fe y Zn	Chelate at 6% y 13,2%	200 cc/100 lt water	1.750 lt/ha	Foliar application
				Lorsban 4E	48% EC	120 cc/100 lt water	3.000 lt/ha	Grape snout beetle, aphid, scale
				SU-120	Liquid detergente	250 cc/100 lt water	8.000 lt/ha	White fly
		Apr-00	Growing fruit	Fert-All Zn	Chelate at 13,2%	150 cc/100 lt water	1.750 lt/ha	Foliar application
		Jun-00	Harvest start	SU-120	Liquid detergente	250 cc/100 lt water	8.000 lt/ha	White fly

		Aug-00	Harvest	SU-120	Liquid detergente	250 cc/100 lt water	8.000 lt/ha	Red mite
Agrícola Villa Alegre La Granja	Los Clérigos	07 al 25-10-99	Set Fruit	SU-120	Liquid detergente	400 gr/100 lt water	2.600 lt/ha	White fly, fumagina
	Los Clérigos 26 Hás.	06 al 10-11-99	Set Fruit	Basfoliar Zn 35 Mn 15	350 gr Zn + 150 gr Mn/kg	200 gr/ 100 lt water	1.600 lt/ha	Foliar application
	Los Clérigos	05 al 21-01-00	Set Fruit	Citroliv Miscible	950 gr/lt	1.50%	2.500 lt/ha	White fly and scale
				Lorsban 4E	480 gr/lt	100 cc/100 lt water	2.700 lt/ha	Scale
	Los Clérigos 26 Hás.	15 al 26-02-00	Set Fruit	Citroliv Miscible	950 gr/lt	1.50%	2.800 lt/ha	Red mite
	Los Clérigos	02-02 al 03-03-00	Set Fruit	Lorsban 4E	480 gr/lt	100 cc/100 lt water	2.000 lt/ha	Scale
	Los Clérigos 26 Hás.	03 al 30-03-00	Set Fruit	SU-120	Liquid detergente	400 gr/100 lt water	4.700 lt/ha	White fly, fumagina
	Chuquicamata	3/15/2000	Set Fruit	Citroliv Miscible	950 gr/lt	1.50%	2.500 lt/ha	Red mite eggs
Los Clérigos 26 Hás.	13-05 al 02-06-00	Harvest	SU-120	Liquid detergente	400 gr/100 lt water	3.855 lt/ha	White fly, fumagina	

TABLE 2: DATA CARD-REGISTRATION OF CLEMENTINE ORCHARDS

Name of Exporter: DEL MONTE FRESH PRODUCE (CHILE) S.A.

Phone number: 51-324123/314218/322247

Fax: 51-311474

Responsible for Program: Christian Gisseleir e Salinas

E-mail: cgisseleire@cl.fresh delmonte.com rlopez@cl.freshdelmonte.com

Name of Exporter or Business Name	Name of Orchard	Region	Province	Town	Responsible of Orchard	Position	Phone	Fax	N° of Lots	Total Area (ha.)	N° of Plants	Harvest starting date
Agrícola Villa Alegre El Sauce	Fundo El Sauce	IV	Elqui	Coquimb o	René López Reyes	Administrator	51- 247777	51-247777	7	148.21	82,255	29-May
Agrícola Villa Alegre La Granja	Fundo La Granja	IV	Limarí	Ovalle	Alfredo Motta P.	Administrator -Ing. Agr.	51- 681005	51-681005	4	41.50	21,550	16-May



TABLE 3: DATA CARD PACKING PROCESS

Name of Exporter: DEL MONTE FRESH PRODUCE (CHILE) S.A.

Name of Packing: CENTRAL REQUINOA

Name of Exporter or Business Name	Name of Orchar d	Date of Process	Washing with Detergent			Rinsing (optional)			Waxing	
			Shower or Immersion	Detergent Brand	Dose	Only Shower	Shower over brush		Brand	Dose
							Amount	Kind		
Agrícola Villa Alegre El Sauce	Fundo El Sauce	7/3/2000	Shower	Etonil	200 cc/100 lt water		10	Horse hair	Imizalil	400 cc/100 lt water
Agrícola Villa Alegre La Granja	Fundo La Granja	7/21/2000	Shower	Etonil	200 cc/100 lt water		10	Horse hair	Imizalil	400 cc/100 lt water

TABLE 1: DATA CARD OF PHYTOSANITARY TREATMENT OF ORCHARD

Name of Exporter: SUBSOL E S.A.

Name of Exporter or Business Name	Name of Orchard	Date of Application	Phenological State	Product	Formulation	Dose	Water Volume	Pest/Disease
Agrícola El Canelillo	El Encierro	06-Jan		Lorsban	Oil	1,8 lt/100 lt water	1.500 lt-3 dosis/ha	scale, mealybug and mite
				Sunspray		19 lt/100 lt water		
		15-Jan		Lorsban	Oil	1,8 lt/100 lt water	1.500 lt-1 dosis/ha	scale, mealybug and mite
				Pyrinex		1,8 lt/100 lt water		
				Sunspray		19 lt/100 lt water		
		17-Jan		Pyrinex	Oil	1,8 lt/100 lt water	1.500 lt-2 dose/ha	scale, mealybug and mite
				Sunspray		19 lt/100 lt water		
		2-Feb		Ecopol	Detergent	9 lt/100 lt water	1.500 lt	washing
24-Mar	Ecopol	Detergent	9 lt/100 lt water	1.500 lt	washing			
Fernando Cerda	San Francisco	Jan-00	Fruit 1 cm diameter	ORXYS	Oil	1 lt/100 lt water	3.000 lt	scale, mealybug and mite
		Jan-00	Fruit 1 cm diameter	Lorsban		100 cc/100 lt water	3.000 lt	scale, mealybug and mite



TABLE 2: DATA CARD-REGISTRATION OF CLEMENTINE ORCHARDS

Name of Exporter: SUBSO
LE S.A.

**Phonenu
mber:** 2422
683
2411
Fax: 220

**Responsible for
Program:** Hermán
Alday

E-mail: halday@subsol
e.cl

Name of Exporter or Business Name	Name of Orchard	Region	Province	Commune	Responsible of Orchard	Position	Phone	Fax	N° of Lots	Total Area (ha.)	N° of Plants	Harvest starting date
Agrícola El Canelillo Rut: 96.813.740-9	El Encierro	V	Quillota	Quillota	Gonzalo Vargas	Administrator	09- 5373968	33-319175	3	24.00	13,320	5-Jun
Fernando Cerda Rut: 4.887.864-4	San Francisco	RM	Melipilla	Mallarauco	Gildo González	Foreman	8312571	8312571	3	2.70	1,498	10-Jun

TABLE 3: DATA CARD PACKING PROCESS

Name of Exporter: SUBSOL E S.A.

Name of Exporter or Business Name	Name of Orchard	Name of Packing	Date of Process	Washing with Detergent			Rinsing (optional)			Waxing	
				Shower or Immersion	Detergent Brand	Dose	Only Shower	Shower over brush		Brand	Dose
								Amount	Kind		
Agrícola El Canelillo	El Encierro	Agricom Polpaico/Packing El Molino	27-Jun	Hypoclorite		200 ppm		6	PVC roller	Fungaflor	100 cc/100 lt
								4	Sponge roller	Waterwax U.E. (cera)	1 lt/ton
				4	Horse hair roller						
Fernando Cerda	San Francisco	Multifruta S.A./Linderos-Buin	29-Jun	Texaclor		200 cc/100 lts		5	Horse hair roller	Teycer K.G.L.	1 lt/ton
								8	Horse hair roller		
								2	Sponge roller	Tiobendazo 1 + Imazalil	10 cc/lt
								3	Rubber		7 cc/lt

TABLE 1: DATA CARD OF PHYTOSANITARY TREATMENT OF ORCHARD

Name of Exporter: UNIFRUTTI TRADERS LTDA.

Name of Exporter or Business Name	Name of Orchard	Date of Application	Phenological State	Product	Formulation	Dose	Water Volume	Pest/Disease
Cítricos Uniagri Ltda.	Gibraltar	25-Oct	2° Flash	Allete	W.P.	400 grs/100 lt water	2.500 lt	Phytophthora
		14-Jan	3° Flash	Pirimon	W.G.	20 grs/100 lt water	2.500 lt	Aphid
		17-Feb	Fin 3° Flash	SV-120	U.K.	40 grs/100 lt water	13.000 lt	White fly and Mealybug
				SV-143	W.G.			
20-Feb	Fin 3° Flash	Sunspray	Aceite	1%	3.000 lt	White fly and Mealybug		
Cítricos Uniagri S.A.	Santa Rosa	Aug-99	After pruning	SV-143	Detergent	40 grs/100 lt water	5.000 lt	Aphid
		Sep-99	Budding	Diazinon	600 EW	17 lt/ha	250 lt	Trunk paint/ants
		Oct-99	35% budding	Citroliv	Oil	0.80%	2.000 lt	Aphid
		Nov-99	Fruit growth	SV-143	Detergent	40 grs/100 lt water	5.000 lt	Aphid and mites
		Dec-99	Fruit growth	SV-143	Detergent	40 grs/100 lt water	5.000 lt	Aphid and mites
		Mar-00	Fruit growth	Oxicup	Cooper oxiclорure	300 grs/100 lt water	2.000 lt	Phytophthora
		Apr-00	Change of color	Oxicup	Cooper oxiclорure	300 grs/100 lt water	2.000 lt	Phytophthora
		May -00	Harvest	Oxicup	Cooper oxiclорure	300 grs/100 lt water	2.000 lt	Phytophthora

TABLE 2: DATA CARD-REGISTRATION OF CLEMENTINE ORCHARDS

Name of Exporter: UNIFRUTTI
TRADERS LTDA.

Responsible for Program: Mauricio
Latrille
(Copiapó)

Phone number: 52-213668/52-
218790

Fax: 52-213668
anexo 275

E-mail: unifcppo@unif
rutti.cl

Responsible for Program: José
Araya

Phone number: 711101

Fax: 711132

E-mail: _____

Name of Exporter or Business Name	Name of Orchard	Region	Province	Commune	Responsible of Orchard	Position	Phone	Fax	N° of Lots	Total Area (ha.)	N° of Plants	Harvest starting date
Cítricos Uniagri Ltda.	Gibraltar	III	Copiapó	Copiapó	Carlos Pacheco	Administrat or	52- 213889		7	38.24	35	16-May
Cítricos Uniagri S.A.	Santa Rosa	IV	Limarí	Monte Patria	Román Aros	Administrat or	711132	7111 32	30	108.00	90,000	8-May



TABLE 3: DATA CARD PACKING PROCESS

Name of Exporter: UNIFRUTTI TRADERS LTDA.

Name of Exporter or Business Name	Name of Orchard	Name of Packing	Date of Process	Washing with Detergent			Rinsing (optional)			Waxing	
				Shower or Immersion	Detergent Brand	Dose	Only Shower	Shower over brush		Brand	Dose
								Amount	Kind		
Cítricos Uniagri Ltda.	Gibraltar	Unifrutti Copiapó	14-Jun	Ducha	Deterfrut	100/100		33	natural	Fomesa	1 lt/ton
								19	synthetic	Waterwax	1.2 lt/ton

Table 1: Prevalence at different stages from harvest to postharvest in clementines
2000 Season
(average number of mites per fruit)

Name of Orchard	Harvesting Bins	"Curado" Bins	Packed Fruits	Prevalence by orchard, 1999
Gibraltar(III)	1.28	0.8	0.27	0.44
Sta Rosa(IV)	0.34	0.29	0	0.13
El Sauce(IV)	0	0.03	0.03	0.006
La Granja(IV)	0	0.17	0	0.003
El Encierro(V)	0.97	0.55	0	0.05
Sn. Francisco(RM)	0.34	0.5	0	0.14
General average	0.49	0.39	0.05	
Average efficacy of process: 89,79%				

Sample=30 fruits in each case

*) Source. Annex 1: Survey and identification of pests associated to clementine, mandarine and tangerine fruits. (P.R.A)

Author: David Castro-FDF

Table 2: Prevalence of *Brevipalpus chilensis* at different stages of the process-clementines-2000 Season

Region	Name of Grower	Name of Orchard	Prevalence of mites average/fruit 1999 season	Monitoring for <i>Brevipalpus chilensis</i> at different stages of process-clementines											
				Harvesting Bins				"Curado" Bins *				Packed Fruits			
				Live mites		Dead mites	Live mites average/fruit	Live mites		Dead mites	Live mites average/fruit	Live mites		Dead mites	Live mites average/fruit
				On Fruit	On Pedicel			On Fruit	On Pedicel			On Fruit	On Pedicel		
III	Unicitricos	Gibraltar	0.44	38	3	14	1.28	13	11	5	0.80	1	7	4	0.27
IV	Cítricos Uni-Agri S.A.	Santa Rosa	0.13	3	8	9	0.34	0	9	4	0.29	0	0	0	0.00
IV	Agrícola Villa Alegre	El Sauce	0.006	0	0	1	0.00	0	1	0	0.03	0	1	3	0.03
IV	Agrícola Villa Alegre	La Granja	0.003	0	0	0	0.00	3	2	1	0.17	0	0	0	0.00
V	Agr. El Canelillo S.A.	El Encierro	0.05	18	11	5	0.97	9	8	8	0.55	0	0	0	0.00
RM	Fernando Cerda	San Francisco	0.14	4	7	0	0.34	6	9	3	0.50	0	0	0	0.00
Total				63	29	29	0.49	31	40	21	0.39	1	8	7	0.05

Fig. 1: Prevalence at different stages of process-clementines

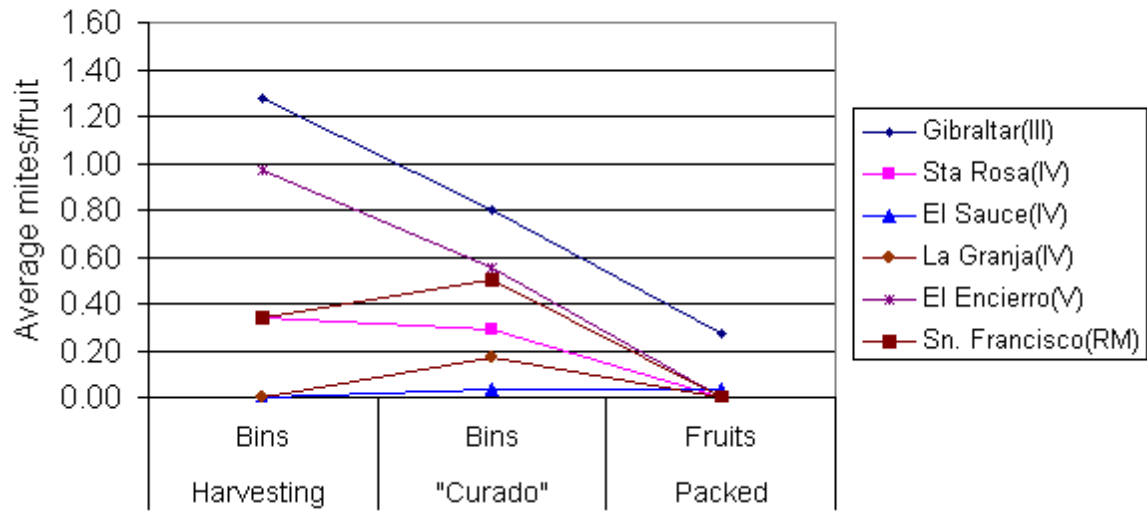
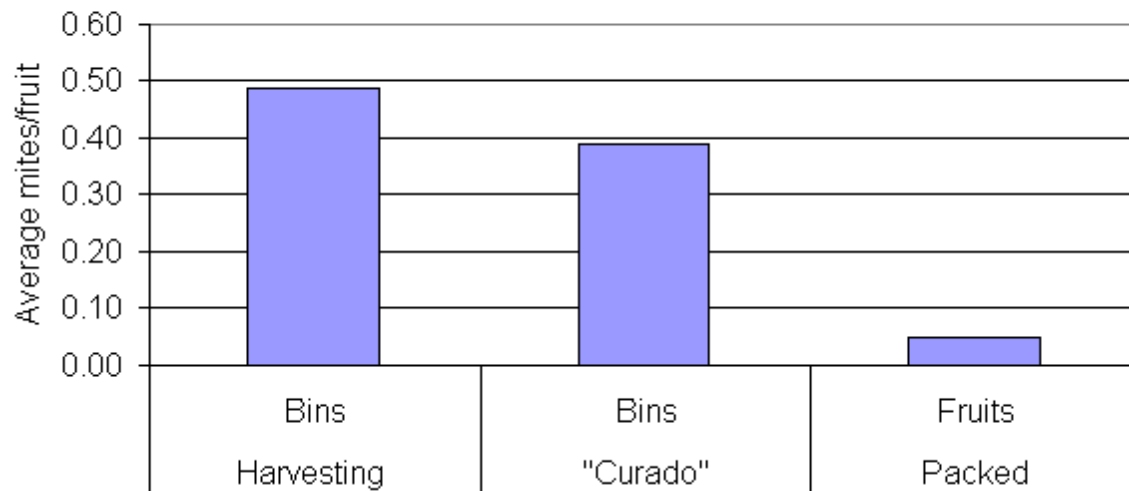


Fig. 2: Average prevalence-Fruit from all surveyed orchards



ANNEX 5

EFFECT OF THE PACKING PROCESS ON
Brevipalpus chilensis
IN CLEMENTINE FRUITS
(COQUIMBO, IV REGION, AND HIJUELAS, V REGION)

(UNIVERSIDAD CATOLICA DE VALPARAISO)

UCV

EFFECT OF THE PACKING PROCESS
ON THE REMOVAL OF
Brevipalpus chilensis **Baker**
ON CLEMENTINE FRUITS
AT COQUIMBO (IV REGION) AND HIJUELAS (V REGION)

CHILE

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UCV

April, 2001

INTRODUCTION

Brevipalpus chilensis Baker has been observed associated with clementine fruits in Chile. Its presence on the fruit does not constitute a pest since the mites - when present - are detected at population levels so low that the mites do not produce any damage nor are any symptoms associated with their feeding on the plant.

The mite's quarantine status for U.S. agricultural authorities has led to efforts aimed at reducing its presence on export fruit. For this purpose, both orchard management and postharvest treatments have been evaluated.

Washing with water and detergent followed by waxing, treatments successfully applied on export cherimoyas against the same *B. chilensis* led us to think that the same procedure could control and eliminate this mite when processing clementine fruits.

This study was designed to evaluate the removal and control of mites on the fruits when introducing these procedures into the normal packing line used for citrus fruits. The aim is to test a commercial processing line's efficacy in the removing mites both on the surface of the fruit and under its pedicel.

METHODOLOGY APPLIED AT COQUIMBO

A trial was made at the Processing Plant of Unifrutti Traders Ltda. clementine packing plant at the port of Coquimbo, IV Region, during the 2000 season harvest.

Clementine fruits coming from two commercial orchards located at El Palqui (IV Region) and Nogales (V Region) were processed.

For this purpose, 302 clementine fruits were harvested on June 20, 2000 and 504 fruits on July 5, 2000.

Both times the fruit were first taken to the Entomology Laboratory of the Agronomy Faculty of Universidad Católica de Valparaíso in order to infest them with *B. chilensis*.

The fruits were placed on transparent plastic trays, the pedicels facing upward; then three privet leaves highly infested with *B. chilensis* (*Ligustrum sp.*) were placed on top of each of them.

The trays with fruits were kept in metal shelves in a room with temperature, humidity and illumination conditions appropriate for the mite's activity.

As the privet leaves dehydrated, the mites started moving onto the fruits, colonized them and started laying eggs. After about 5-6 days, the clementine fruits were infested with mites at various stages of development, both on the surface and under the pedicel.

MITE EGG COUNT

Once the fruits had been infested, the following step was to mark and count the eggs present on the fruit's surface.

The marking of the eggs was made with a fine indelible ink felt-tip pen by drawing a circle around the egg or eggs and writing down the number of eggs inscribed in that circle.

In this way, the number of eggs present on the surface of each fruit was known before their entry into the packing process.

Since it is impossible to make an egg count under the pedicel because this would destroy and/or alter the structure of the pedicel of fruits later to be processed in the packing house, 50 fruits were taken after inoculation and the eggs under the pedicel were counted. The average value obtained was assumed to be the average egg population existing under the pedicel before the fruits were treated.

MOBILE STAGE MITE COUNT

Since the mite's mobile specimen can not be marked on the fruit's surface, an estimation of their number was made for this lot of inoculated fruits. The procedure used to estimate the mobile specimen located under the pedicel was the same as the one applied for an estimate of eggs under the pedicel.

PROCESS IN THE PACKING HOUSE

Once the initial number of eggs and mobile stages, both on the fruit's surface and under the pedicel, had been obtained, the clementine fruit were taken to a commercial packing line belonging to Unifrutti Traders Ltda. situated at Coquimbo, in Chile's IV Region, where a total of 706 fruits underwent the process described below.

At the starting point of the process line, the fruit are bathed in a chlorine water solution; the same solution then carries them to a worm gear that transports them to the washing facility, where they are washed with water and detergent. The detergent foam acts on the fruit. Then they are moved along by rubber rollers to a place where they are rinsed off with high-pressure water. Finally, they are moved along by sponge coated rollers which, together with powerful fans, facilitates drying.

The fruit then undergoes waxing and drying in a forced hot air tunnel. The fruit then go to the selection tables and to calibration before being packed.

In the case of the fruit participating in this trial, instead of packing them, after the selection tables, the fruits were taken away in order to be sent to the laboratory to continue the evaluation process.

EVALUATION OF THE CONTROL OF MOBILE MITE STAGES

Once the inoculated fruits had undergone the packing process, they were again taken to the Entomology

Laboratory at Universidad Católica de Valparaíso. Here the fruit were examined under the stereoscope. First the mobile mite stages on the fruit surface were evaluated, both the live ones and the ones killed by the wax..

Of the mites still present on the fruit surface, those that did not react to a physical stimulus were considered to have been killed by the wax. The post-process population was then compared to the estimated initial pre-process population.

The same procedure was applied to the mobile stages of mites (whether live or dead) present under the pedicel.

EVALUATION OF THE CONTROL OF EGGS

In order to evaluate the effects of the packing process on the eggs present on the fruit surface, by means of the stereoscope every circle that had been marked was checked for the presence of *Brevipalpus chilensis* eggs. .

Among the eggs still present on the surface, those that were covered by wax and had not hatched by the time 70 percent of the untreated control eggs had hatched were considered to be dead. Those eggs whose presence had originally been marked and which no longer were present in the circles were considered removed by the process.

In order to evaluate the effect of the packing process on the eggs located under the pedicel, the pedicel of every fruit was removed and the eggs were counted. The number of eggs that had been covered by the wax was recorded, as well as the number of those that had not been reached. In order to determine the viability of these eggs, an untreated control was kept.

The findings were then compared to the estimated pre-process number of eggs under the pedicel.

RESULTS AND DISCUSSION

According to the results indicated in Chart 1, 100 percent mortality was attained for both eggs and mobile mite stages present on the surface of fruit that underwent the packing process.

Chart 1. Effect of one commercial fruit processing line on the mortality of *Brevipalpus chilensis* Baker in Clementines. (UNIFRUTTI, Coquimbo, IV Region)

	INITIAL POPULATION	FINAL POPULATION	% OF MITES DRAGGED AWAY	% OF MITES KILLED BY WAXING	% LIVE MITES
Eggs on fruit surface	1.195	171	86	14	0
Eggs under pedicel	2.569	1384	46	0	54
Mobile mites on fruit surface	9.472	506	95	5	0
Mobile mites under pedicel	10.923	2.773	75	0	25

This result is consistent with previously gathered information in which the packing process line, the friction among the fruits and/or with the rollers, together with the washing, removed a high proportion of the mites present on the fruit's surface.

The mites, whether mobile or eggs, that were protected by the pedicel represent the greatest difficulty for total removal, although significant reductions could be achieved, especially in the case of mobile mite stages protected by the pedicel.

The Chart also shows that very few mobile stages or eggs managed to remain on the fruit surface, but the wax, which causes their death, trapped all of them.

These data are consistent with those for cherimoya fruits carrying the same *B. chilensis* mite: once the mite is covered by the wax, it is impregnated by it and becomes unable to get free of it (in the case of mobile stages) or unable to hatch (in the case of eggs).

When analyzing the total removal and control figures, including eggs and mobile stages, both on the fruit surface and under the pedicel, a control efficacy of 79.99% could be attained, a figure that is similar to the one reached during the process carried out at Hijuelas, V Region described below in this paper.

This leads to the conclusion that the clementine packing process has an important mitigation effect on *Brevipalpus chilensis* populations present on the fruit.

METHODOLOGY APPLIED AT HIJUELAS

A trial was made at the Processing Plant of the SAFEX Company with its clementine packing plant at Hijuelas (V Region), during the 2000 season harvest.

Clementine fruit coming from a commercial orchard located at El Palqui (IV Region) were processed.

For this purpose 540 clementine fruits were harvested on June 9, 2000 and 554 fruits on July 6, 2000.

On both dates, the fruit were initially taken to the Entomology Laboratory of the Agronomy Faculty of Universidad Católica de Valparaíso in order to infest them with *B. chilensis*.

The fruit were placed on transparent plastic trays, the pedicels facing upward; then three privet leaves highly infested with *B. chilensis* (*Ligustrum sp.*) were placed on top of each of them.

The trays with fruits were kept in metal shelves in a room with temperature, humidity and illumination conditions appropriate for the mite's activity.

As the privet leaves dehydrated, the mites started moving onto the fruits, colonized them and started laying eggs. After about 5-6 days, the clementine fruits exhibited various stages of the mite's development, both on the surface and under the pedicel.

MITE EGG COUNT

Once the fruit had been infested, the following step was to mark and count the eggs present on the fruit's surface.

The marking of the eggs was made with a fine indelible ink felt-tip pen by drawing a circle around the egg or eggs and writing down the number of eggs inscribed in that circle.

In this way, the number of eggs present on the surface of each fruit was known before their entry into the packing process.

Since it is impossible to make an egg count under the pedicel because this would destroy and/or alter the structure of the pedicel of fruits later to be processed in the packing house, 50 fruit were taken after inoculation and the eggs under the pedicel were counted. The average value obtained was assumed to be the average egg population existing under the pedicel before the fruit were treated.

MOBILE STAGE MITE COUNT

Since the mite's mobile stage can not be marked on the fruit's surface, an estimation of their number was made for this lot of infested fruit. The procedure used to estimate the mobile specimen located under the pedicel was the same as the one applied for an estimate of eggs under the pedicel.

PROCESS IN THE PACKING HOUSE

Once the initial number of eggs and mobile stages, both on the fruit's surface and under the pedicel, had been

estimated, the clementine fruit were taken to a commercial packing line belonging to SAFEX at Hijuelas, V Region, where a total of 994 fruits were processed.

At the starting point of the process line, the fruit were bathed in a chlorine water solution; the same solution then takes them to a worm gear which transports them to the washing facility, where they are washed with water and detergent. The detergent foam acts on the fruit. Then rubber rollers transport the fruit to a place where they are rinsed off with high-pressure water. Finally, they are transported by sponge coated rollers which, together with powerful fans, facilitate drying.

The fruit then undergoes waxing and drying in a forced hot air tunnel. The fruit then go to the selection tables and to calibration before being packed.

In the case of the fruits participating in this trial, instead of packing them, after the selection tables the fruits were taken away in order to be sent to the laboratory and continue the evaluation process.

EVALUATION OF THE CONTROL OF MOBILE MITE STAGES

Once the inoculated fruit had undergone the packing process, they were again taken to the Entomology Laboratory at Universidad Católica of Valparaíso. Here the fruit were examined under stereoscope. First the mobile mite stages on the fruit surface were evaluated, both the live ones and the ones killed by the wax.

Of the mites still present on the fruit surface, those that did not react to a physical stimulus were considered killed by the wax. The post-process population was then compared to the estimated initial pre-process population.

The same procedure was applied to the mobile stages of mites (whether live or dead) present under the pedicel.

EVALUATION OF THE CONTROL OF EGGS

In order to evaluate the effects of the packing process on the eggs present on the fruit surface, by means of the stereoscope every circle that had been marked was checked for the presence of *Brevipalpus chilensis* eggs. .

Among still present on the surface, those covered by wax that had not hatched by the time 70 percent of the untreated control eggs had hatched were considered to be dead. Those eggs whose presence had originally been marked and were no longer present in the marked circles were considered removed by the process.

In order to evaluate the effect of the packing process on the eggs located under the pedicel, the pedicel of every fruit was removed and the eggs were counted. The number of eggs that had been reached and covered by the wax was recorded, as well as the number of those that had not been reached. In order to determine the viability of these eggs, an untreated control was kept.

The findings were then compared to the estimate pre-process number of eggs under the pedicel.

RESULTS AND DISCUSSION

According to the results indicated in Chart 1, 100 percent mortality is attained for both eggs and mobile mite stages present on the surface of the fruit that underwent the packing process.

Chart 1. Effect of one commercial fruit processing line on the mortality of *Brevipalpus chilensis* Baker in Clementines. (SAFEX, Hijuelas, V Region)

	INITIAL POPULATION	FINAL POPULATION	% OF MITES DRAGGED AWAY	% OF MITES KILLED BY WAXING	% LIVE MITES
Eggs on fruit surface	3.279	332	90	10	0
Eggs under pedicel	3.486	1.630	53	0	47
Mobile mites on fruit surface	14.448	334	98	2	0
Mobile mites under pedicel	9.394	3.448	63	0	37

This result is consistent with previously gathered information indicating the packing process line, the friction among the fruits and/or with the rollers, together with the washing, removes a high proportion of the mites present on the fruit's surface.

Mites, whether mobile or eggs, that were protected by the pedicel represent the greatest difficulty for total removal, although significant reductions could be achieved, especially in the case of mobile mite stages protected by the pedicel.

The Chart also shows that very few mobile stages or eggs managed to remain on the fruit surface, but the wax, which causes their death, trapped all of them.

These data are consistent with those for cherimoya fruits carrying the same *B. chilensis* mite: once the wax covers the mite, it is impregnated by it and becomes unable to get free of it (in the case of mobile stages) or unable to hatch (in the case of eggs).

When analyzing the total removal and control figures, including eggs and mobile stages, both on the fruit surface and under the pedicel, a removal figure of 81.23% could be attained.

This leads to the conclusion that the clementine packing process has an important mitigation effect on *Brevipalpus chilensis* populations present on the fruit.

ANNEX 6

RISK MITIGATION OF *Brevipalpus chilensis* ON CLEMENTINE AND
MANDARIN ORANGE FRUITS
BY MEANS OF
EFFICACY TEST OF THE PACKING PROCESS

(2001 SEASON)

FUNDACION PARA EL DESARROLLO FRUTICOLA

(FDF)

EFFICACY OF THE PACKAGING PROCESS IN THE MITIGATION OF
Brevipalpus chilensis
RISK IN CLEMENTINES AND MANDARIN ORANGES
2001 Season

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BACKGROUND INFORMATION

In a preliminary study named “Exploratory Sampling at different steps from harvest to post-harvest of clementines, mandarin oranges and tangerines” carried out in the 2000 season by FDF (See Annex 4) it was observed that the normal packing process of these three fruit species significantly decreased the level of infestation by *Brevipalpus chilensis*. The process includes washing the fruit with detergent as well as waxing and its efficacy was determined to be 89.7 percent. In similar studies performed by López and Parra (Universidad Católica de Valparaíso) (See Annex 5), the efficacy in the mite removal varied between 79.9 percent and 81.2 percent.

In a pilot plan (performed by FDF in 2001 and in which 18 orchards were included) carried out according to the proposed Systems Approach, the average elimination efficacy during the packing / postharvest processing was estimated as 78.4 percent.

The normal packing process for citrus fruits was considered fundamental as a mitigation factor for the pest risk. To determine the efficacy of the packing process in the elimination of *B. chilensis* on fruit, a biological test was designed.

OBJECTIVE

- To establish the efficacy level of the packing process in the reduction in the presence of *B. chilensis* on clementines and mandarin oranges.

METHODOLOGY

3.1 Time and Place

The experiments were performed between May and August 2001 in two selected packing facilities, taking into account that the packing process of the species under study is similar in both facilities.

a) Packing Facility: Unifrutti Traders Ltda. (IV Region)

- Packing Process of clementines, clemenule variety.

b) Packing Facility: La Rosa Sofruco (VI Region).

- Packing Process of clementines, 'Clemenule' variety
- Packing Process of mandarin oranges, 'Hermandina' variety

3.2 Procedure

Artificial Fruit Infestation prior to packing

In order to have a sufficient number of mites for the experiments, a laboratory for artificial breeding of *B. chilensis* on privet plants (*Ligustrum sinensis*) was set up. Environmental conditions were temperatures between 23 and 26°C, RH was 40 to 60% and the photoperiod was 14: 10 (L:N).

Each citrus species was artificially infested in the laboratory under the above- mentioned environmental conditions. For this purpose, the clementine fruits (var. clemenules) and mandarin orange fruits (var. Hermandina) were placed in special racks (See photographs in Appendix). Afterwards, one to two infested privet (*Ligustrum sinensis*) leaves per fruit were placed on the fruit, so that the mites could naturally move onto them as the privet leaves were dehydrating. The fruit were left under these conditions for 7 days, a period of time long enough for the mites to naturally distribute on the whole surface of the fruits.

For each packing line, five replications with 60 artificially infested fruits were used, plus a control under the same conditions, which did not undergo the packing process. A completely randomized experimental design was utilized.

Packing process

From each lot, 60 artificially infested clementine and mandarin orange fruits were processed at the packing facilities of Unifrutti (IV Region) and La Rosa Sofruco (VI Region). The fruits went through the following steps:

1. Drencher
2. Stockpiling in shed (curing process)
3. Emptying into chlorinated water and rinsing
4. Application of Detergent (Shower or Immersion)
5. Rinsing with brush under water shower
6. Pre-drying
7. Application of fungicide shower
8. Pre-drying with hot air
9. Application of wax with fungicide
10. Hot air drying tunnel

C. *Brevipalpus chilensis* Detection

Once the packing process was finished, the fruits were taken to the laboratory for evaluation. Mite detection was performed by the "dragging by washing" method.

This method consists of the following:

Materials used:

Metal sieve with 200 mesh.
Metal sieve with 20 mesh.
Stereoscopic 40 x magnifying glass.
Washing container with high-pressure water supplied in shower form.
300 x to 600 x microscope.
Tweezers
Ethanol alcohol for conserving the samples.
Petri dishes.
3 to 5 ml glass tubes.
QUIX brand detergent.
Slides and slide covers.
Electric plate.
Mounting medium (Hoyer, Euparal).

In each case the following procedure was performed:

Step 1: Washing the complete fruit (including its non removed calyx) and

Step 2: The calyx of each fruit was removed and a second washing was performed, this time washing fruit and calyx separately.

The detailed washing procedure consisted of the following (See Appendix):

- a) The sieves were placed one above the other, with the smallest mesh spaces (200 mesh) sieve below. For the first washing the fruits and pedicel disks were placed on the sieves. Then the fruit and pedicel disks were placed separately on the sieves, in suitable quantities to allow them to easily be washed.
- b) Next, the fruit and pedicel disks were sprinkled separately with a liquid soap and water solution (QUIX in the proportion of two teaspoons per liter of water). Each fruit and pedicel disk was then thoroughly washed under high-pressure water from a shower-type nozzle.
- c) Then the fruit or pedicel disks in the 200 mesh sieve were tipped to one side and washed with water at low pressure, letting the sieve's contents collect on one side. Next, with water, the contents were drained onto a Petri disk.
- d) The mites that had come off the fruits or pedicel disks were observed and counted under a stereoscope microscope.

RESULTS

4.1 Efficacy of packing process

Tables 1, 2 and 3 present the results of the efficacy tests in *Brevipalpus chilensis* elimination carried out in the three packing processes. The first and second packing processes were evaluated with clementines, clemenule variety, in the packing facilities of Unifrutti (IV Region) and La Rosa Sofruco (VI Region). The third packing process was performed on mandarin oranges, Hermandina variety, at the La Rosa Sofruco

facility (VI Region).

The Tables show that the mite elimination percentage reached an average of 85.4 percent to 85.9 percent in both processes applied on clementines (La Rosa Sofruco and Unifrutti). As for the mandarin oranges processed at La Rosa Sofruco, mite elimination amounted to an average 89.3 percent.

For each case, a 95 percent confidence interval was used. there is no statistically relevant difference between the range for the averages: 85.4 (79.5 – 91.4); 85.9 (79.9 – 91.8) and 89.3 (83.3 – 95.2) for the different processes evaluated.

SPECIES: Clementines
VARIETY: Clemenules

EXPORTER: Sociedad Agrícola La Rosa Sofruco S.A.
GROWER: Sociedad Agrícola La Rosa Sofruco S.A.
ORCHARD: La Rosa
REGION: VI
PROVINCE: Cachapoal
MUNICIPALITY: Peumo
PACKING FACILITY: Sociedad Agrícola La Rosa Sofruco S.A., VI Región

Table 1: Efficacy test of postharvest treatment (Packing process)

Replications	N° treated mites (1)	Postharvest treatment (packing process) N° mites	N° eliminated mites	Elimination percentage
1	1.694	234	1.460	86,19
2	1.973	103	1.870	94,78
3	948	173	775	81,75
4	1.622	197	1.425	87,85
5	650	136	514	79,08
Total	6.887	843	6.044	85,93

S				6,1
SE				2,7
95%,Confidence interval for mean				(79.9 - 91.8)

(1): Number of survivors in untreated control

Table 2: Efficacy test of postharvest treatment (Packing process)

Replications	N° treated mites (1)	Postharvest treatment (packing process) N° mites	N° eliminated mites	Elimination percentage
1	760	200	560	73,68
2	1.794	163	1.631	90,91
3	1.136	225	911	80,19
4	859	83	776	90,34
5	1.037	80	957	92,29
Total	5.586	751	4.835	85,48

S				8,2
SE				3,6
95%, Confidence interval for mean				(79.5 - 91.4)

(1): Number of survivors in untreated control

SPECIES: Mandarin Oranges
VARIETY: Hermandina

EXPORTER: Sociedad Agrícola La Rosa Sofruco S.A.
GROWER: Sociedad Agrícola La Rosa Sofruco S.A.
ORCHARD: La Rosa
REGION: VI
PROVINCE: Cachapoal
MUNICIPALITY: Peumo
PACKING FACILITY: Sociedad Agrícola La Rosa Sofruco S.A., VI Región

Table 3: Efficacy test of postharvest treatment (Packing process)

Replications	N° treated mites (1)	Postharvest treatment (packing process) N° mites	N° eliminated mites	Elimination percentage
1	1.231	150	1.081	87,81
2	963	112	851	88,37
3	2.239	126	2.113	94,37
4	863	102	761	88,18
5	965	118	847	87,77
Total	6.261	608	5.653	89,30

S				2,8
SE				1,3
95%, Confidence interval for mean				(83.3 - 95.2)

(1): Number of survivors in untreated control

4.2 *Brevipalpus chilensis* distribution on clementine fruits

In Tables 4, 5 and 6, the mites' distribution is presented, both on the fruit's surface and under the calyx, in accordance with the "dragging by washing" detection method applied on artificially infested fruits of the untreated control group and fruit that had followed the packing process.

These tables show that, in the case of the control fruit, after one week in the laboratory for infestation, the distribution ratio of mites detected on the fruit's surface vs. the mites detected under the calyx was 7:1. In the fruit that underwent the packing process, the distribution ratio of mites detected on the fruit's surface vs. the mites detected under the calyx changed to 2:1. This result is explained by the significant elimination of mites on the fruit surface.

Table 4: Detection and Distribution of mites in control and Postharvest treatment Artificially infested fruits

Treatment	Replication	Clementines, Unifrutti IV Region		
		Nº mites Fruits	Nº mites Calyx	Nº mites Total
	1	1.368	326	1.694
	2	1.780	193	1.973
Control	3	829	119	948
	4	1.315	307	1.622
	5	526	124	650
	Total	5.818	1.069	6.887
	1	180	54	234
	2	66	37	103
Postharvest Treatment	3	90	83	173
(Packing process)	4	134	63	197
	5	107	29	136
	Total	577	266	843

Table 5: Detection and Distribution of mites in control and Postharvest treatment
Artificially infested fruits

Treatment	Replication	Clementines, La Rosa Sofruco VI Region		
		Nº mites Fruits	Nº mites Calyx	Nº mites Total
	1	730	30	760
	2	1.482	312	1.794
Control	3	1.049	87	1.136
	4	789	70	859
	5	974	63	1.037
	Total	5.024	562	5.586
	1	95	105	200
	2	107	56	163
Postharvest	3	114	111	225
Treatment	4	46	37	83
(Packing process)	5	61	19	80
	Total	423	328	751

Table 6 : Detection and Distribution of mites in control and Postharvest treatment Artificially infested fruits

Treatment	Replication	Mandarines, La Rosa Sofruco VI Region		
		Nº mites Fruits	Nº mites Calyx	Nº mites Total
	1	1.112	119	1.231
	2	877	86	963
Control	3	2.044	195	2.239
	4	753	110	863
	5	818	147	965
	Total	5.604	657	6.261
	1	81	69	150
	2	71	41	112
Postharvest	3	101	25	126
Treatment	4	58	44	102
(Packing process)	5	103	15	118
	Total	414	194	608

CONCLUSIONS

1. The normal packing process produces a significant elimination of *Brevipalpus chilensis* mites on clementines and mandarin oranges, with average elimination values of 85.4 percent to 89.3 percent.
2. The mites on the artificially infested control fruit showed a 7:1 distribution ratio for the mites detected on the fruits' surface vs. the mites detected under the calyx. After the packing process, the distribution ratio changed to 2:1.

APPENDIX: PHOTOGRAPHS



Clementines, clemenules



Artificial infestation



Clementines, Postharvest treatment
Packing process



Detection process



Detection Analysis

ANNEX 7

EFFICACY TEST FOR

Brevipalpus chilensis

ON CLEMENTINE FRUITS

FUNDACION PARA EL DESARROLLO FRUTICOLA

(FDF)

EFFICACY TEST FOR *Brevipalpus chilensis* DETECTION ON CLEMENTINE FRUITS

1. METHODOLOGY

1.1. Mite detection procedure

A method for detecting and identifying mites was developed based on the protocol used in the "Systems Approach" program for kiwifruit exports. That protocol is based on fruit washing as modified due to the botanical characteristics of this citrus fruit. The procedure generally consists of washing fruit with a solution of water and soap, and using sieves to collect any mites present on the fruit.

D. 1.2. Materials

The materials used were as follows:

Metal sieve with 200 mesh.

Metal sieve with 20 mesh.

Stereoscopic 40 x magnifying glass.

Washing container with high-pressure water supplied in shower form.

300 x to 600 x microscope.

Ethanol alcohol for conserving the samples.

Petri dishes.

3 to 5 ml glass tubes.

QUIX brand detergent.

Slides and slide covers.

Electric hot plate

Mounting medium (Hoyer, Euparal).

1.3. Testing the efficacy of the method

Clementines were artificially infested by placing them next to privet leaves carrying *Brevipalpus chilensis*, for 72 hours at 24°C, so that the mites could move over on to the fruit.

After infestation, the mites were observed under a stereoscope using direct light and their number was counted. Mites distributed on the fruit and on the pedicel disk were marked individually with a pen. Next, the pedicel disk was removed, and the mites below were counted.

After these steps, fruit and pedicel disk were washed separately. The washing process was as follows:

a) The sieves were placed one above the other, with the smallest mesh space (200-mesh) sieve below. Then the fruit and pedicel disk were placed separately on the sieve, in suitable quantities to allow them to easily be washed.

b) Next, the fruit and pedicel disk were sprinkled separately with a liquid soap and water solution (QUIX, two teaspoons per one liter of water). Each fruit and pedicel disk was then thoroughly washed under high-pressure water from a shower-type nozzle.

c) Straight away, the fruits or pedicel disks in the 200 mesh sieve were tipped to one side and washed with water at low pressure, letting the sieve's contents collect on one side. Next, with water, the contents were put on a Petri disk.

d) The mites from the fruit or the pedicel disk were observed under a stereoscope, in the same way as those previously marked on the fruit had been observed under the stereoscopic microscope.

Fruit or pedicels on which some mites remained after the first wash went through the procedure again in order to determine the effectiveness of this double washing.

2. RESULTS

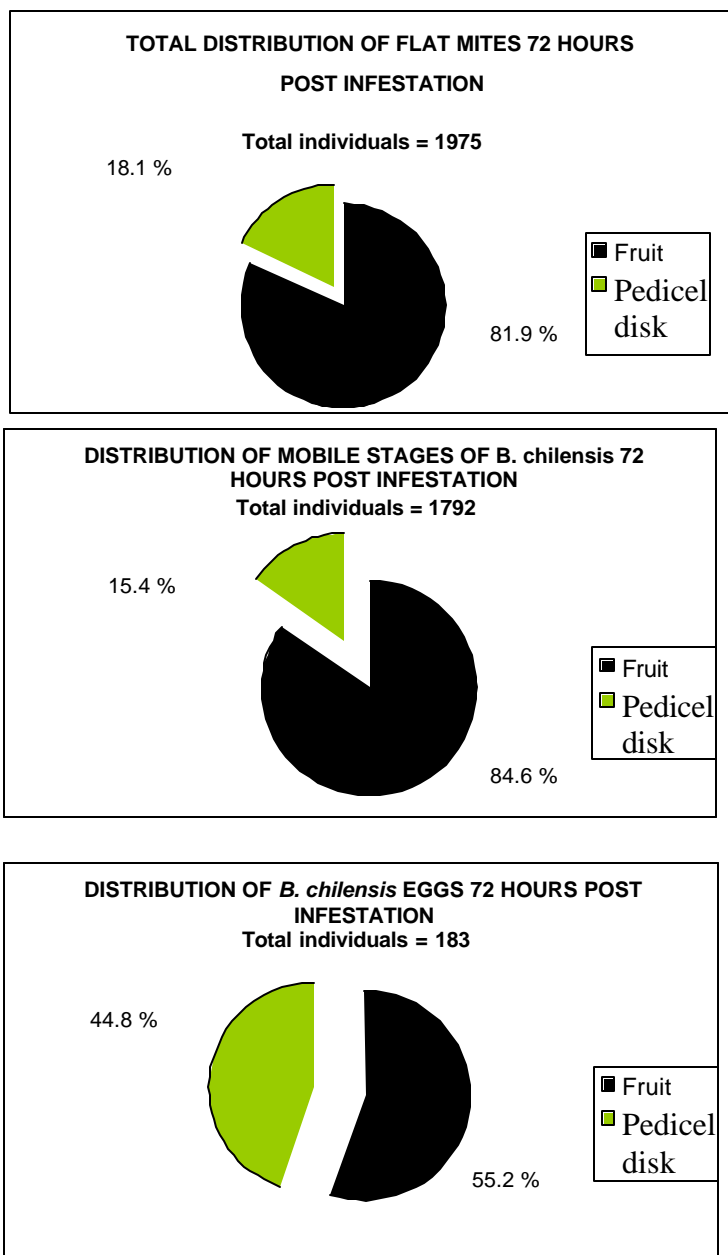
The results of detection efficacy of mites on fruit and pedicel with only one wash, with the second wash and with a double wash (the first wash plus the second wash) are shown in Table 1, which shows that both in the case of pedicel and of fruit, mite detection efficacy increased significantly by double washing, where efficacy reached levels of 97.6 percent for mites on fruits and 87.2 percent for mites on/under pedicel disk. This gives an average efficacy of **95.6 percent** with a 95 percent confidence ranging from 94.5 percent to 98.3 percent.

TABLE 1: EFFICACY TEST FOR *Brevipalpus chilensis* DETECTION ON CLEMENTINE FRUITS

Variable	N° of observations (Fruits/Pedicel disk)	BEFORE WASHING			AFTER WASHING			efficacy (% REMOVAL)		
		N° of mites Mobile stages	N° of Eggs	Total Cases	N° of mites Mobile stages	N° of Eggs	Total Cases	Mobile stages	Eggs	Total Cases
Fruits washed once (A)	189	1.516	101	1.617	102	61	163	93,3	36,6	89,9
Fruits washed twice (Mites not removed in first wash) (B)	47	102	61	163	4	35	39	96,0	42,6	76,1
Double-washed fruits (A+B)	189	1.516	101	1.617	4	35	39	99,7	65,3	97,6
Pedicel disk washed once (A)	189	276	82	358	37	52	89	86,6	36,6	75,1
Pedicel disk washed twice (Mites not removed in first wash) (B)	142	37	52	89	8	38	46	78,3	26,9	48,3
Double-washed Pedicel disk (A+B)	189	276	82	358	8	38	46	97,1	53,6	87,2
RESULTS OF DOUBLE-WASHED Fruits plus Pedicel disk	189	1.792	183	1.975	12	73	85	99,3	60,1	95,6

Figure 1 illustrates the distribution of mites in pedicel disk and fruits 72 hours after artificial infection. This shows that the mites distributed themselves mainly on the fruit (82%) and less on the pedicel disk (18%), unlike eggs that were spread equally between pedicel disk and flowers.

FIGURE 1: DISTRIBUTION OF *Brevipalpus chilensis* ON CLEMENTINE FRUIT 72 HOURS POST INFESTATION



ANNEX 8

SYSTEM APPROACH DE KIWIS CON DESTINO A USA

(USDA-APHIS Regulations, November 2001)

9.17 Kiwi Protocol (Approximate translation from Spanish)

SAG will work with the producers of kiwi to implement a " Systems Approach " that allows for recognition of orchards with low prevalence of *Brevipalpus chilensis* ("Grape flat mite"). The objective is to provide the option of a Joint Inspection by SAG/USDA, as an alternative phytosanitary condition of entry to the mandatory Methyl Bromide fumigation for kiwis exported to the United States.

Kiwi orchards interested in choosing this alternative must have controlled this pest at the field level to register with ASOEX for certification by SAG as low prevalence orchards. In order to grant this certification, SAG will evaluate the condition of each one of the orchards, prior to the beginning of the harvest. The evaluations will be conducted by SAG directly or through accredited companies designated by SAG.

USDA, APHIS will be able to monitor the process of certification by SAG, both at the stage of sampling in the orchard and in the laboratory analysis. To facilitate monitoring, each orchard will have a registration card listing the location of each one of the estates, in addition to the results of the orchard inspections, as well as of the laboratory analyses.

Once SAG has certified an orchard as a low prevalence production area, its fruit will be eligible for the alternative sampling and joint inspection in Chile. Each lot will be sampled on the basis of a percentage table. Additionally, at the site of inspection, the samples of Kiwi will be subjected to a specific process for the detection of mites. For this inspection, 29 fruits will be selected from several inspection units, in proportion to the number of boxes from the orchards represented in the lot.

If, using this inspection system, the Program reaches a level of 25% rejections considering all quarantine pests, including *B. chilensis*, USDA, APHIS will evaluate the predominant cause of the rejection percentage. On the basis of this evaluation, the suspension of sampling and joint inspection will be decided and may result in the reestablishment of fumigation as the only accepted condition for entry into the United States.

In case the conditions of the " Systems Approach " do not give the results that are expected, SAG, USDA-APHIS, and the ASOEX will be able to propose modifications and a new system will be able to be developed in the following seasons.