

Epidemiology of *Spiroplasma citri* in Corsica

P. Brun, S. Riolacci, R. Vogel, A. Fos, J. C. Vignault,
J. Lallemand and J. M. Bové

ABSTRACT. The leafhopper *Neoliturus (Circulifer) haematoceps* has been shown recently to be vector of *Spiroplasma citri*. *N. haematoceps* is present in Corsica and its distribution on the island has been surveyed. While the leafhopper extends from the coastal dune vegetation up to the "maquis" covered hills and mountains of the interior, it has never been found in citrus orchards. Several host plants of this leafhopper have been identified. *S. citri*-infected *N. haematoceps* have been found at certain times in various areas of the east coast of Corsica. *N. haematoceps* individuals naturally infected with *S. citri* are able to transmit the causal agent of stubborn to periwinkle plants, but transmission to citrus seedlings has not yet been attained. Wild host plants harboring *S. citri* are being sought.

The leafhopper, *Neoliturus haematoceps* Mulsant & Rey, is present in Corsica (1), as well as other countries of the Mediterranean area (4), and has been collected recently from different sites on the island.

Since this leafhopper is a vector of *Spiroplasma citri* (3, 5), and stubborn is an important disease for commercial varieties of citrus in orchards or in nurseries, it was necessary to investigate the status of the leafhopper and natural transmission of the disease. Surveys were conducted in various regions of the island in order to collect leafhoppers and determine the occurrence of *S. citri* in both the insect vector and its host plant.

MATERIAL AND METHODS

Research on the presence of *N. haematoceps* in various areas of Corsica has been done using a portable D-Vac suction sampler (D-Vac Corp., Riverside, Calif., U.S.A.) with a 33-cm-diameter intake. Samples with the D-Vac were made by placing the collecting cone as close as possible to the ground vegetation or pressed to the ground over the vegetation for a few seconds. For sampling bushes, the collecting cone was placed near the canopy and then swept up and down.

Leafhopper samples were sorted according to species (6) using a stereomicroscope after anesthesia with CO₂.

Detection of *S. citri* in field-collected insects or plants was conducted by enzyme-linked immunosorbent assay (ELISA) and by culturing the mycoplasma on artificial media (2, 7).

In some wild vegetation areas, as well as in the citrus mother blocks of the San Giuliano Research Station, periwinkles were used as indicator plant for natural transmission of *S. citri*.

RESULTS AND DISCUSSION

In an attempt to find the leafhopper *N. haematoceps* in Corsica, surveys were conducted in various regions of the island, along the coast or in the inland foothills. The main survey took place in the oriental coastal area corresponding to the major localisation of commercial citrus orchards. Samples were obtained from citrus orchards (on trees or on ground weeds), and in other crops, along roads, or from natural vegetation.

The different localities, where *N. haematoceps* was collected are indicated in table 1. In the north zone, as well as in the inland valleys, the populations were very low and each location gave only few individuals (1 to 5 adults). In the other areas, the leafhopper population of *N. haematoceps* was frequently low and only in some locations did the number of individuals rise to 10 or 50, or sometimes up to 100 adults. The "high-level" populations were located

TABLE 1
SITES SAMPLED FOR THE OCCURRENCE OF *NEOALITURUS HAEMATOCEPS*
IN CORSICA

| Area | Location | No. sites sampled |
|-------------------------------|---|-------------------|
| North zone (Bastia) | Arena — Lucciana — Marana — San Stefano | 4 |
| Occidental coast | Ile Rousse — Stareso — Galeria | 9 |
| Interior valleys | Corte — Venaco — Castirla — Ponte Leccia | 6 |
| East coast and Oriental plain | Bravone — Linguizetta — Aleria — Ghisonaccia — Diane — Alzitone — Rottani | 58 |
| South zone (Bonifacio) | Balistra — Ventilegne — Monaccia — Ortolo | 6 |

on the East coast (Bravone-Linguizetta-Diane) or in the South zone (Balistra-Monaccia).

Adults were the main stages collected during our survey; larvae were very scarce and some were collected only in places where relatively high population levels were found. We noticed that adults of *N. haematoceps* were present all year round. Overwintering stages were predominantly females.

In Corsica, *N. haematoceps* can be collected in two typical places—near the seaside on sandy coastal dunes and inland in typical vegetation called “maquis”. Within established citrus orchards in the oriental plain, it has never been possible to recover *N. haematoceps*. On sandy dunes, *N. haematoceps* is collected in places covered with different weeds as shown in table 2. Among these weeds, *Matthiola sinuata* R. Br., is well represented on the dunes and is found all year round. This plant is the preferred host for *N. haematoceps* in this situation; it breeds actively on *M. sinuata*. Adult females lay eggs in the midrib of leaves or near the margin of the blades. Young larvae and adults are able to feed on leaves and stems in spite of the dense trichomes covering these organs.

Salsola kali L. is also found in coastal dunes on bare sandy places near the sea, and also supports leafhopper populations. *N. haematoceps* can be collected year

round from these coastal dunes. Occasional fluctuations of populations may at times lead to a complete disappearance of the leafhopper as in 1985 at the Diane and Bravone sites.

Behind these coastal dunes exists another type of vegetation covering the hills and very well represented in Corsica. The composition of this typical vegetation called “maquis” is indicated in table 3. Within the maquis, *N. haematoceps*, is found only in sunny places where the soil is very poor, sandy, and dry in summer with sparse vegetation. It is possible to collect adults from the sunny side of pathways in the “maquis”, but not from the shaded side. As a whole, *N. haematoceps* populations are low in the “maquis” and most frequently few

TABLE 2
MAIN WEED SPECIES FOUND IN THE
EASTERN COASTAL DUNES OF
CORSICA

| SPECIES | FAMILY |
|----------------------------------|-----------------|
| <i>Pycnocomon rutaefolium</i> L. | Dipsacaceae |
| <i>Anthemis maritima</i> L. | Compositae |
| <i>Silene nicaeensis</i> All. | Caryophyllaceae |
| <i>Cynodon dactylon</i> Pers. | Gramineae |
| <i>Agropyrum junceum</i> P.B. | Gramineae |
| <i>Avena barbata</i> L. | Gramineae |
| <i>Lagurus ovatus</i> L. | Gramineae |
| <i>Crucianella maritima</i> L. | Rubiaceae |
| <i>Cakile maritima</i> Scop. | Crucifereae |
| <i>Matthiola sinuata</i> R. Br. | Crucifereae |
| <i>Asphodelus aestivus</i> L. | Liliaceae |
| <i>Plantago coronopus</i> L. | Plantagineae |
| <i>Eryngium maritimum</i> L. | Umbellifereae |
| <i>Salsola kali</i> L. | Chenopodiaceae |

TABLE 3
MAIN PLANT SPECIES IN THE
"MAQUIS" REGION OF CORSICA

| SPECIES | FAMILY |
|-------------------------------------|------------------|
| <i>Cistus monspelliensis</i> L. | Cistaceae |
| <i>Cistus salvifolius</i> L. | Cistaceae |
| <i>Lavandula stoechas</i> L. | Labiataeae |
| <i>Erica arborea</i> L. | Ericaceae |
| <i>Erica scoparia</i> L. | Ericaceae |
| <i>Arbutus unedo</i> L. | Ericaceae |
| <i>Genista corsica</i> D.C. | Leguminoseae |
| <i>Inula viscosa</i> Ait. | Compositaeae |
| <i>Logfia gallica</i> L. | Compositaeae |
| <i>Odontites lutea</i> Rehb. | Scrophulariaceae |
| <i>Myrtus communis</i> L. | Myrtaceae |
| <i>Phyllerea angustifolia</i> L. | Oleaceae |
| <i>Brachypodium racemosum</i> R. | Gramineae |
| <i>Briza maxima</i> L. | Gramineae |
| <i>Rosmarinus officinalis</i> L. | Labiataeae |

adults are collected from colonized spots (10 to 50 individuals).

Leafhoppers, froghoppers, and planthoppers associated with *N. haematoceps* represent 58 genera and 93 different species. Table 4 indicates the species most frequently collected during the survey.

Comparison between species collected in association with *N. haematoceps* in other countries of the Mediterranean area (Morocco, Syria, Turkey) indicates that 44 species are similar in Corsica with some differences in frequency and abundance within species. Only one species, is well represented with high population levels in Corsica: *Philaenus spumarius* L., which is not found in the other countries except for small numbers in Morocco. In Corsica, two other *Neoliturus* species are also represented besides *N. haematoceps*. Very few adults of *Neoliturus fenestratus* Herris Schaeffer, have been collected inland. *Neoliturus tenellus* Baker was collected in October 1986 for the first time in Corsica, near the sea at Diane on the east coast. A few adults were collected mixed with *N. haematoceps* adults.

The search for *S. citri* in field-collected leafhoppers or plant hosts was carried out in the places where *N. haematoceps* had been collected.

TABLE 4
PLANTHOPPERS, FROGHOPPERS AND LEAFHOPPERS COLLECTED IN
ASSOCIATION WITH *NEOLITURUS HAEMATOCEPS* IN CORSICA

| | |
|--------------------------|---|
| Fulgoroidea—Planthoppers | |
| Delphacidae | <i>Laodelphax striatellus</i> Fallen. <i>Toya propingua</i> Fieber. <i>Ribautodelphax pungens</i> Ribaut. |
| Cicadoidea—Froghoppers | |
| Cercopidae | <i>Philaenus spumarius</i> L. <i>Neophilaenus campestris</i> Fallen. |
| Cicadellidae—Leafhoppers | |
| Agallinae | <i>Agallia ribauti</i> Oss. <i>Agallia venosa</i> Fourc. <i>Austroagallia sinuata</i> M. R. |
| Typhlocybinae | <i>Empoasca vitis</i> Goethe. <i>Ribautiana debilis</i> D. <i>Eupteryx</i> (5 species). |
| Eupelicinae | <i>Eupelix cuspidata</i> F. |
| Aphrodinae | <i>Exitianus capicola</i> Stal. <i>Aphrodes bicinctus</i> Schrank. |
| Selenocephalinae | <i>Phlepsius spinulosus</i> Wgn. |
| Dectocephalinae | <i>Psammotettix</i> (4 species). <i>Euscelis</i> (4 species). <i>Balclutha rosea</i> Scott. <i>Macrosteles quadripunctulatus</i> Kbm. <i>Macrosteles sernotatus</i> Fallen. <i>Neoliturus fenestratus</i> Herris Schaeffer. <i>Neoliturus tenellus</i> Baker. <i>Neoliturus haematoceps</i> Mulsant & Rey. |

Samples of weeds and bushes representing the main botanical species found in the "maquis" or in littoral dunes were collected for *S. citri* detection.

Of 566 samples (468 plants, and 98 insects) analyzed with the ELISA procedure, or sampled for culturing *S. citri*, only 19 insects and 15 plants gave positive results. Positive results indicated that in Corsica *N. haematoceps* is the only species infected with *S. citri*. The two other species of *Neolaliturus* (*N. fenestratus* and *N. tenellus*) gave negative results.

Males and adult females of *N. haematoceps* are found to be naturally infected with *S. citri*. Isolation of *S. citri* from field-collected adults of *N. haematoceps* was possible at any season of the year: from October, November, December, March to June and August. Naturally infected leafhoppers have been collected only in the "maquis" along the oriental plain (Rottani, Ventulella, Bravone, Alzitone, Antisanti, Abazzia) at spots 2 to 12 km from the sea. The distance between the northern site (Ventulella) to the southern site is about 30 km.

Plants representing the main species growing in the "maquis" (20 families, 34 genera and 36 species), and especially within delimited sites where *N. haematoceps* is naturally in-

fectured with *S. citri*, have been tested many times at various seasons without positive results.

Positive results with plants were obtained only by artificial rearing on host plants. Adults of *N. haematoceps* naturally contaminated with *S. citri* and collected in autumn at three sites on the oriental plain (Ventulella, Rottani, Alzitone), could transmit *S. citri* to healthy plants of periwinkle grown in the greenhouse. Until now, no positive results have been obtained in transmission attempts to citrus seedlings. Since periwinkle is a very sensitive plant for detecting natural spread of *S. citri*, periwinkles were established in "maquis (Rottani) for 2 months in summer and for 8 months in a young grapefruit plantation surrounded by wild vegetation and located at 2 km from Ventulella. All of these plants were negative for *S. citri*.

Further research work is necessary for assessing the rate of transmission of *S. citri* by insect vectors to citrus or wild plants.

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