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CICADELLIDAE (HEMIPTERA: AUCHENORRHYNCHA) ASSOCIATED WITH MAIZE CROPS IN NORTHWESTERN ARGENTINA, INFLUENCE OF THE SOWING DATE AND PHENOLOGY OF THEIR ABUNDANCE AND DIVERSITY

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ABSTRACT - The diversity of Cicadellidae (Hemiptera: Auchenorrhyncha) associated with the maize crop in Tucumán province (Argentina) was studied. Samplings in two different sown dates, optimal and late, took place during the 2004-2005 and 2005-2006 growing seasons. The cicadellids were sampled from the coleoptile emergence to the vegetative stage V10. Thirty five species, belonging to six subfamilies, were identified, of which fifteen species were new records to Tucumán province, and nine were registered for the first time inhabiting maize agro-ecosystems. The most abundant and frequent species was *Dalbulus maidis* (DeLong & Wolcott), followed by *Agalliana ensigera* Oman and *Chlorotettix fraterculus* (Berg), which were present in all 44 samples. The diversity indices obtained were H'=1.75 and D=0.47 for the optimal sown date, and H'=0.46 and D=1.52 for the late one, being *Dalbulus maidis* the species with highest relative importance in both sown dates. Curves of seasonal abundance for the five most common species were showed.

KEY WORDS: Cicadellidae; *Zea mays*; Diversity; Seasonal abundance.

INTRODUCTION

Leafhoppers (Hemiptera, Auchenorrhyncha, Cicadellidae) represent one of the more conspicuous pests attacking maize crops in the world; they cause damages that vary from necroses to severe physiological alterations produced by their feeding and/or oviposition habits, and also by their ability to transmit pathogens (DAMSTEEGT, 1981). In reference to the importance of these insects as vectors, NIELSEN (1985) mentioned that there are 151 species in 65 genera, Deltocephalinae being the best represented subfamily, followed by Cicadellinae, Agallinae and Typhlocybinae. NAULT and KNOKE (1981) mentioned the genera *Dalbulus*, *Graminella*, *Euscelidus*, *Stirellus*, and *Exitianus* as major maize disease vectors.

In Argentina there are five maize diseases vectored by Auchenorrhyncha: "Maize Mosaic Virus" (MMV), "Mal de Rio Cuarto Virus" (MRCV) "Maize Rayado Fino Virus" (MRFV), "Maize Bushy Stunt Micoplasm" (MBSM) and "Corn Stunt Spiroplasm" (CSS); last three are persistently transmitted by the corn leafhopper, *Dalbulus maidis,* an important pest in tropical and subtropical areas of America (GAMEZ and LEON, 1985; NAULT, 1985; NAULT and AMMAR, 1989; OLIVEIRA *et al.*, 1998; PARADELL *et al.*, 2001; VIRLA *et al.*, 2004). CSS is the most important due to the steady growth in its incidence in the subtropical areas of the country, and to a slow advance into the temperate areas of high production, approaching the core maize area (GIMÉNEZ PECCI *et al.*, 2002a,b).

Studies regarding the Auchenorrhyncha communities inhabiting crops are mostly conducted in agro-ecosystems where hopper-transmitted diseases are involved, for example sugar-cane in Cuba (HIDALGO-GATO *et al.*, 1999), vineyards or almonds in Spain (TORRES *et al.*, 1998; LA SPINA *et al.*, 2005), plum trees in Brazil (HICKEL *et al.*, 2001), wheat crops and *Citrus* orchards in Argentina (REMES LENICOV and VIRLA, 1993; REMES LENICOV *et al.*, 1999). At the global level, the leafhopper fauna living in maize crops is poorly known and few references exist. REMES LENICOV *et al.* (1997), PARADELL (1995), and PARADELL *et al.* (2001) reported leafhopper species associated with maize crops, but these were mainly supported by the study of specimens deposited in entomological collections, and/or those captured by isolated samples from different localities of the country.

In Argentina, nearly three million ha are sown

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with maize, reaching an annual yield of almost 15 million tons (LORENZATI, 2005). During the last decade, the increment of soybean production in most of the traditional maize sowing area, due to the rise of international prices, moved the commercial maize crop towards northern Argentina (AVELLANEDA *et al.*, 2005). These circumstances enhanced the concurrence of the commercial maize production with the permanent area of distribution of *D. maidis* (PARADELL *et al.*, 2001), causing a high exposure to the diseases transmitted by it.

Tucumán is characterized by a long maize growing season, from October to April. A premature sowing could prevent high densities of *D. maidis* and consequently elevated CSS infections and yield losses, but one of the greatest barriers preventing early corn planting in the region is the lack of soil moisture during spring. Therefore, the optimum time to plant corn occurs during late November and the first ten days of December, when rainfall arrives. On the other hand, delaying crop planting beyond February can result in substantial yield loss.

The knowledge of the diversity in an agroecosystem, and the specific densities during the development period of the crop, is necessary to perform appropriate measures for the control of insect pests and to minimize yield losses (BARRIOS-DIAZ *et al.*, 2004). The lack of information regarding the diversity and seasonal abundance of leafhoppers affecting maize crops in northwestern Argentina prompted us to study these aspects in Tucumán province.

MATERIALS AND METHODS

Study site

The study was carried out in a cornfield of 2 ha, surrounded by wild vegetation placed in El Manantial (Dpto. Lules, Tucumán province) (26° 49' 50.2" S, 65° 16' 59.4" W, elevation: 495 m).

El Manantial is located in the foothills of San Javier Mountains, covered by the subtropical Yunga's rainforest. Mean annual rainfall is 1050 mm with a well defined dry season during winter and with 84% of rains occurring between November-March (spring-summer). Annual average temperature of the warmest month (January) is 24.7°C and, 11.6°C in the coldest month (July); the relative humidity average ascends to 74±9% (DE FINA, 1992).

During the study, the crop received all conventional cultural practices recommended for corn, except that no insecticides were applied. Climatic data (temperature, humidity and precipitation) were provided by the Climatological station at University of Agronomy campus (El Manantial, Tucumán), 1 km from the sampling area (Table 1).

The natural vegetation associated with the crop was represented mainly by monocotyledons: *Brachiaria* sp., *Bromus unioloides* (rescue grass), *Cenchrus echinatus* (southern sanbur), *Paspalum* sp., *Cynodon dactylon* (Bermuda grass), *Digitaria sanguinalis* (crab grass), *Eleusine indica* (goose grass), *Sorghum halepense* (Johnson grass) (Poaceae), *Cyperus* sp. (Cyperaceae), and several species of dicotyledons: *Amaranthus hybridus* (pigweed) (Amarantaceae), *Solanum* sp. (Solanaceae), *Euphorfia dentata* (toothed spurge) (Euphorbeaceae), *Eryngium* sp. (Umbelliferaceae), *Bidens pilosa* (hairy beggarticks) (Asteraceae), *Oxalis* sp. (Oxalidaceae), *Sida rhombifolia* (yellow barleria) (Malvaceae), *Verbena* sp. (Verbenaceae).

Sampling methods

Surveys were performed during 2004/2005 and 2005/2006 growing seasons, in two different sown dates: optimal (late spring, December 10) and late (February 8). During each sown date 11 samplings were taken, which took place twice a week, from coleoptile (Cot) stage to V10 (tenth leaf vegetative stage). Samplings were finished before the reproductive stage because the first growth stages are more susceptible to CSS acquisition (HRUSKA and GOMEZ PERALTA, 1997). During each sampling date, 200 maize plants (20 groups of 10) taken at random were searched for leafhoppers resting or feeding on maize plants. Individual plants were thoroughly inspected for adult insects. During the first phenological stages, the record of leafhoppers took place covering the plants with polyethylene bags, and then using manual vacuum aspirators, following the methodology described by CUADRA and MAES (1990). All samples were taken between 9:00 and 12:00 hs.

Insects were preserved in 70° ethanol and later identified in the Facultad de Ciencias Naturales y Museo (UNLP), where we deposited the voucher specimens.

Data analysis

We used the Spearman coefficient of rank correlation to compare the data obtained between the first and second growing season for each sown date.

TABLE 1 - *Climatic conditions at El Manantial site (Tucumán province) registered during the sampling periods (min: minimum; max: maximum).*

	Optimal sown period			Late sown period		
	mean	min. mean	max mean	mean	min. mean	max mean
Temperature (°C)	24.8	20.0	30.4	22.5	17.9	27.7
Relative humidity (%)	72.3	65.9	91.8	79.2	66.1	94.3
Total precipitation (mm)		301.8			280.4	

Regarding the diversity, we used the Shannon Wiener (H'), the Simpson (D), and Evenness (E) indexes:

$$H' = -\sum pi \times \log2pi \qquad D = 1-\sum (pi)^2 \qquad E = H'/H'max$$

where pi: ni/N, ni: number of individuals of species i and N: number of individuals of all species, H'max: log2S, S: number of species.

The relative importance (RI) for each species was determined, using the following formula:

$$RI = (ni/nt) \times (mi/mt) \times 100$$

where ni: number of individuals of species "i", nt: number of individuals of all species, mi: number of samples containing species "i", and mt: total number of samples.

The species that have a RI>0.5 were considered "frequent", those with RI ≤0.5≥0.001 "scarce", and "rare or occasional" with RI<0.001. A diversity analysis was made discerning the early (coleoptile to thirds leaf stage, cot-V3) and last vegetative stages (fourth to tenth leaf stages, V4-V10); arbitrally we decided to use this division because early stages are more susceptible to CSS (Scott *et al.*, 1977; Hruska and Gomez Peralta, 1997), and because V4 the crop began to be invaded by natural vegetation.

Insect density data were evaluated by analysis of variance (ANOVA), and the means were compared by a Tukey test using InfoStat Professional®. Previously, data were Ln (x+1) transformed.

RESULTS

Composition of Cicadellidae community

Altogether 11,595 individuals of 35 species, representing 6 cicadellid subfamilies (Agallinae, Cicadellinae, Deltocephalinae, Gyponinae, Typhlocybinae and Xerophloeinae) were collected from the maize crops during the two growing seasons (see appendix). Deltocephalinae, with 14 genera and 20 species, was the most species subfamily in the cornfield.

A total of 26 species was identified for the optimal sown period (4,580 individuals), whereas during the late sown date 7,015 individuals belonging 31 species were captured.

The study presents new records for Tucumán province of the following species: *Amplicephalus dubius* Linnavuori 1955, *Atanus angustus* Linnavuori 1959, *Balclutha incisa* (Matsumura 1902), *Balclutha lucida* Butler 1877, *Bucephalogonia xanthophis* (Berg 1879), *Curtara pagina* De Long and Freytag 1976, *Hortensia similis* (Walker 1851), *Paratanus exitiosus* (Beamer 1943), *Protalebrella brasiliensis* (Baker 1899), *Scopogonalia subolivacea* (Stål 1862), *Syncharina punctatissima* (Signoret 1854), *Stirellus bicolor* (Van Duzze 1892), *Stirellus picinus* (Berg 1879), *Tapajosa similis* (Melichar 1925), and *Unerus colonus* (Uhler 1895). Generally these are widespread species, so their occurrence in Tucumán is not surprising. Similarly, *Acinopterus gentilis* (Berg 1879), *B. lucida, Copididonus hyalinipennis* (Stål 1859), *Dechacona missionum* (Berg 1879), *P. brasiliensis, S. subolivacea, S. bicolor, T. similis,* and *U. colonus* are recorded for the first time inhabiting a maize agro-ecosystem.

The distribution of the recorded leafhopper species on the plants is diverse, *D. maidis* and *H. similis* are mostly located in the whorl, *D. missionum* and *E. obscurinervis* on the stems, and the other species predominantly on the leaf blades.

Leafhopper diversity and seasonal abundance patterns

The positive correlation obtained in optimal date (r_s=0.94) and late sown date (r_s=0.87), enabling us to add the results of both sampled growing seasons in order to facilitate interpretation (Fig. 1).

Although the total abundance of leafhoppers be-

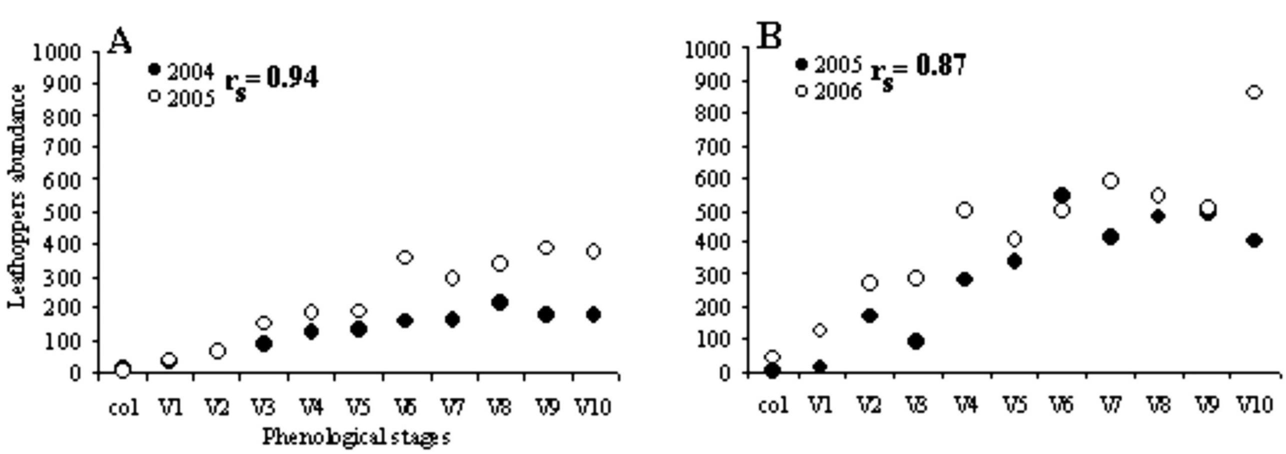

FIGURE 1 - Spearman's Correlation between leafhopper abundance obtained from the optimal (A) and late (B) sowing periods, in the two surveyed growing seasons. (r_s = Spearman coefficient).

TABLE 2 - *Measures of diversity separated by the two sown periods, and considering the early and last growth vegetative stages. (H': Shannon-Wiener diversity index; D: Simpson index; E: Evenness). The mean±SD followed by same letter are not significantly different (ANOVA, p≥0.05).*

	Optimal sown period						Late sown period					
	Abundance (mean±SD)	Richness	H'	H' max	D	E	Abundance (mean±SD)	Richness	H'	H' max	D	E
Total	4580 (5.62±1.22)a	26	1.75	4.7	0.47	0.37	7015 (6.19±0.92)a	31	1.52	4.9	0.41	0.31
Cot-V3	479 (4.34±1.21)a	13	2.09	3.7	0.32	0.56	1000 (5.25±0.93)a	14	1.41	3.8	0.41	0.37
V4-V10	4101 (6.36±0.20)b	25	1.65	4.6	0.64	0.36	6015 (6.73±0.23)b	31	1.53	4.9	0.41	0.31

tween the sown dates in the two growing seasons were different, this difference was not significant (F=1.53, P>0.05, G.L.=1). In contrast, when data from the earlier and last growth stages were compared, abundances during the optimal sown were significantly different (F=19.9, P<0.001, G.L.=1) and also for the late sown (F=17.05, P<0.001, G.L.=1). Generally, abundance was correlated with maize growth (Fig. 2, appendix).

The leafhopper community inhabiting the crop sown at optimal date was more species during the V8 phenological stage, while in late sown crops this parameter is high during V6 (Fig. 2). Except for minor drops, the abundance always increased with growth of the corn plant. The strong increment in the abundance during V9-V10 in the late sown period was associated with the population increment of the corn leafhoppers.

The diversity indices obtained were H'=1.75 and D=0.47 for the optimal sown date, and H'=0.46 and D=1.52 for the late one (Table 2).

Six species were measured as "frequent" for both considered sowing dates. These six species were collected exclusively during the optimal period whereas nine occurred solely in the late sowing date (see appendix). *D. maidis* was the most abundant and frequent species throughout the maize growth and the two sown dates, showing the greatest values of relative importance (RI). Almost 95% of the captured specimens corresponded to six species: *D. maidis*, *A. ensigera*, *C. fraterculus*, *H. similis*, *T. rubromarginata,* and *E. obscurinervis* (Table 3).

The curves representing the densities of the five most important species (considering abundance and frequency) show that the first species colonizing the

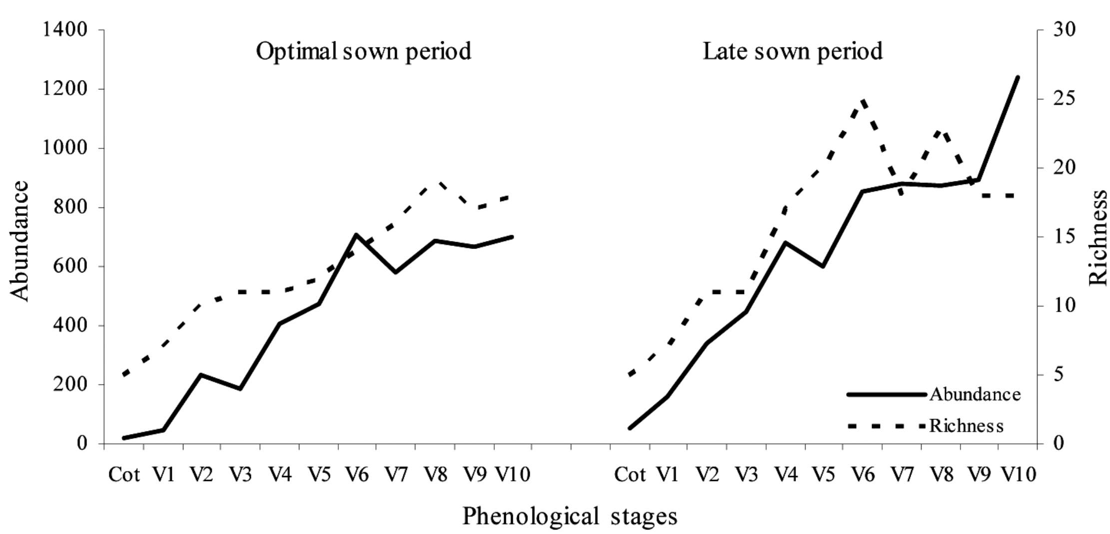

FIGURE 2 - Abundance and species richness of the leafhopper community in the different phenological states of the maize crop, in the optimal and late sown periods.

TABLE 3 - *Occurrence (%) and relative importance (RI) of the most abundant leafhoppers species in a cornfield at El Manantial (Tucumán province).*

	Optimal sown period		Late sown period	
	%	RI	%	RI
Dalbulus maidis	66.5	50.6	75.9	69.3
Agalliana ensigera	14.2	8.7	4.7	2.2
Chloretettix fraterculus	7.3	3.2	7.5	3.8
Hortensia similis	3.3	0.9	4.7	2.2
Tapajosa rubromarginata	3.1	0.7	1.5	0.5
Exitianus obscurinervis	1.3	0.1	0.7	0.08
Others spp.	4.3		5.0	

crop were *D. maidis*, *A. ensigera* and *C. fraterculus* and, after V2 arrived, *H. similis* and *T. rubromarginata* (Fig. 3).

The population growth of the corn leafhopper was always positive. During the early growth stages the vector registered a mean abundance of 0.3 and 0.6 specimens/plant for the optimal and late sown period respectively. At highly developed vegetative stages, *D. maidis* presented densities reaching 1.5 and 2.1 specimens/plant on average for the optimal and late sown dates, respectively.

Despite their continuous presence, most of the more frequent species varied in abundance over the maize growth stage. Remarkable seasonal patterns to be highlighted were the *A. ensigera* population, which in the optimal sown date had a population peak during V2 (0.35 specimens/plant), and the peak shown by the *C. fraterculus* population, registering 0.4 specimens/plant during V4 stage in the late sown period (Fig. 3).

DISCUSSION AND CONCLUSIONS

The maize agro-ecosystem in Tucumán province has an important leafhopper community, consisting of 35 species. Like previous reports by PARADELL (1995), the most representative subfamily of maize agro-ecosystem was Deltocephalinae. For the recorded 35 species, 9 of them are here recorded for the first time living in maize fields in Argentina. In addition, this contribution enlarges the geographical distribution for fifteen species (listed above) to include Tucumán province.

The most abundant and frequent species inhabiting maize crops and throughout both monitored period was *D. maidis*, as was also reported by PARADELL *et al.* (2001).

The Fig. 2 shows general trends in which both the abundance and richness values of leafhoppers increased with the growth of maize plants, as expected. When early and late growth stages were considered, high values were reached before V4 stage. This fact is related with the crop colonization by natural vegetation (mostly herbs). It is well known that taxonomically diverse plant habitats of-

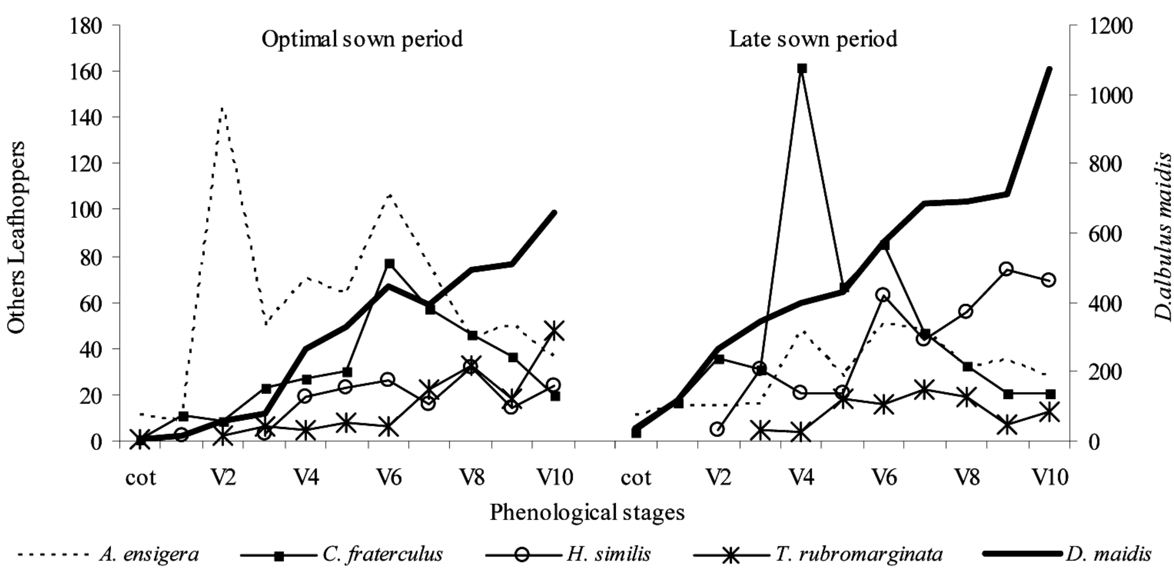

FIGURE 3 - Seasonal abundance of the five most common cicadellids (*D. maidis, A. ensigera, C. fraterculus, H. similis* and *T. rubromarginata*), during the growing seasons and separated by sowing period.

APPENDIX - Number of specimens and their relative importance of the leafhopper species (Cicadellidae) inhabiting the maize agro-ecosystem at El Manantial (Tucumán province) separated by the two sown dates [F: frequent (IR> 0.5), S: scarce (IR ≤ 0.5 ≥ 0.001), and R: rare (IR< 0.001), absent (---)].

Subfamily	Tribe	Species	Optimal sown date	Late sown date
Agallinae		***Agalliana ensigera*** Oman, 1934	650 (F)	327 (F)
Cicadellinae	**Cicadellini**	***Balacha*** sp.	---	1 (R)
		Bucephalogonia xanthophis (Berg, 1879)	3 (R)	1 (R)
		Ciminius platensis (Berg, 1879)	5 (R)	8 (S)
		Hortensia similis (Walker, 1851)	153 (F)	384 (F)
		Plesiommata mollicella (Fowler, 1900)	4 (R)	42 (S)
		Scopogonalia subolivacea (Stål, 1862)	2 (R)	2 (R)
		Syncharina punctatissima (Signoret, 1854)	39 (S)	6 (S)
	Proconiini	***Dechacona missionum*** (Berg, 1879)	36 (S)	68 (S)
		Tapajosa rubromarginata (Signoret, 1854)	142 (F)	104 (F)
		Tapajosa similis (Melichar, 1925)	1 (R)	---
Delthocephalinae	**Acinopterini**	***Acinopterus gentilis*** (Berg, 1879)	---	11 (S)
	Deltocephalini	***Amplicephalus dubius*** Linnavuori, 1955	5 (R)	7 (S)
		Amplicephalus marginellanus Linnavuori, 1955	1 (R)	---
		Haldorus sexpunctatus (Berg, 1879)	2 (R)	9 (S)
		Unerus colonus (Uhler, 1895)	1 (R)	11 (S)
	Euscelini	***Atanus angustus*** Linnavuori, 1959	---	2 (R)
		Atanus coronatus (Berg, 1879)	2 (R)	1 (R)
		Chlorotettix fraterculus (Berg, 1879)	333 (F)	524 (F)
		Chlorotettix neotropicus Jensen-Haarup, 1922	1 (R)	---
		Copididonus hyalinipennis (Stål, 1859)	29 (S)	28 (S)
		Exitianus obscurinervis (Stål, 1859)	62 (S)	52 (S)
		Paratanus exitiosus (Beamer, 1943)	4 (R)	1 (R)
		Planicephalus flavicosta (Stål, 1860)	---	10 (S)
	Hecalini	***Spangbergiella vulnerata lacerdae*** Signoret, 1879	5 (R)	---
	Macrostelini	***Balclutha rosea*** (Scott, 1876)	---	20 (S)
		Balclutha incisa (Matsumura, 1902)	---	16 (S)
		Balclutha lucida Butler, 1877	---	8 (S)
		Dalbulus maidis (DeLong and Wolcott, 1923)	3045 (F)	5323 (F)
		Stirellus bicolor (Van Duzze, 1892)	---	2 (R)
		Stirellus picinus (Berg, 1879)	---	4 (R)
Gyponinae		***Curtara pagina*** De Long and Freytag, 1976	10 (S)	7 (S)
Typhlocybinae	**Alebrini**	***Protalebrella brasiliensis*** (Baker, 1899)	7 (S)	15 (S)
	Empoascini	***Empoasca manubriata*** Young, 1953	33 (S)	19 (S)
Xerophloeinae		***Xerophloea viridis*** (Fabricius, 1794)	5 (R)	2 (R)

ten provide microclimates, greater availability of food sources, alternative hosts, and shelter sites that encourage colonization and population buildup of insect communities (COLL and BOTTREL, 1995; GIANOLI *et al.*, 2006). ALTIERI (1999) mentioned that the presence of weeds might increase the species richness and abundance of phytophagous and associated beneficial insects in agro-ecosystems. MOYA-RAYGOZA (1993) found a heavy association between the leafhopper species richness and the numbers of herb species associated with maize and two of its wild relative species.

The higher abundance and richness were recorded for the late sown date. Despite this, the obtained Shannon-Wiener diversity index for the late sown date showed a lower value than for the optimal period. This might be because *D. maidis* populations rose to an overabundance over the other species (i.e., loss of evenness). According to VIRLA *et al.* (2003), *D. maidis* densities increased remarkably in crops sown near autumn, when most of the cornfields in the region are almost dead and presumably because the insects could not shift from older crops to newly planted ones and therefore concentrated on the scarce yet green maize plants.

The registered densities of *D. maidis* (1.5 and 2.1 specimens/plant on average for the optimal and late sown dates respectively) are similar to the values recorded by DIAZ *et al.* (2005) when mentioned levels reaching a mean of 8.7% and 19.3% of CSS affected plant for an early sown date and a late, to the same site of study.

In an integrated pest management approach, knowledge on the diversity is closely related to many aspects of pest regulation, and allows the arrangement of functional groups that drive key ecosystem processes. Although no definitive support can be obtained from the diversity, richness, and seasonal abundance patterns with only two growing seasons, our description of composition and population changes of the leafhopper community inhabiting maize crops in northern Argentina serves as a reasonable first measure, giving baseline information for people interested on CSS epidemiology, and/or vectors control.

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