Activity 3. Taxonomy of the Springtails (Collembola) Associated with Cotton and Maize of the Cauca Valley, Colombia.

Introduction

The entomofauna associated with crops plays an important role in soil quality and consequently in crop yield. Agricultural practices can affect the diversity and abundance of groups of non-target organisms, among which we highlight the class Collembola, or springtails. These are small (250 microns to 10 mm in length) wingless arthropods that are similar to insects. Springtails are entognathous, have a postantenal organ, a maximum of 8 ocelli on each side of the head (8+8) or completely lack visual organs, and possess 4 antennal segments sometimes subdivided. The thorax has three segments, the first reduced in some groups; the legs have basically four segments. The abdomen has three segments on which specialized appendages are located on segment 1 (collophore), 3 (retinaculum) and 4 (furcula); the genital opening is located on segment 5. Developmental instars have gradual metamorphosis (ametabolous) (Greenslade 1991).

Springtails are ecologically important because of their influence in improving soil structure and accelerating the decomposition of animal and plant material. Most soils contain millions of springtail feces that can be beneficial in slowing the liberation of nutrients essential for plant roots. They also serve as substrate for a large number of microorganisms, stimulating the activity of fungi and bacteria, accelerating the processes of decomposition, and indirectly improving the structure, absorption capacity and fixation of interchangeable bases of the soil (Villalobos 1990; Cutz Pool 2002). On the other hand, springtails are the prey of many arthropods, particularly ants, beetles and predaceous mites, and thereby form a fundamental element of trophic interactions (Palacios-Vargas 2000).

Given the high abundance and diversity of organisms associated with the soil of agroecosystems, it is important to begin to understand the patterns of diversity and ecological function of groups like the Collembola. In Colombia, a study was conducted by the Agronomy Department of the National University of Colombia in Bogotá (Entomological Museum, UNAB) on the families of springtails associated with forage crops in the Department of Antioquia. This study identified 14 springtail families, seven of which constituted first reports of those families in Colombia (Ospina *et al.* 2003). This work was conducted as part of a broader project ("Collembola (springtails) of Colombia") initiated to make a preliminary inventory of the springtail fauna present in Colombia.

This technical report summarizes activities related to the separation, mounting and identification of springtails collected as part of a series of studies conducted in the Cauca Valley as part of the project "Evaluating the Impact of Biotechnology on Biodiversity." The first subproject focused on maize, and was designed to gauge the effect of chlorpyrifos on non-target soil fauna over two consecutive growing cycles (2002B and 2003A). The second subproject focused on cotton, designed to compare the non-target effects of conventional and Bt-transgenic plant protection strategies. Springtails were a dominant fauna collected in the course of these studies. In order to shed higher resolution of the response of this group to the experimental treatments, the specimens collected are being identified to better understand the diversity and function of this fauna in cotton and maize crops of the Cauca Valley.

Objectives

General Objective: Describe the springtail fauna associated with the soil of maize and cotton crops in the Cauca Valley.

Specific Objectives

- Identify the taxonomic families of springtails associated with maize and cotton crops.
- Compare the composition of the springtail fauna present in conventional cotton with that of Bt-transgenic cotton.
- Compare the composition of the springtail fauna captured in pitfall traps (surface-active) and soil cores extracted with Berlese funnels (soil-active).

Establishment and execution of the work plan: This work was based on specimens collected from two localities: (1) the CIAT experimental station, Palmira, located at 3º31'N, 76º21'W with an elevation of 965 m, mean annual temperature of 24ºC and (2) the ICA experimental station, Palmira, located at 3º31'N, 76º19'W with an elevation of 1295 m, mean annual precipitation of 24ºC and relative humidity 76%. Both localities correspond to the Holdrige classification of Dry Tropical Forest. At CIAT, specimens were collected as part of the activities of the project "Response of Non-Target Soil Arthropods to Chlorpyrifos in Colombian Maize", where the two treatments of maize with and without insecticide were evaluated over two consecutive cropping cycles (Subproject 1). At ICA, specimens were collected as part of the activities of the project "Effect of Transgenic Cotton [Bollgard® Bt Cry1A©] on Non-Target Soil Arthropods in the Cauca Valley of Colombia" where two treatments of conventional cotton (DP 5415) and Bt-transgenic Bollgard® cotton (NuCotn 33B) were evaluated (Subproject 2).

Springtails were separated from the original samples of arthropods that had been collected from maize and cotton and stored in 70% ethyl alcohol. While the springtails had already been separated from the other arthropods in the maize samples, they had to first be separated from the mixed arthropod samples collected from the cotton plots. This was performed by examining the samples under a stereoscope, separating all springtails, and returning the remaining arthropods to their original specimen vials.

Preparation and processing of springtail specimens: Springtails were initially sorted into morphospecies, taking into account the form of the body and the presence or absence of scales. These morphospecies were separated into different vials. In order to prepare the specimens for identification, they were fixed by clearing for 5 min in 10% KOH in individual glass wells, followed by submersion in lactophenol. The time in lactophenol varied depending on the size and pigmentation of the specimen; heat was used to accelerate the process as needed. In order to avoid the formation of crystals in the final fixing phase, the specimens were rinsed with Hoyer's solution to totally remove any reactives adhering to the body.

The fixed mounting was done under a stereoscope where dissecting pins were used to arrange the specimen in a drop of Hoyer's solution. The cover slip was placed on top with care to avoid forming bubbles. Finally, the slide was labeled, placed on a warming plate at 45-50ºC for a period of 4 days, after which the slide was examined to confirm that the Hoyer's solution had hardened. Once drying was finished, the excess Hoyer's solution was scraped off and the edges

of the cover slip were sealed with varnish. Finished mounts were stored in slide cases until identification of the specimen.

Identification of springtail specimens: Once the specimens were mounted, they were identified to family using the taxonomic keys of Palacios-Vargas (1990, 1991), Greenslade (1991), Jaensen (2001) and Ospina *et al.* (2003). Identification of genera was done in collaboration with Dr. José G. Palacios-Vargas, springtail specialist in Mexico, using the keys of Jaensen (2002) and Cristiensen and Bellinger (1980a, 1980b, 1980c, 1981). Finally, we (C. Ospina) developed a taxonomic key to the families of springtails associated with maize and cotton crops of the Cauca Valley based on the specimens mounted as part of this study (Appendix 1).

The reference collection produced from this work is housed at CIAT, in the office of the research group "Evaluating the impact of biotechnology on biodiversity." Voucher specimens will also be housed in the collaborating institutions of the Colombian National University in Bogotá (entomological museum UNAB) and Cornell University (CU Insect Collection).

Analysis of information: The information obtained in these studies will be analyzed with the indices of similarity of Jaccard and Sorensen appropriate for qualitative data. The degree of similarity will be determined and compared between crop type (maize vs. cotton) and sampling method (pitfall vs. soil cores extracted with Berlese funnels) at the level of family and genus.

Results

Springtails in maize: Over the two semesters of evaluation, a total of 5,444 specimens were captured from the class Collembola. Of those, 62.5% were captured in the insecticide treatment and 87.0% were captured during the first growing cycle (2002B).

The identified springtails belonged to three orders and six families, each of which has been previously documented in Colombia **(Table 1)**. The most abundant order was Poduromorpha with 27.3 and 5.6 times more individuals than the orders Symphypleona and Entomobryomorpha, respectively. In terms of abundance, no differences were detected between treatments at the level of order. There were statistically significant differences between semesters for each order. While Entomobryomorpha was more abundant in the second semester (2003A), the other two orders were more abundant in 2002B (Subproject 1)

Springtails in cotton: During the cropping cycle of cotton (2003), 229,425 specimens from the class Collembola were captured in pitfall traps, and 9,347 in soil samples (only partially analyzed to date). Of total captures, 59.8% were captured in Bt-transgenic cotton (Pitfall and Berlese), with 60.0% belonging to Poduroporpha. Both Poduromorpha and Symphypleona exhibited differences between treatments, being significantly more abundant in the Bt-transgenic plots (Subproject 2).

Eight families were identified to date. Of those, seven were detected in pitfall traps, and of those six were common to both treatments. The family Neanuridae was only detected in the conventional plots, and also only in pitfall traps **(Table 2)**. Six families were detected in soil samples and of those five were common to both treatments. The family Dicyrtomidae was only

detected in the conventional plots. In addition the family Cyphoderidae was only detected in soil samples (in both treatments).

Table 1. Families and genera of springtails associated with maize at the CIAT experimental station, Palmira, Colombia.

Family	Genus
Hypogastruridae	*Ceratophysella*
Isotomidae	*Isotoma*
Entomobryidae	*Seira*
	Entomobrya
Sminthurididae	*Sphaeridia*
	Denisiella
Bourletiellidae	*Deutosminthurus*

Of the 13 genera identified, eight were common to both treatments and collection techniques. The genera *Brachystomella* and *Arlesia* were only detected in pitfall traps (in both treatments), while the genus *Chypoderus* was only detected in soil samples (in both treatments). The genus *Salina* was only detected in the conventional treatment (in both berlese and soil samples)

Table 2. Families and genera of surface active (pitfall traps) and soil active (soil samples processed in Berlese funnels) springtails associated with cotton at the ICA experimental station, Palmira, Colombia.

		Pitfall		Berlese	
Family	Genus	NuCotn 33B	DP 5415	NuCotn 33B	DP 5415
Hypogastruridae	*Ceratophysella*	1	1	1	1
Brachystomellidae	*Brachystomella*	1	1	0	0
Neanuridae	*Arlesia*	0	1	0	0
Isotomidae	*Isotoma*	1	1	1	1
	Proisotoma	1	1	1	1
	Folsomides msp 1	1	1	1	1
	Folsomides msp 2	1	1	1	1
Entomobryidae	*Seira*	1	1	1	1
	Lepidocyrtus	1	1	1	1
Paronellidae	*Paronella*	1	1	1	1
	Salina	1	1	0	1
Cyphoderidae	*Cyphoderus*	0	0	1	1
Dicyrtomidae	*Calvatomina*	1	1	0	1

(1) Present; (0) Absent.

Arthropod Taxonomic Diversity: In terms of similarity in families between maize and cotton, for pitfall traps the Jaccard and Sorensen indices were 0.33 and 0.50, respectively, in other words 41.5% of the families captured in pitfall traps were common to both systems. In comparing collection techniques within the cotton crop, the similarity indices were 0.75 and 0.85 for Jaccard and Sorensen, respectively, i.e. 80% of families were sampled by both pitfalls and soil cores.

In terms of similarity in genera between maize and cotton, for pitfall traps the Jaccard and Sorensen indices were 0.19 and 0.32, respectively, or 25.5% overlap in genera between systems for pitfall traps. In comparing collection techniques within the cotton crop, the similarity indices were 0.85 and 0.92 for Jaccard and Sorensen, respectively, i.e. 88.5% of families were sampled by both pitfalls and soil cores **(Table 2)**.

A general view of the identified specimens is shown in **Figures 1-10**. These photos are of specimens in the reference collection mounted by C. Ospina and photographed by C. Olaya (CIAT).

Conclusions

- This report on the taxonomic diversity of springtails associated with maize and cotton crops is the first of its kind for any agricultural production system (other than forage crops) in Colombia.
- Of the genera identified to date, only the detection of *Brachystomella* and *Cyphoderus* depended on collection method; captures of these genera were limited to pitfall and soil cores, respectively.
- The similarity in family and genus composition of springtail fauna was low in comparing crop (maize and cotton), but high in comparing collection method (pitfall and soil core in cotton).
- For the first time, a key to the families of springtails associated with maize and cotton crops in the Cauca Valley of Colombia has been made developed, complemented with photos of select morphospecies.

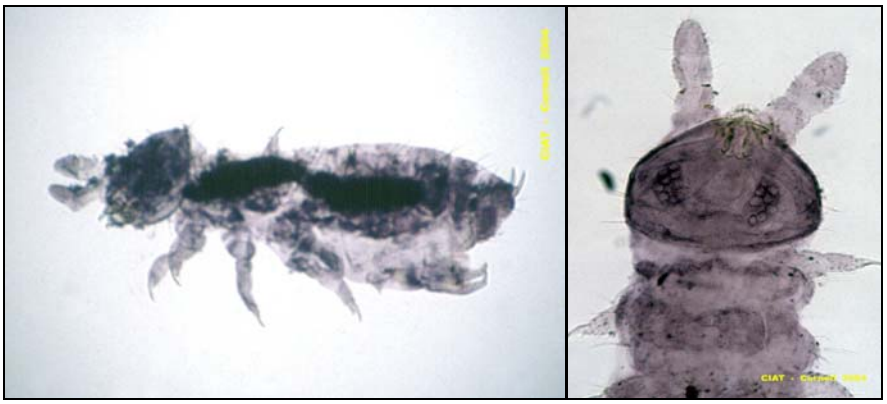

Figure 1. General habitus of *Ceratophysella* (Hypogastruridae), detail of the head, showing the 8 ocelli on each side of the postantenal organ (OPA), as well as the mouthparts formed from the mandibles and maxillae. Species present in maize and cotton.

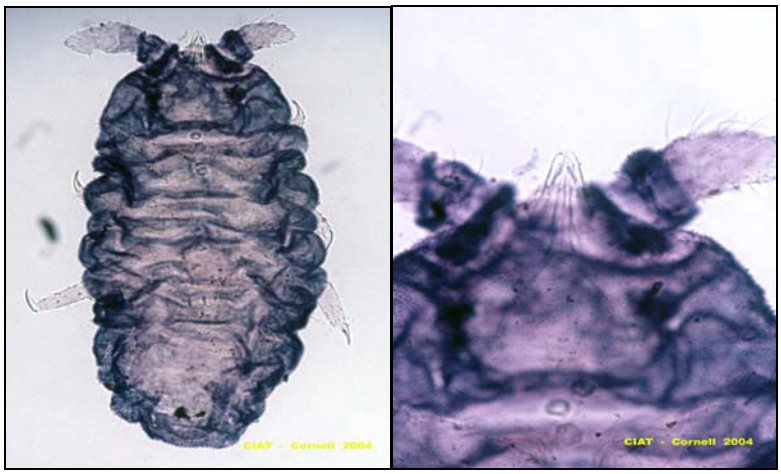

Figure 2. **General habitus of *Arlesia* (Neanuridae), detail of the stiletiform mouthparts. Species present in cotton.**

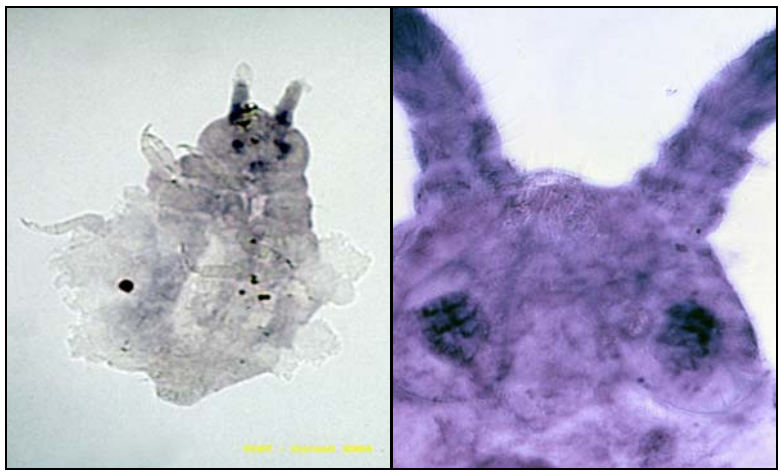

Figure 3. **General habitus of *Brachystomella* (Brachystomellidae), detail of the quadrangular mouthparts. Specimens collected in cotton.**

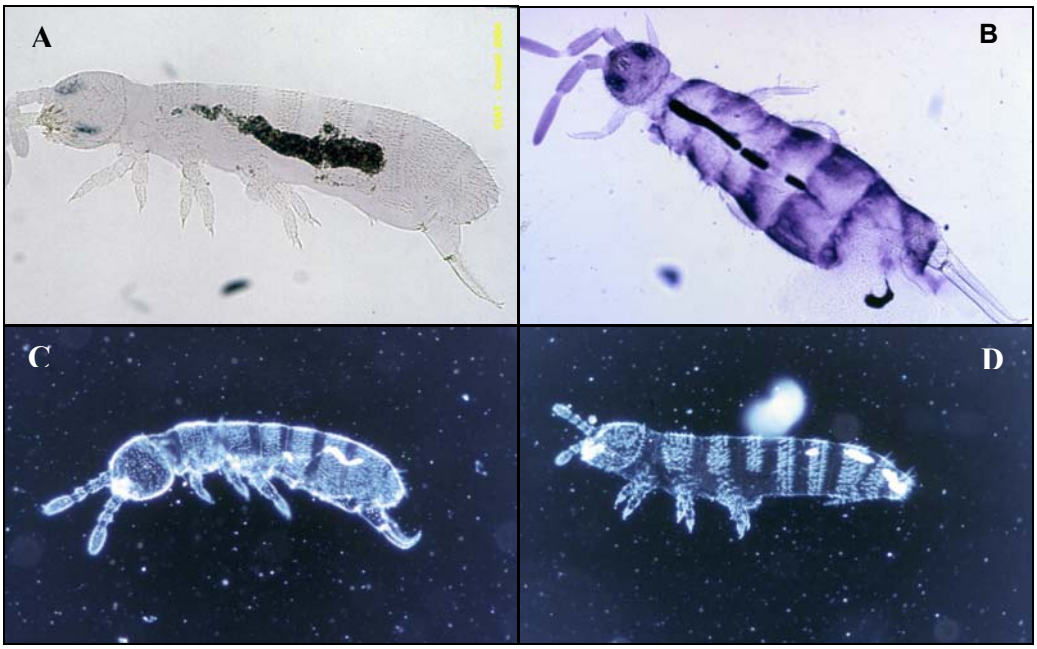

Figure 4. **General view of the genera of Isotomidae: (A) *Isotoma* (specimen from maize), (B) *Proisotoma*,(C) *Folsomides* sp. 1 and (D) *Folsomides* sp. 2 (specimens from cotton).**

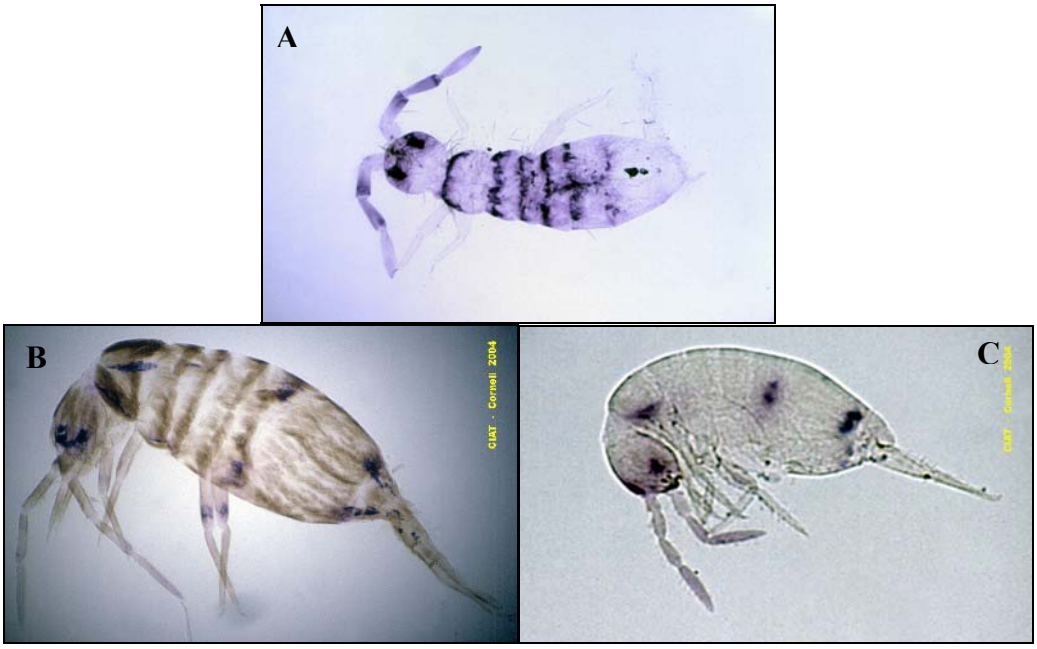

Figure 5. **Habitus of the genera of Entomobryidae: (A) *Entomobrya* (specimen from maize), (B) *Seira* and (C) *Lepidocyrtus* (specimens from cotton).**

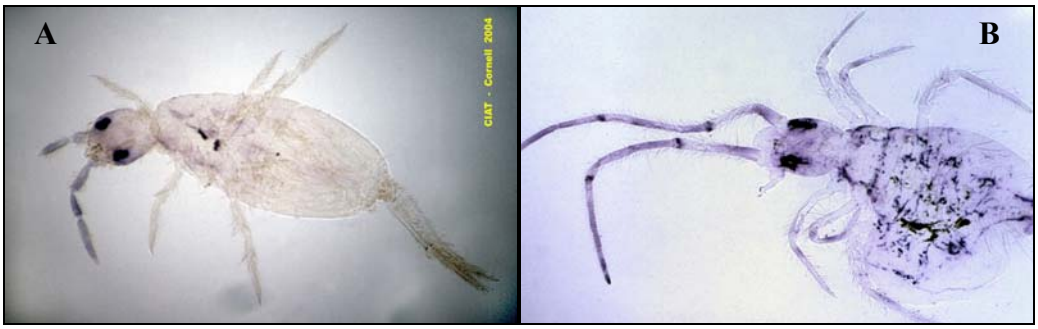

Figure 6. **Habitus of the genera of Paronellidae: (A) *Paronella* and (B) *Salina*, collected from cotton.**

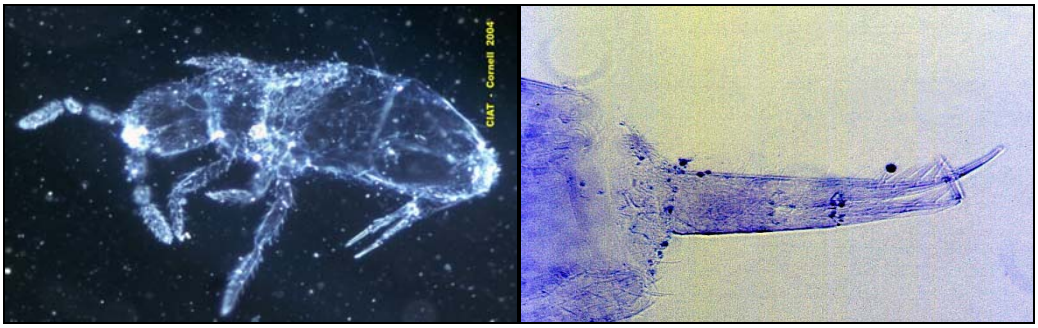

Figure 7. **General habitus of *Cyphoderus* (Cyphoderidae) with detail of the furcula, found in a sample of soil from cotton.**

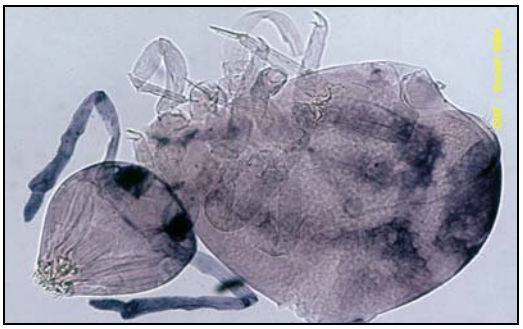

Figure 8. **General habitus of *Calvatomina* (Dicyrtomidae), specimen from maize.**

Figure 9. **General habitus of *Sphaeridia* (Sminthurididae), (A) male and (B) female, specimens collected in maize.**

Figure 10. General view of Deutosminthurus (Bourletiellidae), specimen collected in maize.

References

CUTZ POOL, L.Q. 2002. Colémbolos edáficos de dos agrosistemas de San Salvador, Hidalgo. Seminario de Edafología. México.

CHRISTIANSEN, K.A; BELLINGER, P.F. 1980a. Part 1. Poduridae and Hypogastruridae, The Collembola of North America North of the Rio Grande, Grinnell College, Iowa, p.1-386.

CHRISTIANSEN, K.A; BELLINGER, P.F. 1980b. Part 2. Families Onychiuridae and Isotomidae, The Collembola of North America North of the Rio Grande, Grinnell College, Iowa, November, 1980. p.387-784.

CHRISTIANSEN, K.A; BELLINGER, P.F. 1980c. Part 3. Family Entomobryidae, The Collembola of North America North of the Rio Grande, Grinnell College, Iowa, December, 1980. p.785-1042.

CHRISTIANSEN, K.; BELLINGER, P.F. 1981. Part 4. Families Neelidae and Sminthuridae; Glossary; Bibliography; Index, The Collembola of North America North of the Rio Grande, Grinnell College, Iowa. p.1043-1322.

GREENSLADE, P.1991.Collembola (Springtails). The Insects of Australia: a textbook for students research workers/ the division of Entomology, Commonwealth Scientific and Industrial Research Organization. Cornell University Press, Ithaca, New York. . 2nd ed. Vol. II Chap. 11:252-264.

JANSSENS, F. 2003. Checklist of the Collembola: Families. Department of Biology, University of Antwerp (RUCA), Antwerp, B-2020, Belgium http://www.collembola.org/taxa/collembo.htm

OSPINA, C M., SERNA. F.J, SERNA.S.L., PEÑARANDA.M.R. 2003. Colémbolos Asociados a Pastizales de Tres Zonas de Vida de Antioquia. Trabajo de Grado. UN. Facultad de Agronomía. Bogotá. (Unpublished).

PALACIOS-VARGAS, J.G.1990. Diagnosis y Claves para determinar las Familias de los Collembola de la Región Neotropical. Manuales y guías para el estudio de Microartrópodos. UNAM. Mexico D.F.

PALACIOS-VARGAS, J.G. 1991. Guide to the Springtails of Panamá and Costa Rica. (*Collembola*). *In:* QUINTERO D. y AIELLO A. (Eds.). 1992. Insects of Panamá and Mesoamérica. Selected Studies. Oxford University Press London. 692 p.

PALACIOS-VARGAS, J.G., 2004. Curso de Sistemática y Ecología de Microartrópodos del Suelo. Universidad Nacional de Colombia, Facultad de Agronomía Escuela de Posgrado. Bogotá.

PALACIOS-VARGAS, J.G.; CASTAÑO-MENESES, G.; ET MEJÍA-RECAMIER, B.E. IN LLORENTE BOUSQUETS, J.E.; GONZÁLEZ SORIANO, E.; PAPAVERO, N. 2000. 12 Collembola., Biodiversidad, Taxonomía y Biogeografía de Artrópodos de México: Hacia una Síntesis de su Conocimiento, Volumen II, III Grupos de Hexapoda, México, p. 249-264.

VILLALOBOS, F. 1990.Collembola del Noreste de México. Folia Entomológica Mexicana. 80 5-29 p.

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APPENDIX 1

KEY TO IDENTIFICATION OF THE FAMILIES OF SPIRNGTAILS ASSOCIATED WITH THE MAIZE AND COTTON CROPS IN THE CAUCA VALLE, COLOMBIA.

1. Body elongate (Figure 1) never globular; thorax and first four abdominal segments not fused; furcula present, well developed…… .....…..**2**
1' Body globular (Figure 2); at least the first four abdominal segments fused; furcula always well developed…….....…. **8**

2. Prothorax well developed (Figure 1), with dorsal setae; furcula not well developed……..... .....**3**
2'. Prothorax reduced (Figure 3), without dorsal setae; furcula frequently well developed..... .....**5**

3. With chewing mouthparts; mandibles with molar surface (Figure 4) ..... **HYPOGASTRURIDAE**
3' With modified mouthparts.....…….....**4**

4. Mandibles and maxillae present; maxillae stylifrom (Figure 5).....**NEUANURIDAE***
4'. Mandibles absent; maxillae square and usually with teeth (Figure 6) ....**BRACHYSTOMELLIDAE***

5. Body segments similar length (Figure 7); post-antennal organ (PAO) simple (Figure 8) ....**ISOTOMIDAE**
5' IV Abdominal segment elongated (alargado) (Figure 3), PAO absent.....**6**

6. Dens spined or toothed; mucro square, much shorter than the dens (Figure 9) ....**PARONELLIDAE***
6'. Dens spineless and tootheless.....….....**7**

7. Dens crenulate; mucro short, hook like with 1 or 2 teeth (Figure 10)..... .....**ENTOMOBRYIDAE**
7'. Dens smooth; mucro elongate with variable number of teeth .....**CYPHODERIDAE***

8. Antennae elbowed between segments II y III, segment IV much shorter than III (Figure 12).....…… .....….....**DICYRTOMIDAE***
8'. Antennae elbowed between segments III y IV, (Figure 13), segment IV longer that III.....….....**9**

9. Abdominal segments V and VI fused , males with prehensile antennae (Figure 14), female lacking anal appendages..…… ..... ….....**SMINTHURIDIDAE****
9' Abdominal segments V and VI separate, males with simple antennae; female with anal appendages (Figure 15); mucro spatulate (Figure 16) ..... .**BOURLETIELLIDAE****

* Only family in cotton.
** Only family in maize.

FIGURES

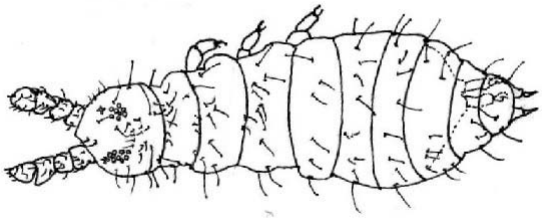

Figure 1. Habitus of Hypogastruridae.

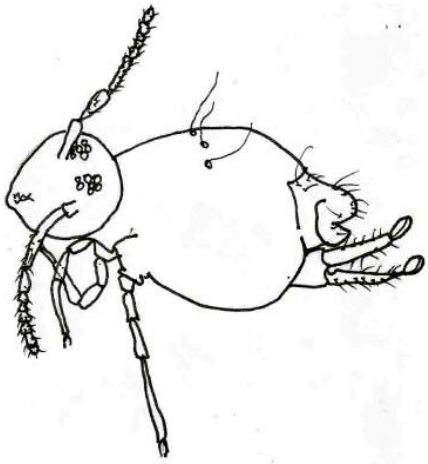

Figure 2. Habitus of Bourletiellidae.

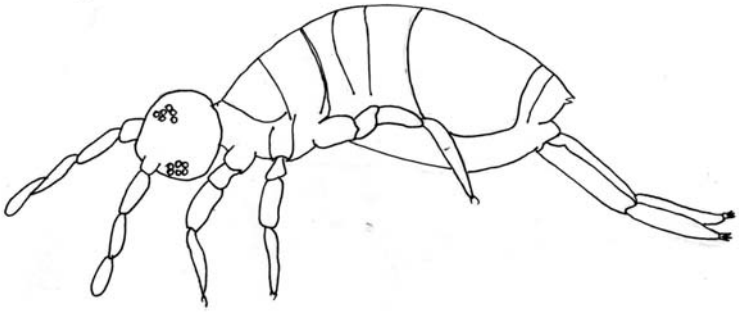

Figure 3. Habitus de Paronellidae.

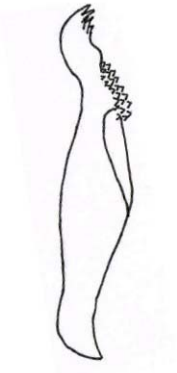

Figure 4. Mandible, *Ceratophysella.*

Figure 5. Mandibles and maxillae modified, Neanuridae.

Figure 6. Maxillae of *Brachystomella.*

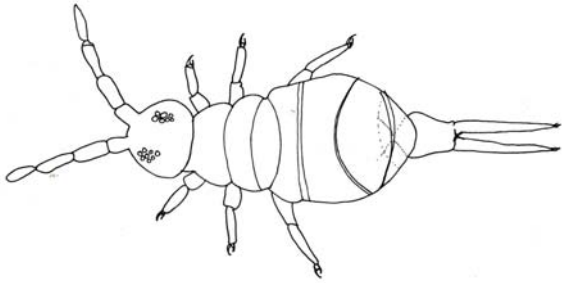

Figure 7. Habitus of *Isotoma.*

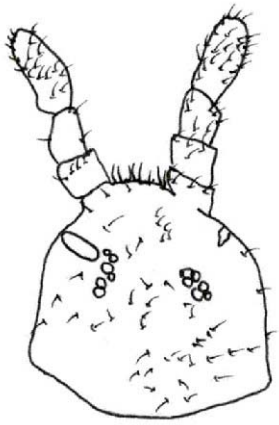

Figure 8. OPA of *Folsomides.*

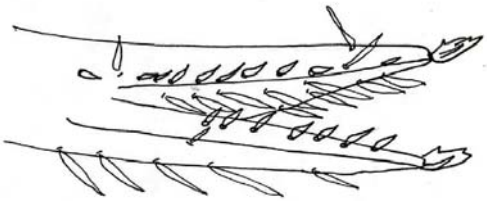

Figure 9. Furcula of *Paronella.*

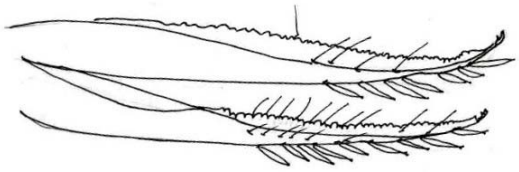

Figure 10. Furcula of *Lepidocyrtus.*

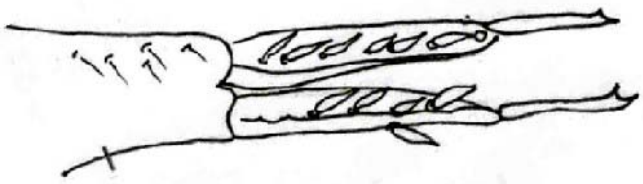

Figure 11. Furcula of *Cyphoderus.*

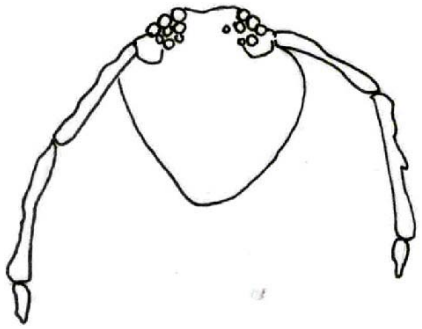

Figure 12. Head of *Dicytoma.*

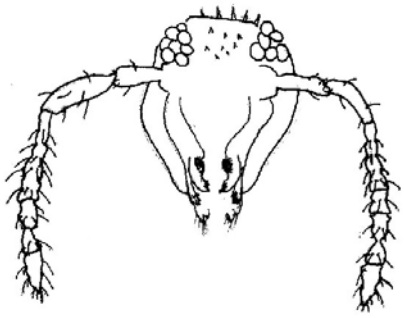

Figure 13. Head of Sminthuridae Ospina *et al* 2003).

Figure 14. Antennae of *Sphaeridia* (Macho) (Ospina *et al* 2003).

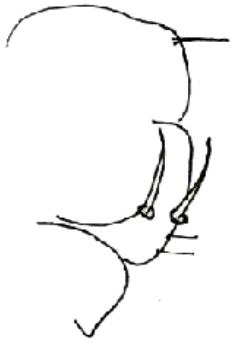

Figure 15. Anal appendages of *Deutosminthurus.*

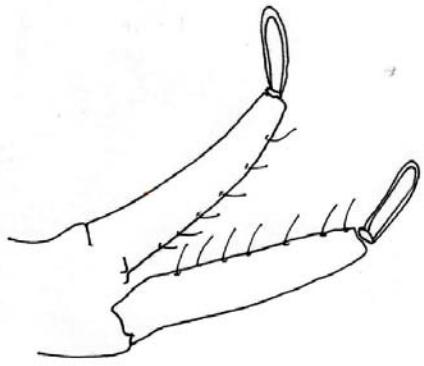

Figure 16. Mucro of Bourletiellidae.

Activity 4. Publications, posters, conferences, training and consultancies.

Publications

Rodríguez, Ch.J.; Castro, U.; Morales, A.; Peck, D.C. 2003. Biología del salivazo *Prosapia simulans* (Walker) (Homoptera: Cercopidae), nueva plaga de gramineas cultivadas en Colombia, Revista Colombiana de Entomología, 29(2):149-155.

Rodríguez, Ch.J.; Peck, D.C. 2004. Diversidad y abundancia de artrópodos del suelo en algodón Bt (NuCotn 33B) y algodón convencional (DP 5415) en el Valle del Cauca. Memorias, XXXI Congreso de la Sociedad Colombiana de Entomología, Bogotá, Colombia. p. 115-124.

Peck, D.C.; Rodriguez, Ch.J.; Gómez, L.A. 2004. Identity and first record of the spittlebugs *Mahanarva bipars* (Hemiptera: Auchenorrhyncha: Cercopidae) on sugar cane in Colombia. Florida Entomologist, 87(1).

Gaviria, J.D.; Rodríguez, Ch.J. 2004. El salivazo o mion de los pastos (Homoptera: Cercopidae) en la Cañicultura Colombiana. Tecnicaña, 8(15). p. 4-12. (ISSN 013-0409).

Publications in Preparation

Rodríguez, Ch.J.; Peck, D.C. 2004. Parámetros poblacionales de *Zulia carbonaria* (Homoptera: Cercopidae) en condiciones controladas sobre *Brachiaria ruziziensis.* SOCOLEN.

Rodríguez, Ch.J.; Peck, D.C. 2004. Biología y hábitos de *Mahanarva andigena* (Homoptera: Cercopidae) en condiciones de casa de malla. Neotropical entomology.

Posters

Rodríguez, Ch.J.; Peck, D.C. 2004. Gauging the effect of transgenic maize and cotton on non-target soil arthropods in Colombia. 8th International Symposium on the Biosafety of Genetically Modified Organisms. September 26-30, Montpellier, France.

Conferences

Rodríguez, Ch.J.; Peck, D.C. 2004. Gauging the effect of transgenic maize and cotton on non-target soil arthropods in Colombia. 8th International Symposium on the Biosafety of Genetically Modified Organisms (September 26-30, Montpellier, France). p. 102-104.

Rodríguez, Ch.J.; Peck, D.C. 2004. Parámetros poblacionales de *Zulia carbonaria* (Homoptera: Cercopidae) en condiciones controladas sobre *Brachiaria ruziziensis*. Resúmenes XXXI Congreso Sociedad Colombiana de Entomología, SOCOLEN. July 28-30, Bogotá, Colombia. p. 43.

Rodríguez, Ch.J.; Mojocoa, A.M.; Peck, D.C. 2004. Efecto de Clorpirifos sobre artrópodos no-blanco del suelo en maíz. Resúmenes XXXI Congreso Sociedad Colombiana de Entomología, SOCOLEN. July 28-30, Bogotá, Colombia. p 35.

Ospina, C.M.; Rodríguez, Ch.J.; Mojocoa, A.M.; Serna, C.F.; Peck, D.C. 2004. Colémbolos asociados a cultivos de maíz y algodón en el Valle del Cauca, Colombia. Resúmenes XXXI Congreso Sociedad Colombiana de Entomología, SOCOLEN. July 28-30, Bogotá, Colombia. p 57.

Gauging the effect of transgenic maize and cotton on non-target soil arthropods in Colombia

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INTRODUCTION

The Colombian Ministry of Agriculture and Development, through the Instituto Colombiano Agropecuario (ICA), designed a scheme to determine the viability of incorporating GMOs into the agricultural production process. In 1998, ICA published resolution 03492 to regulate and establish the procedures for the introduction, production, liberation and commercialization of GMOs. Through Agreement 013/98 and 0002/02, ICA created the National Technical Council for Agricultural Biosecurity (NTC) to function in the regulation of GMOs.

Since the establishment of those regulations, applications have been submitted for *Brachiaria*, carnations, cassava, coffee, cotton, maize, rice, *Stylosanthes* and sugar cane. Of these, only four have been approved to date: (i) carnations for cut-flower production, (ii) cotton for commercial production, (iii) rice for small scale field trials, and (iv) maize for biosecurity tests (Diaz 2003).

At present, the biosafety information available to researchers and regulators in Colombia stems from studies conducted in other countries that largely represent temperate regions. That ex-situ experience has to be effectively transferred to the tropical and developing country arenas if we are to successfully gauge the magnitude of GMO effects on the abundance, diversity and ecological function of non-target arthropods.

OBJECTIVE

Evaluate and compare the impact of GMO and non-GMO plant protection technologies on non-target soil arthropods in Colombian maize and cotton.

MATERIALS AND METHODS

Due to delays in the approval of Bt-transgenic maize, an initial study was conducted on the soil insecticide chlorpyrifos in conventional maize. The research was conducted at the International Center for Tropical Agriculture (CIAT), located at 3º31' N, 76º21' W, 956 m elevation, mean annual rainfall 1000 mm, mean temperature 24ºC, and Holdridge life zone classification Dry Tropical Forest. There were eight experimental plots (43 x 43 m each) evaluated over two consecutive cycles of maize (second semester 2002 and first semester 2003). The two treatments were maize (commercial hybrid "Master" from Syngenta) with and without insecticide incorporated at planting to control soil-active lepidopteran pests, in particular the impact of *Spodoptera frugiperda* (Noctuidae) as a cutworm.

In a second phase of activities, in collaboration with ICA's division of Agricultural Regulation and Protection, field studies were initiated in cotton to establish the effects of Bollgard® technology (Bt-transgenic cotton insecticidal to lepidopteran pests). The first of three consecutive cycles (first semester 2003), in rotation with soybean, was conducted at the ICA research station in Palmira, located at 03°31'N, 76°19'W, 975 m elevation, annual precipitation 1295 mm, mean temperature 24ºC, relative humidity 76%, and Dry Tropical Forest. There were 24 experimental plots (15 x 15 m each) with four replicates of six treatments based on plant material (Bollgard® technology represented by the var NuCont 33B with the Cry1A(c) gene and conventional technology represented by var DP 5415) and insecticide regime (conventionally applied insecticides, insecticides to control non-lepidopteran pests, and Bt-based insecticides). Because economic thresholds were never reached in the first cycle, no insecticides were applied and the data were analyzed as two plant variety treatments.

Fig. 1. Pitfall traps showing (A) fixed component, (B) removable component and (C) lid.

Information was gathered from two types of samples: pitfall traps to sample surface-active arthropods (maize and cotton) and soil cores extracted with berlese funnels to sample soil-active arthropods (only cotton). Pitfall traps were located between plants within the rows; eight were put out in each experimental plot (Fig. 1) and these were opened to sampling for a 24-hour period each week. Soil samples were taken with a cup cutter (10 cm diam, 10 cm depth) every 2 wk from within the row between plants (Fig. 2). Four samples were taken from each experimental plot. Samples were placed in berlese funnels for 24 hours, then arthropods were sorted from the debris and stored in 70% ethyl alcohol until analysis (CIAT 2003; Mojocoa 2003; Rodriguez & Peck 2004). The statistical model used for the analysis of the data was a completely randomized block design.

Fig. 2. Field collection of samples for berlese extraction of arthropods (A) cup cutter, (B) soil sample, (C) berlese funnels.

With this design an ANOVA will be used to determine differences in abundance among treatments and determine the effect of their interactions. In addition, for the most abundant groups we will conduct an analysis of the area under the population curve (accumulated insect-days) to determine differences among treatments during the trial. We will also compare the diversity and abundance among treatments using various indices of taxonomic diversity, dominance and equity.

RESULTS

Surface-active arthropods (pitfalls): In the two-cycle maize study a total of 11,850 arthropods were captured and sorted from pitfall traps, representing five taxonomic classes and 18 orders; 58.7% of individuals were captured in the insecticide plots (Table 1). Poduromorpha and Acarina were the most abundant orders with 37.8 and 19.7% of specimens, respectively. Treatments had a significant effect on two orders, Acarina (more abundant with insecticide) and Thysanoptera (more abundant without insecticide). There were significantly more pitfall captures in the first cycle compared to the second.

In cotton, 438,934 specimens were captured in the first cycle, representing eight classes and 20 orders (Table 1); 54.3% of individuals were captured in NuCotn 33B (Bt-transgenic) plots. Sixty-five different species have been identified and only three of these were not present in both NuCotn 33B and DP 5415 (Table 2). The most abundant class was Collembola with 52.3% of total captures (Table 1). Poduromorpha, Hymenoptera and Acarina were the most abundant orders with 50.4, 29.7 and 17.2% of specimens, respectively. Treatments had a significant effect on two orders, Collembola and Isopoda, each more abundant in NuCotn 33B.

Table 1. Number of individuals and composition of invertebrate classes caught in pitfall traps in maize (2002-2003) and cotton (2003).

Taxonomic class	Maize			Cotton		
	With insecticide	Without insecticide	Total	NuCotn 33B	DPL-915	Total
Arachnida	1,979	1,298	3,214	37,321	38,877	76,998
Chilopoda	10	50	30	8	2	8
Collembola	3,467	2,027	5,444	138,061	91,364	229,425
Diplopoda	102	94	196	17	26	43
Insecta	1,462	1,514	2,966	62,668	70,443	133,108
Malacostraca	0	0	0	112	48	160
Nematoda	0	0	0	25	108	133
Oligochaeta	0	0	0	0	1	1
Symphyla	0	0	0	27	27	54
Total	6,961	4,989	11,850	238,295	200,639	438,934

Table 2. Abundance of invertebrate orders (mean number of individuals captured per evaluation date) in maize (2002-2003) and cotton (2003).

Order	Maize		Cotton	
	With insecticide	Without insecticide	NuCotn 33B	DPL-915
Araeine	11.38±1.23 a	7.44±0.59 b	10.65±1.79 a	8.08±0.35 a
Araneae	4.57±24.61 a	2.53±9.66 a	4.98±25.59 a	2.03±1.48 b
Blattaria	0.02±0.15 a	0.01±0.09a	0	0.03±0.18
Chilopoda[1]	0.10±0.39 a	0.15±0.55 a	0.13±0.54 a	0.11±0.41 a
Coleoptera	3.60±6.46 a	2.49±4.21 a	3.38±6.02 a	2.67±4.82 a
Dermaptera	0.10±0.45 a	0.04±0.24 a	0.09±0.44 a	0.05±0.25 a
Diplopoda[1]	0.82±1.79 a	0.76±1.64 a	0.25±0.69 b	1.37±2.23 a
Diptera	0.91±2.07 a	0.85±1.85 a	0.43±0.97 b	1.36±2.85 a
Entomobryomorpha	3.19±3.62 a	3.26±4.83 a	1.22±1.81 b	5.35±5.03 a
Hemiptera	0.98±2.43 a	0.67±1.79 a	1.43±2.79 a	0.38±0.75 b
Homoptera	0.31±0.63 a	0.43±0.86 a	0.36±0.67 a	0.38±0.84 a
Hymenoptera	4.32±8.37 a	5.91±15.06 a	5.71±15.89 a	4.49±8.21 a
Isopoda	0.52±1.06 a	0.63±1.42 a	0.56±1.42 a	0.58±1.06 a
Lepidoptera	0.02±0.13 a	0.02±0.13 a	0.01±0.09 a	0.03±0.16 a
Nematoda	0.14±0.41 a	0.19±0.50 a	0.13±0.42 a	0.20±0.50 a
Neuroptera	23.66±112.35 a	12.48±84.12 a	34.77±136.23 a	0.26±1.05 b
Oligochaeta	0.72±1.86 a	0.60±1.41 a	1.02±2.13 a	0.26±0.71 b
Orthoptera	0.05±0.22 b	0.19±0.62 a	0.20±0.60 a	0.04±0.20 b
Poduromorpha	0.75±1.46 a	0.60±0.97 a	0.81±1.54 a	0.53±0.79 a
Symphyla[1]	11.38±1.23 a	7.44±0.59 b	10.65±14.79 a	8.08±9.35 a
Symphylosoma	4.57±24.61 a	2.53±9.66 a	4.98±25.59 a	2.03±1.48 b
Thysanoptera	0.02±0.15 a	0.01±0.09a	0	0.03±0.18
Unidentified	0.10±0.39 a	0.15±0.55 a	0.13±0.54 a	0.11±0.41 a

For each row and crop, means followed by different letters are statistically different at $P<0.05$ (Tukey-Kramer test for multiple comparisons).[1]Taxonomic class.

Soil-active arthropods (berlese): To date, 44% of the samples collected in the first cycle of cotton have been evaluated, numbering 80,541 specimens representing 11 classes and 21 orders (Table 3). The most abundant classes were Arachnida and Insecta with 65.0 and 20.4% of total specimens, respectively. The most abundant order was Acarina, with 65% of total captures and 1.2 times more abundant in DP 5415.

Diversity indices: In maize, the species richness index (S) was not significantly between treatments or between semesters. The Shannon diversity index and Simpson dominance index were significantly different between semesters but not between treatments. In terms of species similarity, the Jaccard index showed that 97 and 91% of orders were in common between treatments and semesters, respectively.

In cotton, the species richness, Shannon and Simpson indices were not significantly different between the treatments NuCotn 33B and DP 5415. Values were 14.1, 1.0 and 0.4, respectively. The Jaccard index showed that 80% of orders were in common between the two treatments.

Table 3. Number of individuals and composition of invertebrate classes extracted from soil cores in cotton (2003).

Taxonomic class	NuCotn 33B	DP 5415	Total
Arachnida	24,127	28,218	52,345
Chilopoda	36	44	80
Collembola	4,709	4,638	9,347
Diplopoda	14	42	56
Diplura	48	70	118
Insecta	7,679	8,717	16,396
Malacostraca	46	63	109
Nematoda	12	20	32
Oligochaeta	155	215	370
Protura	3	4	7
Symphyla	729	952	1,681
Total	**37,558**	**42,983**	**80,541**

CONCLUSIONS

- These studies have identified a high abundance and diversity of soil-active and surface-active fauna associated with the cotton crop under the conditions of the Cauca Valley, Colombia.
- Pitfall traps are an appropriate method for measuring the abundance of surface-active arthropods and comparing their activity and diversity across treatments.
- Extracting soil cores with berlese funnels is an adequate method for measuring the abundance of soil-active arthropods and comparing their activity and diversity across treatments.
- The various indices of taxonomic diversity, richness, dominance and equity are useful tools for comparing ecological communities and will allow us to make long-term comparison of the effects of different plant protection technologies under the conditions of the Cauca Valley, Colombia.
- The abundance differences observed between treatments in the first cycle of cotton should be studied in more detail to define how GMOs affect those differences. The protocols established in the first cycle will therefore be implemented in two additional cycles to better describe abundance effects over time, and to gather information to compare differences in species composition of key groups such as the springtails.
- Although abundance and diversity differences may exist in response to GMO technology, it is important to determine whether the magnitude of those differences is ecologically relevant, i.e. have an effect on ecological function or overall soil health.

REFERENCES

DIAZ, A.L. 2003. Situación de los organismos modificados geneticamente en la República de Colombia. Informe al Consejo Técnico Nacional de Bioseguridad (CTN). pp. 8.

CIAT (CENTRO INTERNACIONAL DE AGRICULTURA TROPICAL). 2003. Annual Report Project PE-1. Integrated Pest and Disease Management in Major Agroecosystems. Cali, Colombia. p. 43-52.

MOJOCOA, M. 2003. Efecto del uso de clorpirifos en maiz (*Zea mays* L.) sobre los artrópodos no-blanco del suelo. Undergraduate research thesis, Ingeniero Agrónomo. Facultad de Ingeniería Agronómica, Universidad del Tolima, Ibagué, Colombia. 44 p.

RODRIGUEZ, CH. J. & D.C. PECK. 2004. Diversidad y abundancia de artrópodos del suelo en algodón Bt (NuCotn 33B) y algodón convencional (DP 5415) en el Valle del Cauca. In Memorias, XXXI Congreso de la Sociedad Colombiana de Entomología, Bogotá, Colombia. p. 115-124.

ACKNOWLEDGMENTS

We thank Ana Luisa Diaz (ICA), Gerson Fabio V. (CIAT), John Losey and Leslie Allee (Cornell).

8th International Symposium on the Biosafety of Genetically Modified Organisms (September 26-30, 2004, Montpellier, France)

Training Received

Visiting Scholar in the Department of Entomology. Jairo Rodríguez, IPM Project. NYSAES, Cornell University, 15 Sep.-15 Dec., 2003.

Workshop on Gene Flow Analysis and Environmental Briosafety. Jairo Rodríguez, IPM Project. CIAT, Cali, Colombia, 7-9 Oct. 2004.

Course "Taxonomia de Coleopteros: Sacarabaeidae de Importancia Agricola (Coleoptera: Scarabaeidae "Pleurosticti)." Caludia, M. Ospina, Paola Soelo, Anyimilehidi Mazo, IPM Project. CIAT, Cali, Colombia, 6-10 Sep. 2004. (Professor: Jhon César Neita Moreno, Universidad Nacional, Bogotá).

IV Curso de Educación Continuada Museo Entomológico UNAB. Sistemática y Ecología de Microartrópodos. Claudia M. Ospina, IPM Project. Universidad Nacional, Bogotá. 3-14 May 2004.

Curso Bioecología y MIP del Salivazo. Jairo Rodríguez, IPM Project. CIAT, Palmira, Colombia, 16-20 February 2004.

Bayer CropScience, Integrated Pest Management in Cotton. Jairo Rodríguez, IPM Project. Buga-Valle, 30 April 2004.

CORPOICA, Palmira. CORPOAGRO – IV Encuentro de Actualización Agrícola. Seminario 9. Palmira, 4 Sept. 2004.

Thesis Completed

Mojocoa, A M. 2004. Efecto del uso de Clorpirifos en Maíz (*Zea mays* L.) sobre los artrópodos no-blanco del suelo. [Universidad del Tolima, Ibagué, Colombia]. 43 pp.

Thesis in Progress

Mazo, V.A. Efecto del Algodón Bollgard® sobre la diversidad y abundancia de artrópodos del suelo durante su segundo año en el Valle del Cauca. (Bióloga). Universidad del Valle, Colombia.

Proposals funded in 2004

- Assessing the impact of biotechnology on diversity: effect of transgenic maize on non-target soil organisms, (Biotechnology and Biodiversity Interface). US$ 99,360, Cornell, 2002-2005.
- Protocol for the Monitoring and resistance Development to de Bollgard® Technology in Colombia. US$ 30,960, CoaCol, 2004-2005.

Proposals submitted in 2004

- La respuesta y recuperación de los artrópodos no-blanco del suelo frente al uso del algodón transgénico. US$ 77,985, Colciencias, 2005-2006.

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