SOIL PESTS - CASSAVA AND OTHER CROPS

Activity 1. Integrated control of subterranean pests in South America.

Introduction

Soil pests cause serious economic losses to many important crops in South America. Until about 20 years ago white grubs and burrower bug *Cyrtomenus bergi* were not considered important pests in Latin America. However, in the recent past outbreaks are no longer seasonally restricted as before and are present in many agro-ecosystems, such as hillsides, tropical lowlands, including savannas and forest margins in Central and South America (King & Saunders, 1984; Posada, 1993; Londoño & Pérez, 1994; Pardo, 1994; Corpoica, 1996; Shannon & Carballo, 1996). Inappropriate agricultural methods like inadequate cropping patterns, burning of harvest residues, deforestation, cultivation of marginal land, discontinuance of tillage, loss of floral and faunal biodiversity, reduction of organic material, ill-timed and excessive applications of synthetic pesticides and elimination of natural enemies of pests are considered to be the key factors responsible for the increased pest status of white grubs and *C. bergi* (Posada, 1993; Londoño, 1994). The frequent use of highly toxic soil pesticides can lead to development of resistance in pests and is additionally very often ineffective. The link between pesticide use and soil and water contamination is well documented, as well as its threat to farmers' and consumers' health (e.g. Mervyn, 1998).

Five outputs were envisaged in our project in order to accomplish the major objective, that is to identify the key pest species and to develop appropriate IPM strategies for their effective control:

I. Problem diagnosis from the farmers' perspective and identification of local existing knowledge and cultural practices.
II. Description of pest problem (taxonomic identification of the white grub species complex, rapid identification of larvae, distribution and occurrence of species, yield loss and damage estimates).
III. Characterization of potential biological control agents (search for natural enemies, identification and propagation, laboratory and field evaluation).
IV. Establishment of network for soil arthropod research (electronic compilation of relevant data, communication network, website).

I. Problem diagnosis by means of farmer interviews.

Methodology: The problem diagnosis from the farmers' perspective is a key tool for a thorough identification of the pest problem. We interviewed 172 farmers in the departments of Quindío (99), Risaralda (52) and Northern Cauca (21). Farms were identified with the help of the local Umatas, rural meetings where farmers were asked about their problems with soil pests and in an arbitral manner. On all selected farms one of the following crops were cultivated: cassava, onion, and pasture. We included cassava because this crop is of increasing importance in the coffee zone and because of CIAT's expertise in this crop. Onions have a long tradition in the hillsides of La Florida, La Bella and La Colonia, three villages close to Pereira in a range of 1600 – 2000 meters above sea level (asl).

The questionnaire focused on farmers' perception of the principal pest species and their management practices, such as use of pesticides, crop rotation, intercropping, previous and adjacent crops, degree of damage, soil management and other factors. The great majority of the persons who were interviewed were only temporary 'majordomos' or tenants and did not have sufficient background knowledge for fully answering all questions. For this reason we visited additional 29 farms near Montenegro (Quindío), where the farmers confirmed soil pests as one of the major pest problems.

Results and Discussion: Fifty-two farmers considered trips as their major pest problem, followed by the hornworm (*Erinnyis ello*) (32) **(Figure 1)**. Only 16 farmers mentioned white grubs and six *C. bergi* as a problem. In contrast, onion farmers only considered burrower bug and white grubs as important insect pests. These results indicate that farmers' perception of a pest problem is apparently related to its visibility. When harvesting, onion farmers find the burrower bug directly feeding on the onions and can thus relate directly the damage to the pest. However, in cassava *C. bergi* is not present when the root develops to a mature tuber. Likewise white grubs are not present throughout the entire growing season. Thus it is not surprising that farmers considered the trips or the hornworm as the most important pests although we rarely observed the presence of these insects. Almost no pest species were reported on pasture. We frequently observed that farmers confounded burrower bug damage with *Phytophthora* root rot.

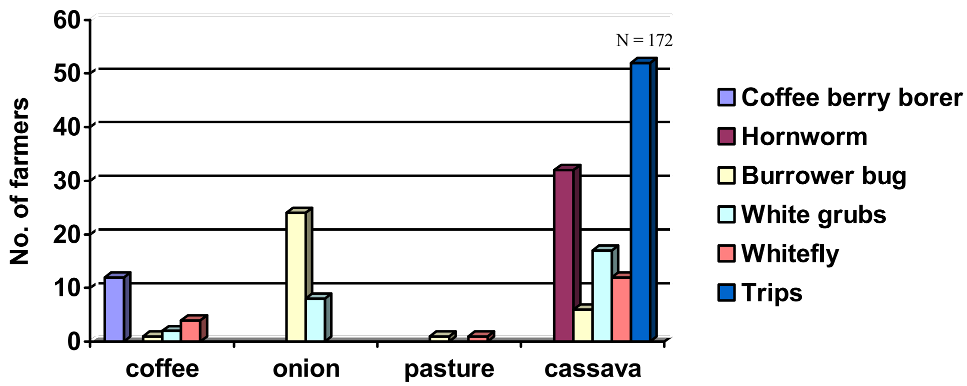


Figure 1. Farmers' perception of key pests.

Consequently only six farmers (all crops) complained about yield losses greater than 76% due to white grub attack; 15 farmers reported losses in a range of 51 – 75%, and 15 farmers estimated the losses between 25 – 50%. In the case of burrower bug only five farmers reported losses between 51 – 75% on onion **(Figure 2**).

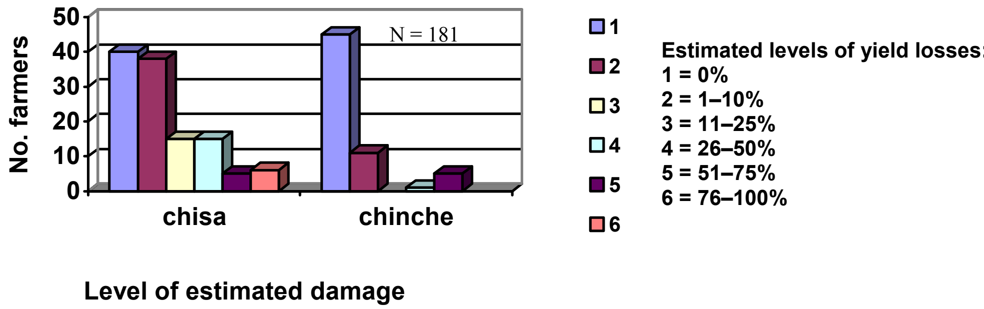


Figure 2. Estimated level of yield losses due to soil pests of cassava in Quindío, Risaralda and Cauca. The interviews were realized in the second semester off 2002.

Of the 29 farmers who recognized the presence of soil pests, 71% applied insecticides for control of white grubs and/or burrower bug; 14% used biological control and another 14% combined insecticides and with biological and organic methods (*e.g.* entomopathogenic fungi, lime, mulch). 82% were convinced that their applied treatment (chemical and/or biocontrol) is effective , only 17% denied the efficiency. Use of insecticides was the predominant control strategy (86%), and carbofuran was the most frequently used active ingredient (32%). 10% of the interviewed farmers used other strategies such as combination of onion and detergent. 5% of them applied a combination of an insecticide and entomopathogenic fungi.

Training of farmers: In order to enhance the perception of the soil pest problem together with SENA Armenia we organized a two days farmers' training workshop on identification of white grubs and burrower bugs. In December 2003 we will organize a follow-up meeting to evaluate the adoption rate of this knowledge.

II. Description of pest problem.

Methodology: In September 2002 we started stratified surveys (frequency once a month) in cassava, potatoes, onion, and pasture for soil pests to study the fluctuation (time and spatial) of scarabids. The crops were selected according to their economic importance in each region: Cassava in Northern Cauca (990 m asl) and in Pereira (1400 m asl), onion in La Florida and La Colonia in Risaralda (1660 m and 2000m asl, respectively), potatoes in North and East Antioquia (2100 - 2800 m asl) and in Cundinamarca at the Centro Agropecuario Marengo (CAM) (2547 m asl) and in Cogua (3200 m asl). Additionally, we monitored pastures in each zone because of its high susceptibility to white grub attack. First results indicate that 10 samples of 100 x 100 x 15 cm per site are an appropriate sample size. We selected this sampling method because it provides more accurate data on abundance and diversity of white grubs than the monolith (30 x 30 x 25 cm) (Pardo 2002). Moreover, we collected adult beetles at weekly intervals by means of light traps in each region (North of Cauca, Pereira, and La Florida).

Collaboration with partner institutions: In order to be able to realize stratified surveys we established the cooperation with Research Center of Corpoica "La Selva" in Rionegro (Antioquia), the National University of Bogotá and University of Caldas, Manizales. This cooperation is crucial for fulfilling the project output of identification of the pest species complex in various agroecological zones. CIAT focused activities on surveys in the North of Cauca and gave logistic support for surveys in Risaralda such as transportation and supplies where the University of Caldas participated in forms of two undergraduate theses.

Moreover, the project collaborates with two German institutions. The Federal Institute of Biological Research (Germany), that identified fungi and other entomopathogens and Kiel University that collaborated in improving our activities with entomopathogenic nematodes.

In December 2002 we received a call from the Hacienda Córcega (1330 m asl) (Quimbaya, Quindio), one of the biggest coffee producers in this zone. White grubs had severely attacked coffee grains that were stored in sacks for seedlings next to a maize field. The administration of the hacienda collaborates in executing the field surveys and is willing to conduct research activities in order to develop control strategies. We also receive logistic support from SENA Armenia sending us the sampled material for identification.

Results

Santander de Quilichao: Until August 2003 at CIAT's experimental station in S/Quilichao we collected in soil samples pasture 2,434 larvae, 46 pupae, 114 adult scarabs, and 61 parasitoids. This corresponds to a density of 6.3 larvae, 0.2 pupae, 1.6 adults and 0.6 parasitoids per m^2. In the cassava plots we found 769 larvae, 10 pupae, 10 adult scarabs, and 13 parasitoids, corresponding to a density of 5.9 larvae, 0.1 pupae, 0.1 adults and 0.2 parasitoids per m^2.

In the light trap we collected in total 17,355 specimens, belonging to the following scarab genera: *Cyclocephala, Plectris, Dyscinetus, Aphodiinae, Anomala, Phyllophaga, Pelidnota, Lygirus, Sericini, Podischnus, Strategus,* and *Lycomedes.*

Eleven months of light trap data indicate a response of scarabids to the short but not to the long rainy seasons, with a distinct peak in flight activity around October/November but not in March-May. During October/November we collected more than 50% of our annual collection.

The most abundant scarabid genus was *Cyclocephala* (50%), followed by *Plectris* (28%), *Aphodiine* (9%), *Dyscinetus* (2.7%), *Anomala* (2.3%), *Phyllophaga* (2%), and *Lygirus* (1.8%). Predominant species were *Cyclocephala amazonica* (28.3%) and *Plectris* sp. 2 (17.4%) **(Figure 4)**. *Cyclocephala amazonica* was trapped throughout the entire observation period. In contrast, high numbers of *Plectris* sp. 2 were trapped in October and a smaller peak was recorded in June. In general, all species peaked in October.

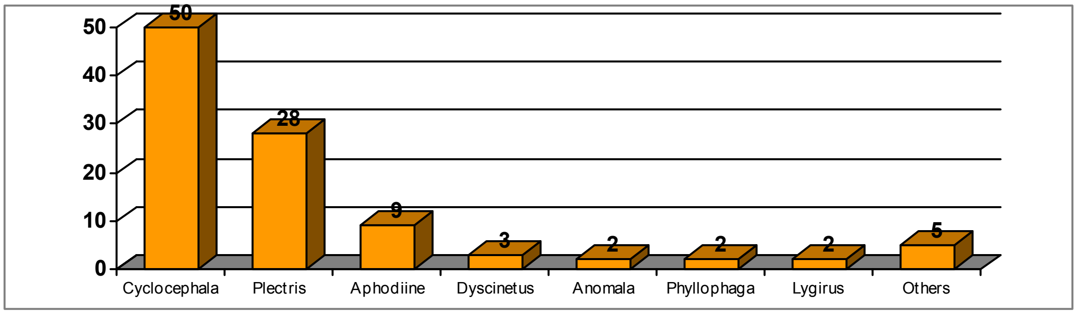


Figure 3. Most abundant Melolonthidae species (adults) collected from September 2002 to June 2003 in light traps in Quilichao.

Cyclocephala amazonia was the most abundant white grub larvae found in pasture and cassava. *Phyllophaga* sp. and *Leucothyreus* followed in pasture. The sequence was reverse in cassava where *Leucothyreus* was more abundant than *Phyllophaga* sp. *Plectris* spp., a genus of presumed economic importance was almost absent. We did not observe any damage due to white grub attacks, neither on cassava nor on pasture. *Cyclocephala amazonia* is commonly not considered as a pest. The presumed important pest genera *Phyllophaga* and *Plectris* were detected in high numbers in pasture only in January and February.

In the initial phase white grubs from Risaralda and Northern Cauca were taken to our laboratory Quindío at CIAT for adults' identification. However, we observed that the larvae development was delayed or the pupae were malformed. We hypothesized that these abnormalities were due to the fact that they were removed from their original habitat. Hence, we established an on-farm laboratory in Caldono (Cauca) where we can study their life cycle and feeding behavior. In this lab we are observing *Phyllophaga menetriesi*, *Plectris fassli, Plectris pavida, Anomala undulata, Anomala sp., Cyclocephala* pos. *lunulata* and *C. amazonia*. The farmer Manuel Trujillo and his son Huber are running this lab in participatory research manner. Both have been collaborating working many years with CIAT and have received a training in the work with white grubs.

Quindío: In the light trap on the Hacienda Córcega in Quimbaya (Quindío) we found the following white grub complex: *Phyllophaga menetriesi, Phyllophaga* sp.*, Anomala cincta, A. undulata* and other species whose the economic importance is unknown. On the big cassava, coffee or maize plots *P. menetriesi* was mainly recorded. According to the administrator, it occurs primarily in December and March, causing up to 100% damage in cassava and maize. Employees reported that they found up to 20 larvae per m^2 during this period. In January and February we collected 8 and 7 larvae per m^2, respectively, in a maize field intercropped with cassava.

Risaralda: In La Florida we collected 1,934 adult scarabids between October 2002 and June 2003. The subfamily Rutelinae harbored 65% of the captured beetles, followed by Dynastinae with 26%. *Anomala cincta* and *A. undulata* were the most abundant species (**Figure 5).**

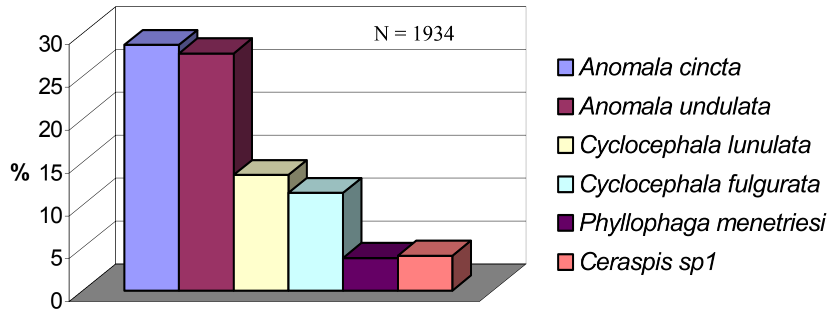


Figure 4. Scarabid adults caught in light traps in La Florida (Risaralda) from September 2002 to August 2003.

In La Colonia we collected in total 1,962 adults. *Cyclocephala fulgurata* and *C. lunulata* were the dominant species, followed by *Anomala cincta* and *A. undulata* (**Figure 6**).

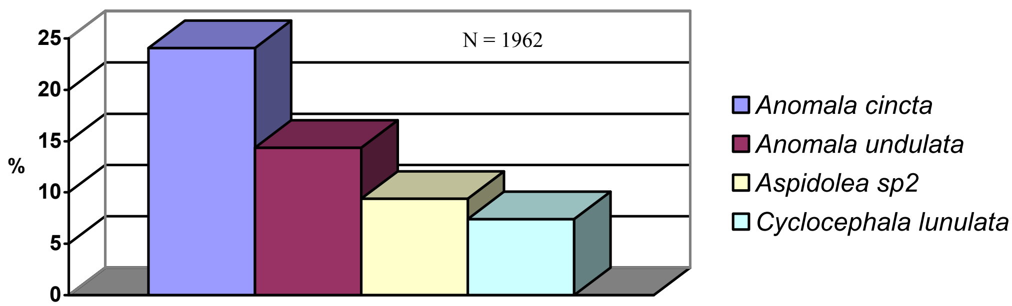


Figure 5. Scarabid adults caught in light traps in La Colonia (Risaralda) from September 2002 to August 2003.

During the surveys in onion and pasture fields we collected in total 1,949 white grub larvae. 1,255 larvae (64,4%) were collected in pasture and 694 (35.6%) in onion. The average density of white grub larvae were 10.4 and 5.7 larvae per m^2 in pasture and onion, respectively.

The white grub complex consisted of 11 species belonging to the subfamilies Melolonthinae, Dynastinae and Rutelinae. **Figure 7** shows that Dynastinae sp. 1 dominated in pasture, followed by un identified species of Macrodactylini, Melolonthinae and *Phyllophaga*. The most abundant species in onion was Dynastinae sp. 1 (284), followed by Rutelinae sp. 1. In onions we found a similar distribution, however, Dynastinae sp.1 was more dominant **(Figure 8).**

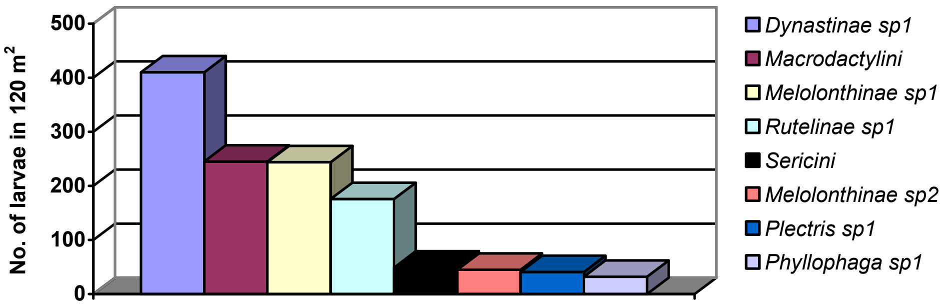


Figure 6. **No. of scarabid larvae collected in pasture in La Florida from September 2002 to August 2003.**

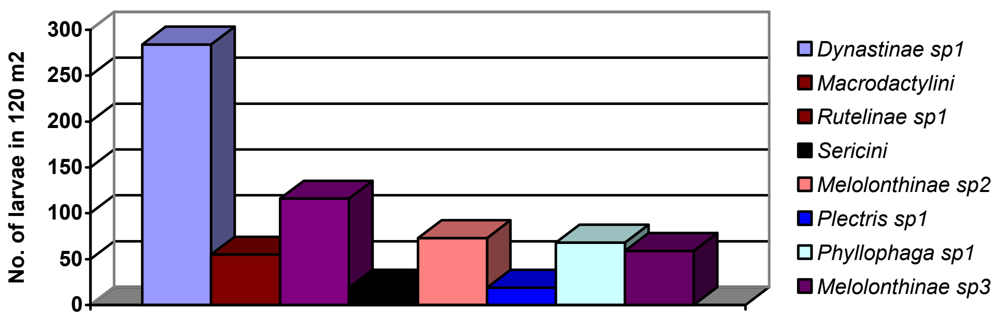


Figure 7. **No. of scarabid larvae collected in onions in La Florida from September 2002 to August 2003.**

In the Pereira region the most frequently collected adult scarabs in pasture were *C. lunalata* (69%). The other species were*: Anomala cincta* (20%), *A. undulata* (11%), *P. menetriesi* and *C. fulgurata* (2%). In the light trap in cassava we again found *Cyclocephala lunalata* (51%) to be the most abundant species. Other species were *A. undulata* (32%), *Anomala* sp. 1 (8%), *A. cincta* (5%). *Phyllophaga menetriesi* and *C. fulgurata* were present in the same portion as in the pasture samples (2%).

In cassava plots near Pereira we identified 1,858 larvae of eight white grub species. An unidentified species of the tribe Rutelinae was present in greatest abundance (42.4%), followed by *Cyclocephala* (18.8%) and *Plectris* sp. 2. In pasture we collected 2,834 larvae of 10 species, where *Cyclocephala* dominated (38.3%), followed by Macrodactylini (23,1%). Larvae associated with cassava showed three peaks: The first in December (Ø 12 grubs per m^2), the second in March (Ø 100 grubs per m^2) and in May (Ø 80 grubs per m^2). Highest numbers of larvae in pasture were recorded in November (Ø 27 grubs per m^2) and April (Ø 34 grubs per m^2).

Antioquia: Adult scarabs and larvae were collected in pasture and in potato fields. We installed six light traps in the municipalities of Rionegro (2,100 m asl), El Carmen de Viboral (2,258 m asl), San Vicente (2,300 m asl), La Union (2,460 m asl), Santa Rosa (2,486 m asl), Entrerrios (2,437 m asl) and San Andres de Cuerquia (1,211 m asl). Until June we revised about 80,000 specimens and separated them into 28 species.

In the savanna of Northern Antioquia (2,600 – 2,800 m asl) we have identified adults of scarab 15 species: Ancognatha scarabaeoides, A. humeralis, Cyclocephala sexpunctata, C. gregaria, Heterogomphus dilaticollis, H. schoenerri, H. chevrolati, H. rubripennis, Megaceras pos. morpheus, Astaena sp., Golofa eacus, Anomala sp., Plectris sp. and one unidentified species. Ancognatha scarabaeoides was the most abundant species (almost 37,000 specimens), followed by C. sexpunctata (18,000). In La Union, a village in the savanna of East Antioquia (2,400 – 2,600 m asl) we identified 11 species: A. scarabaeoides, A. humeralis, C. sexpunctata, C. gregaria, H. dilaticolis, H. schoenerri, A cincta, Plectris sp., Astaena sp., Isonichus sp. and Golofa sp. As in the North of Antioquia, A. scarabaeoides was the predominant species (10,000 captured adults); Astaena sp. was collected less frequently. The savanna of Northern Antioquia corresponds to a moderate cold climate zone (2,100 – 2,300 m asl). Here we collected the following 20 species: A. scarabaeoides, A. humeralis, C. sexpunctata, C. gregaria, C. fulgurata, Cyclocephala sp., Phyllophaga obsoleta, Astaena sp., H. dilaticollis, H. chevrolati, G. eacus, Isonychus sp., Plectris sp., Anomala cincta, A. pos. undulata, Anomala sp., Macrodactylus sp., Anatista lafertei, Callistetus cupricollis and one unidentified species. Again, A. scarabaeoides (5,400 specimens) dominated in abundance, followed by C. sexpunctata and P. obsoleta with 1,300 specimens captured each and Isonychus sp. with 1,000 adults trapped.

Up to now we have revised 500 m^2in potato fields. We found tuber damage in 250 m^2 of survey, but not necessarily caused by insects. We checked a total of 19,946 tubers, corresponding to 40 tubers per m^2. Of these, 8.6% showed symptoms of insect attack and 0.1% due to other reasons. White grubs caused 5.3% of the damage, followed by Guatemalan potato moth (Tecia solanivora) (3,2%), and black cutworm (Agrotis ipsilon) (0.1%). White grubs were present at an average density of 7.6 larvae per m^2. We additionally sampled 440 m^2 in Kikuyo (Pennisetum clandestinum) and found white grubs and cutworms in 50.7% and 1.6% of the area, respectively.

So far we were unable to identify larvae to species level, underlining the need for development of suitable keys for their identification.

Cundinamarca: 1,038 larvae of Ancognatha sp. and Heterogomphus dilaticollis (Dynastinae) and Clavipalpus pos. ursinus (Melolonthinae) were collected between September 2002 and May 2003 at the Centro Agropecuario Marengo (CAM, Cundinamarca). During the first four months we observed very low population densities (average of 5 larvae per m^2). In February 2003, populations increased to an average of 21.6 larvae per m^2 and maintained this density during the following two months. Variation was very high; in some cases we collected up to 168 larvae in m^2. Larvae of Ancognatha sp. and H. dilaticollis were associated with organic material and/or plant parts in decomposition, suggesting that this white grub complex does not feed on living plants or may prefer organic material to living plants.

In Cogua (3,200 m asl, Cundinamarca) we sampled from October 2002 until August 2002 in total 350 larvae of Ancognatha sp. and Heterogomphus dilaticollis (both subfamily Dynastinae). With 8.7 larvae per m^2 the greatest density was recorded in December 2002. Thereafter densities decreased to a minimum of 2.2 larvae / m^2 in July 2003. The average larval density was lower in Cogua than at CAM (8.7 larvae / m^2), indicating a preference for pasture as habitat.

In June 2003 we found in a fallow plot larvae of Clavipalpus pos. ursinus associated with decomposing plant material, suggesting that this species has a facultative saprophagous behavior as strategy for survival when principal host plants are rare.

In the light trap in pasture at CAM we collected in total 7,008 adult scarabids. Ancognatha scarabaeoides was by far the predominant species (98%); other specimens were A. ustulata and Heterogomphus dilaticollis (subfamily Dynastinae) and Manopus bigutatus (subf. Melolonthinae). The majority of the beetles was trapped in January and March, and lowest numbers were recorded in February, March and May. In Cogua we only could make three surveys. We detected five species of three genera of Dynastinae: Ancognatha scarabaeoides, A. ustulata, H. dilaticollis, Astaena pos. tarsalis, and A. vulgaris. 90% of 910 collected adults corresponded to A. scarabaeoides, corroborating the results at CAM where A. scarabaeoides strongly dominated the scarab diversity. Interestingly, larvae of Clavipalpus pos. ursinus, Manopus bigutatus, Astaena sp. and Lacioccala sp. were never found in Cogua or at CAM despite their presence as adults in the light traps.

The white grub complex in Cundinamarca shows a much more reduced species biodiversity than in other similar agro-ecological zones. In Cauca or in the North of Antioquia up to 45 species have so far been identified.

Field-collected larvae were transferred to the lab for establishing colonies. In doing so we obtained important data on biology and behavior of the larvae. For instance we can prove that Ancognatha scarabaeoides, A. ustulata and Heterogomphus dilaticollis are not obligatory pest species since all larvae of the three species developed from egg to adult on a pure diet of organic material, corroborating our field observations. When feeding larvae of these three species with carrots, they always clustered on spots where the food was already in the process of decomposition. In contrast, Clavipalpus pos ursinus always focused on fresh parts of carrots and moving to the soil surface when the food presented symptoms of decomposition.

These observations are important indicators for the potential pest status of white grubs in the Savannah of Bogotá and corroborate results from the earlier study by Ruiz and Posada (1986) who claimed that “the mixture out of soil, decomposed wood and dung were the only substrate where they complete their life cycle”.

We morphologically described larvae of Ancognatha scarabaeoides, A. ustulata, Clavipalpus pos. ursinus, H. dilaticollis and Ceraspis sp.

Discussion of survey results from the three agro-ecological zones: In general, our data show that the biodiversity of the white grubs complex is greater in warmer (S/Quilichao) and reduced colder agro-ecological zones (Andean zones of Antioquia and Cundinamarca). Another important finding is that the complex of noxious white grubs is limited. *P. menetriesi* is the most important pest species in Quindío and Risaralda. Its economical importance is well documented (*e.g.* King 1984). In Antioquia in zones over 2000 m asl the pest complex is restricted to *P. obsoleta* (Vallejo *et al*. 1998, Vallejo *et al*. 2000). In Antioquia and in Cundinamarca *Ancognatha scarabaeoides* is the most abundant white grub (up to 98%). However, our observations that this species feeds mostly on decomposing organic material seriously questions its pest status though more data on the biology and ecology of *A. scarabaeoides* is needed. However, there is a need for a more detailed study of the feeding behavior of this insect.

III. Characterization of potential biological control agents.

Search for native entomopathogenic organisms in Cauca, Risaralda and Panama: We collected 320 soil samples in Panama (31), and in Colombia in Quindío (61), Risaralda (135), Caldas (40) and Cauca (53) and isolated entomopathogenic nematodes by means of the *Galleria* bait method **(Table 1)**.

Table 1. Sites where soil samples were collected for isolation of entomopathogenic nematodes.

Country	Department	Municipality	Date	Crop
Panamá	El Valle de Antón	Coclé	Oct-02	onion, peanuts
	Ocú	Veraguas	Oct-03	cassava
	Sioguí	Chiriquí	Oct-03	cassava
	Cerro Punta	Estac. IDIAP	Oct-03	onion
Colombia	Quindío	Quimbaya	Mar-03	cassava, plantain, maize
	Risaralda	Santa Rosa	Mar-03	onion
		Pereira	Mar-03	pasture, cassava, onion
			May-03	onion
			Feb-03	pasture, onion, cassava, peas
		Dosquebradas	Mar-03	pasture
		La Florida	Jun-03	onion
	Caldas	Manizales	Jun-03	guamo, breadfruit tree, maize, beans, cassava, avocado, plantain, mango, mandarin, lemon, passion fruit, coffee
	Cauca	S/Quilichao	Mar-03	maize, pasture, cassava

The three nematode strains collected in Panama were shown to be saprophytic. In Colombia we isolated 16 strains from samples taken in various crops, 10 of them showed lethal effects on *Galleria* in sand. Thereafter we verified these observations, following Koch's postulates. Pathogenic strains will be sent to Kiel University (Germany) for identification as soon as we will obtain the exportation permit from the Colombian authorities.

In S/Quilichao no entomopathogenic EPN or bacteria were collected but the following fungi were isolated from 850 white grubs: 3 x *Metarhizium anisopliae*, 7 x *Fusarium* sp. and 3 x *Paecilomyces* sp. The unidentified fungi will be sent to Germany for identification. *M. anisopliae* and *Paecilomyces* sp. were isolated from white grubs found in cassava, *Fusarium* sp. in pasture.

In preliminary experiments we tested *M. anisopliae*, a strain of *Paecilomyces*, and *Beauveria bassiana* against larvae (third instar) of *P. menetriesi* with 20 replicates for each strain. The white grub control was 100%. It is of interest for future work to repeat these experiments at greenhouse or field level. Moreover, other white grubs should be included in these tests.

Risaralda: In fields near Pereira and in La Florida 500 white grubs per locality were collected and brought to the lab for further studies of potential pathogens. Later, these pathogens were identified by experts of Cenicafé. 50% of the collected larvae in Pereira showed symptoms of a bacterial infection **(Table 2)**. This rate seems to be extraordinarily high since the larvae were collected on several plots. Hence, it can be excluded that all the 250 white grubs were from a spot where bacteria were highly concentrated. Moreover, we have never observed such a high extent of bacterial infections in larvae sampled in the other surveys. Hence it is possible that the larvae collected in Pereira were contaminated in the lab.

Table 2. Frequency of various entomopathogens associated with white grubs in Pereira collected from pasture and cassava.

Entomopathogens	No. of Observations
Bacteria	
Bacillus popilliae	250
Fungi	
Metarhizium spp.	9
Beauveria bassiana	2
Trichoderma	50
Fusarium	20
Nematodes	
Mermithida sp.	2 nematodes in one grub

In La Florida we found 30 larvae attacked by entomopathogens. Eight were infected by *Metarhizium* spp., 10 by *Trichoderma* spp. and five by *Fusarium* spp. and none by bacteria.

Antioquia: We sampled in potato and pasture fields for white grubs (10 m^2 per plot) and took the larvae to the laboratory in order to identify them and their natural enemies. We observed a high mortality of the larvae due to infection by pathogens. Mortality was higher in East Antioquia than in the North. The rate of mortality is summarized in **Table 3**. After evaluation in the lab, microbial organisms turned out to be main cause for white grub mortality. The most frequent pathogens we found were the bacteria *Bacillus popilliae* and the fungus *Metarhizium anisopliae* **(Table 4)**.

Table 3. Mortality due to pathogens of collected white grub larvae sampled in three agroecological zones in Antioquia from October 2002 to June 2003.

Region	No. of grubs alive	No. of dead grubs	%
North 2600-2800 m asl	548	432	78
East 2600-2800 m asl	624	566	90
East 2100-2400 m asl	648	613	94

We collected a total of 502 white grubs from the pasture Kikuyo, in three ecological zones in Antioquia. 93.7% of the grubs taken from the cold North Antioquia (2800 m asl) died in the lab, 57.4% from the cold East (2600 m asl) and 86.7% from the West, the latter characterized by a moderate cold climate. Frequency of mortality factors were similar to the ones in potatoes, however, with an increased frequency of *Bacillus popilliae* and hymenopteran and dipteran parasitoids.

Table 4. Mortality factors of white grubs (D = Dynastinae and M = Melolonthinae) collected in three regions in Antioquia from potato fields and pasture from October 2002 to June 2003.

Mortality factors	North (2600-2800 m)		East (2600-2800 m)		East (2100-2400 m)	
	D	M	D	M	D	M
Bacillus cereus	4	0	9		2	2
Bacillus larvae	4	0	1		0	0
B. lenthimorbus	3	0	1		0	0
B. popilliae	70	1	72		30	9
B. sphaericus	10	0	6		7	0
Clostridium sp.	9	0	7		6	2
Pseudomonas sp.	0	0	1		0	0
Ma-1	9	0	241		54	1
Ma-2	0	0	2		54	0
Ma-3	0	0	0		1	1
Ma-4	3	0	0		0	0
Cladosporium sp.	0	0	0		1	0
Paecilomyces sp.	0	0	2		1	
Fusarium sp.	0	0	0		3	
Synnematium sp.	0	0	0		0	
Ectoparasite	0	0	0		2	
Endoparasite	0	0	0		2	
Protozoans	0	0	1		0	
Nematodes	2	0	0		37	
In process	2	0	0		1	
No visible reason	96	6	65		57	
Other reasons	188	16	132		174	
Total	400	23	540		432	

Cundinamarca: Approximately 100 white grub larvae showing disease symptoms were brought to the lab for further identification of the pathogens. 22% of these larvae were collected in Cogua, 32% at CAM and 46% in Subachoque. Entomopathogenic fungi were recorded most frequently (43%), followed by bacteria (40%), nematodes (21%) and protozoa (2%). The higher numbers of diseased larvae found in Subachoque and at CAM might be related to the fact that the potato fields in Subachoque had passed through a two years fallow period and at CAT samples were taken in pasture. Such conditions are possibly more suitable for both white grubs and entomopathogens than the pesticide-treated crops at Cogua.

60% of the 100 diseased larvae belonged to the genus *Ancognatha*, 33% were *Clavipalpus* pos. *ursinus* and 6% were *Heterogomphus dilaticollis*. Although the rate of natural infection was highest in *Ancognatha* spp., the mortality rate in *C. ursinus* was considerably higher compared to the other infected white grubs, possibly explaining why this white grub species is much less frequent in Cundinamarca than in other zones.

So far we have isolated 35 fungal strains that we store in a ceparium. To date the following species/strains have been identified in collaboration with our partners from BBA Darmstadt (Germany): *Metarhizium anisopliae* (4 isolates), *Fusarium* spp. (3), *Beauveria bassiana* and *Verticillium* spp. (2 isolates each) and *Aschersonia* spp. (1).

We additionally isolated 29 bacterial strains. In collaboration with Corpoica, Rionegro, we identified 15 isolates as *Bacillus popilliae*, three as *B. sphaericus*, two as *Clostridium* sp. 1, one as *B. larvae* and one as *B. cereus*. Bacterial-infected white grubs were soft and of a white, brown, or black color. The majority of these grubs had a strong odour, probably due to fermentation caused by the bacteria.

In total 16 white grubs were attacked by, presumably entomopathogenic, nematodes and we managed to rear four strains on *Galleria mellonella*. Presently these samples are in the process of purification and identification since some of them were associated with saprophytic organisms (mites, bacteria or other nematodes). White grubs attacked by nematodes were in general transparent and of slightly reddish color. The nematodes clustered close to the spiracles or the legs. When the grubs were stored in maturation chambers, in most of the cases the nematodes were unable to pass through the cuticle. Often grubs turned black, possibly due to proliferation by other saprophytic microorganisms. In these cases it was almost impossible to recover infective nematode juveniles. We observed that the mouth of the grub cadavers is the main exit for the nematodes. We managed to identify two nematode populations belonging to the genera *Mesorhabditis* and *Steinernema*.

Discussion of recollection of entomopathogenic organisms in three agroecological zones: The poorest collection of pathogens was in S/Quilichao, the most numerous in Antioquia. The poor presence of pathogens in S/Quilichao may be explained by the dry climate. The difference in pathogen collections may be due to factors other than agroecological conditions. In Cundinamarca the survey was conducted by only one student, while Corpoica could carry out a more extensive survey. Pathogen collections in Risaralda fell between the two extremes. Therefore, it is suggested to continue the survey of soil pests affected by pathogens including the registration of climatic factors and soil components (*e.g.* organic material).

Another important task results from the fact that many collected isolates still have to be identified. Here, we can count on the collaboration with German specialists.

Tritrophic interactions between burrower bug, host plants and entomopathogenic nematodes (*Heterorhabditis megidis*): The entomopathogenic nematode *Heterorhabditis megidis* is attracted by cues from maize plants, but not by those of its insect host, the burrower bug *Cyrtomenus bergi*. These are the conclusions of a research project carried out by partners at Hannover University. The objectives of the study were:

1. To investigate the searching behavior of entomopathogenic nematodes (EPN) associated with *C. bergi.*
2. To study possible plant- and/or host-related cues that guide the EPN.

In preliminary experiments we recorded moderate levels of EPN-induced mortality (on average 32%) in adult *C. bergi*, confirming that *H. megidis* can successfully parasitize and kill burrower bugs. In our experiments we only used adult bugs and selected *H. megidis* due to its cruiser behavior. Maize was used as the host plant. Experiments were conducted in a sand-filled olfactometer, similar to the one used by Boff *et al*. (2001) **(Figure 8**).

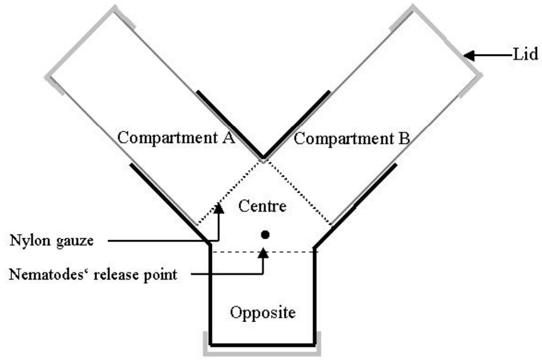


Figure 8. Sand-filled olfactometer (after Boff *et al.* 2001).

We conducted six experiments:
1. *C. bergi* vs. sand
2. Maize plants vs. sand
3. Maize plants + *C. bergi* vs. sand
4. *C. bergi* vs. maize plants
5. Maize plants + *C. bergi* vs. maize plants
6. Maize plants + *C. bergi* vs. *C. bergi*

The nematodes were significantly attracted by the maize seedlings but not by *C. bergi* adults. Interestingly the behavior of the EPN did not differ when bug-infested and non-infested plants were offered simultaneously. The EPN reacted indifferent when exposed to *C. bergi* or sand. Possible reasons for these findings are: (i) as a root-sucking insect, *C. bergi* does not produce enough cues, compared to root-chewing insects, for attraction of EPNs, and (ii) *H. megidis*, an EPN species from the Northern hemisphere, is not well adapted to *C. bergi* as a host. In ongoing experiments we are investigating the response pattern of EPN species/strains native to Colombia.

Evaluation of three native entomopathogenic nematodes against *Phyllophaga menetriesi* larvae: Third instar of larvae of *P. menetriesi* are hard for EPNs to kill. The insect's thick integument and layers of hair are probably efficient defense mechanisms against an EPN attack. Additionally, it seems that *P. menetriesi* larvae posses an efficient, yet to be identified immune response that kills EPNs after penetration. These are the main results of evaluating the virulence of three native Colombian EPN strains. The tested strains were: *Heterorhabditis* sp HNI 0100 (Cenicafé), *Heterorhabditis* sp. (CIAT) and *Steinernema* sp SNI 0198 (Cenicafé), applied at concentrations of 7,000 and 13,000 infective juveniles per ml. Penetration rate was observed after 5 and 10 days. We used *P. menetriesi* as a test species because it is one of the most harmful white grub species in Colombia. White grub larvae were collected in fields in Cauca and Risaralda. The strains from Cenicafé showed slightly higher penetration rates than the CIAT strain. Concentrations did not affect the penetration rate of all tested EPN strains. Curiously we found less EPN in the grub after 10 days than after five. Mortality was higher after 10 days in the case of HNI 0100 at both concentrations and *Heterorhabditis* (CIAT) at high concentration.

In ongoing studies we are evaluating more EPN species/strains. Moreover, younger instars of white grubs will be tested, and for this we are in the process of establishing lab colonies of several white grub species.

Why are some white grubs easier to control than others?

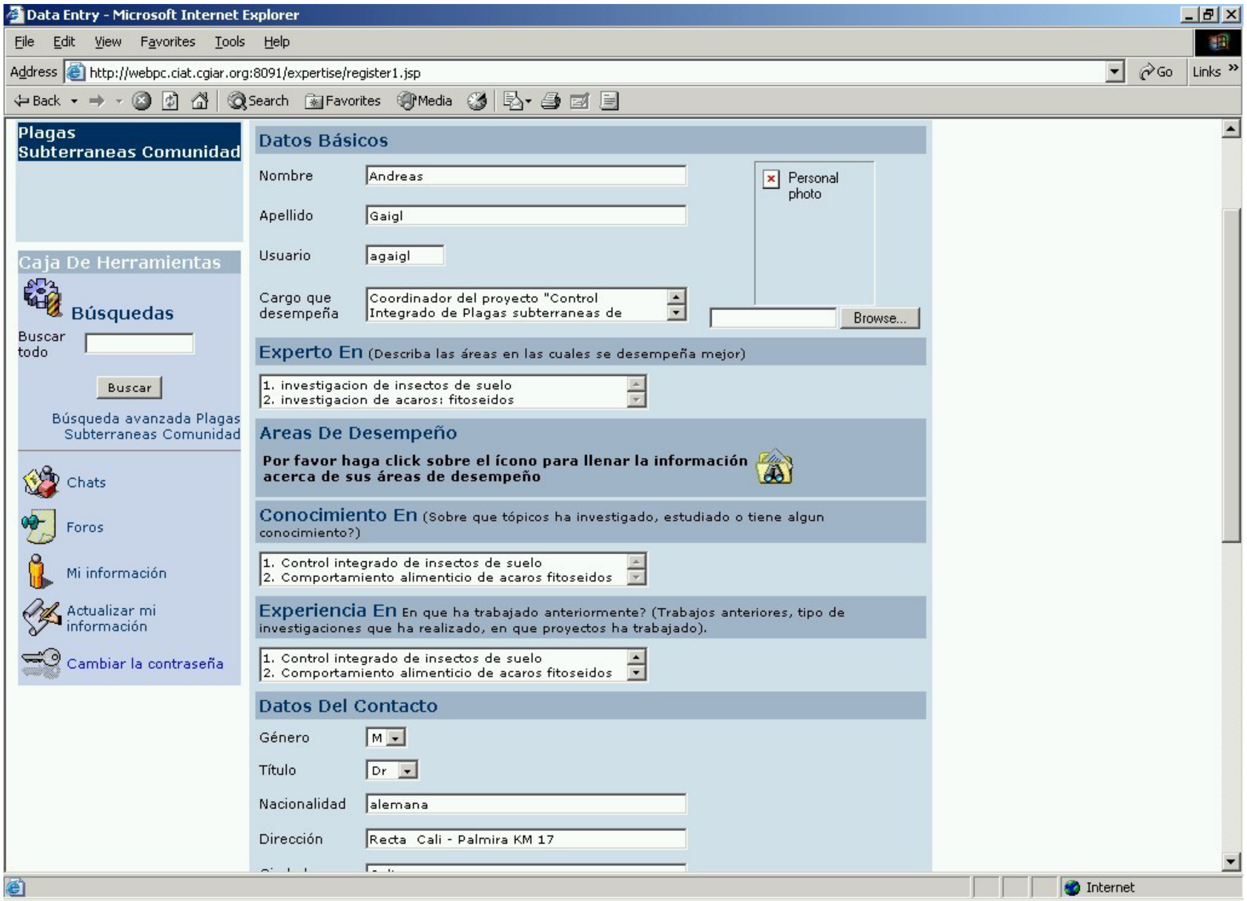
Field surveys suggested that the most susceptible targets for entomopathogens are white grubs from the subfamily Dynastinae. In contrast, Melolonthidae were less infected. The reasons for this possibly the fact that we collected much more Dynastinae than Melolonthidae larvae, thereby increasing the chance to find diseased Dynastinae. However, the are several additional factors that might be responsible for an overall reduced susceptibility of Melolonthidae to entomopathogens:

1. Physical barrier: The integument of *P. menetriesi* (Melolonthidae) is thicker than that of *Cyclocephala* (Dynastinae).
2. Resistance: We assume that *P. menetriesi* can activate an immune response that kills nematodes that have penetrated into the host.
3. Evade antagonists: We have observed during our field surveys that *Phyllophaga* spp. dwell deeper in the soil than *Cyclocephala* spp. or other Dynastinae or Rutelinae.

IV. Establishing a network of soil arthropod researchers in Latin America.

So far research on subterranean pests in Colombia as well in South America have not been properly coordinated. For this reason CIAT developed an Internet-based platform, which is easy to access, and where researchers can exchange information on soil pests, share their experience, and coordinate their activities (http://webpc.ciat.cgiar.org:8091/expertise/index.jsp).

We presented this database at the SOCOLEN congress this year. Our Internet community to date counts 35 members and we want to extend the group. For this reason the network will be presented on the Round Table of Soil Pests in Mexico in October 2003. Presently the database contains a directory of all members. However, during the next weeks we will include more data on literature and general information on subterranean pests.



Outlooks

- We will continue with the surveys in Northern Cauca and Antioquia. We decided to switch from Risaralda to Quindío where we have identified farmers who have repeatedly experienced out breaks of soil pests on their farms. They are willing to support our work by collaborating in the surveys. Moreover, next, we will conduct several participatory on-farm experiments in Quindío.
- Establishing of white grub colonies for experiments with entomopathogens.
- Testing of four fungal strains against *Cyrtomenus bergi* in the laboratory and greenhouse.
- Field experiments with commercially available entomopathogenic fungi for control of *C. bergi* (asparagus, Caldas).
- Testing intercropping practices with repellent plants against soil pests (Montelindo, Universidad de Caldas).
- Evaluation of commercial EPN products against *C. bergi* in the greenhouse.
- Evaluation of several native and exotic EPN species/strains against various white grub species in the laboratory.
- Evaluation of the economic damage (i.e. yield loss trials) of white grubs in participatory on-farm experiments (Quindío).

Theses completed or near completion

César A. Zuluaga (U. Nacional/Bogotá):	Recognition of white grubs, *Cyrtomenus bergi* and their natural enemies (Cundinamarca). Completed.
Maria Paulina Quintero (Univalle):	Pathogenicity of entomopathogenic nematodes in *Phyllophaga menetriesi.* Completed by the end of 2003.
Lina María Serna: (U. de Caldas):	Recognition of white grubs, *Cyrtomenus bergi* and their natural enemies in Pereira. Completed by the end of 2003.
Nelly Villegas (U. de Caldas):	Recognition of white grubs, *Cyrtomenus bergi* and their natural enemies in La Florida, Risaralda. Completed by the end of this year. Completed by the end of 2003
Lisa Struck (Hannover University):	Tritrophic interactions between burrower bug, host plants and entomopathogenic nematodes. Completed.

Thesis underway

Germán Andrés Calverto (Universidad Autonoma):	Establishment of rearing methods of whitegrubs.
Juliana Jaramillo (M.Sc., Hanover):	Pathogenicity of entomopathogenic fungi against *Cyrtomenus bergi* and white grubs in semi-controlled experiments.
Ana M. Caicedo (M.Sc., Hanover):	Pathogenicity of entomopathogenic nematodes against *Cyrtomenus bergi* in semi-controlled experiments (initiated in October 2003).
Anuar Morales (M.Sc., Cornell Univ.)	Use of pheromones for the study of soil pests.

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