Short Research Article

Olfactory attraction of rhizome borer (Coleoptera: Curculionidae) to banana genotypes inoculated with entomopathogenic fungus

.

ABSTRACT

The species: *Cosmopolites sordidus* (Germar) (Coleoptera: Curculionidae) stands out as one of the main pests of banana. The damage to the crop is caused by the larvae of this pest, when they feed on the plant tissues. The aim of this study was to investigate the olfactory responses of *C. sordidus* for different varieties and the possible olfactory interference after application of *Beauveria bassiana* fungus on the crop. The research was conducted at the Phytosanitary Clinic in the Agriculture Sector of the Center for Humanities, Social and Agrarian Sciences of the Federal University of Paraíba, located in Bananeiras - PB, Brazil, from January to July 2017. The experimental design was completely randomized, with four treatments represented by banana varieties (Apple, Silver, Nanica and Pacovan) with 11 replications each. The bioassays were distributed in two stages; the first stage was an evaluation of the attractiveness of banana genotypes and rhizome and pseudostem tissues. In the second, the attractiveness of C. sordidus to the tissues contaminated with the fungus *B. bassiana* was investigated. The Prata banana plantation was less attractive to *C. sordidus*, the highest preference was to theNanica banana plantation. The most attractive tissue was pseudostem. Given the conditions under which the study was conducted, the banana plantation and the plant tissues analyzed present an olfactory influence under *Cosmopolites sordidus*. The Nanica banana plantation is the most susceptible to insect attack. The application of the fungus *Beauveria bassiana* on banana baits does not interfere with the odour conditions exerted the nanica banana plantation under *C. sordidus*.

Keywords: Plant resistance, biological control, Olfactometer, Beauveria bassiana

1. INTRODUCTION

The species beetle: *Cosmopolites sordidus* (Germar) (Coleoptera: Curculionidae) stands out as one of the main banana pests, occurring in practically every region of the planet [1]. Injuries to the crop are caused by the larvae of this pest, which when feeding on plant tissues build galleries causing the interruption of the sap and the rotting of the plant and thus leading to decreased productivity.[2].

The use of systemic insecticides is considered one of the main forms of control for this pest [3]. However, one of the major problems encountered in chemical control is the translocation of the active ingredient of these insecticides via sap to the fruits, which are mostly marketed "in natura" [4]. In this context another form of control which has been widely used is the use of attractive baits, produced with pieces of pseudostem split in half. These baits are based on the attraction exerted by the volatile substances present in the banana pseudostem and rhizome. [5].

Use of attractive baits may be enhanced after spraying with entomopathogenic fungus: *Beauveria bassiana* (Bals.) Vuill, this way the fungus acts against insects which served as parasite spreading agents to other parts of the banana plantations. The fungus *B. bassiana* is one of the most effective fungi and studied in biological control. [6]. This fungus as well as other entomopathogenic fungi penetrate the host via the integument, causing the death of insects due to mycotoxin production, and due to vegetative growth promoting mechanical blockage of the digestive tract and other physical damage due to mycelial growth. [7]Several studies have been performed to detect *C. sordidus* olfactory preference to different banana varieties. [8,9,10,11]. However, studies are needed to provide information on C. sordidus olfactory preference for entomopathogenic fungi. Since such agents may cause biological interference in the treated material. This hypothesis was also studied by Magalhães et al., [12] Where, evaluating the interactions between entomopathogens, parasitoids and predators, he observed that bioinsecticides may influence the choice of parasitoids by changes in host characteristics such as color, shape, texture or odor.

Thus, the objective of this study was to investigate the *C. sordidus* olfactory responses for different varieties and the possible olfactory interference after *B. bassiana* fungus application in the culture.

2. MATERIALS AND METHODS

The research was carried out at the Phytosanitary Clinic in the Agriculture Sector of the Center of Human, Social and Agrarian Sciences of the Federal University of Paraíba, located in Bananeiras, Paraiba, Brazil. At the same site, *C. sordidus* specimens were obtained. The capture took place by means of tile baits, made from pieces of banana pseudostems. The insects were kept in the laboratory in plastic containers, measuring approximately 10 cm high by 80 cm wide, with 1 cm radius perforations on the sides, containing as a food source and shelter fresh pseudostem pieces, changed every five days The four banana genotypes analyzed were: Apple, Silver, Nanica and Pacovan (these are kinds of banana), from which two parts of the plant (pseudostem and rhizome) were used. Later the most preferred variety was inoculated with the fungus: *B. bassiana* for possible verification of the preference after inoculation by the banana tree borer. The strain of the entomopathogenic fungus *B. bassiana* was isolated from a mummified specimen of *C. sordidus*, found in the Rural community of Roma, Bananeiras district. The fungus was produced in Petri dishes in BDA culture medium using the methodology described by Alves. [7].

For evaluation of *C. sordidus* olfactory response to banana genotypes, the rhizome and pseudostem of each genotype were reduced in dimensions of approximately 2 cm, arranged in a multiple arena similar to that described by Botelho *et al.* [13].Which features a central arena and four side arenas as options to choose from. The bioassays were performed during the night, a time associated with greater activity of the banana tree. [14].

The experimental design used was a completely randomized design. The bioassays were composed of four treatments represented by banana varieties (Apple, Silver, Nanica and Pacovan) containing 11 repetitions and distributed in two stages; where the first one evaluated the attractiveness to the banana genotypes and the rhizome and pseudostem tissues. In this evaluation the tissues and each vegetable were grouped in isolation on the olfactometer, where two arenas were filled with fresh tissue of one genotype and the other two by cotton wicks soaked with distilled water as a control. Subsequently, four *C. sordidus* adults not sexed were placed in the central arena and remained for 40 minutes exposed to the volatiles released by the tissues of the analyzed plants.

In the second stage to evaluate the attractiveness of *C. sordidus* to tissues contaminated with the fungus: *B. bassina,* the arenas were filled with fresh tissue (pseudostem) of the Nanica banana plantation, this time using only, detected in the first stage of the bioassays as the genotype and the tissue with the highest preference for the insect. For the contamination of banana tissues, the fungus was diluted in water and the tissues dipped in this solution for conidia adherence in one minute. Then a 10-minute interval was waited for subsequent attractiveness check by the insect.

The parameters evaluated were preference, non-preference and individuals with no response to odors. For this, the insects found in three conditions were quantified: on fresh rhizome and pseudostem tissues, found on the control and those that remained in the release arena. Insects not in these conditions were disregarded.

The results for banana olfactory response to volatile banana genotypes were analyzed using the non-parametric X2 test (chi-square).

3. RESULTS

The results for *C. sordidus* olfactory preference (Table 1) showed a high attractive effect of the insect to the pseudostem plant tissue, with only values of olfactory preference below the expected frequency.

Apple cultivar has low olfactory influence on pseudostem tissue, with values of 36.36%, below the expected frequency (40%). For cultivar Prata, there was a high influence on C. Sordidus, when they had an olfactory preference of 81.81%. The indices observed for the cultivar Nanica showed a preference of 81.81%. The results show that the insects had a higher preference for pseudococcus also for cultivar Pacovan, with values of 45.45%.

Table 1. Attractiveness of *C. sordidus* **for pseudostem tissue in banana varieties**

Pseudotem tissue					
Observed Frequency (Expected) (%)					
Varieties	Preference	No preference	No answer	X²	Value de (P)
Apple	36,36 (40)	18,18 (40)	45,45 (20)	44,6	-0,001
Silver	81,81 (40)	9,09 (40)	9,09 (20)	73,53	-0,001
Nanica	81,81 (40)	9,09 (40)	9,09 (20)	73,53	-0,001
Pacovan	45,45 (40)	9,09 (40)	45,45 (20)	57,01	-0,001

C. sordidus olfactory preference (Table 2), for rhizome plant tissue, presents low olfactory preference values, below the expected frequency for the cultivars Apple, Prata and Pacovan, only the cultivar Nanica presents values above the expected frequency of 40%.

Table 2. Attractiveness of *C. sordidus* **for rhizome tissue in banana varieties**

Rhizome tissue

Observed Frequency (Expected) (%)					
Varieties	Preference	No preference	No answer	X²	Value de (P)
Apple	18,18 (40)	9,09 (40)	72,72 (20)	174,8	-0,001
Silver	9,09 (40)	18,18 (40)	72,72 (20)	174,8	-0,001
Nanica	54,54 (40)	0,09 (40)	36,36 (20)	58,48	-0,001
Pacovan	36,36 (40)	0,09 (40)	54,54 (20)	99,8	-0,001

The apple variety rhizome tissue was not attractive to the insect, and the expected frequency (40%) was higher than the observed frequency (18.18%). Similarly, the cultivar Prata presented values below the expected frequency (9.09%). Olfactory preference for cultivar Nanica was 54.54%, the expected frequency for tissue. In this tissue, the cultivar Pacovan presented 36.36%, obtaining values below the expected frequency.

As it was observed in Table 5, *C. sordidus* showed preference by pseudostem tissues when they were inoculated by the fungus B. bassiana.

Table 3. Attractiveness of C. sordidus in **tissues of inoculated Nanica Banana plantation and without application of the fungus** Beauveria bassiana.

Observed Frequency (Expected) (%)				
Pseudostem with application of *B. bassiana*	X²	Pseudostem without application	X²	Value of (P)
60,0 (50)	2,0	40,0 (50)	2,0	(0,001)

4. DISCUSSION

Among the various managements used for the control of agricultural pests, varietal resistance has been researched and pointed as a viable alternative in the control of *C. sordidus* [15, 16]. We evidenced in this study the olfactory influence exerted by different plant tissues and genotypes, analyzed on *C. sordidus*. The attractiveness of Insect by the plant is produced due to the secondary volatile compounds released by the plant as semiochemicals present in the rhizome and pseudostem [17, 8].

Facundo *et al.* [18] states that the various banana genotypes have different volatile compounds that vary according to variety. According to Oliveira *et al.,* [11], the volatiles found in banana genotypes, whether varieties or hybrids, have the same attractiveness for *C. sordidus*. However, there is variation in the composition and concentration of volatiles that may vary among banana genotypes, reflecting the responses of *C. sordidus* adults to each genetic material [18, 19].

We corroborate in this study the high attractiveness of the volatile compounds of Nanica Banana plantation for both plant tissues analyzed. The Nanica Banana Plantation is considered one of the most susceptible to attack by this insect, with losses of up to 80% of production, these losses are also affected in the Prata banana plantation, with a smaller proportion around 30% [20].

Infestations of this pest in banana plants can be reduced by planting less attractive plantations, which resulted in lower insects demand for the plant and consequently less oviposition and emergence of new individuals [1]. Another efficient type of control widely used by producers is the use of entomopathogenic fungi traps. The use of these fungi for the biological control of banana broth as with other agricultural pests is a viable alternative that reduces environmental impacts caused by the use of chemicals such as insecticides [20]. The use of alternative pest controls also reduces the use of chemicals, reducing the amount of toxic waste to humans in banana consumption and at the time of application [21].

5. CONCLUSION

Given the conditions under which the study was conducted, it can be concluded that the banana varieties and plant tissues analyzed have an olfactory influence under *Cosmopolites sordidus*. The Nanica banana variaty is the most susceptible to Insect attack or presenting high olfactory preference for pseudostem and rhizome. The application of the fungus *Beauveria bassiana* on banana baits does not interfere with the odour conditions exerted the nanica banana plantation under *C. sordidus*.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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