Facultad de Ciencias Naturales y Exactas
Universidad del Valle

COMPOSTING TO CONTROL THE LEAF-CUTTING ANT *ATTA CEPHALOTES* L. (HYMENOPTERA: FORMICIDAE)

Inge Armbrecht
Universidad del Valle

Mavir Montoya-Correa
Universidad del Valle

María Cristina Gallego-Ropero
Universidad del Cauca

James Montoya-Lerma
Universidad del Valle

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Resumen

Las hormigas cortadoras de hojas, *Atta* y *Acromyrmex*, son los herbívoros más importantes en los trópicos. En este estudio se comparan los efectos de dos métodos no químicos sobre la sobrevivencia y actividad de nidos de *Atta cephalotes* (Formicidae:Myrmicinae). Un total de 83 nidos activos de *A. cephalotes* fueron asignados de forma aleatoria a uno de tres tratamientos: 1) incorporación de un material compostable al suelo; 2) remoción y mezcla de la capa superficial del suelo (método convencional) y 3) placebo o control (sin tratamiento). Después de tres meses, los porcentajes de nidos eliminados con cada uno de los tratamientos fueron 26,5; 9,6; y 3,6; respectivamente. Un análisis de varianza de medidas repetidas mostró una diferencia significativa entre las colonias de hormigas sometidas a los tratamientos compostado y remoción mecánica en relación con el placebo. Esto debido a la destrucción de las pistas de forrajeo, de las aberturas, cámaras y túneles. Sin embargo, el más fuerte efecto fue observado con el compostado. Basado en estos resultados sobre los nidos de *A. cephalotes* y su efecto potencial de enriquecer el suelo, el tratamiento compostado puede representar una práctica sustentable y ecológicamente amigable en el control de hormigueros de *Atta.*

Palabras claves: hormiga plaga, compostado, orgánico, control hormigas.

Abstract

Leaf-cutting ants, *Atta* and *Acromyrmex*, are the most important defoliating herbivores in the tropics. In this study, field assays were conducted to compare the effects of two non-chemical methods on survival and activity of *Atta cephalotes* (Formicidae: Myrmicinae) nests. A total of 83 *A. cephalotes* active nests were randomly assigned to one of three treatments: 1) compostable material introduced and processed into the soil of ant nests; 2) conventional mechanical mixing of the ant nests soil; 3) untreated ant nests. After three months the percentages of dead nets were 26.5, 9.63 and 3.61 using manure compost, mechanical, and control, respectively. A repeated measurement analysis of variance showed significant difference between ant colonies for both treatments, composted and mechanical mixing, with respect to the untreated treatment, given the disruption of foraging trails, mound openings, chambers and tunnels. The strongest effect was observed with the compostable material. Based on its effect on *A. cephalotes* (Formicidae: Myrmicinae) nests and potential soil enrichment, compostable treatment could be a sustainable and ecologically sound management practice if proper conditions are given at the site.

Keywords: ant pests, compost, organic method, ant control.

1 Introduction

In the Neotropics, *Atta* and *Acromyrmex* (Myrmicinae), comprising 24 and 15 species, are widely distributed leaf-cutting ant species [25]. Leaf-cutting ants build their nests in the soil and are fungi growers that do not directly consume plants but cut leaf sections to prepare a substrate upon which they culture the symbiotic *Leucoagaricus gongylophorus* fungus [24; 19]. Although ecologically important as functional herbivores, their highest impact is due to their elevated ecological and behavioural adaptability, traits that make them serious pests to agricultural and silvicultural systems in several Latin American countries [6; 9; 26]. Small landholders perceive leaf-cutting ants as one of the most difficult and defiant pests, affecting the profitability of many of their cash crops [10]. Many publications address their management and control [3; 11].

Methods for controlling leaf-cutting ants are mechanical destruction of nests through physical (drenching or firing up the nests), cultural (deep plough, crop traps, and use of resistant or tolerant varieties), chemical insecticides (powders, fogging, baits), or biological control (predators, fungal antagonists and entomopathogens) [6]. Synthetic insecticides are the most commonly used control method, but they have adverse effects for human and environmental health. This undesirable situation urges generating alternative, environmentally friendly control methods [1]. Promising plant extracts are currently being tested [22], albeit it must be recognised that their use is highly expensive and time consuming.

In Colombia, *Atta cephalotes* is one of the most widely disseminated leaf-cutting ant species [15; 8]. It is frequently found in forests, agriculture, and urban areas. Its presence in agricultural and forest areas is usually associated to losses in crops like cassava (*Manihot esculenta*), maize (*Zea mays*), cotton (*Gossypium* spp.), cacao (*Theobroma cacao*), and forest nurseries. However, reports are circumstantial and not well documented, usually supplying no quantitative data. Indeed, people assume its pest status as a fact. Furthermore, although even less well documented, several machinery and livestock accidents occurred due to their enormous subterranean colonies [23]. In urban areas, leaf-cutting ants are responsible (or at least perceived so) for attacks on ornamental trees and damage or deterioration of buildings and social premises [17].

Eight years ago, a novel approach to controlling leaf-cutting ants was devised by O. Mora [18], by exploiting their biology and applying several ecological practices using a type of compost. He named it *"arrierón"* (after *arriera*, a common name given to leaf-cutting ants in Colombia). The core of this technique is the use of organic waste material (forest litter and poultry manure) combined with lime and molasses and yeasts aimed at controlling all types of leaf-cutting ants while, at the same time, improving soil fertility and enhancing physical properties of the soil. In 2006, Chaves [5] carried out a pilot study with 14 nests to test Mora's technique [5; 18]. Although this author found mortality and reduction of ant nests, the low replication of the study prevented drawing solid conclusions. Consequently, this study sought to evaluate the efficiency of the organic compost (henceforth composted product) on reducing the proliferation and activity of *A. cephalotes* colonies in field conditions. Our final goal was to validate the composted product as an alternative method to control leaf-cutting ants.

2 Materials and methods

2.1 Study area

The study was carried out in 100 ha in the main campus of Universidad del Valle, located in Meléndez, Cali, Department of Valle del Cauca (3°24'56" N; 76°30'10" W). The life zone has been considered as Tropical Dry Forest [7]. Annual rainfall fluctuates between 1.100 and 1.500 mm, displaying a bimodal pattern (April-May and October-November) [14]. Remnant isolated trees, representing the flora of natural Tropical Dry Forest [2] and ornamental (usually introduced) plants surround the university campus.

The fresh and shaded environmental setting is suitable for the establishment and dissemination of animal fauna, among them *A. cephalotes*, the only leaf-cutting ant species present and also the most conspicuous herbivore in the area. Some of the buildings have been selected by the ants as nesting places.

2.2 Ant nests

A census of all active *A. cephalotes* nests was carried out on the campus in February 2006. The ant nests had not been previously treated with insecticides. To test the viability of each nest, a person jumped on the entrance and looked for the appearance of soldiers. Each living nest was labelled and ant nest attributes (size and number of surface openings, number and width of ant trails) were assessed at once (March 2006) before the application of the treatments and, then monthly for three consecutive months. For each nest, the most distant mound openings were located and the nest area was estimated (in square meters). To assess ant activity, the number of foraging ants on a random but active trail was counted by using a hand counter. Observations were made during a period of one minute, and the average of three counts was recorded. Width of trails was evaluated at distance of 50 cm from each mound opening with a detectable ant trail. Finally, the total number of mound openings was counted. Observations and measurements were made on sunny days, the first measurement one week before treatments were established.

2.3 Treatments

Three groups of nests were tested. (1) The first treatment was the "compost", which represented a compostable mixture of organic (leaf litter, poultry manure, molasses, and yeasts) with inorganic (lime and water) matter placed on the top of the nests. The materials were applied in approximately the following amounts (and following their relative proportions): 5-kg pack of leaf litter, 15 kg of poultry manure, 5 kg of agricultural lime, 1 kg of molasses (which can be replaced by sugar), 1 kg of yeast, and 25 litres of water. These materials were employed for every 25 m^2 of nest area at soil surface, concentrating activity at the nest's central conglomerate. Molasses and yeast were previously dissolved in the water. To apply the materials, the soil, chambers, and fungus were mechanically shovelled by using a 1.5-m long sharpened shovel. Then, the materials described were evenly spread over the nest and immediately covered with a black plastic sheet for the following 30 days after which the plastic was removed. (2) The second treatment, or "mechanical mixing", consisted of physically digging up the whole nest and mixing it with the soil (at a depth of

30-50 cm or until ant fungus chambers were detected) [16]. No special effort was placed on finding and eliminating the queen in any of the treatments. (3) A third group of nests was designated as placebo or "control" or untreated nests without any intervention. The response variables described above (nest area, number and area of openings, number of trails and trail width, and number of soldiers coming out per minute after disturbance) were measured four times: at the beginning and then one, two, and three months after the experiment was established.

2.4 Experimental design and data analysis

A randomized block design was used with the treatments described above: (1) compost-treated nests, (2) mechanically shovelled nests, and (3) the untreated nests. Groups of three neighbouring nests were taken as blocks and treatments assigned randomly to the individual nests. The total result was: 30 nests assigned to receive the compost treatment; 30 as mechanically shovelled nests, and 23 as untreated nests.

The experiment was conducted under rain-fed conditions. The number of dead nests (with no signs of ants or activity) was counted 30, 60 and 90 days later. Treatment effects over the four time periods were evaluated by repeated measurement analysis of variance (ANOVA) by using the average nest area of surviving colonies and the average areas of all nest openings. Data from the four months were pooled and then transformed into logn+1 to comply with the assumption of normality. Variances were checked for homogeneity by using Levene tests. All analyses followed the procedures of Zar [27] and were performed using the SPSS statistical program, version 14.0 for Windows [21]. Chi square analyses using Past [12] were performed to test whether the proportion of surviving nests per treatment was different along the four time counts over the four time periods.

3 Results and discussion

A total of 83 nests were selected on the basis of their activity and affected an important nest area, totalling 1857 m^2. Nest size area ranged from 0.85 m^2 to 190.58 m^2 and the number of mound openings fluctuated between 4 and 114. Twenty-two, eight, and three nests (corresponding to 26.5, 9.63 and 3.61 percent) belonging, respectively, to the composted-treated nests, mechanically shovelled, and untreated nests were found dead on the last evaluation in June 2006. Both, the compost and mechanical treatments induced high stress in all colonies given the destruction of foraging trails, mounds, openings, chambers, and tunnels. Overall, the response variables decreased sharply over time for both compost and mechanically treated nests while they tended to stay relatively constant in the control nests.

Given that a smaller number of nests was evaluated in the control, the proportion of surviving nests throughout the study time was pooled out, and similar results were obtained. Surviving nests within composted-treated nests come from a population not having the same ratio as the untouched control ($\chi^2 = 32.79$, df = 3, $P < 0.00001$) and the mechanical mixing ($\chi^2 = 26.57$, df = 3, $P < 0.00001$) (Figure 1). These results show that relatively less ant nests survived in the composted treatment with respect to the other treatments. However, the ratio between mechanical mixing and the control ($\chi^2 = 1.25$, df = 3, $P = 0.7$) was not statistically different.

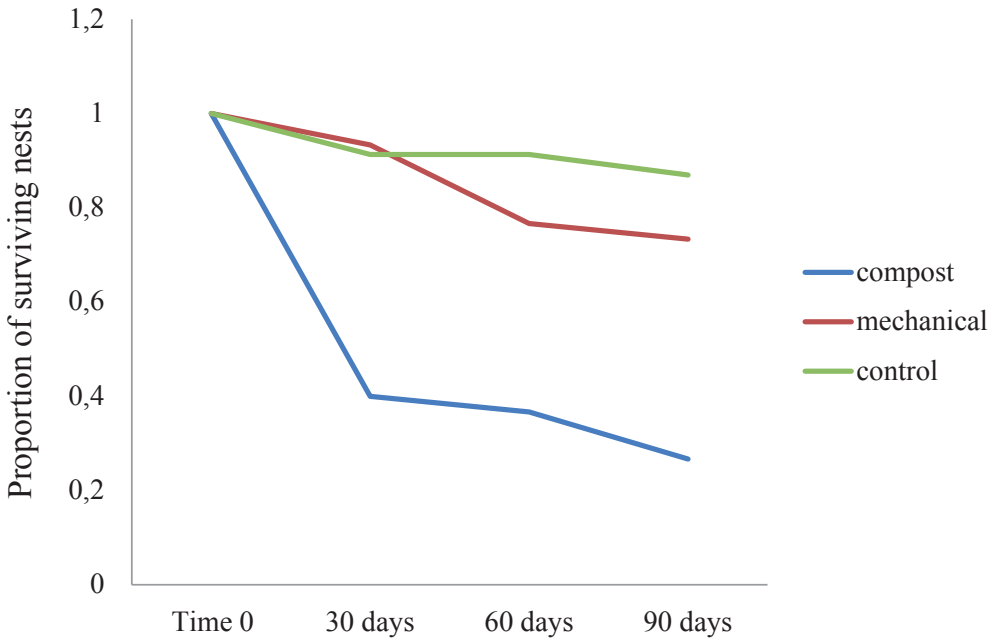

Figure 1. Proportion of Atta cephalotes nests surviving after setting on three treatments in Cali, Colombia in 2006. Treatments consisted of: 1)"Compost" (blue line) in which a compostable material was introduced and processed into the soil of ant nests; 2)"mechanical" conventional mechanical mixing of the ant nests soil; 3) "Control" or untreated ant nests. Time 0, at the setting of the experiment and 30, 60, and 90 days after.

The negative effect of the compost on ant nests was probably due to the massive microbial and gaseous disruption of the colony and its symbiotic fungi. It is, therefore possible to speculate that the substantial reduction in ant nest size and activity observed during the first month corresponds to the high ammonia emissions of the first stage of composting [20]. These events were sufficient for the destruction of small nests up to areas of 25 m^2 or about two or more years old [13]. However, our study did not test for these possible explanations.

During the final stage, the compost cools, ending in the production of mature compost, which may permit the gradual recovery of those ant nests that survived, especially large ones. In a laboratory experimental assay, inhibition of fungal growth was documented with brown rot fungus due to competition for resources from the high microbial diversity in active compost [4]. Thereby, a study should be conducted to determine whether or not a correlation exists between ammonia emissions, increased temperature, presence of yeasts, and reduction of fungus size and ant population.

This study also found that the average decrease in all response variables was due to the death of several colonies (measurements with zero values) and not necessarily to a gradually diminishing response of the ant colonies. Although the medians showed that there were statistically significant reduction effects of the compost treatment over all the response variables measured (Table 1), the areas of those ant nests that survived the treatments (which were just about one fourth for the compost treatment) actually increased (Figure 2). For instance, the initial average area of those nests that eventually died was 13 m^2, 15 m^2, and 16 m^2 for compost, mechanically shovelled, and untreated

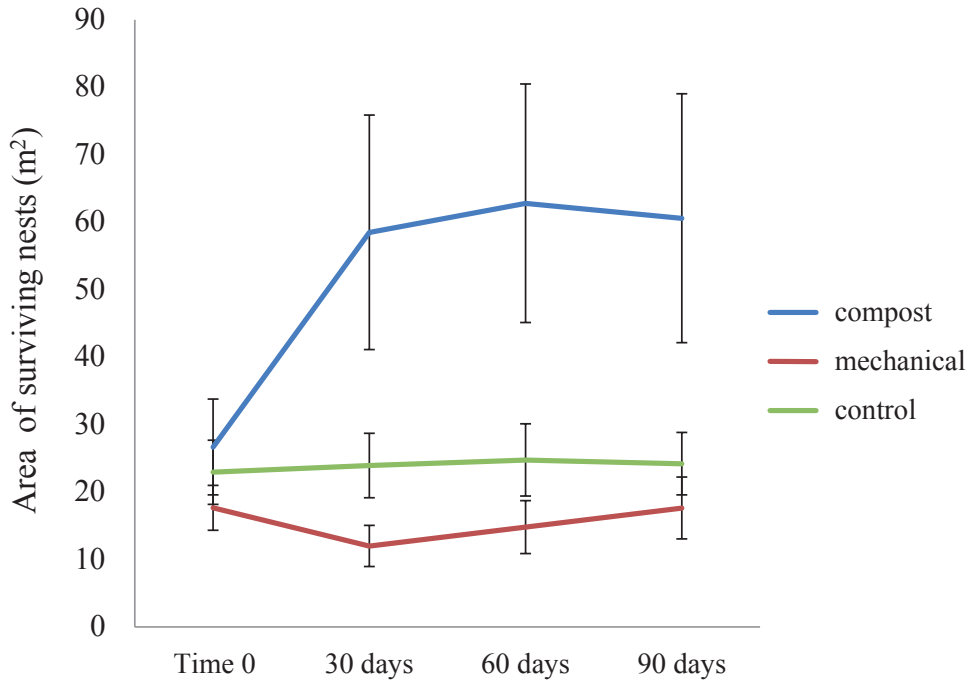

Figure 2. Nest area of Atta cephalotes nests surviving the treatments.Standard error bars are shown for each mean. The number of nests (n =30 for month 0 except for the "control" in which n = 23) varies from 8 to 28 in further months. See Figure 1 for a short description of the treatments.

Table 1. *Medians* of five response variables measured from A. cephalotes nests under three treatments (compost, mechanical mixing of the ant nest soil, and control nests without any disturbance) obtained in June 2006 after four months of the experiment setting at Universidad del Valle, Meléndez Campus.*

	Treatment			Kruskal	
Variable	**Compost**	**Mechanical mixing**	**Control**	**Wallis, χ^2; df=2**	***P***
Nest area (m^2)	0.0	3.2	13.6	11.6	0.003
Number of trails/nest	0.0	1.0	1.0	14.3	0.001
Sum of trail width/nest (cm)	0.0	1.5	3.0	13.3	0.001
Number of openings/nest	0.0	11.0	16.5	13.6	0.001
Sum area of the openings/nest	0.0	39.0	65.8	11.0	0.004
Number of soldiers/min after disturbance	0.0	0.0	6.0	7.6	0.022

*median:" defined as the numerical value separating the higher half of a sample".

nests, respectively (ranging between 1.4 and 44 m^2). On the contrary, the average area of the nests surviving after three months of treatment was significantly larger for the compost (63 m^2; n = 8) than in the mechanically shovelled (19 m^2; n = 22) or untreated treatments (24 m^2; n = 20) (repeated measurements ANOVA, $F_{2, 47} = 9.56$, $P < 0.0001$) (the range of nests that survived was between 4.3 and 190.6 m^2). These surviving nests appeared larger

because new holes were dug outside the borders of the black plastic sheet covering the nest surface, presumably as a response of the ants to chemical and physical stress. Our results are consistent with Chaves [5], where 33% of her compost-treated nests increased the area up to 200%. Again, in this study, nests grew faster in the compost-treated because the new openings made by the ants were all located at the boundaries of the transparent plastic cover of her study (outside the "treatment" zone) so that, if a nest survives, it "appears" to be larger just because of this spatial distribution of the new openings. In our study area, we noticed that the composted nests were not active by 2011 (pers. obs.), although no data is available to support that this is due to the composting.

Overall, this study suggests that ecologically sound methods, like mechanical removal and composting, are viable methods to reduce populations of leaf-cutting ants. Further, it is necessary to emphasize the rationale of this agro-ecological practice, which, besides achieving control of the leaf-cutting ants, seeks to manage this pest while providing benefits to the soil (increased fertility). . On the other hand, a broad range of methods to control leaf-cutting ants has been tested. Usually, these methods are costly and many are harmful and have toxicological and environmental side effects. In consequence, current environmental protection laws restrict the sale and use of several of these products. Therefore, improving the composted method is an alternative option for leaf-cutting ant control, given the relatively easy application, easy availability of the ingredients, and the high effectiveness against small to medium size nests. Hence, this composted method fulfils the multiple criteria to be an alternative control.

4 Conclusions

The results of this study show that the composting treatment caused a rapid decrease of ant nests, up to 73% of the *A. cephalotes* nests found in the study area; hence, it is concluded that the compost treatment can be used as an alternative method to control *A. cephalotes* nests.

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Author's address

Inge Armbrecht
Department of Biology, Universidad del Valle, Cali - Colombia
inge.armbrecht@correounivalle.edu.co.

Mavir Montoya-Correa
Department of Biology, Universidad del Valle, Cali - Colombia
mavir29@hotmail.com

María Cristina Gallego-Ropero
Department of Biology, Universidad del Cauca, Popayán - Colombia
macrigaro@yahoo.es

James Montoya-Lerma
Department of Biology, Universidad del Valle, Cali - Colombia
james.montoya@correounivalle.edu.co