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Entomofauna involved in pollination and biocontrol of pests in cashew (*Anacardium occidentale* L.) orchards in the Marahoué region (Bouaflé, Côte d'Ivoire)

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Abstract

The aim of the present study is to evaluate the association between the vegetative and reproductive structure of cashew trees and the habitat of beneficial insects, in order to improve the agronomic performance of orchards and income of cashew producers. Consequently, observations were made in three types of orchards of different ages (0-5 years, 5-10 years and over 10 years), each occupying an area of 1 hectare. For each age group and in each orchard, an arrangement of 10 consecutive cashew trees chosen at random in the direction of the north-east diagonal was set up. In all the orchards sampled, over 60.68% of the expected species diversity was observed. A total of 184 pollinating bees were collected, with the Apidae family the most diverse, and marked by high activity from *Apis mellifera* and *Meliponula bocandei*. As for ants, 124 individuals were collected, with the Formicinae family the most diversified, marked by high activity of the species *Oecophylla longinoda*. Foraging activity of bees and ants was significantly higher in orchards over 10 years old than in orchards 0-5 years and 5-10 years old. To maximize the productivity of all orchards, it is essential to implement compensatory actions such as the installation of beehives and the introduction of plants favored by beneficial ants.

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Introduction

Cashew (*Anacardium occidentale* L.), a plant native to northeastern Brazil (Trevisan *et al.*, 2006), was planted in northern Côte d'Ivoire in the early 1960s because of its rapid growth and hardiness in combating deforestation, bush fires and soil erosion (Soro *et al.*, 2011; Bassett *et al.*, 2018). For this agriculturally-oriented country, cashew has become a fruit cash crop whose production has only increased to the point where cashew has become the second most exported crop after cocoa (Bassett *et al.*, 2018). Today, this crop is the main source of cash income for over 5,000,000 people, including 500,000 smallholders in 20 of the 31 regions (Djaha *et al.*, 2012).

However, numerous studies have revealed low yields in Ivorian orchards and very low quality of raw cashew nuts (Stéphane *et al.*, 2020; Stéphane *et al.*, 2021; Yepié *et al.*, 2023).

Indeed, cashew nut cultivation in Côte d'Ivoire has developed autonomously, without the support of the state or the assistance of agricultural research and extension structures (A.F.D, 2010; Bassett, 2017). This autonomous development has led to: the use of unimproved nuts, production techniques that are still traditional, the use of pesticides whose consequences on the environment and useful fauna have yet to be assessed, and above all the persistence of phytosanitary problems, particularly insect pest attacks (Djaha *et al.*, 2014). However, Côte d'Ivoire is the world's leading producer, ahead of India and Vietnam, respectively the world's second and third largest producers, whose orchard productivity and nut quality are even better (AFP, 2016; FIRCA, 2018). Thus, the leading position occupied by Côte d'Ivoire justifies intensive land use for the establishment of orchards in order to fill the gaps created by palliative measures that are not being taken (FIRCA, 2018). Furthermore, the activities following the establishment of cashew orchards can have the potential to reduce the quantity and diversity of many organisms, especially during maintenance work requiring the use of pesticides (Cissé *et al.*, 2021; Yao *et al.*, 2020). In view of these facts, scientific investigations are needed to assess the impact of cashew orchards on the survival of useful fauna, particularly organisms involved in pollination and biocontrol of pests, in order to help growers, choose technical itineraries capable of conserving this heritage and the health of the ecosystems that support them. It is with this in mind that the present study has set itself the general objective of assessing the population of beneficial insects associated with the vegetative and reproductive structure of cashew trees. The specific aim is to determine: (i) the population of insects involved in pollination; (ii) the population of insects involved in bio-control of pests. The hypothesis is therefore that the floral preference of bees and the biocontrol of pests by ants make these organisms excellent bioindicators of the agronomic performance of cashew seedlings. The results of the present study can contribute to the implementation of beekeeping in orchards and help to better manage ant populations for more efficient management and improved cashew nut production in Côte d'Ivoire.

Materials and methods

Study site

The study was carried out in the Marahoué region, west-central Côte d'Ivoire, one of the pioneering cashew-growing regions. It is located 60 km from Yamoussoukro (political capital) and 310 km from Abidjan (economic capital). This area straddles the forest and savannah zones, with cashew nut production ranging from 10,000 to 20,000 tons/year. In addition, the village of Attossé (Fig. 1) was chosen as the host site for the study because it represents one of the areas in the region with a large area under cashew cultivation but characterized by low productivity per tree.

Choice of plots and sampling device

The present study is based on the selection of 3 types of orchards in different age brackets (0-5 years, 5-10 years and over 10 years), each covering 1 hectare. For each age group and in each orchard, 10 cashew trees were randomly selected in a northeast diagonal direction. Selections were made consecutively without skipping trees until all 10 trees were reached (Fig. 2).

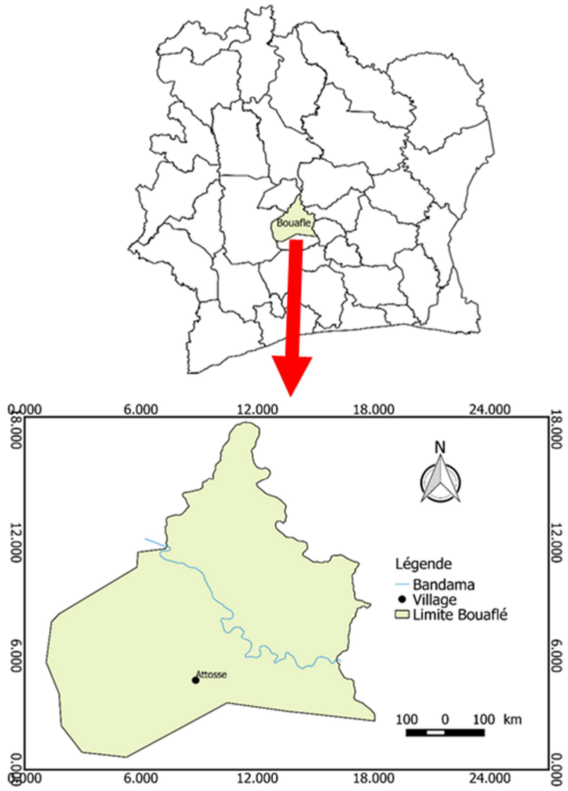

Fig. 1. Study site

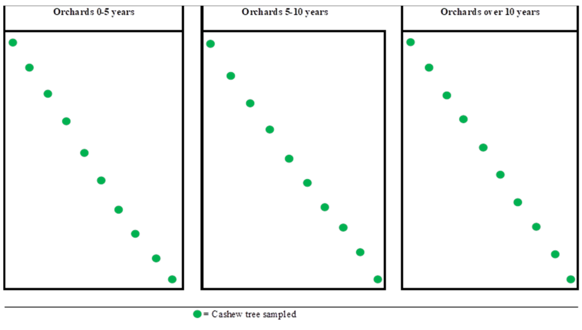

Fig. 2. Sampling device

Capture of insects associated with pollination (bees)

The mowing net, which is an active trap, was used to catch the bees visible on the flowers during the day. These captures were made at the following times: 9 a.m., 12 p.m., and 3 p.m. corresponding to the times when bee activity becomes maximum according to Abou-sharra *et al.* (2014). The bees were captured for a period of 5 minutes, on each of the four main branches of each tree in the device, and kept in a bottle containing 70° alcohol. Each tree produces 4 samples per sampling hour, and 12 samples per day. Once all the specimens had been counted, they were grouped together and identified morphologically to species level, using determination keys (Eardley, 2004; Eardley *et al.*, 2010) under an Olympus SZ61 binocular loupe. However, the analysis of bee visit frequencies was based on the activity of two species, *Apis mellifera* and *Meliponula bocandei*, which can be identified as they move about. They are also the main pollinators of cashew trees (Silué *et al.*, 2022).

Capture of insects involved in biocontrol of pests (ants)

Ants were sampled at each sampling point that means on the main branch per tree, in accordance with the method of Peng *et al.* (2010). The technique consisted in gently shaking the branches with a 1-meter stick, in order to induce the ants to move. Each tree produces 1 sample per day. Supplies captured with entomological forceps were first preserved in labeled vials containing 70° alcohol and then sent to the laboratory for identification. Genera were determined using the key of Bolton (1994) and Fisher and Bolton (2016). Species were determined, where possible, using the Bolton identification key (1976, 1980, 1982 and 1987) and a reference collection deposited at the Lamto ecological station (Yeo, 2006).

Data analysis

Observed richness (Sobs) was obtained by counting insect species after identification.

EstimateS software version 9.1.0 was used to obtain the estimated species richness (Chao 2), Simpson's diversity, and its Equitability. Then, a Pearson correlation was established between the parameters studied (insect activity, parasite pressure and agronomic performance) using software (PAST) version 3.0.9 at a significance level of 0.05. Prior to this, all data were analyzed using Levene's test for homogeneity of variance (data distribution test) before multiple comparisons were made between orchards. In the case of a normal distribution, Tukey's paired test or one-way analysis of variance (ANOVA) on the repeated measurement was required for multiple comparison. Otherwise, the non-parametric Kruskal-Wallis multivariate analysis of variance or Mann-Whitney U test was used for comparison. In addition, all the parameters measured in the orchards were compared with those of control cashew trees selected by (Silué *et al.*, 2022) and recognized as high-yielding cashew trees.

Results

Insect populations involved in pollination (bees)

Bee sampling efficiency

In all the orchards sampled, more than 60.68% of the expected specific diversity was observed. However, sampling efficiency was significantly higher in orchards over 10 years (78.98±3.06%), followed by orchards 5-10 years (42.33±6.33%) and orchards 0-5 years (19.53±3.02%) (Mann Whitney, *p = 0.0036*). This observed coverage rate indicates the high efficiency of the sampling method used (Table 1).

Diversity of bee populations

A total of 184±7.01 pollinator bees were collected. According to orchard age, bee abundance was significantly higher in orchards of more than 10 years (129±5.58 individuals), followed by those of 5-10 and 0-5 years corresponding to 16±1.03 and 39±4.25 individuals respectively (Mann Whitney, *p = 0.0012*). This sampled pollinating bee population was made up of 2 major families (Apidae and Halictidae) subdivided into 10 species and 8 genera. The Apidae family was more diverse (7 species) than the Halictidae family (3 species) (Table 2).

Table 1. Coverage rate for bee sampling

	Orchards 0-5 years	Orchards 5-10 years	Orchards over 10 years	Total	p-value
Coverage rate (%)	$19.53±3.02^c$	$42.33±6.33^b$	$78.98±3.06^a$	60.68±2.23	0.0036

Values with different letters on the same line are significantly different (p < 0.05) using the Mann Whitney test.

Table 2. Collected bee species and corresponding diversity parameters

Families	Species	Orchards 0-5 years	Orchards 5-10 years	Orchards over 10 years	Total	p-value
Apidae	*Apis mellifera*	16±1.88	36±2.04	91±5.63	143±8.32	-
	Dactylurina staudingeri	0±0.00	0±000	4±0.43	4±0.74	-
	Hypotrigona sp.1	0±0.00	0±0.00	2±0.06	2±0.15	-
	Meliponula beccarii	0±0.00	1±0.43	7±0.51	8±0.56	-
	Meliponula bocandei	0±0.00	1±0.22	19±2.01	20±1.09	-
	Xylocopa albiceps	0±0.00	1±0.51	1±0.02	2±0.01	-
	Xylocopa olivacea	0±0.00	0±0.00	1±0.04	1±0.02	-
Halictidae	*Acunomia* sp.1	0±0.00	0±0.00	2±0.36	2±0.13	-
	Crocisaspidia chandleri	0±0.00	0±0.00	1±0.05	1±0.01	-
	Lasioglossum sp.1	0±0.00	0±0.00	1±0.06	1±0.05	-
Abundance (individuals)		$16±1.03^c$	$39±4.25^b$	$129±5.58^a$	184±7.01	0.0012
Observed species richness (Sobs)		1^a	4^b	10^c	10	0.02
Estimated specific richness (Chao2)		5.12	9.45	12.66	16.48	-
Simpson's diversity indices		-	0.21^b	0.39^a	0.48	0.041
Equitability indices		-	0.7^a	0.61^a	0.55	0.68

Values with different letters on the same line are significantly different (p < 0.05) using the Mann Whitney test.

Table 3. Coverage rates for ant sampling

	Orchards 0-5 years	Orchards 5-10 years	Orchards over 10 years	Total	p-value
Coverage rate (%)	$54.34±4.20^a$	$55.55±3.09^a$	$50.71±3.50^a$	50±2.93	0.056

Values with different letters on the same line are significantly different (p < 0.05) using the Mann Whitney test.

Species richness was significantly higher in orchards over 10 years (10 species) than in orchards 5-10 years (4 species) and 0-5 years (1 species) (Mann Whitney, p = 0.02).

Simpson's diversity indices were significantly higher in the more than over 10 years orchard (0.48) than in the 5-10 years (0.39) and 0-5 years (0.21) orchards (Mann Whitney, *p = 0.041*). Conversely, equitability evolved in the opposite direction, with the highest index in orchards 5-10 years (0.7), followed by those over 10 years (0.61) (Table 2).

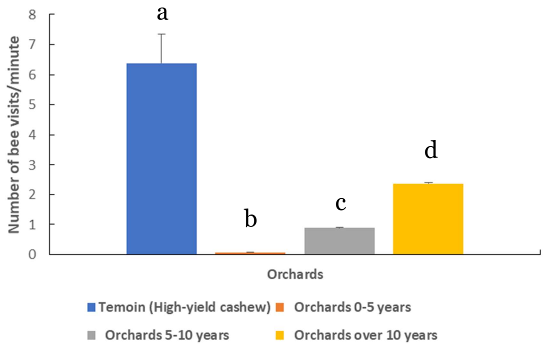

Fig. 3. Frequency of bee visits

Values with different letters on the same line are significantly different (p<0.05) using the Mann Whitney test.

Frequency of bee visits

Fig. 3 shows the frequency of bee visits to the various orchards. Bee foraging activity was significantly higher in the orchard over 10 years old (2.36±0.04 visits/minutes), followed by orchards 5-10 years old (0.89±0.01 visits/minutes) and orchards 0-5 years old (0.08±0.01 visits/minutes) (Mann Whitney, *p = 0.0081*). However, the foraging activity of bees from the three 3 orchards studied was significantly lower than that of control cashew trees (6.38±0.96 visits/minutes).

Insect populations involved in pest bio-control (ants)

Ant sampling efficiency

In all three types of orchards sampled, more than 50±2.93% of the expected specific diversity was observed. Sampling efficiency was significantly higher in orchards 5-10 years (55.55±3.09%) than in orchards 0-5 years (54.34±4.20%) and over 10 years (50.71±3.50%).

However, no difference was observed between the sampling efficiencies of the different cashew orchards (Mann Whitney, *p = 0.056*) (Table 3). This observed coverage rate indicates that the sampling method used was highly effective in all the orchards sampled.

Table 4. Collected bee species and corresponding diversity parameters

Subfamily	Species	Orchards 0-5 years	Orchards 5-10 years	Orchards over 10 years	Total	p-value
Formicinae	*Oecophylla longinoda*	5±0.62	15±1.01	33±2.03	53±0.00	-
	Camponotus acvapimensis	3±0.31	10±1.22	15±1.01	28±0.00	-
Myrmicinae	*Lepisiota* sp.1	2±0.02	3±0.35	1±0.01	6±0.00	-
	Crematogaster sp.1	10±1.07	3±0.68	3±0.34	16±0.00	-
	Pheidole sp.1	5±0.57	6±0.81	10±0.98	21±0.00	-
Abundance (individuals)		25±3.08^{c}	37±4.16^{b}	62±6.01^{a}	124±8.60	0.009
Observed species richness (Sobs)		5^{a}	5^{a}	5^{a}	5	0.81
Estimated specific richness (Chao2)		9.2	9	9.86	100	
Simpson's diversity indices		0.71^{b}	0.82^{a}	0.76a^{b}	0.8	0.043
Equitability indices		0.21^{a}	0.28^{a}	0.26^{a}	0.3	0.76

Values with different letters on the same line are significantly different (p < 0.05) using the Mann Whitney test.

Diversity of ant populations

For all the orchards sampled, 124 ants were collected. The distribution of ant abundance was significantly higher in orchards over 10 years (62±6.01 individuals) than in orchards 5-10 and 0-5 years, with 37±4.16 and 25±3.08 individuals respectively (Mann Whitney, *p = 0.009*).

The collected ant population was composed of 2 subfamilies (Formicinae, and Myrmicinae) subdivided into 5 species and 5 genera. The Formicinae subfamily, represented by 3 species, was considerably more diverse than the Myrmicinae (2 species) (Table 4). However, for the different types of orchards, all 5 species were represented. Thus, no difference was observed between the species richness of these orchards (Mann Whitney, *p = 0.81*). As for Simpson's diversity, it was significantly higher in orchards 5-10 years (0.82) than in orchards over 10 years (76) and 0-5 years (0.71) (Mann Whitney, *p = 0.043*). On the other hand, equitability varied very little from one type of orchard to another (Mann

Whitney, $p = 0.076$), with values of 0.28, 0.26 and 0.21 respectively for orchards 5-10 years, over 10 and 0-5 years (Table 4).

Discussion

Evaluation of sampling efficiency using the coverage rate indicates that over 60.68% of expected bee species and 50% of expected ant species were collected. This indicates that the sampling methods used were effective and that the samples obtained are representative of the communities in the orchards.

However, the ant sampling method proved less effective than the bee sampling method. This suggests that a large number of ant species went undetected compared to bee species.

Moreover, compared with control cashew trees, beneficial insect activity in the three different types of orchards was lower. These observed differences could be explained by the good health of the inflorescences and flowers due to the foliage of these preferred cashew trees (controls), which provide ideal shelter for the allies (Silué *et al.*, 2022). Thus, for those orchards already in place with less improved varieties, the consideration of palliative measures, namely the installation of hives and the intrusion of plants preferred by beneficial ants such as mango would be necessary for an improvement in orchard productivity (Houngbo *et al.*, 2018; Tuo *et al.*, 2020; Coulibaly *et al.*, 2024).

A total of 184 bee pollinators comprising 10 species subdivided into 2 families (Apidae and Halictidae) and 8 genera were collected. Diversity, species richness, abundance and visit frequency of foraging bees were higher in orchards over 10 years. This indicates that the seedlings in these orchards would be in a healthier state, with conditions more favourable to pollinator activity, than those in young orchards (5-10 years and 0-5 years). Orchard seedlings over 10 years could therefore have very low parasite incidence rates, and consequently high recruitment of vegetative and floral organs and good quality floral resources (nectars and pollens) (Masawe *et al.*, 2006; Chipojola *et al.*, 2013; Da Silva Santos *et al.*, 2022). In terms of the taxonomic structure of the 2 families of foraging bees collected from cashew flowers (Apidae and Halictidae), the Apidae are distinguished by their high species diversity. Among these species, *Apis mellifera* and *Meliponula bocandei* showed very high abundances. This explains the low values observed for equitability and Simpson's index in the three types of orchards sampled. These results corroborate those of Silué *et al.* (2021) and Silué *et al.* (2022), who showed that cashew trees are visited by large numbers of bees.

However, *Apis mellifera* followed by *Meliponula bocandei* are the most efficient pollinators in terms of the number of times they visit the flowers, the number of pollen grains they carry and the contact they make with the pistil on 98.74% of visits. In addition, studies by Kojobe (2006) have shown that among the bee species involved in pollination, these two species have the greatest demand for food resources (nectar and pollen) for brood maintenance (larvae, queen, colony individuals).

As for ants, 124 individuals of 5 species divided into 2 subfamilies and 5 genera were collected. This second category of beneficial insects was also more abundant in orchards over 10 years. This is probably because cashew trees in orchards over 10 years old offer good-quality shelter and more interesting extrafloral resources than those in the 2 young orchards (Peng *et al.*, 2010; Anato *et al.*, 2015). Analysis of the taxonomic structure of the ants showed that *Oecophylla longinoda* (African weaver ants) dominated the other ant species in the different orchards sampled. This explains the low equitability values of Simpson's index observed in all three orchards. Studies carried out by a number of authors have shown that the *Oecophylla longinoda* ant species has the particularity of building satellite nests between leaves, and represents fierce defenders of their territory against any intruder. Thus, they develop effective strategies for hunting in groups on tree foliage, on the ground at the foot of the trees on which they nest and capture the insect pests found there (Dwomoh *et al.*, 2009; Crozier *et al.*, 2010; Olotu *et al.*, 2013; Anato *et al.*, 2015; Diame *et al.*, 2015).

Conclusion

In the Marahoue region, cashew orchards are characterized by the presence of plants from less advanced varieties, which harbor a high specific diversity of beneficial insects such as bees and ants. However, older orchards have higher species richness, abundance and frequency of visits by pollinating bees and pest control ants. As far as bees are concerned, the Apidae family is characterized by a wide variety of species, with a particularly marked presence of species such as *Apis mellifera* and *Meliponula bocandei*, both of which play an essential role in pollination. As far as the ant population is concerned, the species *Oecophylla longinoda*, often referred to as African weaver ants, which exerts strong pressure on pests, stood out for its predominance. Nevertheless, to optimize the productivity of the various orchards, it is essential to adopt compensatory measures such as setting up beehives and introducing plants appreciated by beneficial ants.

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