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Akissi Evelyne Kouakou
Université Félix Houphouët Boigny, UFR Biosciences, Laboratoire des Milieux Naturels et Conservation de la Biodiversité, 01 BP V 34 Abidjan01, Côte d'Ivoire

Kanvaly Dosso
Université Nangui Abrogoua, UFR des Sciences de la Nature, Laboratoire d'Ecologie et du Développement Durable, 02 BP 801 Abidjan 02, Côte d'Ivoire.

Yves Roisin
Université Libre de Bruxelles, Evolutionary Biology and Ecology, Faculty of Sciences, 50 av. F.D. Roosevelt B-1050 Brussels, Belgium.

Souleymane Konate
Université Nangui Abrogoua, UFR des Sciences de la Nature, Laboratoire d'Ecologie et du Développement Durable, 02 BP 801 Abidjan 02, Côte d'Ivoire

Kouassi Phillipe Kouassi
Université Félix Houphouët Boigny, UFR Biosciences, Laboratoire des Milieux Naturels et Conservation de la Biodiversité, 01 BP V 34 Abidjan01, Côte d'Ivoire

Corresponding Author:
Akissi Evelyne Kouakou
Université Félix Houphouët Boigny, UFR Biosciences, Laboratoire des Milieux Naturels et Conservation de la Biodiversité, 01 BP V 34 Abidjan01, Côte d'Ivoire

Soil-feeding termite diversity and abundance in a natural tropical humid forest (Tai National Park, Côte d'Ivoire)

Akissi Evelyne Kouakou, Kanvaly Dosso, Yves Roisin, Souleymane Konate and Kouassi Phillipe Kouassi

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Abstract
This study aimed to contribute to the soil-feeding termite knowledge in the primary, secondary and swampy forest inside Taï National Park (Côte d'Ivoire). The objective was to assess the diversity and abundance of soil-feeding termite species in these different habitats. With a standardized method, data were collected on soil-feeding termite from habitats. The results showed a significant variation of soil-feeding termite subfamilies between the habitat types. 45 soil feeding termites species and morpho species have been recorded and identified. They belong to the Termitidae family, 3 subfamilies (Apicotermitinae, Cubitermitinae, and Termitinae), 28 genera and 428 occurrences. The highest occurrence of species (70.60%) belongs to the Apicotermitinae subfamily followed by the Termitinae (16.14%) and the Cubitermitinae (13.25%). This study has identified some new species belonging to the Apicotermitinae subfamily in the Tai National Park using the anatomy of the gut of workers. These new species will be described later.

Keywords: soil-feeding termite, diversity, enteric valve, Taï National Park, Côte d'Ivoire

Introduction
Termites play important roles in the functioning and dynamic of tropical ecosystems [23, 27]. They are also known as good bio-indicators in tropical ecosystems because of their sensitivity to habitat disturbance causing changes in their species richness, composition and functional characteristics [16, 18]. Termites are important in soil fertility for plant growth [1] and soil porosity [20]. Termite species are generally settled into four principal feeding groups, which are: fungus growers, wood feeders, grass feeders and soil feeders [25]. Soil-feeding termites are social insects living and feeding in soil [9]. Their biomass is the highest in the tropical gallery forest of savannah [37]. Soil-feeding termites are very sensitive to habitat disturbance and are the most affected by land degradation [10], so that they represent very good biological indicators. The majority of these termites feed directly on the soil including the Apicotermitinae, a subfamily of the higher termites (Termitidae), which is especially diverse in the African tropics [9]. However, there are many difficulties in their identification, because among them, there are the soldierless, which can be identified by gut dissection to isolate the enteric valve. This method is adequate to easily identify soil-feeding termites without soldiers. Indeed, one of the most variable and extraordinary structures in the termite gut is the enteric valve which is often well developed although extremely variable in some of the phylogenetically derived termites, the Termitidae, and most developed in the subfamily Apicotermitinae [14]. This valve is relatively insignificant in the wood feeders [14] but very important in identifying the worker caste of termite genera from soils of Africa and the Middle East [35]. However, almost nothing is known about soil-feeding termites living in Taï National Park using enteric valve morphology for their identification. The first preliminary study carried out on termites in this park did not make an accurate assessment of the different trophic groups [36]. So, there is a lack of information about soil-feeding termites groups living in this area. Thus, this study aimed to provide a recent list of the soil-feeding termites with or without soldiers living in the Taï National Park because species can change from a habitat to another. Three habitat types inside the Taï National Park (primary forest, secondary forest and swampy forest) have been chosen. Specifically, soil-feeding termites' richness and occurrences within three different habitat types have been determined.

Also, the soil-feeding termite composition list was also established in the different habitat types. And the soil-feeding termite diversity within the three different habitat types was compared.

Material and methods

Study area

This study was carried out in the Taï National Park (TNP) during the rainy season from March to September 2019. This Park is located in the south-western part of Côte d'Ivoire (5°09'- 6°08' N and 6°48'-7°26'W). It is one of the last remnants of the original west African dense forest block. This Park has a current area of 536.700 ha [33] and has been included in the biosphere reserve network since 1978 then was registered on the UNESCO world heritage list in 1982. The climate is a typically subequatorial. According to climate data collected until the 1960, the annual precipitation average was 1150 mm and the temperature was 27◦C [19]. But, during the time of this study, the climate was characterized by a long rainy season from February to November and a short dry season from December to January, the annual precipitation average was 2050 mm and the temperature was 28◦C. Three habitat types (a primary forest, a secondary forest, and a swampy forest) differing in their vegetation structure were visited.

The primary forest (PF: 05 50'472" N and 007 20'779" W) is a closed plant formation, very heterogeneous in its floristic composition and in the size of the plants that compose it. The upper stratum is made of emergent trees that can reach 50 m in height and the lower stratum is less than 10 m and consists of a dense undergrowth of shrubs. The secondary forest (SeF: 05 82'786" N and 007 38'872" W) results from the felling of cocoa and coffee plantations in 1999 and 2000 in the application of a government decision to consist of destroying plantations within national parks and reserves in the country. The swampy forest (SF: 05 49'941" N and 007 20'424" W) is a plant formation on waterlogged soils, the stratification is simplified, with a homogeneous, low, dense upper storey, poor in lianas and an undergrowth rich in herbaceous plants.

Sampling methods

Five 5000 m² (100 m × 50 m) plots were delimited in each habitat type. A standardized method designed for rapid assessment of termites diversity [24, 29, 32] was used. Indeed, three transects 200 m² (100 m × 2 m) separated by 15 m between them were established in each plot. Each transect was subdivided into 20 no contiguous 5 m² (5 m x 1 m) quadrats in order to standardize the sampling effort. In each quadrat, 12 surface soil samples (12 cm wide and 10 cm deep) were dug out and sorted to collect soil-feeding termites encountered. The soil was hand-sorted in situ, and the soil-feeding termite was collected and kept in 90% ethyl alcohol. In order to sample all 20 quadrants in one day, four trained collectors were deployed.

A systematic capture had consisted of researching soil-feeding termites surrounding each plot. This systematic sampling was to excavate the accumulations of litter and humus at the base of trees and between buttress roots, the hollows inside tree stumps, in the dead woods, branches and twigs, the soil within and beneath very rotten woods, subterranean nests, mounds cartons sheeting and runways on vegetation, and arboreal nests up to 2 m above ground level.

Specimens identification

The identification was made in the laboratory using a binocular magnifying glass. Soil-feeding termite species were identified or their morpho species were numbered using the standard determination keys [5, 6, 7] and the books [21, 29, 35]. The identification of the soldierless specimens had required dissection of the enteric valve, which is an organ present in the digestive tract of soil feeding termite workers. The section of the gut containing a part of the first proctodeal segment (P1), the enteric valve, and a part of the third proctodeal segment (P3) was cut using a scalpel. This section of the gut is placed in a glass vial containing 85% ethanol and then immersed in a Branson ultrasonic stripper (B-22-4 Fisher Scientific, Pittsburgh, PA) for several seconds. The muscle and tissue of the inner lining of the gut are observed with a microscope. Images of the enteric valve structures are observed and photographed. The images are compared with existing images to determine the species [3] or morphospecies.

Data analysis

The occurrence (presence-absence) data were used because the number of individuals could be misleading when dealing with social animals, which are patchily distributed [30]. The occurrence was defined as the number of encounters per transect, where the presence of one species in a quadrat represented one encounter [30]. The species richness was determined by the number of species observed in a plot. The three transects in each plot were combined for statistical analysis. The comparison of mean species richness and mean subfamilies richness in the three habitat types were tested using the R software version 4.0.2. Variations of soil feeding species richness across habitat types were tested using ANOVA1 and the least significant difference (LSD) post hoc comparison test was used to detect differences between them. The second-order no-parametric estimator Chao2 was used as the estimator of the species richness. This estimator takes into account the distribution of species among sections and only needs, for its calculation, the number of species found in just one section and the number of species in exactly two [12]. This estimator was calculated using Estimate S 9.1.0. The accumulation curves represent the evolution of the number of species (species richness) according to the sampling effort (expressed in sampled sections). The observed (Sobs) and estimated (Chao2) species accumulation curves were constructed using Estimate S 9.1.0, after randomizing the sample order 500 times to ensure the statistical representation of the target assemblage [11]. The sample coverage, which is the ratio of the observed species richness (Sobs) as percent of the estimated species richness (Chao2) in each habitat has been determined. The assessment of Jaccard's similarity index of soil-feeding termite assemblages between habitat types was defined using Estimate S 9.1.0. The Shannon index and the evenness were used to measure the diversity of the soil-feeding termite assemblage. They were computed by using the "Past" software version 4.07. This measure tends towards 0 when a taxon has largely dominated by a stand and is equal to 1 when all taxa have the same occurrence.

Results

Sampling efficiency

The values of sampling coverage were high for all habitats. The highest value was found in the swampy forest, followed by primary and secondary forests. The total species richness and uniqueness species were the highest in the primary forest, followed by the secondary forest and swampy forest (Table 1). There was a low dissimilarity between species encountered in the primary and secondary forest, these two habitats shared many species in common compared to the species in the swampy forest. On the other hand, there was a high dissimilarity between species found in a primary and swampy forest. The secondary and swampy forest shared almost the same number of common species (Table 2). The accumulation curves of the observed (Figure 1A) and estimated (Figure 1B) were all are nearing of the asymptote.

Table 1: Species richness (Sobs and Chao 2), Sample coverage and Uniques species of soil-feeding termite in three habitats. (uniques =species collected only once in each habitat).

Habitat types	Sobs	Chao2	Sampling coverage	Uniques species
Primary forest	39	42.24	92	5
Secondary forest	34	38.95	87	3
Swampy forest	18	18	100	1

Table 2: Community complementarity between habitats. Data in cells are reported as follows: common species/different species/1-Jaccard's similarity index (%).

Habitat types.	PF	SeF	SF
PF	×	32/13/30%	15/30/64%
SeF		×	27/18/51%
SF			×

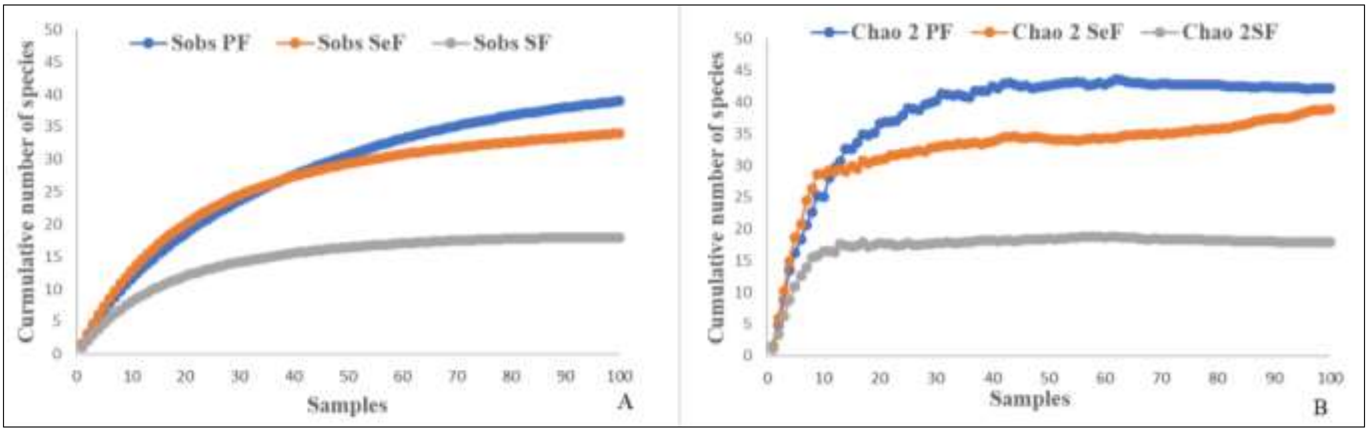

Fig 1: Accumulation curves of the observed (A) and estimated species richness (B) in the three habitat types. (PF: Primary Forest); (SeF: Secondary Forest); (SF: Swampy Forest).

Taxonomic structure of the recorded soil-feeding termite

Overall, 45 soil-feeding termite species and morphospecies were collected and identified. They belong to the Termitidae family, 3 subfamilies (Apicotermitinae, Cubitermitinae, and Termitinae) and 28 genera (Table 3). There was a significant variation between subfamilies occurrences in the three habitat types according to the ANOVA 1 (F= 8.9377; df= 2; $p < 0.05$). The highest occurrence of species (293 representing 70.69%) was observed in the Apicotermitinae subfamily, followed by the Termitinae (67 representing 16.14%) and the Cubitermitinae (55 representing 13.25%) (LSD test, $p < 0.05$; n = 3).

Table 3: List species and morphospecies collected and their frequency of occurrences in the visited habitat types.

Subfamilies	Termites species	Soldier caste	Primary forest	Secondary forest	Swampy forest
Apicotermitinae	*Acholotermes* sp.	-	0	0	9
	Adaiphrotermes sp1.	-	3	8	5
	Adaïphrotermes sp2.	-	7	1	3
	Aderitotermes cavator (Sands, 1972)	-	1	8	4
	Aderitotermes sp1.	-	2	8	8
	Aderitotermes sp2.	-	3	3	8
	Aderitotermes sp3.	-	12	9	10
	Allognathotermes sp.	+	3	3	9
	Alyscotermes sp.	-	4	9	12
	Amicotermes ivorensis (Sands, 1972)	-	2	4	4
	Anenteotermes sp.	-	3	12	2
	Anenteotermes polyscolus (Sands, 1972)	-	5	6	2
	Apicotermes sp.	+	0	4	0
	Astalotermes quietus (Silvestri, 1914)	-	4	7	3
	Astalotermes sp1.	-	0	4	2
	Astalotermes sp 2.	-	3	3	4
	Astalotermes sp 3.	-	5	6	2
	Astalotermes sp4.	-	0	12	2
	Astratotermes sp.	-	2	1	11
	Duplidentitermes sp.	+	3	1	9

	Eburnitermes sp.	+	2	1	0
	Hoplognathotermes sp.	+	1	0	0
	Kaktotermes sp.	-	8	0	0
Cubitermitinae	*Apilitermes* sp.	+	1	5	0
	Basidentitermes potens (Silvestri, 1914)	+	3	3	0
	Basidentitermes sp1.	+	3	4	0
	Basidentitermes sp2.	+	1	5	
	Cubitermes sp.	+	2	0	0
	Euchilotermes sp.	+	0	3	0
	Lepidotermes sp.	+	1	1	0
	Megagnathotermes sp.	+	5	4	0
	Noditermes sp.	+	4	8	0
	Orthotermes depressifrons (Silvestri, 1914)	+	3	1	0
	Orthotermes sp.	+	1	1	0
	Unguitermes sp1.	+	0	2	0
	Unguitermes sp2.	+	4		
	Thoracotermes sp.	+	4	0	0
Termitinae	*Angulitermes* sp.	+	12	0	0
	Cephalotermes sp1.	+	2	4	0
	Cephalotermes sp2.	+	2	2	0
	Pericapritermes appelans (Silvestri, 1914)	+	24	8	0
	Pericapritermes silvestrianus (Emerson, 1928)	+	2	1	0
	Pericapritermes sp1.	+	3	1	0
	Pericapritermes urgens (Silvestri 1914)	+	2	2	0
	Promirotermes holmgreni (Silvestri, 1912)	+	2	0	0

Soldier caste: - absent, + present.

Mean species richness in the three-habitat types.

The variation between mean species richness significantly differs in the three habitat types according to the ANOVA 1 (F= 4.1915; df= 2; p =0.01). The mean species richness in the three habitat types was respectively (30.8±5.9), (33±5.1) and (21.8±5.4) in the primary forest, secondary forest and swampy forest (LSD test, p < 0.05; n = 3) (Figure 2).

Soil-feeding termite subfamilies variation across the visited habitats.

Globally, the mean subfamilies richness of soil-feeding termites significantly varied across the three habitat types according to the ANOVA 1 (F= 8.9377; df= 2; p < 0. 05). The highest mean of subfamilies richness was observed in the Apicotermitinae (21.13±4.12) followed by the Cubitermitinae (8.53±5.03) and the Termitinae (3.40±2.06) (LSD test, p < 0.05; n = 3) (Figure 3).

Soil-feeding termite species diversity in the habitat types

The secondary forest showed the highest number of individuals (165), followed by the primary forest (154) and the swampy forest (109). The Shannon index in the secondary forest was higher (3.32) than in the primary forest (3.25) and the swampy forest (2.71), but the evenness was the highest in the swampy forest (0.90). It also appears that the evenness in the three habitat types was between (0.74 - 0.90), therefore above 0.5 either 50% (Table 4).

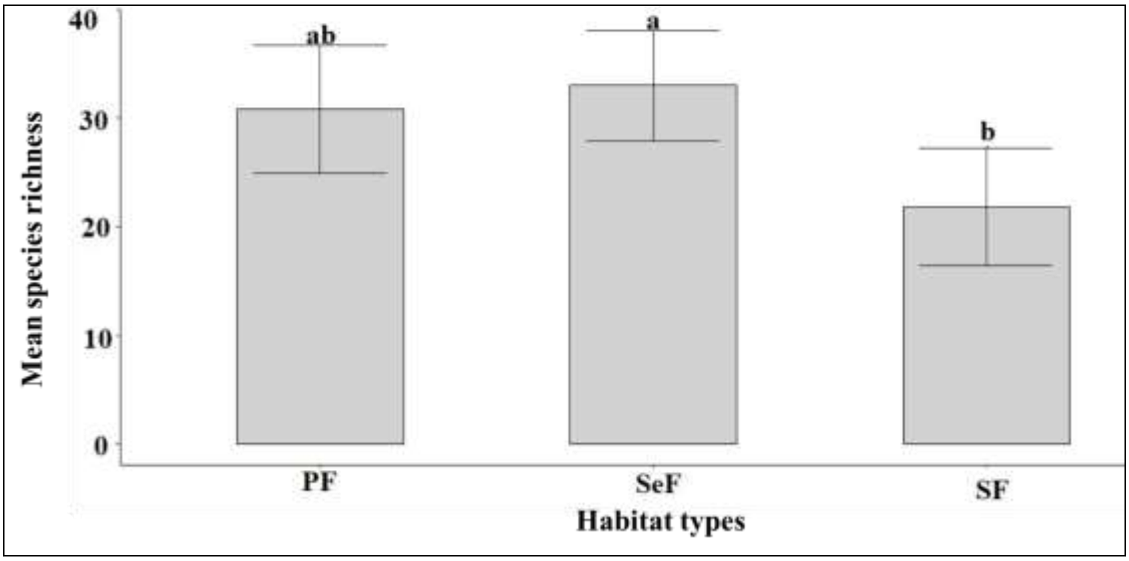

Fig 2: Mean species richness (±SE) of soil-feeding termite collected inside three habitat types.

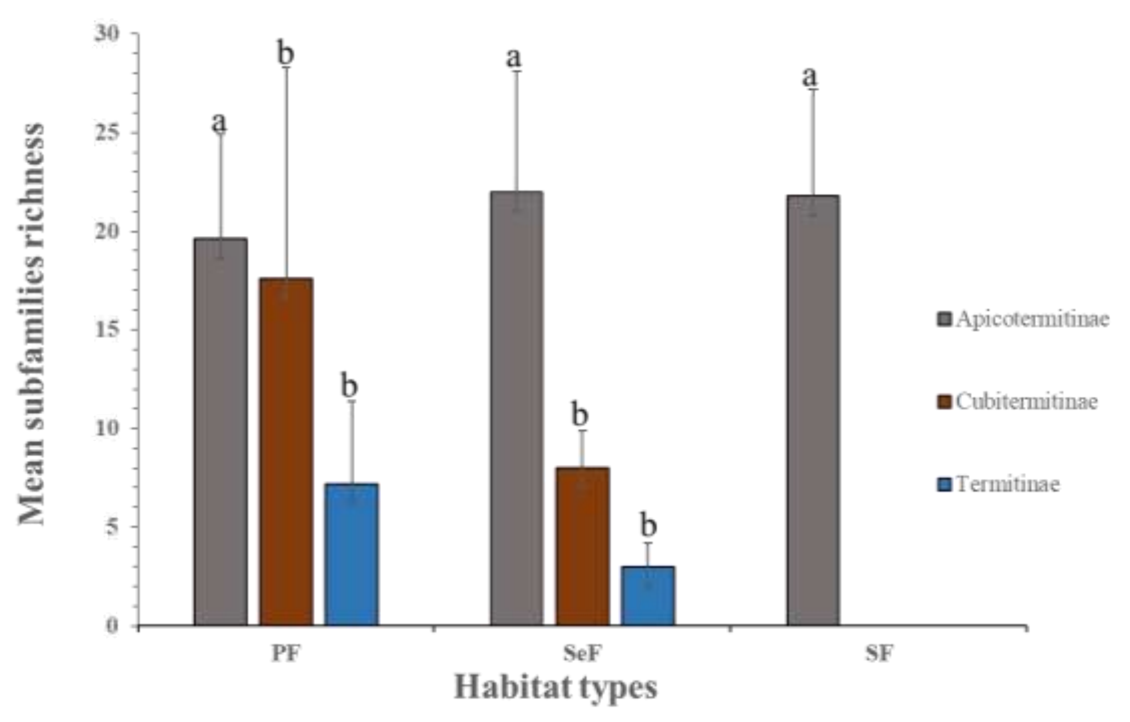

Fig 3: Mean subfamilies richness (± SE) of soil-feeding termite collected inside three habitat types in the Taï National Park, Côte d'Ivoire

Table 4: soil feeding termite diversity index in different habitats

	PF	SeF	SF
Individuals	154	165	109
Shannon (H')	3.25	3.32	2.71
Evenness (E)	0.74	0.89	0.90

Discussion

The accumulation curves of the observed and estimated species richness had presented the sampling method used in this study was efficient, so that the soil-feeding termite assemblage was efficiently described. This was illustrated by the plateau attained by all observed species accumulation curves as well as by high values of sample coverage. In addition, we combined two methods: the rapid assessment of termite diversity and the systematic capture, which consisted of researching soil-feeding termites surrounding 50 m and 100 m of each plot in a habitat type, so the species richness was observed would be observed increase.

Forty-three soil-feeding termite species and morpho species were identified within all the visited habitats. Furthermore, there was a significant variation between occurrence of the subfamily in the three habitat types. The highest occurrence of species was observed in the Apicotermitinae subfamily followed by the Termitinae and the Cubitermitinae. This observation was probably due to the fact that among soil-feeding termites, this subfamily is particularly rich in species [22] and was counted for more than one third of the total local termite species richness [8]. This result was observed in the termite communities in sugarcane plantations in southeastern Brazil [26]. Also, in our samples, a lot of soldierless soil-feeding termite species have been collected. They are ranked in the Apicotermitinae group with practice valve enteric dissection of the workers. This result is different from Lamto reserve [16, 17, 18] and the Comoé National Park [29] using only morphometric method to identify termites. Indeed, these authors, had identified three principal species which were *Adaiphrotermes* sp., *Aderitotermes* sp. and *Astratotermes* sp. on the basis of the size of their abdomen. Also, their studies had revealed the highest occurrence of these species in the savannah. Some species such as *Acholotermes* sp. had been collected only in the swampy forest, *Apicotermes* sp., *Euchilotermes* sp., *Unguitermes* sp., in the secondary forest and *Angulitermes* sp**.,** *Cubitermes* sp., *Eburnitermes* sp., *Hoplognathotermes* sp., *Kaktotermes* sp., *Promirotermes holmgreni* only in the primary forest. This result could confirm the hypothesis that soil-feeding termite species can change from a habitat to another. This fact was observed in a comparative study on the termites species richness in two classified forests in South-Western Côte d'Ivoire [2]. According to the results obtained by these authors, *Cubitermes* sp. and *Thoracotermes macrothorax* were recorded only in the cavally classified forest but *Dicuspiditermes* sp. and *Ophiotermes* sp. were recorded only in the "haute dodo" classified forest. The significant variation between mean species richness and mean subfamilies richness in the habitat types were probably due to the organic matter in the three habitats according to the different vegetation type. Indeed; soil-feeding termites are the organisms linked exclusively to decomposing organic particles in the humic fraction of the soil [38]. Also, the majority of species in this study has been collected in the 10 cm of soil which seemed to be rich in organic matter [9].

The secondary forest has recorded the highest specific richness and the Shannon index compared to the primary and the swampy forest, certainly because it would be possible to find in the secondary forest vegetation with a microclimate to the soil-feeding growth. Indeed, the secondary forest is an old cocoa plantation of the study area. So, the residues of organic matter from the decomposition of these old cocoa fields could provide the soil more nutrient to the soil feeders growth compared to the two other habitat types including the primary and swampy forest. The equitability index above 50% indicates a good distribution of individuals between the species which have been recorded in the three habitat types, this result was obtained in the Yoko Forestry Reserve [4].

Conclusion

This study had shown a significant number of the soil-feeding termite belonging Apicotermitinae subfamily in the three habitat types. It reveals that the majority of the soil- feeding termite species collected in Tai National Park belonging to the subfamily Apicotermitinae. With using enteric valve morphology for their identification, there are certainly some species that deserve special attention because they could be a new species that will be described. So, it is therefore important to combine molecular analyses to the morphology and the gut dissection methods to identify the soil-feeding termites clearly living exactly inside this park.

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