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## Full Length Research Paper

# Diversity of ectomycorrhizal fungal fruit bodies in Comoé National Park, a Biosphere Reserve and World Heritage in Côte d'Ivoire (West Africa)

Linda Patricia Louyounan Vanié-Léabo<sup>1,2\*</sup>, Nourou Soulemane Yorou<sup>3</sup>, N'Golo Abdoulaye Koné<sup>4</sup>, François N'Guessan Kouamé<sup>1,5</sup>, André De Kesel<sup>6</sup> and Daouda Koné<sup>1,2</sup>

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The key role of ectomycorrhizal (EcM) fungi in ecosystems functioning has been demonstrated worldwide. However, their diversity, spatial distribution, fruiting phenology and production as influenced by climatic parameters variability remain poorly understood in tropical African forests. Weekly surveys were conducted from April to early October 2014 at the Comoé National Park (CNP), Côte d'Ivoire (West Africa) in 09 permanent plots established in *Isoberlinia doka* (IW), *Uapaca togoensis* (UW) and Mixed (MW) woodlands. Non metric multidimensional scaling (NMDS) of EcM fungi abundance was run to assess the influence of environmental parameters on fungi distribution using the package VEGAN. Hierarchical clustering based on dissimilarity and indicator species analysis were run to characterize fungi communities. Analyses were computed with the statistical program R. A total of 123 EcM fungi species belonging to 23 genera and 09 families were collected at CNP. Simpson diversity (1-D) and evenness were 0.97 and 0.54, 0.97 and 0.61, 0.96 and 0.52 for IW, MW and UW respectively. Yet, weekly-based species accumulation curves did not reach an asymptote. Stem density of *U. togoensis* Pax (UTDen) and *I. doka* Craib & Stapf were the most important tree parameters influencing EcM fungi distribution (respectively  $r^2 = 0.92$  / p-value = 0.002 and  $r^2 = 0.83$  / p-value = 0.018). Two sites groups were distinguished and four indicator species were identified.

**Key words:** EcM fungi, fruit bodies, diversity, indicator species.

## INTRODUCTION

Productivity, diversity and composition of plant communities have been demonstrated indirectly and

directly influenced by belowground micro-organisms from which plant symbionts play a key role (Van Der Heijden

et al., 2008; Van Der Heijden and Horton, 2009). Globally, over 90% of terrestrial plants depend upon an ecological relationship with soil fungi for their growth and regeneration (Smith and Read, 2008; Singh et al., 2011; Dickie et al., 2014). This relationship termed mycorrhiza is the most prevalent symbiosis on Earth, including cultivated plants, herbaceous species and forest trees. Generally, autotrophic plants provide carbohydrates to their fungi partners, which in turn improve host performance by enhancing mineral nutrient uptake from soil, especially nitrogen (N) and phosphorus (P). Symbiotic fungi enhance plant tolerance to environmental stress caused by low soil water potential, toxic heavy metals, salinity, herbivores and root pathogens (Smith and Read, 2008; Singh et al., 2011; Dickie et al., 2014). Among mycorrhizas types, ectomycorrhiza (EcM) is the most advanced one (Moore et al., 2011) involving mostly higher plants and fungi (Piepenbring, 2015). Thus, EcM fungi have an important position in the plant-soil interface (Ceulemans et al., 1999) worldwide, playing a key role in the growth and regeneration of forest trees, and in ecosystems functioning.

However, the global biodiversity is under decline since the 19th century due to serious climate, environmental and ecological changes through human activities around the globe. The global climate system is actually modified by increased greenhouse gases (GHG) in the atmosphere subsequently to unrestrained deforestation, fossil fuel combustion and other anthropogenic activities (WMO, 2007). Few key parameters of global change are among other trend towards warming (increasing temperature), increase of atmospheric CO<sub>2</sub> and disturbance in the distribution, seasonality and amount of rainfalls. It is predicted that Earth surface temperature will increase from 0.3°C to 1.7°C under scenario RCP2.6 by the end of the 21st century (2081–2100) whilst the atmospheric carbon level is continuously increasing (IPCC, 2014). Though the impact of global change on ecosystems is not yet adequately addressed, it is expected that many changes in global biodiversity and ecosystem functions will occur. High temperature is expected to alter tree phenology, plant growth and distribution toward migration and adaptation ecozones (Montoya and Raffaelli, 2010) but also to increase the length of the growing season (Walther et al., 2002; Morin et al., 2007), and the aboveground growth and reproductive effort of plants (Hollister et al., 2005). At the other side, elevated atmospheric CO<sub>2</sub> and nitrogen will likely increase the rate of net photosynthesis by 40 to 80% (Körner et al., 2005), the allocation of carbon to the plant roots (Janssens et al., 2005) and the production of leaves, wood and coarse roots (Hyvönen et al., 2007). It

is actually difficult to predict the exact response of plant diversity to climate change as many investigations are still needed to understand the resilience, adaptation and/or migration following fluctuation of climatic parameters.

As both partners are living more or less obligatory and intimately, any possible change that affect host plants is also expected to influence the symbiotic fungi. In temperate and boreal zone, rainfall and moisture availability have been demonstrated as critical to EcM fruiting and natural production (O'Dell et al., 2000; Gange et al., 2007; Kauserud et al., 2010). Furthermore, long term observations of fungal phenology in temperate forests reveal that fruit bodies production and temporal changes are strongly influenced by either increasing temperature (Kauserud et al., 2008; Kauserud et al., 2010) and/or rainfalls (Krebs et al., 2008). Due to their vital role in forest ecosystems and the sensitivity of their respiration to high temperature and strong seasonality (Vargas et al., 2010; Bahram et al., 2012), EcM fungi represent best candidates to investigate for a better understanding of ecosystems response to global warming and especially in carbon sequestration capability (Simard and Austin, 2010; Orwin et al., 2011; Büntgen et al., 2012; Büntgen et al., 2013; Boddy et al., 2014). Unfortunately, the response of EcM communities to global warming and environmental changes is scarcely addressed in tropical zones and especially in tropical. In Sudanian woodlands of Africa, a strong variability has been noticed regarding species richness and community structure throughout the fruiting season (Yorou et al., 2001). Nevertheless, the authors failed to link species composition, community structure and productivity patterns of EcM with either the local temperature or soil humidity. To our knowledge, that study is the only one in tropical Africa addressing the impact of climate parameters on wild EcM fungi phenology and productions. Now, knowing temporal change in the phenology and production distribution, and their determinants is essential in the valorisation of natural productions of wild edible EcM fungi that amounts to thousand tons annually and involves many rural women (Yorou et al., 2001, 2014; Boa, 2004). However, a prerequisite to climate impact assessment is the analysis of EcM fungi diversity and the evaluation of possible other natural underlying mechanisms of richness pattern (Tedersoo and Nara, 2010). It has been demonstrated that the impacts of atmospheric carbon dioxide enrichment is more clear on fruit bodies than on below-ground tips (Andrew and Lilleskov, 2009; Pickles et al., 2012). Therefore, this study aims to (1) assess the diversity (species richness and community structure) of

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EcM fungi species through fruit bodies diversity and (2) assess the spatial variability of the community composition following habitat characteristics (plant and soil parameters) at local scale. We hypothesised that (1) African protected areas harbour a great diversity of EcM fungi with many species likely new to sciences, and (2) host plants and soil structural parameters drive the communities of EcM fungi.

## MATERIALS AND METHODS

### Study site

The Comoé National Park (CNP) is located in the North-East of Côte d'Ivoire (8°32' - 9°32'N, 3°01' - 4°24'W) between the towns of Bouana and Dabakala, and south of the border with Burkina Faso. The CNP covers about 11 500 km<sup>2</sup> (Hennenberg, 2004) and is presently one of the largest national park in West Africa (Poilecot et al., 1991). Initially erected as a game park since 1926 ('Refuge Nord de la Côte d'Ivoire') and then established as national park in 1968, Comoé was approved in 1983 and declared as Biosphere Reserve and World Nature Heritage by the UNESCO (Hennenberg, 2004).

The park is located on the large granite stand of West Africa and is characterized by a smooth and level relief. Soils are impoverished sandy to loamy ferralsols above Precambrian granites with small areas of lateritic crusts or banks outcrop at some places (Hennenberg et al., 2005). The climate is a Guineo-Congolian/Sudanian transitional type, a sub-humid tropical climate (Chidumayo et al., 2010) with mean annual rainfall of 1 011 mm falling mainly between March and October. The mean annual temperature is 26.5 to 27°C (Kouloubaly, 2008). CNP vegetation is transitional ranging from forests to savannas including riparian grasslands (Poilecot et al., 1991; Hennenberg et al., 2005).

### Selection of habitat types and establishment of permanent plots

One-week exploratory survey was undertaken within the accessible parts of the park in November 2013 to identify appropriate study sites. Based on available vegetation maps (Poilecot et al., 1991; Lauginie, 2007), three habitat types were selected with regard to; (1) The presence and abundance of known EcM partners trees, members of Caesalpiniaceae and Phyllantaceae (to ensure collection of symbiotic fungi and assess partners influence on fungal species distribution) and (2) the distance to the Ecological Research Station of Comoé, our base camp (for rapid handling of fragile specimens during hot and wet season).

The different habitat types were at least 300 m away from one another and included:

Habitat type 1: *Isoberlinia doka* Craib & Stapf Woodland (IW);

Habitat type 2: Mixed Woodland (MW);

Habitat type 3: *Uapaca togoensis* Pax Woodland (UW).

In each selected habitat type, three permanent plots of 30 m × 30 m each have been established by mean of a hectometer, making a total of nine plots (Figure 1). They have been labelled *FiPi* with *Fi* representing the habitat type and *Pi* the plot. All nine (09) plots have been geo-referenced by recording the coordinates of each corner with a GPS Garmin GPSMAP® 62stc (Garmin International Inc., Olathe, KS, USA). Plots within a habitat type were spaced at least by 10 m one another, according to tree partners' presence and density (Table 1).

### EcM fungal fruit bodies collect and handling

EcM fungal fruit bodies (EFFB) were collected in each plot following parallel bands of 2 m large. To avoid missing short living species, each plot was visited once a week from April to early October 2014 as implemented by Yorou et al. (2001). We recorded the nearest EcM partner trees to each sampled fruit body and geographic coordinates using GPS Garmin GPSMAP® 62stc (Garmin International Inc., Olathe, KS, USA). To facilitate future comparison and morphological identification of species, technical photographs of most representative fruit bodies per species (at different development stage, when applicable) were taken on field and at the base camp using a Canon EOS 1000D digital cameras. Fresh macroscopic features were then recorded from specimens, using standardized descriptions sheets (size, shape; colour and any change with time; presence/absence of ephemeral structures; type of hymenophore, its colour and organization; etc.) developed for tropical African fungi (De Kesel et al., 2002; Eyi Ndong et al., 2011). Afterwards, Fruit bodies per collection were counted, weighted, labelled and representative specimens were dried at 40°C for 24 h. Labelled collections were conserved with basic ecological data (habitat type, substrate, putative nearest partner tree, exposition to sun, etc.) as herbarium materiel at the WASCAL GSP Climate Change and Biodiversity, University Felix Houphouet-Boigny (Côte d'Ivoire).

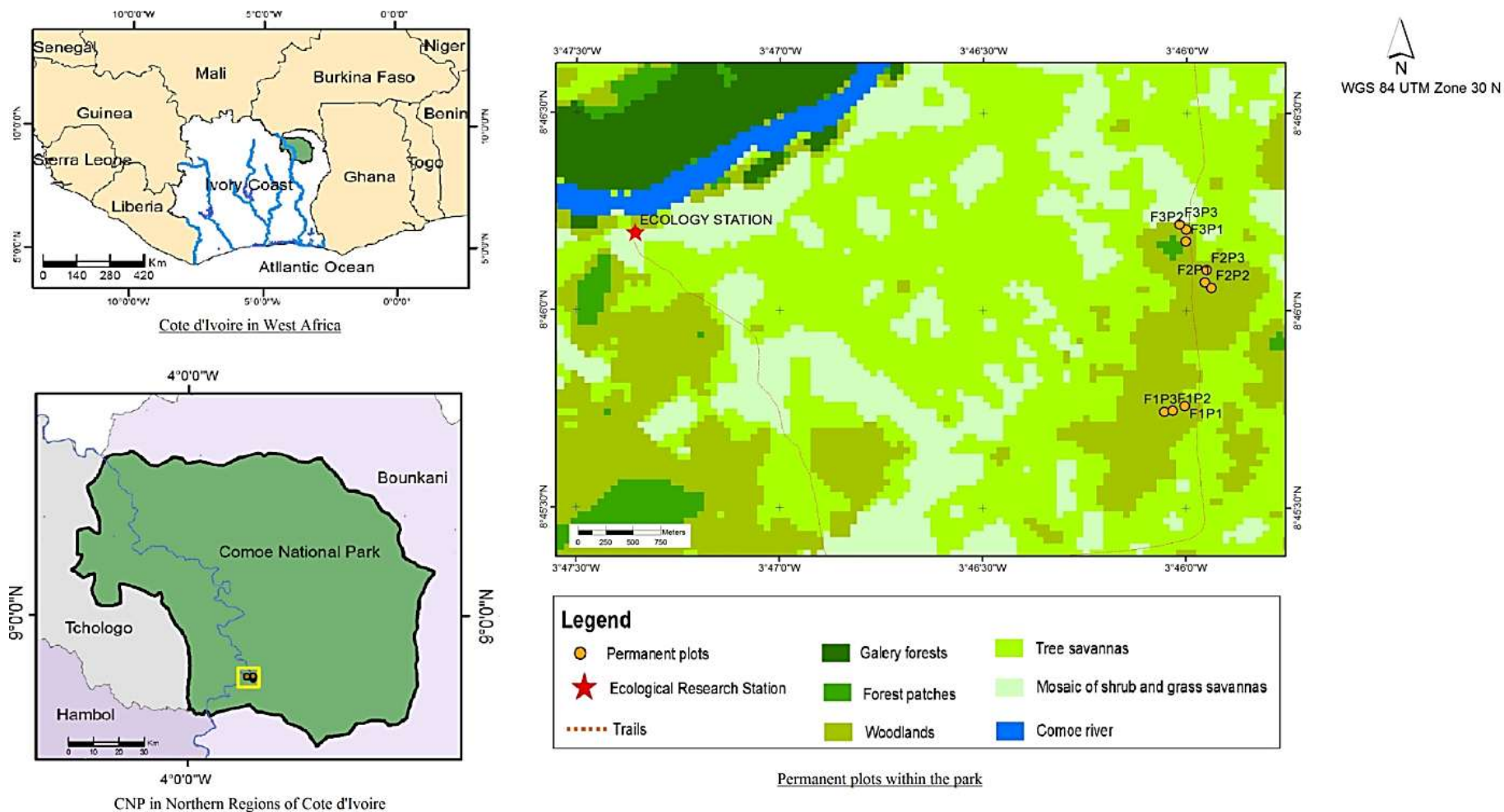
The identification of collected fungal species was performed based on morphological features at Botanic Garden of Munich in Germany and Botanic Garden Meise in Belgium by experts (De Kesel and Yorou, personal communications). Appropriate keys and numerous illustrated monographs on fungi of Central and Western Africa (series of "Flore Iconographique des Champignons du Congo" and "Flore illustrée des Champignons d'Afrique Centrale") were used. These series include monographs on *Amanita* spp. (Beeli, 1935), *Boletineae* and *Cantharellus* spp. (Heinemann, 1954, 1959, 1966), *Scleroderma* spp. (Dissing and Lange, 1963) and *Russula* spp. (Buyck, 1993, 1994, 1997) and *Lactarius* spp. (Heim, 1955). An additional monograph on *Lactarius* spp. (Verbeke and Walley, 2010) was also used. Species names and nomenclatural aspects were checked in index fungorum (<http://www.indexfungorum.org/Names/Names.asp>). Moreover, molecular-based identification of representative specimens per species was performed (Gardes and Bruns, 1993; Maba et al., 2013) at both abovementioned research institutes. Results of molecular analysis along with metabarcoding analyses of composite soil samples (for belowground fungi diversity assessment) will be presented in a manuscript in preparation.

### Habitat types characterisation

Biotic and abiotic variables were collected to assess their possible influence on EFFB occurrence and spatial distribution.

First, systematic inventory of plant species and total canopy cover estimation within plots were performed in April 2014 according to the phytosociological method (Braun-Blanquet, 1932). Primary identification of plants specimens were done with field guide (Arbonnier, 2004) and completed with collected herbarium materials by experts from the Laboratoire de Botanique of the University Felix Houphouet-Boigny in Abidjan, Côte d'Ivoire. However, for statistical analyses, only woody species with diameter at breast height (dbh) equal or above ( $\geq$ ) 10 cm were considered. Therefore, in addition to plant species richness, structural parameters (number of stems and dbh per species and per plot) were recorded.

Second, soil cores were collected with a 10 cm × 10 cm - 10 cm depth auger at each corner and the center of each plot at mid-rainy season (late July). All five cores were mixed to make a composite soil which was air-dried and passed through a 2-mm sieve. Three



**Figure 1.** Location of Comoé National Park (north east of Côte d'Ivoire) and established permanent plots within it (south west of the reserve).

composite soils were thus made per habitat and 200 g per sample were used to assess soil granulometry, pH and minerals contents. Chemical parameters assessed were pH (H<sub>2</sub>O), Carbon (C), Nitrogen (N), soil organic carbon (SOC), ratio C/N, Total Phosphorus (TotalP), Available Phosphorus (AvailP), Calcium (Ca) and Potassium (K).

Physical parameters referred to soil texture: Clay, fine and coarse Silt, fine and coarse Sand. They were determined as follows:

1. pH (H<sub>2</sub>O) measurement was performed with a soil solution at a ratio 2/5 (Duchaufour and Blum, 1997).

2. Determination of extractable cations' content was achieved according to standard NFX 31-130 (AFNOR, 1999).

3. Determination of organic and total carbon: The total carbon content in soil is determined after dry combustion. The soil's organic carbon content is calculated

**Table 1.** Positions of permanent plots within habitat types in Comoé National Park (CNP), Cote d'Ivoire

Habitat type	<i>Isoberlinia</i> Woodland			Mixed Woodland			<i>Uapaca</i> Woodland		
Plot	F1P1	F1P2	F1P3	F2P1	F2P2	F2P3	F3P1	F3P2	F3P3
Latitude (dd)	8.76264	8.762447	8.762408	8.767876	8.7676	8.768387	8.769594	8.770105	8.7703
Longitude (dd)	-3.7667	-3.76719	-3.76754	-3.76588	-3.766	-3.76581	-3.76668	-3.76665	-3.767
Altitude (m)	235.13	233.17	232.64	230.40	230.79	248.19	216.23	213.81	213.62

dd: decimal degrees; m: meters

according to the method NF ISO 10694 (AFNOR, 1995).

4. Particle size determination by sedimentation - the pipette method following the standard method NF X 31-107 (AFNOR, 2003).

## Data analysis

### *EcM fungal fruiting bodies diversity assessment*

Basic estimators and indices were calculated to assess the diversity of fungi species as reflected by EFFB at plot and habitat type level. They included also similarity between plots and habitat types as well as the number of shared species to compare communities.

### *Observed species richness and diversity assessment*

Presence/absence data of EFFB was used to determine (1) the observed species richness (SR: number of species) and composition (SC: list of species) per habitat type; (2) the total observed species richness and composition as cumulative data of all habitat types. Thereby, the frequency of occurrence (percentage of total weeks during which a species was recruited) of fungal species was used to highlight the contribution of each species in the community (Horton and Bruns, 2001). The relative frequency of each species was calculated as the percentage of total frequency.

Assessment of fungi diversity and evenness of frequency of species within habitat types was achieved respectively by computing Simpson's Index of Diversity (1 - D) and Simpson's Evenness with the program Ecological Methodology (Krebs and Kenney, 2002). Simpson's Index of Diversity (1 - D) refers to the probability that two individuals randomly selected from a sample will belong to different species. Its value ranges between 0 and 1, greater value corresponding to high diversity).

### *Sampling representativeness: Species accumulation curves and similarity assessment*

Sample-based species accumulation curves were constructed in EstimateS ver. 9.1.0 (Colwell, 2013) using presence/absence (incidence) data. The sample order was randomized 500 times without replacement for the statistical representation of the EcM fungi community. In this study, "sample" referred to frequency of survey, a week-interval, against which Observed and Estimated Chao 2 species accumulation curves were plotted.

The similarity of our sampling to the fungi community was estimated by measuring the autosimilarity (Cao et al., 2002) between plots of each habitat type. This was calculated as mean Jaccard coefficient computed with EstimateS ver. 9.1.0 software. Autosimilarity index varies from 0 (no species common to plots) to 1 (same species composition in plots). Constructed week-based species accumulation curves, Simpson's Index of Diversity (1 - D) and Simpson's evenness along with autosimilarity index served to assess the sampling representativeness of fungal communities

of study sites.

### *Habitat characterisation*

**Floristic richness and dendrometric parameters assessment:** Number of stems and dbh per species underwent basic statistical analyses as follows:

1. Plant species density ( $D_i$ ), the number of stems per species per plot surface in square meters ( $m^2$ ), converted later in hectares (ha);
2. Individual stem basal area ( $BA_i$ ).  $BA_i = \pi \times 10^{-4} \times (dbh_i/2)^2$ , where tree dbh in cm and  $BA_i$  in  $m^2$ . This formula is simplified as:  $BA_i = 0.00007854 \times (dbh)^2$ ;
3. Species basal area ( $BA_{sp}$ ) that equals to the sum of all  $BA_i$  of stems of the same plant species within a plot, converted later in hectares (ha);
4. Total basal area (TBA), summing up the all calculated  $BA_{sp}$  within a plot;
5. Species relative dominance (SRD):  $SRD\% = (BA_{sp}/TBA) \times 100$ .

### *Soil chemical and physical analysis*

Soil parameters evaluation was performed according to standard method as follows:

1. Determination of pH ( $H_2O$ ) and content of extractable cations ( $Ca^{2+}$ ,  $K^+$ ,  $NH_4^+$ ) was performed by reading directly the digital display of the pHmeter or spectrophotometer;
2. Determination of organic and total carbon:  $M.org = 1.724 \times C.org$  with  $M.org$  = organic matter (mg / kg);  $C.org$  = organic carbon (mg/kg)
3. Particle size determination by sedimentation using the pipette method. Content of different fractions was determined as follows:

$$C + St\% = [(Pc + s) - (p1) - (Pb)] \times 5000 \times k/Pe \times Fh \quad 1$$

$$C\% = [(Pa) - (P1) - (Pb)] \times 5000 \times k/Pe \times Fh \quad 2$$

$$FSt\% = (C + St)\% - C\% \quad 3$$

$$TSd\% = (Tt - P1) \times 100/Pe \times Fh \quad 4$$

$$CSd\% = (Tc - P1) \times 100/Pe \times Fh \quad 5$$

$$FSd\% = (Tf - P1) \times (100/Pe) \times Fh \quad 6$$

$$CSt\% = TSd \times (CSd + FSd) \times Fh \quad 7$$

With C = clay;  $P_{C+St}$  = T are weight + clay + silt; St = silt; P1 = weight of empty tare (capsule); FSt = fine silt; P2 = Weight of empty tare + white; TSd = total sand;  $Pb = P2 - P1$ ; CSd = coarse sand;  $k = 20N/V$ ; FSd = fine sand; V = volume of the pipette; CSt = coarse silt; Pe = aliquot intake; Tt = cap weight + the total sand;

**Table 2.** Richness of EcM fungi within selected habitat types

Fungi parameters	<i>Isoberlinia</i> Woodland (IW)	Mixed Woodland (MW)	<i>Uapaca</i> Woodland (UW)	Total
Numbers of fruit bodies	1565	513	736	2814
Numbers of species	75	65	56	123
Numbers of genus	21	15	16	23
Numbers of family	9	6	6	9

Fh = humidity factor; Tc = cap weight + coarse sand; Pc = cap weight + clay; Tf = cap weight + fine sand.

The texture of each soil was determined using TRIANGLE, A Program For Soil Textural Classification (Gerakis and Baer, 1999). That texture determination followed percentage of particles within studied soils.

### Gradients effectiveness

Analysis of variance (Anova) test at  $\alpha < 0.05$  was performed to assess the effectiveness of gradient among soil and plant data. It was performed at habitat type level for both variables using package lawstat of R software (Hui et al., 2008). When requirement of distribution and homogeneity of variance were not met, Kruskal-Wallis test (Kruskal and Wallis, 1952) was performed in R software. Afterward, significant gradient (s) underwent a preliminary analysis to check collinearity between them and clarify the ordination. One variable among all highly collinear ones was conserved in the subset of the ordination. That preliminary analysis has been performed with software Statistica 7.1 (StatSoft France, 2006).

### Ectomycorrhizal fungi fruit bodies spatial distribution

To visualize the spatial distribution of EFFB, non-metric multidimensional scaling NMDS ordination was performed based on a matrix of fungi species relative frequency per plot using function *metaMDS* of package Vegan (Oksanen et al., 2015) of R software version 3.3.0 (2016-05-03). Fungi relative frequencies were first transformed by Wisconsin double standardization using function *Wisconsin* to improve ordination. A distance matrix generated by Bray-Curtis dissimilarity index with function *vegdist* was used as input for the NMDS whilst function *metaMDS* used Jaccard index.

Then, main environment variables (host communities and soil parameters) influencing the fungi communities structure were evidenced by fitting them the ordination plot using function *envfit* of the Vegan package. Statistical significance was based on 999 random permutations and plotting was limited to most significant variables with argument *p.max* set at 0.1.

To better visualize the similarity of habitat types, a hierarchical clustering based on Bray-Curtis dissimilarity index was conducted in R software version 3.3.0 (2016-05-03) using function *hclust* and average-linkage. Subsequently, each fungi community was characterized by conducting indicator species analysis using the MULTIPATT function in the R package Indicspecies (De Cáceres and Legendre, 2009; De Cáceres and Jansen, 2015). Indicator Value (IndVal) index (Dufrene and Legendre, 1997) was computed to measure the association between a species and a site group. Statistical significance of association was tested by running 999 random permutations. In addition, the specificity (the so-called IndVal Component A) and the fidelity (second component B of IndVal) of a species as indicator of a target site group were inspected. Component A or specificity refers to “the probability that the surveyed site belongs to the target site group given the fact that the species has been found” whilst component B refers to “the

probability of finding the species in sites belonging to the site group” according to Dufrene and Legendre (1997) and De Cáceres and Legendre (2009). Final, ecological distance between generated site groups was calculated by Jaccard index using the R package Fossil (Vavrek, 2011).

## RESULTS

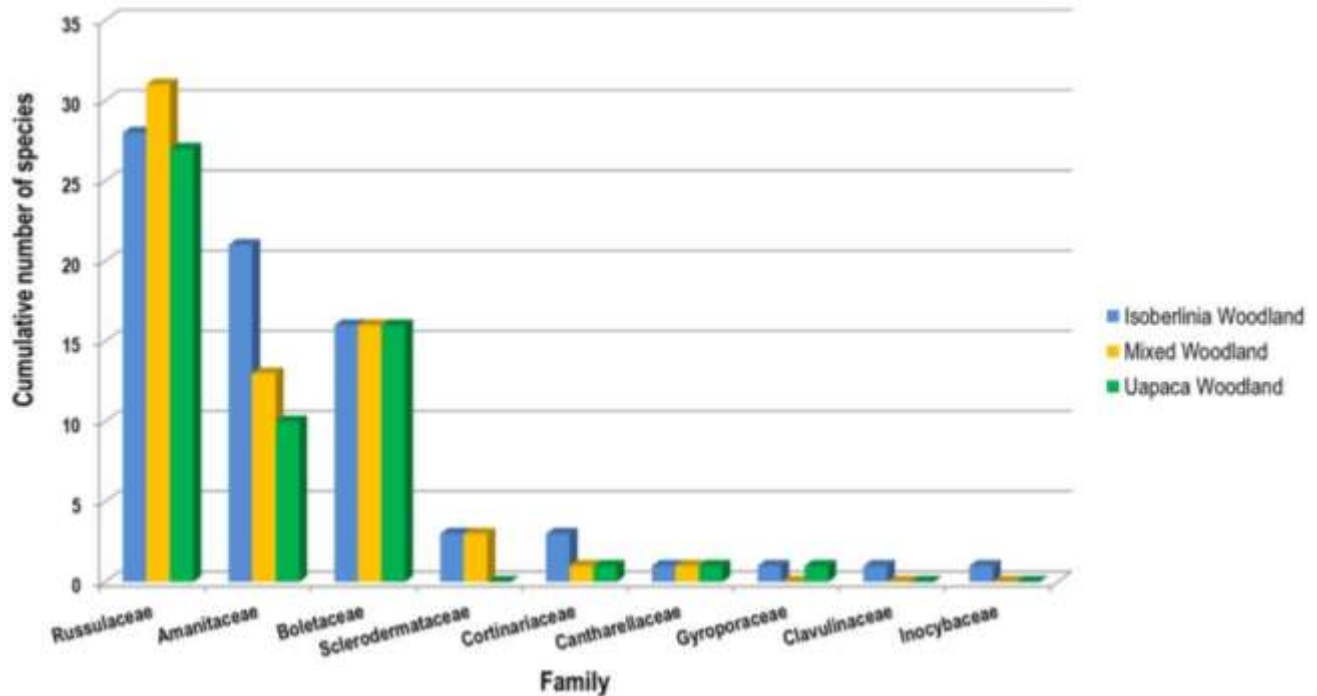
### EcM fungi diversity

#### Observed species richness and diversity indices

EcM fungal fruiting started in mid-May and was still continuing in early October making a cumulative total of 21 weeks of occurrence. In total 2814 fruit bodies have been collected and were sorted into 123 species belonging to 23 genera and 09 families (Table 2). The most frequently recorded family was Russulaceae with 53 species composed of 36 *Russula* species, 11 *Lactifluus* species and 6 *Lactarius* species. The second frequently observed family was Boletaceae represented by 13 genera with a total of 32 species. The Amanitaceae ranked third most important recorded family with a total of 26 species. The less recorded other families included Cantharellaceae, Cortinariaceae, Gyrosporaceae, Inocybaceae, Sclerodermataceae and Clavulinaceae. These families were represented each by only one genus with respectively 1, 3, 1, 1, 5 and 1 species (Figure 2). From the total species richness, 57 taxa (46.34% of the total) were identified up to species level with 19 of them being related to known species from temperate and tropical zones. The remaining 66 species (53.66% of the total) were identified only at the genus level with some of them suspected new to science (Supplementary Table 1).

The most frequent species per habitat type included *Russula congoana* Pat. (13 weeks corresponding to the relative frequency of 2.53%), *Amanita aff. craseoderma* (11 weeks, relative frequency = 2.14%) and *Lactarius tenellus* Verbeken & Walley (10 weeks, relative frequency = 1.95%) in IW; *Amanita annulatovaginata sensu lato* Beeli and *Lactarius tenellus* (both with 8 weeks, relative frequency = 1.56%) in MW; *Cantharellus addaiensis* Henn. and *Amanita aff. subviscosa* Beeli (both with 11 weeks, relative frequency = 2.14%), *Amanita aff. virosa* and *Amanita strobilaceovolvata sensu lato* Beeli (both in 10 weeks, relative frequency = 1.95%) in UW.

22 species were found common to the three habitat



**Figure 2.** Families representativeness per habitat type.

types and represented 17.89% of total observed species richness (Supplementary Table 1). On the other hand, 72 species accounting for 58.53% of the species richness were specific to one habitat type. Many of these specific species were observed and collected only once from May to early October 2014 (Supplementary Table 1) and are unique species. Specific species such *Inocybe* sp 1 and *Cortinarius* subgenus *telamonia* sp 1 have been picked under *Isoberlinia doka* trees in IW. Meanwhile, *Russula annulata* R. Heim, *R. discopus* R. Heim (a rare species) and *Veloporphyrellus africanus* Watling were collected beneath *Uapaca togoensis*. Finally, 29 species (23.58%) were shared by two habitat types. In addition with species common to all habitat types, 38 species were shared by IW and MW (e.g. *Amanita afrospinosa* Pegler & Shah-Smith, *Lactarius saponaceus* Verbeken); 28 species shared by IW and UW (e.g. *Gyroporus castaneus* (Bull.) Quél., *Amanita strobilaceovolvata* sensu lato) and 29 species shared by MW and UW (e.g. *Amanita aff. rubescens* Pers., *Boletus loosii* Heinem).

### Similarity and sampling representativeness

Computed Simpson's Index of Diversity  $1 - D$  of IW was 0.97 with an autosimilarity index calculated to 0.40. Therefore, plots in IW were found non-similar likewise for plots within habitat types MW and UW with respectively 0.33 and 0.29. In those latter habitat types, higher diversity indices were respectively 0.97 and 0.96.

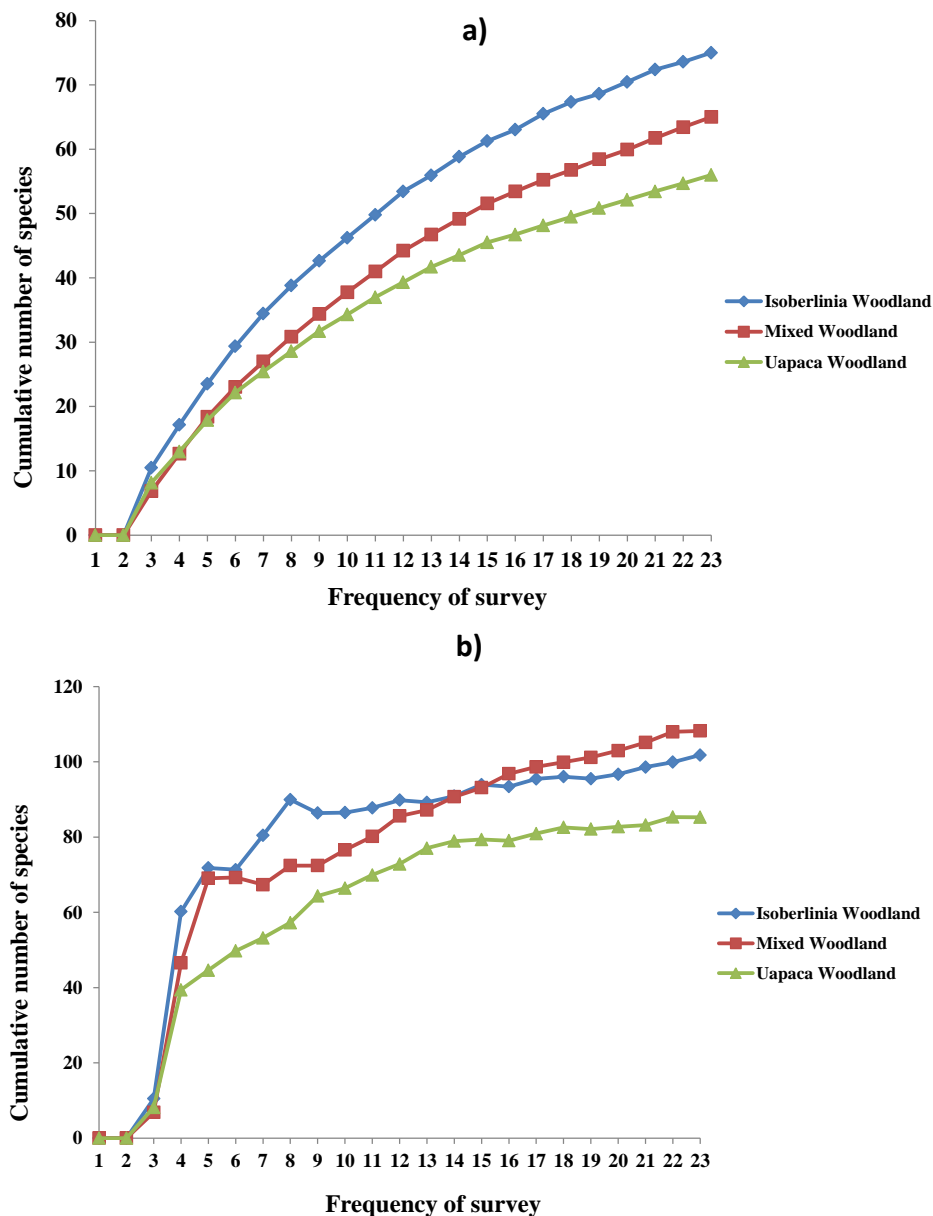
Weekly-based species accumulation curves of the different habitat types have almost the same shape in observed and estimated species richness (Figure 3). Accumulation curves of IW were generally above those of the other habitat types through weeks except for the estimated species richness where curve of MW outdid the other curves from the fourteenth week till the end of the survey. Globally, all curves were ascendant and did not reach an asymptote of total richness.

Sample coverage highlighted the percentage of species detected by our study on the overall estimated species richness. Thus, 75.25% of species was detected in IW against 81.88% in MW and 58.78% in UW (Table 3). Furthermore, 38, 32 and 36 unique species have been collected in the different habitat types respectively.

### Habitats characterisation

#### Floristic and dendrometric parameters

A cumulative number of 822 stems belonging to 49 woody species with  $dbh \geq 10$  cm were detected for all habitat types. Those species belonged at least to 20 families, knowing that 6 species identity was undetermined. 18, 19 and 31 species were inventoried respectively in IW, MW and UW. The total density and basal area of all tree species, and the dendrometric parameters (density and SRD) of cores EcM forest trees in each plots is provided in Table 4.



**Figure 3.** Week-based accumulation curves of observed (a) and estimated (b) species richness of EcM fungi during fruiting season 2014 (mid-May to early-October). Aug. = August, Sept.= September, Oct.= October.

**Table 3.** Sampling representativeness estimators. Sample coverage: proportion of observed species richness ( $S_{obs}$ ) as per cent of estimated species richness ( $S_{est}$ ); Auto-similarity: mean similarity between plots of the same habitat type; Uniques: number of species collected only once during the whole period

Habitat type	Number of fruit bodies	Observed species richness $S_{obs}$	Estimated species richness Chao 2 ( $S_{est}$ )	Sample coverage	Autosimilarity	Simpson's Index of Diversity 1- D	Simpson's Evenness	Uniques
<i>Isoblerlinia</i> Woodland	1542	75	99.67	75.25	0.49	0.77	0.06	38
Mixed Woodland	502	65	79.38	81.88	0.41	0.94	0.25	32
<i>Uapaca</i> Woodland	775	56	95.27	58.78	0.36	0.91	0.19	36

**Table 4.** Mean values of density of woody species, Species relative dominance (SRD) of identified EcM trees and total basal area per habitat.

Plant parameters		<i>Isoberlinia</i> Woodland	Mixed Woodland	<i>Uapaca</i> Woodland
Cumulative number of stems (three plots)		276	246	300
Forest tree species richness SR		18	19	31
Total tree density TD (stem/ha)		3066.66	2733.33	3333.33
Total basal area TBA (m <sup>2</sup> /ha)		179.75	158.43	186.89
Mean canopy cover		66.67	73.33	80
EcM tree partners density(stem/ha)	<i>Isoberlinia doka</i>	171.11	5.56	0.00
	<i>Monotes kerstingii</i>	35.56	18.89	23.33
	<i>Uapaca togoensis</i>	10.00	167.78	153.33
	<i>Isoberlinia doka</i>	62.29	3.68	0.00
EcM tree partners SRD (%)	<i>Monotes kerstingii</i>	10.28	4.13	6.57
	<i>Uapaca togoensis</i>	0.99	53.48	40.50

**Table 5.** Soil chemical and physical parameters variations per habitat type.

Soil parameters	Habitat type			F	Chi-square	Df	p-value
	IW	MW	UW				
pH	6.7 ±0.14	6.52±0.4	6.78±0.2		2.0392	2	0.36
Carbon (%)	1.96±0.09	1.85±0.15	1.71±0.13		4.3922	2	0.11
Nitrogen (%)	0.09±0.05	0.09±0.01	0.12±0.02	0.495		2	0.63
Available Phosphorus (ppm)	1.34±0.32	1.63±0.12	1.20±0.12		3.5862	2	0.17
Calcium (cmol/kg)	1.71±0.42	1.45±0.31	1.07±0.17	3.078		2	0.12
Potassium (cmol/kg)	0.06±0.04	0.09±0.03	0.07±0.03	0.936		2	0.44
Clay (%)	8.67±2.08	10±2.64	9.33±0.58		0.85797	2	0.65
FineSilt (%)	9.33±3.51	5±0.00	8.66±3.05		5.7275	2	0.06
CoarSilt (%)	44.33±12.1	42.66±3.05	45.67±5.86		0.29132	2	0.86
FineSand (%)	34.33±8.14	37±2.64	33.67±3.79		1.1954	2	0.55
Type of soil	Silt loam	Silt loam	Silt loam				

Kruskal-Wallis test demonstrated that plant richness and total basal area did not differ significantly from one another habitat type (chi-squared = 1.55, p-value = 0.46 and chi-squared = 0.62, p-value = 0.73, respectively). Considering EcM tree partners, density and SRD of *I. doka* differed significantly between habitat types (chi-squared = 6.72; p-value = 0.03), IW harboring the highest values. Density and SRD of *U. togoensis* were also significant (F = 20.73, p-value = 0.002 and chi-squared = 5.95, p-value = 0.05 respectively), decreasing from MW to UW and finally IW. At the opposite, the density and SRD of *Monotes kerstingii* does not significantly differ from one another habitat type (chi-squared = 0.62, p-value = 0.73 and chi-squared = 2.51; p-value = 0.28 respectively).

#### Soil chemical and physical parameters

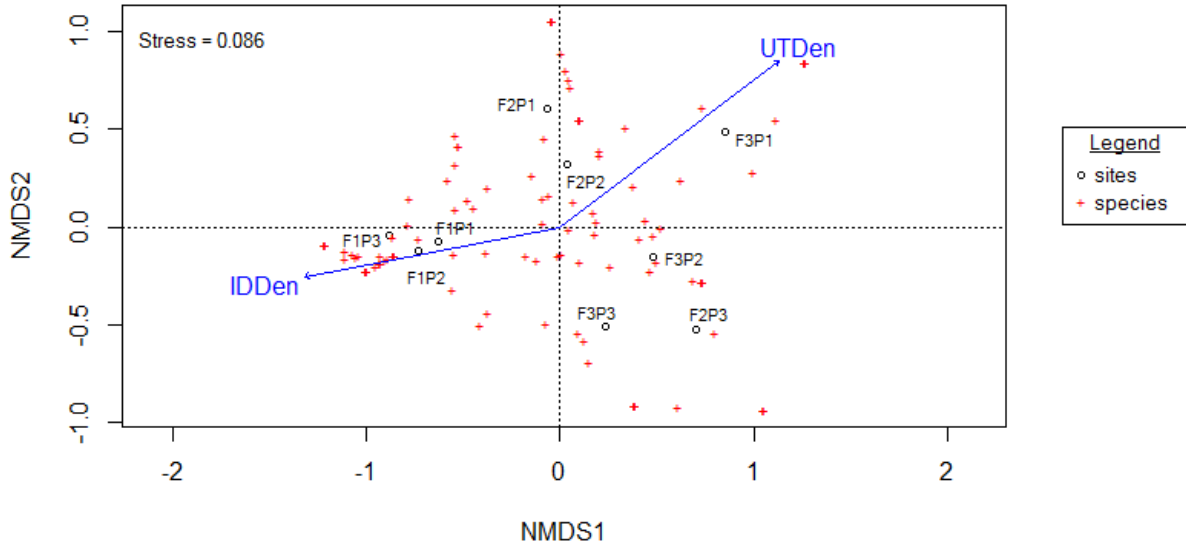
pH (H<sub>2</sub>O) measurement indicated that soils in all plots

were generally neutral, ranging from 6.52 to 6.78. As for texture analysis, soils in plots were generally silt loamy with regard to soil particles size (Table 5). However, differences among both chemical and physical parameters of the different habitat types were not significant at 0.05, pointing out an absence of soil gradient.

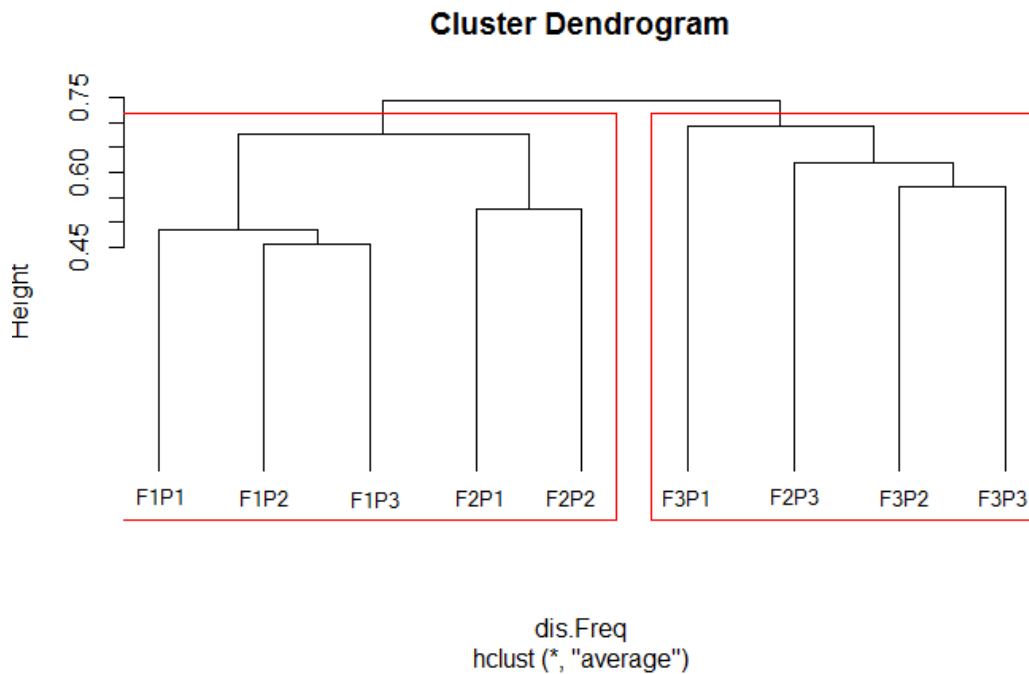
#### Ectomycorrhizal fungi fruit bodies spatial distribution

In absence of soil gradient between habitat types, soil variables were excluded from initial environmental matrix that was finally reduced to 05 plant variables after multicollinearity test. Those variables were plant species richness (PlantSp), total basal area (TBA), *I. doka* density (IDDen), *M. kerstingii* Density (MKDen) and *U. togoensis* Density (UTDen).

Environment variables fitting into NMDS result indicated that *I. doka* Density (IDDen) and *U. togoensis*



**Figure 4.** EcM fungi distribution at Comoé National Park according to stem density of *Uapaca togoensis* and *Isoberlinia doka*.



**Figure 5.** Hierarchical clustering of permanent plots based of dissimilarity.

density (UTDen) are the main statistically significant variables driving the EFFB spatial distribution (Figure 4). UTDen was positively correlated with both axes ( $r^2 = 0.92$ ;  $p$ -value = 0.002) whilst IDDen was negatively correlated to the first axis only ( $r^2 = 0.83$ ;  $p$ -value = 0.018).

Hierarchical analysis of study sites evidenced two sites groups (Figure 5). The first group (G1) encompassed all

plots of habitat 1 (*Isoberlinia* woodland IW) and the two first plots of the second habitat, Mixed woodland (MW). The second group is composed of the remaining plot of habitat 2 (MW) and all plots of the third habitat *Uapaca* Woodland (UW). The indicator species analysis showed that 04 species were significantly associated to just one group on a total of 123 species. 03 species were associated to G1 and 01 species to G2 (Table 6).



**Table 6.** List of indicator species associated to each site group.

Site group	Component A	Component B	Stat	p.value
Group 1 #sps. 3				
RusCon	0.9573	1.0000	0.978	0.013 *
Pulve1	0.9057	1.0000	0.952	0.028 *
AmaXa	0.8276	1.0000	0.910	0.040 *
Group 2 #sps. 1				
AcfVir	1	1	1	0.013 *

Significance codes: 0 '\*\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05 '.' 0.1 ' ' 1 ' ' RusCon: *Russula congoana*; Pulve1: *Pulveroboletus* sp 1; AmaXa: *Amanita xanthogala*; AcfVir: *A. cf virosa*

## DISCUSSION

### EFFB richness

Mycological monitoring within Comoé National Park (CNP) shed light on very specious habitats where almost all known EFFB families were represented. As already mentioned in various paleo and neotropical regions (Sanon et al., 1997; Riviere et al., 2007; Bâ et al., 2012; Henkel et al., 2012; Onguene and Kuyper, 2012), dominance of Russulaceae and specifically of genus *Russula* was also observed. Among the other frequently recruited families in tropical regions, Cantharellaceae was represented, in prospected habitats, by only one species member of genus *Cantharellus*, *C. addaiensis*. In the contrary, four *Cantharellus* species (*C. floridulus* Heinem., *C. platyphyllus* Heinem., *C. cf. platyphyllus* Heinem. and *Cantharellus* sp.) were reported in traditional systems of fallows dominated by many confirmed EcM tree partners near the city of Korhogo North western part of Côte d'Ivoire (Ducouso et al., 1999). This difference may be due to higher number of tree partners in that area, namely *Afzelia Africana* Sm. ex Pers., *Anthonotha crassifolia* (Baill.) J. Léonard, *Berlinia grandiflora* (Vahl) Hutch. & Dalziel, *I. doka* and *U. togoensis*. Likewise in genus *Clavulina*, only one species was detected in *Isoberlinia* woodland (IW) suggesting that other species may have been overlooked or mistook for saprotrophic species. Few species belonging to genera *Inocybe* and *Cortinarius* were also found in CNP. This supports the trend observed in other tropical regions (Onguene and Kuyper, 2002; Riviere et al., 2007; Onguene and Kuyper, 2012), and strengthens the idea that those species might be adapted to temperate and boreal zones (Buyck et al., 1996). However, the paucity of studies in tropical woodlands and forests comparative to temperate and boreal ones should be considered. Yet, the abundance of EcM fungi species was highlighted at continental level. In West Africa, Sanon et al (1997) found 37 EcM fungi during rainy season 1994 and 1995 in savanna and open riparian forests in southwestern

Burkina Faso. 126 EcM species were censured after various surveys in different areas of Benin, ranging from protected areas to farms (Yorou, 2010). In Southern Guinea rainforests, Diédhiou et al. (2010) identified 39 EcM fungal taxa. In central Africa, Onguene et al (2012) reported the collect of 100 EcM fungi in forest habitats of South Cameroon during a three-year survey. Numerous species have been also collected in Congo and are documented in two series, "Flore Iconographique des Champignons du Congo" and "Flore illustrée des Champignons d'Afrique Centrale". Highest species richness and number of EFFB were found in IW. According to Nara et al., 2003, such values reflected host development stage. Indeed, highest cumulative values of tree partners' stems density and basal area were found in plots of IW. Some of those tree species were estimated aging more than 200 years with regard to their dbh (Tederloo, personal communication). In disturbed areas of tropical zones, EcM Fabaceae and Dipterocarpaceae stands (*I. doka* and *M. kerstingii* respectively in our case) are considered climax stands which establishment is facilitated by *Uapaca* spp. (Lawton, 1978; Högborg and Pearce, 1986; Onguene, 2000; McGuire, 2007; Tederloo et al., 2011; Onguene and Kuyper, 2012). According to Poilecot et al. (1991), CNP is included of 93.3 % of fire climax vegetation from which 6.7 % is made of woodlands. Indeed, understorey vegetation in IW and MW were burned either totally or partially according to plot by the annual fire that passed in December 2013, four months before our arrival at the park. However, no plot in UW was burnt. Moreover, EcM fungi species belonging to genus *Scleroderma* previously described as characteristic of disturbed and elevated soil temperature areas (Ingleby et al., 1985; Nara et al., 2003) were collected within burnt plots of IW and MW. Three of the five *Scleroderma* species were recruited in IW and the latter two in MW. Consequently, IW is likely older than the others whilst UW is the youngest and MW at an intermediate stage. This assumption is strengthened by the different proportions of *U. togoensis* and presence/absence of *I. doka* in the different habitats. First, IW harboured many stems of the EcM tree partners *Monotes kerstingii* Gilg and *Uapaca togoensis* but it is dominated by *Isoberlinia doka*. Second, few stems of *I. doka* were censured in MW whilst the tree species is completely absent from UW plots. Another support of that assumption is the presence of *Inocybe* sp. and the number of species of genus *Cortinarius* in IW are other supports of that assumption since those EcM fungi were depicted late successional symbionts (Nara et al., 2003).

### Sampling representativeness

Sampling representativeness assessment demonstrated that a large number of symbiotic fungi were not detected in the different habitats monitored. This result is

corroborated by the important values of unique species that reflected rare species. That number of observed rare species give an estimate of the number of unseen species (Chiarucci et al., 2011) as captured by the estimated species richness in each habitat. That result is a support of the limitation of fruit body based study of EcM fungi species (Horton and Bruns, 2001; Taylor, 2002). Nevertheless, climate impact is more appreciable on fruit bodies than on below-ground tips (Andrew and Lilleskov, 2009; Pickles et al., 2012).

### Spatial distribution of symbiotic fungi

Phytosociological study of permanent plots evidenced important floristic richness and especially numerous stems with dbh above 10 cm. EcM tree partners thrive in dominant and sometimes almost mono-dominant stands. Such habitats have been demonstrated as niche for abundant EcM fungi. *I. doka* and *U. togoensis* were the main dominant species in prospected habitats. Sites grouping were correlated with their density more than stands age. Indeed, though only stems with dbh above 10 cm were considered in data analysis, numerous juveniles and sprouts were present within plots. This was favorable to the establishment of both early- and late-successional EcM fungi. In addition, the grouping also reflected fire impact within study sites evidencing the “drought-tolerant” capacity of some collected fungi species. There is therefore an urgent need to monitor such disturbed stands to adequately address that assumed capacity.

Indicator species analysis evidenced four species associated to site groups (three species associated with G1 and one species with G2). Those species, *Russula congoana*, *Pulveroboletus* sp 1 and *A. xanthogala* were good indicators of G1 and *A. cf virosa* was for G2 taking into account specificity and fidelity. Indeed, those species were collected either exclusively in plots assigned to each group or predominantly in them. Association of *R. congoana* and *A. xanthogala* to *I. doka* was also documented in Benin by De Kesel et al. (2002). Those species are mentioned in literature as edible fungi in various part of Africa (Boa, 2004). As for the two remaining indicators species, they need to be characterised and compared to available monographs and / or keys to ascertain their identity at species level. However, they are likely associated to *U. togoensis*.

### Conclusion

A six-month monitoring of EFFB ascertained their occurrence at Comoé National Reserve in the Sudanian climatic zone of Côte d'Ivoire. Woodlands of the reserve harboured high plant species diversity from which known EcM tree partners were frequently dominant. In these habitat types estimated of more than 200 years old, 123

EFFB species fruited and were collected. Their abundance and spatial distribution were significantly correlated to the stem density of *U. togoensis* and *I. doka* that were respectively the dominant species in each site group. *R. congoana*, *Pulveroboletus* sp 1 and *A. xanthogala* were good indicators of site group G1 and *A. cf virosa* for G2. However, further studies on contrasting soil types, fungal and forest succession, site microclimate as well as fire impact are needed to improve the understanding of fungal community dynamics in West African woodlands.

### Conflict of Interests

The authors have not declared any conflict of interests.

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## SUPPLEMENTARY DATA

**Table 1.** Relative frequency of occurrence of EcM fungi species within woodlands of Comoé National Park.

Distribution class	Taxon	Family	IW	MW	UW	
Common to all habitat types	<i>Amanita aff. subviscosa</i>	Amanitaceae	1.56	0.58	2.14	
	<i>Amanita annulatovaginata sensu lato</i>	Amanitaceae	0.58	1.56	0.39	
	<i>Amanita sp 13</i>	Amanitaceae	1.17	0.78	0.78	
	<i>Amanita xanthogala</i>	Amanitaceae	0.39	0.19	0.19	
	<i>Cantharellus addaiensis</i>	Cantharellaceae	1.75	0.97	2.14	
	<i>Cortinarius sp 1</i>	Cortinariaceae	0.19	0.19	0.39	
	<i>Lactifluus aff. emergens</i>	Russulaceae	0.78	0.19	0.39	
	<i>Lactifluus luteopus</i>	Russulaceae	0.19	0.78	1.56	
	<i>Phylloporus ampliporus</i>	Boletaceae	0.39	0.58	0.19	
	<i>Pulveroboletus sp 1</i>	Boletaceae	1.17	0.39	0.19	
	<i>Pulveroboletus sp 2</i>	Boletaceae	0.58	0.78	1.36	
	<i>Russula aff. cellulata</i>	Russulaceae	0.39	0.19	0.58	
	<i>Russula aff. ochrocephala</i>	Russulaceae	0.58	0.78	0.97	
	<i>Russula cellulata</i>	Russulaceae	0.78	0.58	0.39	
	<i>Russula cf amoenolens</i>	Russulaceae	0.78	0.39	0.58	
	<i>Russula cf flavobrunnea</i>	Russulaceae	0.97	0.39	0.58	
	<i>Russula cf grisea</i>	Russulaceae	0.19	0.19	0.19	
	<i>Russula ciliata</i>	Russulaceae	0.19	0.39	0.19	
	<i>Russula congoana</i>	Russulaceae	2.53	0.78	0.19	
	<i>Russula sp 10</i>	Russulaceae	0.58	0.19	0.78	
	<i>Russula sp 11</i>	Russulaceae	0.19	0.19	0.39	
	<i>Xerocomus sp 4</i>	Boletaceae	0.19	0.19	0.19	
	Shared by two habitat types	<i>Amanita congolensis</i>	Amanitaceae	0.00	0.78	0.97
		<i>Amanita sp 12</i>	Amanitaceae	0.19	0.39	0.00
		<i>Amanita aff. virosa</i>	Amanitaceae	0.00	1.36	1.95
<i>Amanita masasiensis</i>		Amanitaceae	0.19	0.19	0.00	
<i>Amanita sect. lepidella sp 1</i>		Amanitaceae	0.39	0.19	0.00	
<i>Amanita sect. lepidella strips xanthogala sp 1</i>		Amanitaceae	0.19	0.00	0.19	
<i>Amanita sp 5</i>		Amanitaceae	0.19	0.19	0.00	
<i>Amanita sp 8</i>		Amanitaceae	0.19	0.00	0.39	
<i>Amanita strobilaceo-volvata sensu lato</i>		Amanitaceae	0.58	0.00	1.95	
<i>Boletus loosii</i>		Boletaceae	0.00	0.97	0.78	
<i>Boletus sp 2</i>		Boletaceae	0.39	0.00	0.58	
<i>Gyroporus castaneus</i>		Gyroporaceae	0.19	0.00	0.58	
<i>Lactarius afroscrobiculatus</i>		Russulaceae	0.00	0.19	0.39	
<i>Lactarius saponaceus</i>		Russulaceae	0.19	0.19	0.00	
<i>Lactarius tenellus</i>		Russulaceae	1.95	1.56	0.00	
<i>Lactifluus aff. heimii</i>		Russulaceae	0.97	0.19	0.00	
<i>Lactifluus sp 4</i>		Russulaceae	0.19	0.19	0.00	
<i>Octaviana ivoryana</i>		Boletaceae	1.17	0.19	0.00	
<i>Rubinoboletus cf balloui</i>		Boletaceae	0.00	1.17	0.19	
<i>Rubinoboletus cf griseus</i>		Boletaceae	0.39	0.39	0.00	
<i>Russula cf sesenagula</i>		Russulaceae	0.58	0.39	0.00	
<i>Russula sect. griseineae</i>		Russulaceae	0.00	0.19	0.19	
<i>Russula sect. archaeina</i>		Russulaceae	0.39	0.19	0.00	
<i>Russula sp 7</i>		Russulaceae	0.19	0.19	0.00	
<i>Scleroderma sp 2</i>		Sclerodermataceae	0.58	0.19	0.00	
<i>Sutorius sp 1</i>	Boletaceae	0.39	0.00	0.39		

Table 1. Contd.

	<i>Tylopilus</i> sp 1	Boletaceae	0.00	0.19	0.39
	<i>Xerocomus subspinulosus</i>	Boletaceae	0.39	0.39	0.00
	<i>Amanita</i> aff. <i>craseoderma</i>	Amanitaceae	2.14	0.00	0.00
	<i>Amanita</i> cf <i>crassiconus</i>	Amanitaceae	0.00	0.97	0.00
	<i>Amanita</i> sp 1	Amanitaceae	0.19	0.00	0.00
	<i>Amanita</i> sp 2	Amanitaceae	0.00	0.19	0.00
	<i>Amanita</i> sp 3	Amanitaceae	0.19	0.00	0.00
	<i>Amanita</i> sp 4	Amanitaceae	0.00	0.00	0.19
	<i>Amanita</i> sp 6	Amanitaceae	0.19	0.00	0.00
	<i>Amanita</i> sp 7	Amanitaceae	1.75	0.00	0.00
	<i>Amanita</i> sp 9	Amanitaceae	0.19	0.00	0.00
	<i>Amanita</i> sp 10	Amanitaceae	0.19	0.00	0.00
	<i>Amanita</i> sp 11	Amanitaceae	0.19	0.00	0.00
	<i>Amanita subviscosa</i>	Amanitaceae	0.39	0.00	0.00
	<i>Boletellus linderi</i>	Boletaceae	0.97	0.00	0.00
	<i>Boletellus longipes</i>	Boletaceae	0.00	0.58	0.00
	<i>Boletus pallidisimus</i>	Boletaceae	0.00	0.19	0.00
	<i>Boletus</i> sp 1	Boletaceae	0.00	0.00	0.19
	<i>Clavunila</i> sp 1	Clavulinaceae	0.19	0.00	0.00
	<i>Cortinarius</i> aff <i>violaceus</i>	Cortinariaceae	0.39	0.00	0.00
	<i>Cortinarius</i> subgenus <i>telamonia</i> sp 1	Cortinariaceae	0.58	0.00	0.00
	<i>Inocybe</i> sp 1	Inocybaceae	0.58	0.00	0.00
	<i>Lactarius</i> sp 1	Russulaceae	0.00	0.00	0.19
	<i>Lactarius</i> sp 2	Russulaceae	0.00	0.00	0.19
Specific to one habitat type	<i>Lactarius</i> sp 3	Russulaceae	0.00	0.39	0.00
	<i>Lactifluus flammans</i>	Russulaceae	0.00	0.39	0.00
	<i>Lactifluus gymnocarpoides</i>	Russulaceae	0.00	0.00	0.19
	<i>Lactifluus pelliculatus</i>	Russulaceae	0.00	0.00	0.97
	<i>Lactifluus</i> sp 1	Russulaceae	0.00	0.19	0.00
	<i>Lactifluus</i> sp 2	Russulaceae	0.00	0.00	0.19
	<i>Lactifluus</i> sp 3	Russulaceae	0.19	0.00	0.00
	<i>Lactifluus volemoides</i>	Russulaceae	0.00	0.39	0.00
	<i>Phylloporus</i> cf <i>rhodophaeus</i>	Boletaceae	0.58	0.00	0.00
	<i>Porphyrellus</i> sp 1	Boletaceae	0.19	0.00	0.00
	<i>Pulveroboletus</i> sp 3	Boletaceae	0.00	0.00	0.58
	<i>Russula</i> aff. <i>flavobrunnea</i>	Russulaceae	0.19	0.00	0.00
	<i>russula</i> cf <i>annulata</i>	Russulaceae	0.00	0.00	0.39
	<i>Russula</i> cf <i>mairei</i>	Russulaceae	0.00	0.39	0.00
	<i>Russula</i> cf <i>ochrocephala</i>	Russulaceae	0.39	0.00	0.00
	<i>Russula</i> cf <i>subfistulosa</i>	Russulaceae	0.00	0.00	0.39
	<i>Russula discopus</i>	Russulaceae	0.00	0.00	0.19
	<i>Russula oleifera</i>	Russulaceae	0.00	0.19	0.00
	<i>Russula</i> sp 1	Russulaceae	0.00	0.19	0.00
	<i>Russula</i> sp 2	Russulaceae	0.00	0.19	0.00
	<i>Russula</i> sp 3	Russulaceae	0.19	0.00	0.00
	<i>Russula</i> sp 4	Russulaceae	0.39	0.00	0.00
	<i>Russula</i> sp 5	Russulaceae	0.00	0.19	0.00
	<i>Russula</i> sp 6	Russulaceae	0.00	0.00	0.19
	<i>Russula</i> sp 8	Russulaceae	0.00	0.00	0.19
	<i>Russula</i> sp 9	Russulaceae	0.19	0.00	0.00
	<i>Russula</i> sp 12	Russulaceae	0.19	0.00	0.00

Table 1. Contd.

<i>Russula</i> sp 13	Russulaceae	0.00	0.19	0.00
<i>Russula</i> sp 14	Russulaceae	0.00	0.00	0.19
<i>Russula</i> sp 15	Russulaceae	0.00	0.00	0.19
<i>Russula</i> sp 16	Russulaceae	0.58	0.00	0.00
<i>Russula</i> sp 17	Russulaceae	0.19	0.00	0.00
<i>Russula</i> sp 18	Russulaceae	0.00	0.00	0.19
<i>Scleroderma</i> cf <i>cepa</i>	Sclerodermataceae	0.58	0.00	0.00
<i>Scleroderma</i> cf <i>citrinum</i>	Sclerodermataceae	0.00	0.19	0.00
<i>Scleroderma</i> sp 1	Sclerodermataceae	0.00	0.58	0.00
<i>Scleroderma</i> aff. <i>verrucosum</i>	Sclerodermataceae	0.19	0.00	0.00
<i>Tubosaeta heterosetosa</i>	Boletaceae	0.39	0.00	0.00
<i>Tylopilus griseus</i>	Boletaceae	0.00	0.00	0.19
<i>Tylopilus niger</i>	Boletaceae	0.39	0.00	0.00
Boletoid sp 1	Boletaceae	0.00	0.00	0.19
<i>Tylopilus</i> sp 2	Boletaceae	0.00	0.58	0.00
<i>Tylopilus</i> sect. <i>chromapes</i> sp 1	Boletaceae	0.00	0.00	0.19
<i>Veloporphyrellus africanus</i>	Boletaceae	0.00	0.00	0.97
<i>Xerocomus</i> sp 1	Boletaceae	0.00	0.19	0.00
<i>Xerocomus</i> sp 2	Boletaceae	0.00	0.00	0.19
<i>Xerocomus</i> sp 3	Boletaceae	0.19	0.00	0.00
<i>Xerocomus</i> sp 5	Boletaceae	0.00	0.19	0.00
<i>Xerocomus</i> sp 6	Boletaceae	0.78	0.00	0.00
<i>Xerocomus</i> sp 7	Boletaceae	0.00	0.19	0.00

IW: Isoberlinia Woodlands; MW: mixed woodland; UW: Uapaca Woodlands.



Figure 1a, b. *Russula congoana*.



Figure 2a, b. *Amanita xanthogala*.



Figure 3a, b. *Pulveroboletus* sp 1.



Figure 4a, b. *Amanita* cf. *virosa*.



*Full Length Research Paper*

# The attitudes and practices of local people towards wildlife in Chebera Churchura national park, Ethiopia

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**Human activities that affect wildlife and their habitats are pervasive and increasing. Understanding the effects of humans on wildlife populations, as well as devising strategies to ameliorate these effects, is an increasing challenge for resource managers. Commitment of local communities to protected areas is also essential for conserving biodiversity, but little is known about local people attitudes toward biodiversity conservation. Therefore, this paper provides an empirical assessment of local people activities and their attitudes that affect wildlife and their habitats around Chebera Churchura National Park, Ethiopia from 2012 to 2014. Nine villages around the park were selected for this study. A total of 354 households were selected randomly for interview. A semi-structured questionnaire survey, focus group discussions and direct field observations were carried out in the nine selected villages. Among the various human activities recorded, firewood collection, bushfires setting fire, hunting, livestock grazing and farming were having great impacts on biodiversity conservation in the Park. Among the respondents, 51.2% reportedly used the park for livestock grazing, 50.2% for firewood and fodder collection, 15.6% for wild honey and spices collection, 23.1% for timber, 2.6% for wild meat and 2% for farming in and along the boundaries of the Park. Most respondents had positive attitudes towards the conservation of wildlife. A combined strategy aimed at improving local participation in wildlife conservation initiatives, initiation of public education and awareness campaigns and provision of alternative sources of income for the local people will reduce the threat, and contribute to improve conservation of wildlife in Chebera Churchura National Park.**

**Key words:** Human activities, resource use, wildlife conservation.

## INTRODUCTION

Throughout history, human factors have been major drivers of biodiversity loss (Hackel, 1999). Ninety-nine percent of the IUCN Red List species are threatened by these factors (IUCN, 2003). Biodiversity loss is more

pronounced in developing countries, which are more dependent on natural resources as their primary source of income (Wilfred, 2010). In developing countries, pressure on natural resources is growing in tandem

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with rapidly growing human populations (Oladeji et al., 2012). Wildlife species, which are important to humans, decline or disappear as wildlife habitat is cleared for anthropogenic activities (Oladeji et al., 2012). Habitat loss and fragmentation affect the survival of wildlife in various ways including influencing the behavior, abundance, distribution of animals, as well as reducing the extent of usable habitats and degrading habitat quality (Masanja, 2014). In developing nations, firewood remains the major source of energy for cooking, heating and lighting. Until 2010, around 2.8 billion people mainly in developing nations relied on traditional use of biomass for cooking and heating (Bonjour et al., 2013). The over utilization of wood products by rural human communities aggravates the degradation of the habitats of wildlife, and is the major threat to protected areas in developing countries today (Masanja, 2014). Wildfire is a common phenomenon across the African continent (Archibald et al., 2012). Humans are regarded as the main source of wildfire globally, accounting for 59 to 95% of ignitions (FAO, 2007). Studies from east, west, and southern Africa reported that burning protected areas was mostly for livelihood-related, notably range management, thatch production, predator-cover reduction, gathering, hunting and agricultural production (Gandiwa et al., 2014). Agriculture remains a predominant livelihood activity in most parts of Africa (Coad, 2007; Gundogdu, 2011). In Ethiopia, expansion of agricultural practices, settlement and increasing pressure of human and livestock populations are major threats in several protected areas (Tadesse and Kotler, 2013). Local communities' perceptions of protected areas influence the kinds of interactions people have with them, and thereby conservation effectiveness (Ramakrishnan, 2007). Their perceptions of protected areas management play also an important role in their attitudes toward them (Anthony, 2007). Therefore, understanding residents' perceptions about conservation is the key to improve the protected areas-people relationship if protected areas are to achieve their goals (Weladji et al., 2003). Many factors influence the perceptions of the protected areas held by residents living in their periphery. These include the history of park management, the degree of awareness of protected areas existence, the education level, the reference to future generation (Bauer, 2003) and the gender and ethnicity (Mehta and Heinen, 2001). The understanding of all these factors is important to improve the relationship between local residents and protected areas and will improve people awareness about biodiversity conservation within these areas. Therefore, collecting baseline information on various human activities and their attitudes is a vital step in managing the impact of human activities on biodiversity conservation. Chebera Churchura National Park (CCNP) in Ethiopia is a conservation area where the impact of human activities on conservation of wildlife has not been studied as is the case in many other parts of Ethiopia. Effective conser-

vation measures cannot be achieved successfully without clear information about the impact of human activities on conservation of wildlife. Therefore, this study aimed to investigate the potential impact of various human activities and their attitudes on conservation of wildlife in the CCNP, and to obtain useful information to enable recommendations to be made regarding better management of the Park.

## MATERIALS AND METHODS

### Study area

Chebera Churchura National Park (6°39'-7°09'N and 36°27'-36°57' E) is located 580 km south- west of Addis Ababa (Figure 1). It covers an area of 1,215 km<sup>2</sup>, and lies within the western side of the Central Omo Gibe Basin. The mean annual rainfall of the area is 2,154 mm. There are two main seasons, the dry season from December to February and the wet season from March to November. Four main rivers and their tributaries drain the area. The area is rich in floral and faunal biodiversity, consisting of 37 species of large mammals, 18 species of small mammals and 137 bird species (Demeke and Afework, 2013).

### Methods

The present study was carried out through a questionnaire survey and focus group discussions (Newmark et al., 1994; Maddox, 2003), to collect primary data among the households in the study area. The questionnaire had both open and close ended questions to get information about anthropogenic activities in the study area. It was also supplemented with field observations of various aspects of resource use, benefits from wildlife and the associated costs. A preliminary survey was conducted in August, 2012 prior to the actual data collection period. This helped to (i) identify the boundaries of the park, (ii) decide the number of villages/sites based on purposive sampling method and (iii) have a general understanding of the overall situations like anthropogenic activities and problems related to crop damage and livestock loss in CCNP. The questionnaire was pre-tested on randomly selected 62 individuals of varying age, sex and social economic activities among the local communities. These individuals were not included in the main sample group. This helped to identify the various anthropogenic activities in the area and to modify the questionnaire accordingly. Nine villages from 25 Peasant Associations were selected based on the information gathered using the preliminary survey: (i) the distance from the Park, (ii) problems related to crop damage and livestock loss, (iii) dependence of local people on the Park and (iv) encroachment within the Park area. A total of 354 households were selected randomly for the interview. The age of participants ranged from 18 to 72 years. The questionnaire was administered to all households, of which 230 (65%) were males and 124 (35%) were females. Interview questions were written in English, but all interviews were translated and conducted in Dawuro local language to reduce misunderstandings during the interviews, due to cultural and language differences, through back-translation of the interview script (Müller, 2007). Eighteen local people, consisting of two residents in each of the 9 study village were recruited and trained to administer the questionnaires.

To ensure accuracy, the same translators were used and the same interview questions. The questionnaire was administered to farmers within their area of farming and/or residence (Hill, 2000), at a random manner based on first come, first-serve basis (Newmark et al., 1994), and alternating male and female respondents as

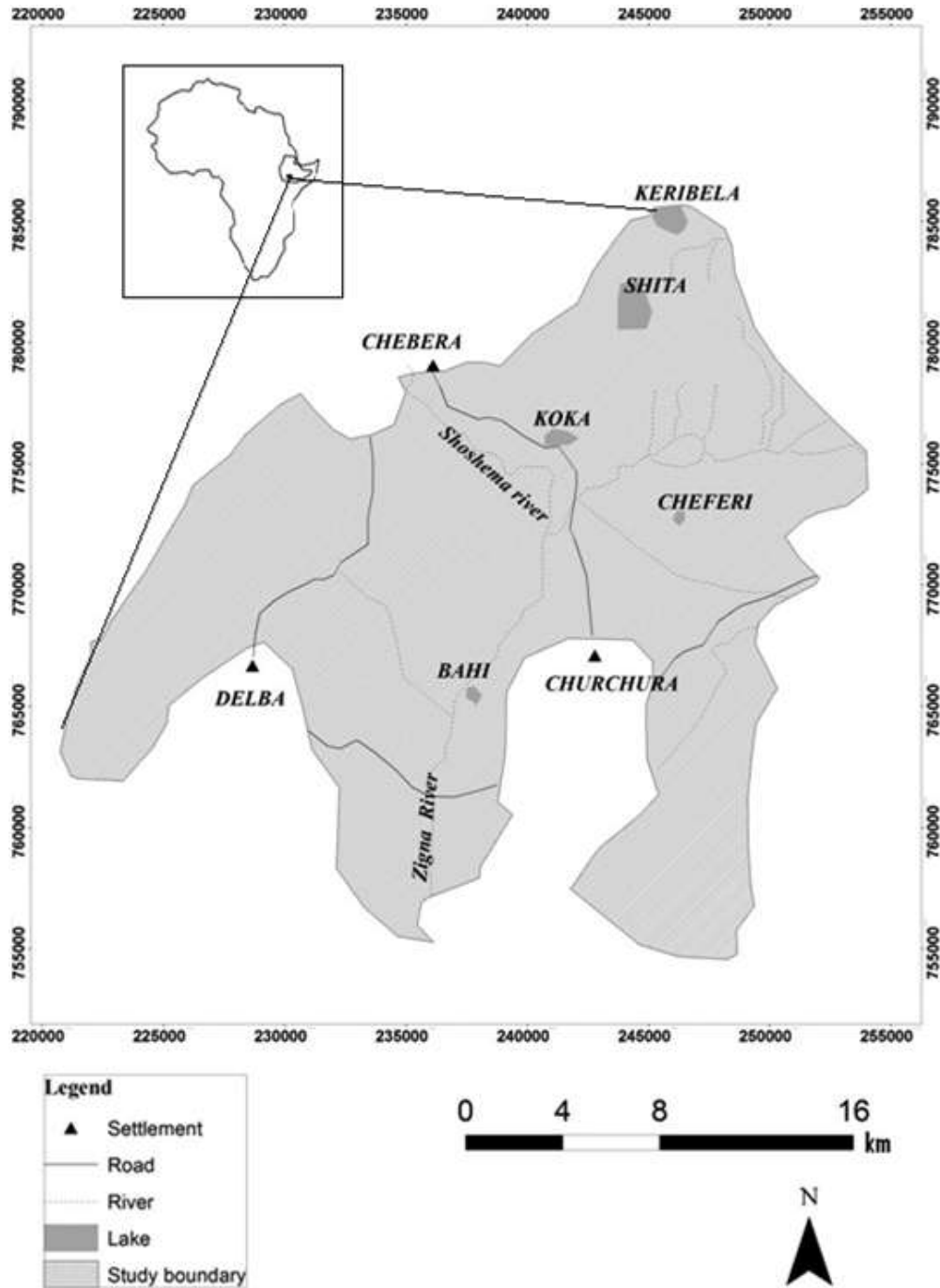


Figure 1. Location map of the study area.

much as possible and different age groups. In some households, the head was interviewed and other people present in a house usually helped in the recall. Respondents answered each attitudes statement according to their strength of agreement by the following attitudes level scores: 1= for strongly agree, 2=agree, 3= neutral, 4= disagree, and 5= strongly disagree (Likert, 1974). The villages covered were Chebera, Serri, Dalba, Yore, Shita, Churchura, Chewda, Gudumu and Adabachew, ranging from 0 to 5 km apart from the boundary of the Park. The questionnaire consisted of a series of semi-structured questions focusing on two main areas of

interest. These include: (i) demographic data and (ii) various anthropogenic activities (grazing, wildfire setting, hunting, firewood gathering, agricultural activities and other socio- economic activities, which could have negative impacts on wildlife in the area). Data were collected using a semi-structured survey design, following a similar format used by Maddox (2003). Data were analyzed using SPSS computer software version 20 (SPSS Inc, IL, U-S-A). Appropriate statistical methods such descriptive statistics and frequency were used to analyse the data. To test for the mean differences in attitude toward conservation of biodiversity among

**Table 1.** Socio-demographic profile of the respondents and their attitude towards CCNP.

Parameter		Attitude towards the conservation area (%)				
Category	Variables	Number of respondents	%	Positive%	Negative%	No idea%
Gender	Male	230	65	63.0	30.5	6.5
	Female	124	35	39.6	41.8	18.6
Age group	≥18	29	8.2	69.7	27.4	2.9
	20–29	55	15.5	63.9	30.2	5.9
	30–39	85	24	60.5	33.4	6.1
	40–49	104	29.4	57.9	35.5	6.6
	50–59	53	15	53.8	39.3	6.9
	>59	28	7.9	50.7	40.4	8.9
Education	Illiterate	194	54.8	44.5	50.1	5.4
	Primary level education	96	27.1	60.2	33.8	6.1
	Secondary level education	28	7.9	71.4	23.3	5.3
	Informal education	36	10.2	45.5	47.1	7.4
Marital status	Married	286	80.8	61.2	33.2	5.6
	Single	20	5.6	70.2	25.3	4.5
	Divorced	37	10.5	45.3	51.2	3.5
	Widowed	11	3.1	39.5	55.6	4.9
Family size	1–3	123	36.8	49.5	45.3	5.2
	4–6	200	60	34.7	59.3	6.0
	7–10	8	2.3	30.2	64.3	5.5
	>10	3	0.9	31.6	65.0	3.4

age groups and gender of the respondents, a non parametric test was conducted on all the test variables. Spearman rho was used to evaluate the correlation among the attitudes of the respondents toward the conservation area.

### Focus group discussion

Focus group discussions method was used to reinforce the data collected through the questionnaire. Group discussions were organized to obtain direct first-hand information through spontaneous responses from the respondent, and the discussions solicited information about local community attitudes of biodiversity conservation. Two focus group discussions sessions were conducted in each of the study village, and the group size in each discussion site varied from 15 to 21. The participants were invited to discuss issues according to their convenience. Park staffs, village leaders, local elders, primary school teacher in the village, other government employers and students have participated to discuss their experience concerned with conservation and to gather their information on wildlife in the area. During such group discussions, the researcher initiated the discussion by stating some of the observations and responses from people interviewed and from questionnaires. Information collected from group discussions were collated and summarized using text analysis method, and presented in a narrative fashion. Thus, the information acquired was

triangulated through questionnaire interviews, focus group discussions and field observations.

## RESULTS

### Socio-demographic characteristics

The socio-demographic characteristics (sex, age-groups, religious groups, occupation and educational background) of the respondents living in the communities surrounding CCNP are presented in Table 1. Out of the 354 respondents, 65% were males and 35.02% females. The youngest respondent was 18 years old, and the oldest 72 years. Majority (68.9%) of the respondents were between 29-49 years old, while 8.1 and 7.9% of the respondents were less than 20 years and older than 59 years old, respectively. Most of the respondents (80.8%) were married, 5.6% were single, 10.5% divorced and 3.1% were widowed. Most of the respondents (54.8%) were illiterate, 10.2% had informal education, 27.1% had primary education, 7.9% had secondary education and

**Table 2.** Percentage of resource utilization by the respondents of different villagers in Chebera Churchura National Park.

Village	n (354)	Respondents (%) involved in different activities						
		CF %	LG %	FW & FC %	WH & SC %	WM %	TC %	OT %
Chebera	48	0.5	51.5	58.5	5.7	2.1	20	0
Sirri	21	4.3	53.4	52.9	21.3	1.2	25.5	0.2
Dalba	44	0.1	50.0	49.5	16.3	1.5	30.3	0.0
Yora	42	2.4	55.7	46.6	12.4	2.1	32.5	0.1
Shita	39	1.4	52.9	48.7	4.5	1.3	27.5	7.3
Churchura	48	3.2	57.5	56.8	26.2	5.6	25.4	1.7
Chewda	34	2.1	45.3	43.6	17.3	2.4	13.5	0.5
Gudumu	48	2.2	48.9	46.9	15.8	5.9	17.8	0.4
Adabacho	30	1.9	45.7	47.9	20.5	1.7	15.3	0.3
Mean		2.0	51.2	50.2	15.6	2.6	23.1	1.2

CF= Crop farming, LG= Livestock grazing access, FW & FC= Firewood gathering, fodder collection and thatching houses, WH & SC= Wild honey and spices collection, WM= Wildmeat access, TC= Timber collection for different purpose, OT= Other benefits.

none had gone beyond secondary level education. There were significant differences in the educational status among the respondents ( $\chi^2 = 98.16$ ,  $df = 3$ ,  $P < 0.05$ ). The majority of respondents (55.4%) had positive attitudes towards the conservation area, while 38.6% had negative attitudes. Most of the better-educated groups (65.8%) had positive attitudes than less-educated respondents (45%), the difference were significant ( $\chi^2 = 27.5$ ,  $df = 3$ ,  $P < 0.05$ ). Most (60%) of the respondents had 4 to 6 family members. On the other hand, 36.8, 2.3 and 0.9% of the respondents had 1 to 3, 7 to 10 and > 10 family members, respectively (Table 1). Respondents with large family size had more negative attitudes towards the conservation area than those with small family size. Mixed farming (crop cultivation and livestock rearing) was the main means of livelihood of most of the respondents (76.8%) in CCNP, and only 12.7% depend on crop farming. Among the respondents, 10.4% claimed to have been involved in one or more secondary occupations. The majority of respondents (51.1%) had a positive attitude towards the conservation area, but on an average 40.5% had negative attitude. The major livestock reared by the local communities are cattle (44.7%), goat (17.1%), sheep (16.4%) and pack animals including donkeys (17.1%), mules (7.6%) and horses (6.9%) in nine villages surrounding the park.

### Resource utilization

Local communities are dependent on a number of natural resources in the Park for their livelihoods (Table 2). Majority of the respondents acknowledged getting benefits from the Park, 51.2% using the conservation area for livestock grazing, 50.2% for firewood and fodder collection, 15.6% for wild honey and spices collection, 23.1% for timber collection and 2% for farming along the

boundaries of the Park.

### Livestock grazing

Majority of the livestock of the respondents (82.3%) grazed inside and around the Park, 39.0% inside the Park and 43.3% in the buffer zone of the Park. Only 17.6% of the local people have own grazing land. The villages differed significantly ( $\chi^2 = 48.34$ ,  $df = 8$ ,  $P < 0.05$ ) regarding livestock grazing in the study area (Table 3).

### Firewood and timber extraction

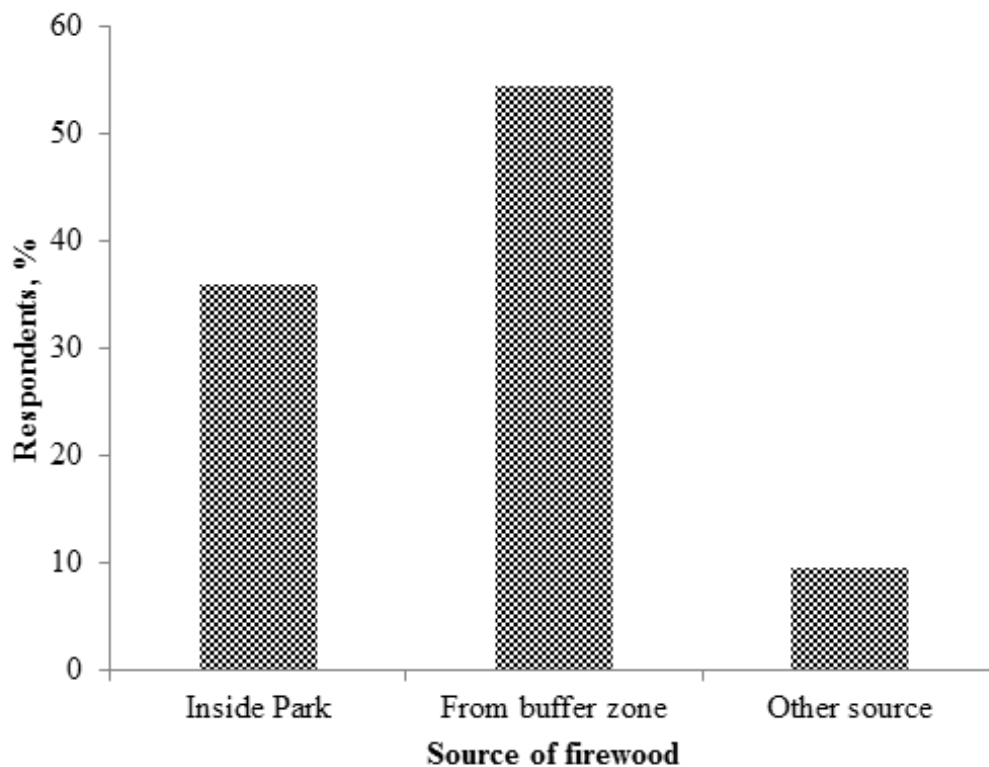
The main source of energy for the people around CCNP was firewood. Among the respondents, 35.9% collected firewood and construction materials from the interior of the park, 54.4% from the buffer zone, and 9.6% from other sources like the farm area (Figure 2).

### Illegal hunting

Majority of the respondents (80.43%) indicated that it was easy to obtain wild meat in the locality, while the remaining 19.57% stated that it was not easy. The difference was significant ( $\chi^2 = 49.85$ ,  $df = 1$ ,  $P < 0.05$ ). Most respondents across the nine study villages reported that illegal hunters commonly used wire snaring (54.8%) and firearms (45.5%). The least reported illegal hunting methods were poisoning (1.24%), hunting with dogs (0.17%) and fire (0.12%) (Table 4). Poisoning, mostly using herbicides and pesticides, was used in retaliation against large carnivores such as spotted hyena and lion as a way to reduce livestock–carnivore conflicts. Wildfire was used to drive animals towards snares and to make

**Table 3.** Grazing sites of villagers in Chebera Churchura National Park.

Village	Livestock grazing (%)				
	n (354)	Distance from the Park (km)	In the Park %	In the buffer zone %	Own grazing land %
Chebera	48	1–2	41.5	52.4	6.51
Sirri	21	0–2	39.3	48.9	11.7
Dalba	44	3–5	40.5	45.8	13.7
Yora	42	0–2	44.4	43.8	11.7
Shita	39	3–5	37.4	38.9	23.6
Churchura	48	1–3	45.6	47.5	6.8
Chewda	34	0–2	36.9	37.7	25.4
Gudumu	48	2–3	32.0	34.6	33.4
Adabacho	30	2–4	33.4	40.4	25.9

**Figure 2.** Different sites of fire wood and timber collection by the respondents.

hunting by dogs easier. A total of 22 wildlife species, including large herbivores and carnivores were reported to be illegally killed in the CCNP ecosystem. Most of the respondents reported that African buffalo (*Syncerus caffer*) (71%), African elephant (*Loxodonta africana*) (45%), common warthog (*Phacochoerus africanus*) (33%), waterbuck (*Kobus ellipsiprymnus*) (25%) and bushbuck (*Tragelaphus scriptus*) (21%) were the most abundant, preferred and commonly hunted animals.

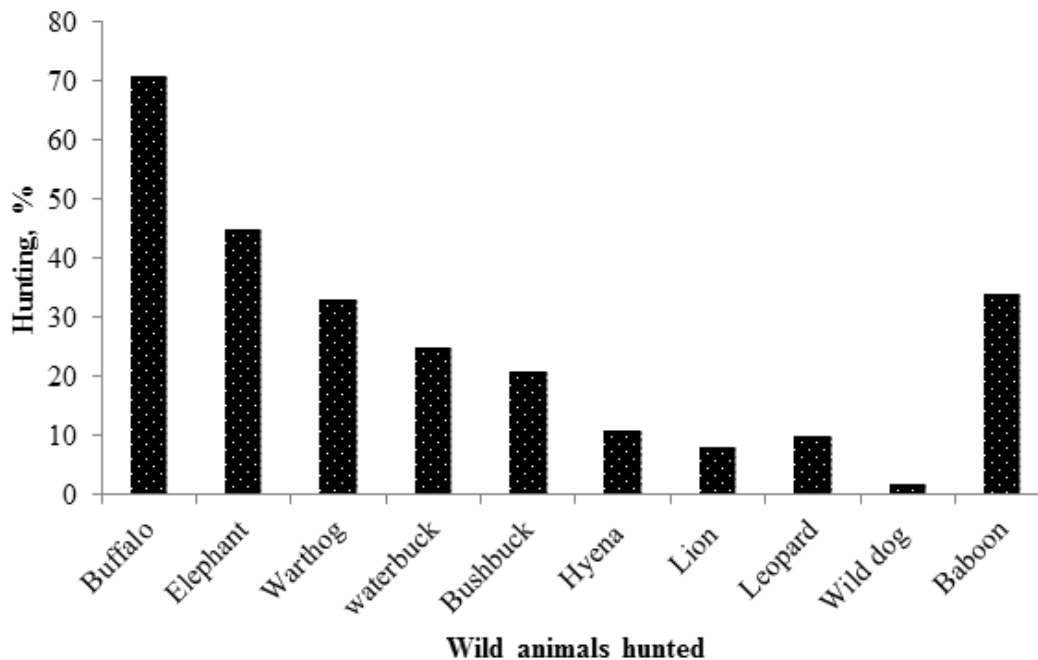
In addition, Anubis baboon (*Papio anubis*) (34%), spotted hyena (*Crocuta crocuta*) (11%), leopard

(*Panthera pardus*) (10%), lion (*Panthera leo*) (8%) and black-backed jackal (*Canis mesomelas*) (2%) were also illegally killed in the CCNP ecosystem (Figure 3). The respondents highlighted five reasons for illegal hunting of wild animals: (i) source of protein for domestic consumption (40.4%), (ii) reduction of crop damage and livestock predation (47.9%), (iii) sale to enhance income (25.5%), (iv) reduction of threats to humans (21.7%), (v) use for traditional/cultural ceremonies (2.0%). The differences were significant ( $\chi^2 = 59.98$ ,  $df = 8$ ,  $P < 0.05$ ). Across the nine study villages, the differences in

**Table 4.** Percentage of illegal hunting methods among different villages in Chebera Churchura National Park area

Villages	Common illegal hunting methods (%)				
	Snares	Firearms	Poisoning	Wildfire	Hunting with dogs
Chebera	69.4	26.4	3.6	0.5	0.3
Sirri	56.9	40.7	2.2	0.2	0.1
Dalba	58.7	47.8	1.7	0.1	0.2
Yora	56.5	48.0	0.2	0.0	0.0
Shita	53.8	45.7	0.4	0.0	0.0
Churchura	39.3	61.5	2.1	0.3	0.3
Chewda	54.7	45.2	0.0	0.0	0.0
Gudumu	55.1	44.5	0.0	0.0	0.4
Adabacho	48.7	49.8	1.0	0.0	0.2
Mean	54.8	45.5	1.24	0.12	0.17

(Total percentage exceeds 100 for each ward because the respondents were allowed to give multiple answers).

**Figure 3.** Percentage of wild animals hunted in Chebera Churchura National Park area.

perceptions of respondents concerning illegal hunting in the CCNP ecosystem during 2012 to 2014 were significant ( $\chi^2 = 51.23$ ,  $df = 8$ ,  $P < 0.05$ ). Of the respondents, 61.6% thought illegal hunting activities had decreased, 20.8% indicate that it had increased, and only 17.5% thought there had been no change. The main reasons given for the perceived decline of illegal hunting in the CCNP ecosystem were: (i) fear of arrest and imprisonment (73.1%), (ii) strengthened law enforcement operations (40.3%), (iii) fear of Park Rangers (22.2%), and (iv) lack of firearms to use in illegal hunting (17.9%).

### Wildfire

Most of the respondents (97.9%) stated that wildfires were of anthropogenic origin, 88.7% of the fires being deliberately set by the local people, largely hunters (83.5%) or farmers (16.5%) to clearing vegetation for cultivation.

Accidental wildfires resulted from collection of wild honey (76.6%) and cooking on farms (23.2%). Most of the farmer-respondents set fires once a year, usually during the dry season (December to February).

### Illegal farming

The local communities surrounding the CCNP had been using the Park area for agricultural purposes even before the establishment of the Park, especially the communities of Chebera, Sirri, Shita, Yora and Churchura villages. Crops farmed included maize (*Zea mays*), teff (*Eragrostis tef*), banana (*Musa acuminata*), mango (*Mangifera indica*), avocado (*Persea americana*), ginger (*Zingiber officinale*) and 'Enset' (*Enset ventricosum*).

### Focus group discussion

The discussants have revealed that activities such as illegal hunting, livestock grazing, wildfire, fuel wood gathering, illegal farming practice, wild honey and green chilli collection were performed by the local people. Most participants agreed that the local communities benefited from the Park resources. Most of them described the shortage of private grazing land and decreased farmland holding due to the Park around them. This could have increased pressure on the Park area resources for livestock grazing and agricultural expansion. They also emphasized additional farmland should be provided as compensation and sharing of resource should be allowed. Some of the discussants noted that previously they used to hunt different wild animals and minimize their threat. However, at present, the negative effects of the animals are on the increase. As a result, some of the discussants were dissatisfied with the existence of CCNP. They considered the Park as a limiting factor in improving their livelihood. Some of them also stated that CCNP has been responsible for their restricted access to resources in the area and further calmed forced relocation. Few discussants considered the Park as useless. They also felt that Park staff members do not like communities around the Park boundaries. But most discussants had positive attitude towards wildlife for its importance to attract tourists, hunting opportunities during drought, enjoyment derived from viewing wildlife and its value for future generation.

## DISCUSSION

### Socio-demographic variables

Many factors such as different economic, legal, social and ecological concerns affect the attitude of local people on conservation issues (Adams and Hulme, 2001). Among the socio-demographic factors examined in the study area, education and age were important predictors of the relationship between local communities and the protected area. Education is one of important factor in understanding the role of protected areas in conservation, and hence influences the attitudes of local people towards

conservation (Mishra et al., 2003). Findings of this investigation also indicated that respondents who were educated had more positive attitude towards conservation than those with less or no education. In the study area, the younger residents tended to have higher educational levels than the older respondents, and this influenced the level of understanding of the importance of wildlife conservation among the educated people (Anthony, 2007; de Boer et al., 2013). Level of education is a major factor in obtaining better employment opportunities and subsequently alternative livelihoods. Local people with higher educational levels participated in agricultural activities as well as other activities like anti-poaching crusades, tour guiding and working in local government organizations. Such activities tended to reduce their dependence on resources from the protected area.

The above are consistent with the assertion by Akama et al. (1995) that the level of education varied inversely with the level of negative attitudes towards the reserve and conservation activities. A low level of awareness regarding conservation issues and protected area management practices with the lack of involvement of the local community in decision making processes might also be an important determinant of negative attitudes of the local people towards the present study area. As a factor, age has a significant influence on the attitudes of local people towards conservation. Younger people have tended to show positive attitudes toward conservation than the elderly, probably due to the fact that younger respondents were more educated than adults. Older respondents felt that the Park would threaten their livelihoods by reducing opportunities farm expansion as well as access to pasture land, fuel-wood and extraction of forest products. Similar results were reported for older residents in around five protected areas in Tanzania (Newmark et al., 1993). Occupations of the respondents also had some effect on their attitudes. Livestock holding was an important predictor of the relationship between local communities and the protected area. Those with higher numbers of livestock tend to have negative attitudes towards the protected area than those with fewer numbers of livestock. People with more cattle are more likely to interact with the protected area through restrictive, prohibitive and punitive laws. They are likely to be arrested and fined if found with livestock in the protected area.

### Livestock grazing

Most of the local people around CCNP were dependent on subsistence agriculture and livestock rearing for their livelihoods. Livestock usually intensely compete with wild animals for the same habitat resources, including forage and water, and this might have strong impacts on wildlife (Masanja, 2014). Cleaveland et al. (2002) indicated that



interactions between livestock and wildlife populations are a key issue in livestock economies worldwide, particularly in east and southern Africa, where many communities live in close contact with wildlife. Most of the respondents considered the Park as their communal pasture area and they did not agree that livestock should be banned from accessing the Park. It was observed that livestock encroached up to 5–10 km inside the Park to graze and access surface water during the dry seasons and this might affect wildlife management practices. Extensive livestock encroachment in the predominantly wildlife grazing zone might lead to their direct or indirect interactions with enhancing chances of disease transmission (Rinderpest, Bovine TB and foot and mouth disease) and competition for forage resources, as well influence wildlife habitat use. For example, the mineral spring water locally known as 'hora', which was used by wildlife, especially African buffaloes and elephants, is largely encroached by expanding livestock. As a result, wild animals avoid use of 'hora' during the day time. The negative impact of livestock activity on range quality of wildlife might reduce the potential for ecotourism by reducing the feasibility of wildlife viewing as indicated by Oladeji et al. (2012). The increase in livestock numbers in the Park also resulted in increases in livestock depredation by large carnivores and created conflict between local people with wildlife and the Park managements.

### Firewood extraction

Firewood was the main source of energy for domestic purposes in the study area. Firewood extraction might have a negative impact on wildlife because trees provide a habitat for a wide range of wildlife (Bonjour et al., 2013). It might reduce feeding grounds and mating sites of wildlife in the Park. Alternative fuel sources such as cattle dung, farm trees and agricultural residue were also used by the local people, and this might help reduce pressure on firewood extraction from the Park. It was observed that in some villages, people travel long distances (about 3 km) and spent more than two hours to collect firewood. The problems of firewood extraction were more acute in the wet season of the year. They also use thatch as roofing material, timber for house construction and furniture and tree fodder as livestock feeds. This dependence on trees in the Park might increase conflict between local people and Park officials and further affect wildlife management practices in the study area.

### Illegal hunting

Illegal hunting is a serious threat to the conservation status of many wildlife species in Africa (Coad, 2007). The CCNP, like many of Africa's protected areas, is also under increasing pressure from hunting. This study has

revealed that illegal hunting was fueled by various factors, including the need for wild meat for household consumption and commercial trade in wild animal products due to few alternative livelihood options and retaliate killings. The local people believed that hunting of destructive wildlife is a good way to scare off crop raiding animals and reduce depredation of their livestock. The link between illegal hunting and human–wildlife conflict reported in this study is consistent with earlier reports from elsewhere in Africa (Coad et al., 2010). For example, in eastern and southern Africa, demand for more land for crop and livestock production has increased antagonism between humans and wildlife, leading to illegal hunting (Wilfred, 2010). A large percentage of respondents admitted hunting crop-raiding animals and expressed great dissatisfaction with the Park authorities for not doing anything to prevent crop raiding and predation of livestock. Focus group discussions also indicated that farmers around CCNP regarded hunting as one of the effective methods to protect crops and livestock from wildlife. Respondents who admitted hunting in the Park had farms located near the Park edge and are therefore more likely to be affected economically by crop raiding and depredation of livestock. Focus group discussions also revealed that animal products commercially traded included ivory from African elephant (*Loxodonta africana*), skins from species such as leopard (*Panthera pardus*), lion (*Panthera leo*) and Colobus monkey. The mane of lions and leopard skin were used to make helmets for male dancers during cultural ceremonies. Skin of large antelopes (e.g. bush-buck (*Tragelaphus scriptus*) and African buffaloes (*Syncerus caffer*) were used to make traditional beds for adults and mats for drying grains, and for making traditional bags for storage and carrying grains. Ivory was also used to make traditional dancing rings worn during the cultural ceremonies.

Results of the present study suggest that illegal hunting is strongly linked to distance of village from the Park boundary. This relationship has also been reported in studies from other part of Africa (Coad et al., 2010), with higher levels of poaching by people living nearer the protected areas than those living farther away. Most of the study villages adjacent to CCNP were within 1 to 3 km of the reserve boundary, and these were the most problematic villages as far as poaching was concerned. Being closer to the reserve might be advantageous because of easy or cost-effective access to wildlife resources. Greater distances mean increased time, effort and costs for hunters to find wildlife and transport meat or other wildlife products to homes or selling points. During the study period, it was observed that large numbers of African elephants were found close to the Park headquarters, possibly for security reasons as reported by Balakrishnan and Ndhlovu (1992). Focus group discussions indicated that wild honey collecting is one cause of illegal hunting. Wild honey collection provided legal reasons for entering the reserve, but managers and

rangers complained that the majority of people use this as a pretext for indulging in illegal activities, like hunting, once inside the Park. Illegal hunters preferred a range of animal species with different body sizes. African buffaloes (*Syncerus caffer*), African elephants (*Loxodonta africana*), waterbuck (*Kobus ellipsiprymnus*), bushbuck (*Tragelaphus scriptus*), bush pig (*Potamochoerus larvatus*), common warthog (*Phacochoerus africanus*), leopard (*Panthera pardus*) and lions (*Panthera leo*) were among the most targeted and preferred species. A variety of illegal hunting methods were used in the CCNP. Common hunting methods included snares, firearms, poisoning and hunting with dogs. Snares were the most commonly reported hunting method in CCNP, probably because they are, easy to use and versatile. Respondents were suggested that with increased law enforcement efforts in the area, illegal hunters were likely to switch to less detectable methods such as snaring, which is a particularly undesirable hunting technique from a conservation perspective. Snare were difficult to locate, making trapping challenging to control, as well as killing non-target species (Lindsey et al., 2012; Martin, et al., 2012). Firearms were most commonly used to hunt large mammals such as African elephants, buffaloes and common warthogs. In this study, crop and livestock losses might derive illegal hunting at a higher level. Poverty might also be a reason for hunting wild animals in the area. As indicated by the respondents, wild meat is the cheapest source of protein, representing an important source of meat for the poorest households around the CCNP. Lindsey et al. (2011) reported in south-eastern Zimbabwe that key drivers of the wild meat trade included poverty and food shortages, failure to provide benefits to communities, inadequate investment in anti-poaching in some areas under wildlife management and weak penalty systems that do not provide sufficient deterrent to illegal wild meat hunters. The results of this study are consistent with studies in the Serengeti national park of Tanzania, which indicated that most local people considered wild meat and wildlife body parts as sources of protein and means of generating income (Nielsen et al., 2013). Most of the respondents indicated that illegal hunting activities in CCNP between the years 2012 to 2014 due to strengthened law enforcement in the Park.

### Wildfire

This study indicated that incidence of natural wildlife (e.g. lightning) wildfires were lower than anthropogenically-caused wildfires. Activities such as wild honey extraction, land clearing for shifting cultivation, obtaining good quality pasture for livestock, controlling harmful wildlife such as snakes and creation of fire-killed wood at the boundaries of the Park increased the occurrence of fire during the dry seasons. Honey collectors used many areas of the Park for traditional bee-hive hanging; a practice frowned upon by Park management because of

its potential to cause fire outbreaks that might ravage the Park and degrade the habitat quality of wildlife. According to the respondents, they set fires to protect themselves and their livestock from predator attacks. Fires were also set along roads in the Park to clear footpaths for ease of walking and visibility. Human encroachment near protected areas contributed to increase in fire occurrences in Gonarezhou National Park in Zimbabwe (Gandiwa et al., 2013). Field observations also indicated that during the dry season the study area was left as a fire mosaic, with different areas burnt at different times. Regular burning has significant ecological effects on the wildlife living in the affected ecosystems. Some of the direct effects of fire on fauna in ecosystems include direct and indirect of smaller mammalian species and reptiles (Klop and van Goethem, 2008). In CCNP, some animals like baboons, bush pigs (*Potamochoerus larvatus*) and snakes have been recorded to have died as a result of wildfires.

### Illegal farming

Illegal farming is another source of anthropogenic pressure in CCNP. Focus group discussions revealed that historically, encroachment of wildlife habitats for agricultural activities and illegal hunting have occurred in the Park. The local communities had encroached into the Park area for shifting cultivation over a long period of time before the establishment of the Park. Human population growth around the Park and poverty might be the main reasons for clearing of land for agriculture as away of increasing crop output because of limited alternative survival strategies which increasingly causing destruction and outright loss of some important habitats in the Park ecosystem. Field observations in CCNP indicate that the growth rate of cultivated areas was high at the periphery of protected areas. This might also be the cause of human-wildlife conflict around the Park area. Chebera Churchura National Park harbors many large mammal, birds and other wild animal species. Therefore, it can serve as an important area for conservation of the country's wildlife and tourist attraction in the future. There is a need to improve understanding of the ecological, social and cultural dimensions of conflict situations in the area, to mitigate anthropogenic impacts in CCNP. The findings further suggest the need to initiate long-term monitoring to analyze trends in the incidences of human impacts on wildlife resources. A combined strategy aimed at improving local participation in wildlife conservation initiatives, initiation of public education and awareness campaigns and provision of alternative sources of income for the local people will reduce the threat, and contribute to improve conservation of wildlife in Chebera Churchura National Park.

### Conflict of Interests

The authors have not declared any conflict of interests.

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