



EFFECTS OF HABITAT TYPES ON DUNG BEETLE (COLEOPTERA: SCARABAEINAE) COMMUNITY STRUCTURE IN A PROTECTED AREA OF CENTRAL BELIZE

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ABSTRACT

Dung beetles are detritivorous insects that predominantly feeds on mammal dung. Through their feeding behavior they perform important ecological services. Due to their sensitivity to changes in the physical and biotic factors in a habitat, they are considered as important biological indicators to document effects of habitat change on biodiversity, especially mammal diversity and abundance. Dung beetle community structure in habitats of Belize is poorly known. Dung beetle community structure across three habitats of Monkey Bay Wildlife Sanctuary (MBWS), Belize was studied. Community structure attributes such as species richness, abundance and diversity were studied across a lowland savanna (LS), lowland broadleaf forest (LBLEF) and riparian forest (RF). LBLEF had the highest community attributes such as Simpson's index of diversity (1-D), Shannon diversity Index (H), Margalef richness index and Fishers alpha diversity. Tunneler guild and small beetles dominated the assemblage in MBWS. *Canthidium ardens* abundant and recorded only from LS preferred open habitats such as savanna. *Onthophagus maya*, *O. yucatanus*, *O. crinitus panamensis* and *Scatimus ovatus* abundant and present only in forest habitats preferred closed habitats such as forests. *O. cyclographus* equally abundant in LS and LBLEF was a generalist species with respect to habitat preference. The study proves that LBLEF with its complex vegetation structure supports more faunal diversity with respect to mammals and dung beetles than LS and RF. LBLEF is the largest habitat type in Belize and the most threatened with respect to habitat conversion. Therefore, it is important to protect our LBLEF, as conversion of such forests can negatively impact biodiversity.

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INTRODUCTION

Dung beetles are an abundant and speciose group of insects that are an important component of the terrestrial ecosystems worldwide. They are detritivorous beetles, predominantly feeding on mammal dung, although few species also feeds on carrion, fungi and rotten fruits (Halffter and Matthews, 1966). Through their feeding behavior they perform vital ecological services. Dung beetles positively influence hydrological properties of the soil by increasing water infiltration, soil porosity, and reduces surface water runoff (Dabrowski et al., 2010); enhance secondary seed dispersal (Griffiths et al., 2016) reduce population of flies and helminth worms (Bergstrom et al., 1976; Bishop et al., 2005); reduces greenhouse gas emissions from cattle farms (Forgie et al., 2018) and enhances nutrient recycling (Bang et al., 2005; Losey and Vaughan, 2006). Due to their sensitivity to changes in physical and biotic factors such as mammal diversity and abundance in a habitat, they are considered as important

biological indicators to document effects of habitat change on biodiversity (Audino et al., 2014; Nichols et al., 2009; Tonelli et al., 2017). Dung beetles employ different strategy for feeding and breeding based on which they are classified into three broad functional guilds. Telecoprids or rollers, roll balls of food on the surface of soil to some distance from the source of resource, where they bury them; paracoprids or tunnelers bury dung balls in tunnels below or beside dung pads and endocoprids or dwellers feed and breed immediately below or in the dung pad (Halffter and Mathews, 1966).

Among the various environmental factors that influence dung beetle community structure, the type of vegetation is one of the most important (Halffter, 1991; Andresen, 2005). Vegetation of a habitat influence the temperature, humidity, light intensity, availability of niche and abundance and diversity of mammals in a habitat (Alkemade et al., 2012; Halffter and Arellano, 2002; Klein, 1989; Navarrete and Halffter, 2008; Nichols et al., 2009). Dung beetle community structure attributes such as species richness, abundance, diversity, and guild structure are influenced by these environmental variables (Filgueiras et al., 2016; Gardener et al., 2008; Gill, 1999; Klein, 1989; Nichols et al., 2007; Shahabuddin, 2010). As habitats change, species composition of dung beetles changes

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(Campos, 2013; Davis *et al.*, 2001). In the Neotropics, the greatest diversity of dung beetles occurs in closed forest areas, mainly due to the higher availability of food resources and nesting conditions (Hanski and Cambefort, 1991). But open habitats such as savanna also harbors their own unique species of dung beetles (Duraes *et al.*, 2005; Louzada *et al.*, 2010; Spector and Ayzama, 2003).

Belize is a Central American country located on the Caribbean coast of northern Central America at 17°15' north of the equator and 88°45' west of the Prime Meridian on the southern part of Yucatán Peninsula. Belize has a land area of 22,965 km². It is bordered to the north by Mexico, to the south and west by Guatemala and east, by the Caribbean Sea. Belize is part of the Mesoamerican biodiversity hotspot which occupies less than 0.5% of the terrestrial planet and is thought to contain 17% of all terrestrial species (Goodwin *et al.*, 2013).

As of mid 2014, some 35.8% of Belize's land territory is under protected status (Cherrington, 2014). Belize's national protected areas system comprises some 103 individual protected areas (LIC 2014), representing 73 ecosystem types identified across the country. But biodiversity in these protected areas especially that of insects is not well documented. Only few studies exist on dung beetle community structure in habitats of Belize (Latha *et al.*, 2016 a,b). In the present study dung beetle community structure in three different habitats (lowland savanna, lowland broadleaf forest and riparian forest) of Monkey Bay Wildlife Sanctuary was studied. Community attributes such as species richness, diversity, abundance, guild structure and beetle sizes across these habitats were investigated. We propose that dung beetle community structure will vary across these habitats and that forest habitats will have more abundance and diversity than savanna habitat.

MATERIALS AND METHODS

Study Area

The study area, Monkey Bay Wildlife Sanctuary (MBWS), established in 1990 was amongst the first of the private protected areas established for the purpose of wildlife conservation, education and biological connectivity within Central Belize. It encompasses an area of 1,150 acres (465ha). MBWS provides biological connectivity with the Manatee Forest Reserve to the south across the Sibun River and the northern protected areas. MBWS along with Runaway Creek Nature Preserve another private protected area, and Peccary Hills Reserve provide stepping stones in the only remaining corridor route that is still forested and forms a central link in the Central Belize Biological Corridor (Fig 1). Monkey Bay is becoming increasingly bounded by mechanized agriculture to the west, north, and even east (Directory of protected areas, 2011).

The sanctuary is characterized by five distinct habitat types such as Lowland savanna, Broken Ridge forest, Lowland Broadleaf Forest, Cohune Ridge and Riparian Forest (Fig 2). Collections were done in lowland savanna (LS), lowland broadleaf forest (LBLF) and riparian forest (RF). LS in MBWS are fire influenced ecosystem on infertile acidic soil with a continuous herbaceous layer of native grasses and sedges and scattered trees such as *Pinus caribaea* (Farjon and Styles) (pine), *Acoelorrhaphe wrightii* (Griseb. and H.Wendl.) H.Wendl. ex Becc. (palmetto), *Byrsonima crassifolia* (L.)

Kunth (craboo), *Curatella americana* L. (yaha or sandpaper tree), Melastomataceae Juss. and *Quercus oleoides* Schltld. And Cham. (live oak) amongst the most structurally conspicuous non-herbaceous elements (Goodwin *et al.*, 2013, Laughlin, 2002). They form the third largest ecosystem in Belize (Fig 3A).

The LBLF in the study area is comprised of distinct layers. The bottom layer called the understory, contains shrubs such as ferns, small palms and heliconias. Above the understory is the canopy which is an interconnected layer of trees growing at a similar height and connected by vines and lianas. Trees in the canopy include *Brosimum alicastrum* Swartz (Breadnut), *Terminalia Amazonia* (J.F.Gmel.) Exell (Nargusta), *Krugiodendron ferreum* (Vahl) Urban (Ironwood), *Aspidosperma macrocarpon* Mart. (Milady) and *Calophyllum antillanum* Britton (Santa Maria). Above the canopy is the emergent layer which is made of tall trees which surpass the height of the canopy layer. These are often *Swietenia macrophylla* King (Mahogany), *Enterolobium cyclocarpum* (Jacq.) Griseb. (Guanacaste), *Ceiba pentandra* (L.) Gaertn. (ceiba) and *Manilkara zapota* (L.) P.Royen. (sapodilla). This is the largest ecosystem in Belize. LBLF in the study area is a secondary forest, part of which was cleared several decades ago for grazing (Fig 3B).

RF of MBWS extend to the edges of Sibun River and contain several broadleaf trees and *Guadua longifolia* (E.Fourn) R.W.Pohl (spiny bamboo). Abundant fruit trees such as *Ficus yoponensis* Desv. (wild fig) and *Inga edulis* Mart. (bri bri) along the river bank provide habitat for numerous riparian species including the endangered *Tapirus bairdii* Gill (Baird's tapir), *Alouatta pigra* Lawrence (Yucatan black howler monkey) and common *Iguana iguana* Linnaeus (iguana) (Fig 3C). *Dasylops virginiensis* Boddaert (white tailed deer), *Peccary tajacu* Linn (Collared peccary), *Urocyon cinereoargenteus* Schreber (grey fox) are other mammals reported from MBWS.

Dung beetle sampling

Dung beetles were collected using baited pit fall traps. Four collections were made during the 2015-16 period. Two wet season collections were made in the months of November and December, 2015 and two dry season collections were made in the months of February and March, 2016. Each collection effort involved placing five pitfall traps in each of the three habitats under study, which were LS, LBLF and RF to trap the dung beetles. The traps consisted of plastic basins containing a mixture of mild detergent solution and salt which served as a killing and preservative agent. The basins were buried with their rim in level with the surrounding substrate and each trap was topped with a plastic cover supported by branches obtained from nearby trees. The purpose of the plastic cover was to prevent desiccation during sunny days and inundation during periods of rain. Approximately 100 grams of bait, fresh pig dung or rotten meat was placed (alternately) on a wire mesh over the plastic basin. After a period of 24 hours, all insects were collected and sorted into glass vials containing 70% ethanol. Vials were appropriately labeled with date, pit number, site and bait type. Stereomicroscopes available at the University of Belize's laboratory were used to identify the beetles to species level with the use of taxonomic keys and by comparing with verified specimens from previous studies. Beetles were sorted based on sizes as small (< 10mm) and

large (≥ 10 mm) and into their functional guild as tunnelers, rollers and dwellers.

Data Analysis

Both wet and dry season collections were pooled (n=20x3) for the purpose of analyses. Sample based species accumulation curves were plotted to assess sampling adequacy (Gotelli and Colwell, 2001). Dung beetle species richness estimation was done to compare observed species richness (Sobs) to estimated species richness using nonparametric species richness estimator Chao 1 (Gotteli and Colwell, 2001). Inventory completeness was measured as the percentage of observed species with respect to the number of species predicted by the estimator. Species accumulation curve and species richness estimation was done using EstimateS v.9. Alpha diversity indices such as Shannon diversity Index (H), Dominance (D), Simpson's index of diversity (1-D), Margalef richness index, Fishers alpha diversity were computed for each habitat (Harper, 1999). Bray-Curtis similarity coefficient (Bray and Curtis, 1957) was used to quantify and compare the similarity of dung beetle species composition among habitats. All the diversity attributes were computed using PAST3.

Variation in abundance between species, guild, and beetle sizes were statistically analyzed. Since the data significantly deviated from normal distribution, non-parametric test, Kruskal- Wallis was used to compare variation in abundance between species and guild. Differences with a p-value <0.05 was compared using Mann-Whitney U test. Variation in abundance between beetle sizes was compared using Mann-Whitney U test. All statistical analysis were done using SPSS 21.

Table 1. Dung beetle abundance, functional guild, beetle sizes, Chao 1, Shannon diversity Index (H), Dominance (D), Simpson's index of diversity (1-D), Margalef's richness index, Fishers alpha diversity values for Lowland savanna (LS) Lowland broadleaf forest (LBLE) and Riparian forest (RF) habitats of Monkey Bay Wildlife Sanctuary during 2015-16 study period. Functional guild: T= Tunneler, R=Roller, D=Dweller; Beetle Size: S= Small, L= Large.

SPECIES	Functional guild	Size	LS	LBLE	RF
<i>Ateuchus laetitiae</i>	T	S	0	3	0
<i>Canthidium ardens</i>	T	S	17	0	0
<i>Canthidium centrale</i>	T	S	0	4	0
<i>Canthon cyanellus</i>	R	S	0	0	1
<i>Copris laeviceps</i>	T	L	0	1	0
<i>Coprophaneus sp1</i>	T	L	1	0	0
<i>Coprophaneus telamon corythus</i>	T	L	1	1	2
<i>Dichotomius maya</i>	T	L	1	0	0
<i>Eurysternus caribaeus</i>	D	L	0	1	0
<i>Eurysternus mexicanus</i>	D	L	0	12	0
<i>Onthophagus batesi</i>	T	S	0	3	5
<i>Onthophagus crinitus panamensis</i>	T	S	0	17	1
<i>Onthophagus cyclographus</i>	T	S	8	18	5
<i>Onthophagus incensus</i>	T	S	0	0	10
<i>Onthophagus maya</i>	T	S	0	44	157
<i>Onthophagus sp 1</i>	T	S	0	2	0
<i>Onthophagus yucatanus</i>	T	S	0	29	7
<i>Pseudocanthon perplexus</i>	R	S	0	0	3
<i>Scatimus ovatus</i>	T	S	0	15	0
Total abundance			28	150	191
Species richness			5	13	9
Chao1			8	14.5	9.5
Shannon diversity H			(62.5%)	(89.65%)	(94.73%)
Dominance D			1.018	2.022	0.7954
Simpson index of diversity (1-D)			0.4541	0.1689	0.6815
Margalef Richness			0.5459	0.8311	0.3185
Fishers alpha diversity			1.2	2.395	1.523
Fishers alpha diversity			1.772	3.417	1.961

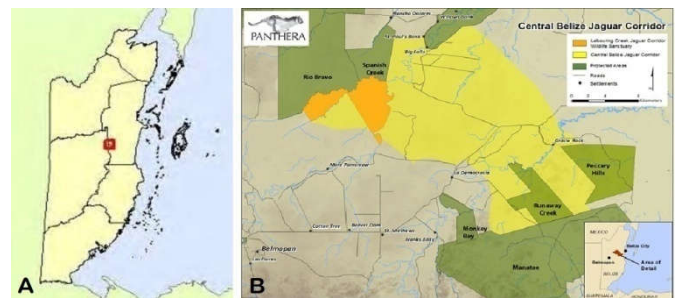


Fig 1 A. Location of Monkey Bay Wildlife Sanctuary in Belize; B. Map of Central Belize Corridor showing Monkey Bay Wildlife Sanctuary.

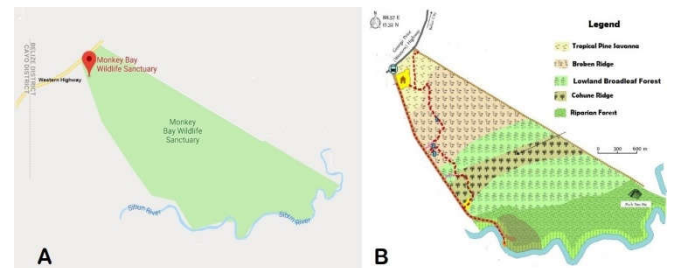


Fig 2 A. Map of Monkey Bay Wildlife Sanctuary; B. Map showing habitat types in Monkey Bay Wildlife Sanctuary



Fig 3 Habitat types under study in Monkey Bay Wildlife Sanctuary, A. Lowland Savanna, B. Lowland Broadleaf Forest, C. Riparian forest

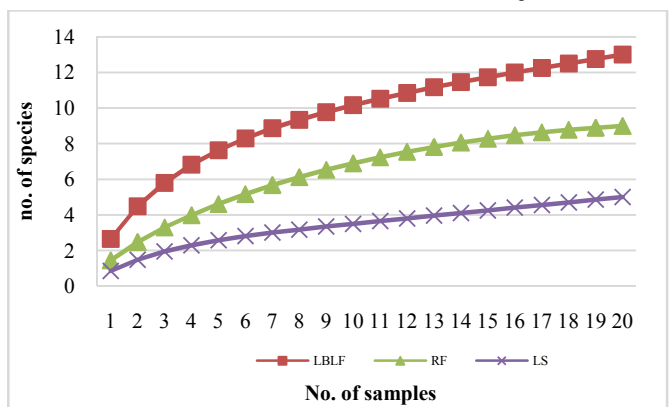


Fig 4 Sample based species accumulation curve (Mao Tau) for Lowland Savanna (LS), Lowland Broadleaf Forest (LBLE) and Riparian Forest (RF) of Monkey Bay Wildlife Sanctuary, Belize for the 2015-16 study period

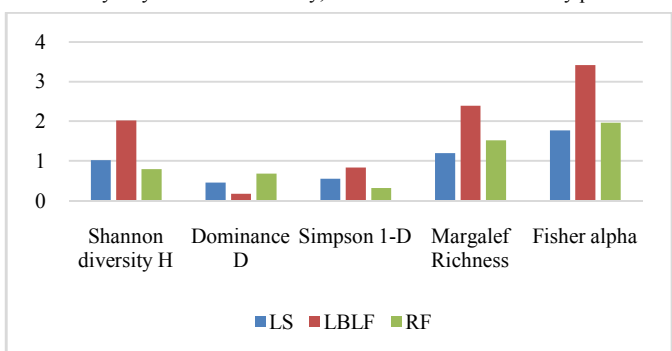


Fig 5 Shannon diversity (H), Dominance (D), Simpson's index of diversity (1-D), Margalef Richness and Fishers alpha diversity values for Lowland Savanna (LS), Lowland Broadleaf Forest (LBLE) and Riparian Forest (RF) of Monkey Bay Wildlife Sanctuary, Belize for the 2015-16 study period

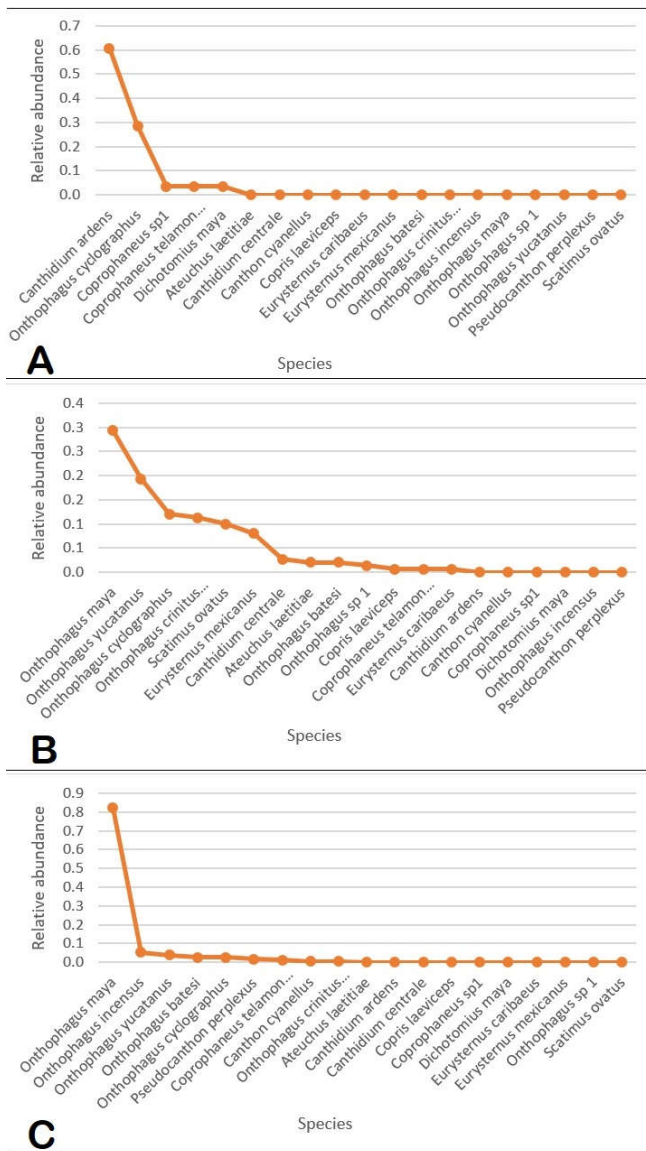


Fig 6 Species Abundance curve for dung beetles in habitats of Monkey Bay Wildlife Sanctuary, Belize for the 2015-16 study period; A. Lowland Savanna, B. Lowland Broadleaf Forest, C. Riparian Forest

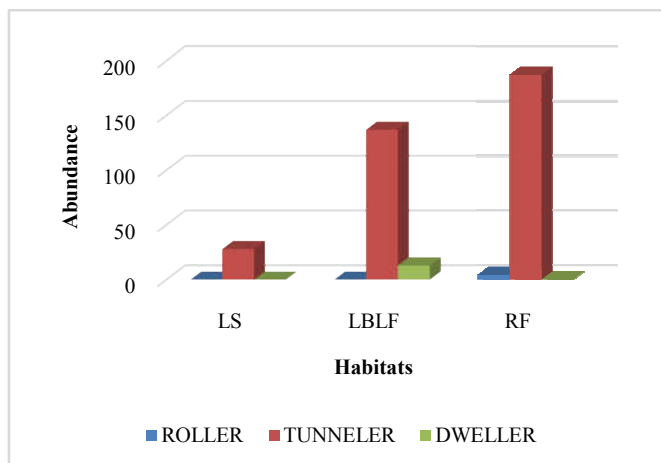


Fig 7 Guild abundance of dung beetles in Lowland Savanna (LS), Lowland Broadleaf Forest (LBLF) and Riparian Forest (RF) of Monkey Bay Wildlife Sanctuary, Belize for the 2015-16 study period

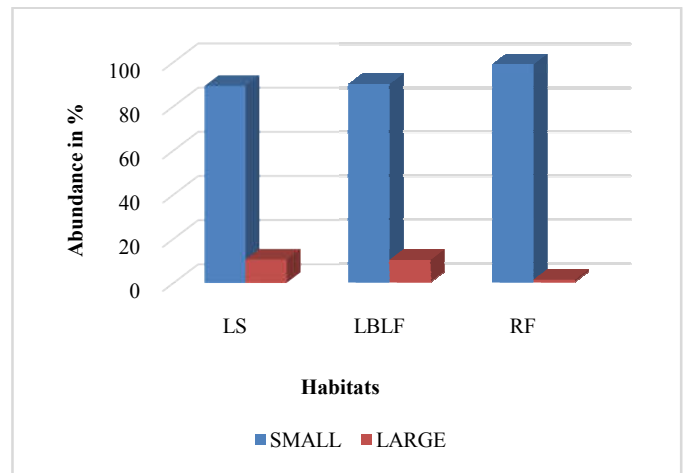


Fig 8 Abundance of small and large dung beetles in Lowland Savanna (LS), Lowland Broadleaf Forest (LBLF) and Riparian Forest (RF) of Monkey Bay Wildlife Sanctuary, Belize for the 2015-16 study period

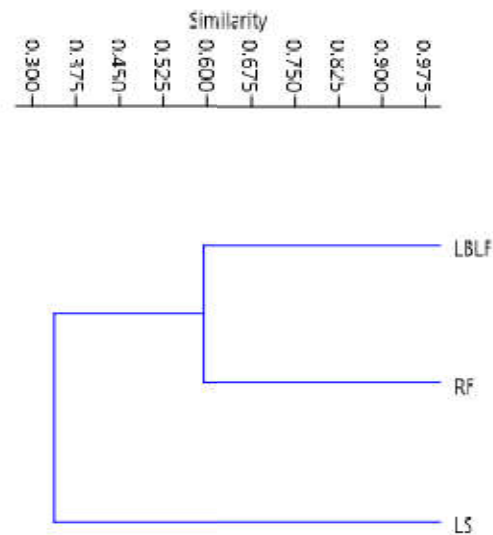


Fig 9 Cluster diagram of Bray Curtis Similarity Index between Lowland Savanna (LS), Lowland Broadleaf Forest (LBLF), Riparian Forest (RF) of Monkey Bay Wildlife Sanctuary, Belize for the 2015-16 study period

RESULTS

A total of 369 beetles belonging to 19 species and 10 genera were collected from MBWS (Table 1). Abundance was highest in RF and lowest in LS. Species richness was highest in LBLF and lowest in LS. Sample based species Accumulation curve for LS and LBLF was continuously rising indicating that more species could be found with additional sampling effort, but for RF was approaching asymptote indicating sampling sufficiency (Fig 4). Nonparametric richness estimator Chao 1 showed sampling completeness of 62.5% for LS, 89.65% for LBLF and 94.73% for RF. Highest abundance was observed in RF, followed by LBLF and LS. Overall species abundance varied significantly between habitats ($H=26.953$, $df= 2$, $p=.000$). Pairwise comparison of abundance between LS and LBLF ($p=.0000$), LS and RF ($p=.002$) and LBLF and RF ($p=.016$) showed significant difference. Shannon diversity, Simpson's diversity index (1-D), Fishers Alpha diversity and Margalef Richness values were highest in LBLF. Dominance D was highest in RF (Table 1, Fig 5). Species abundance curve for the three habitats are shown in Fig 6. Species abundance

curve showed a steep slope and a long tail of rare species for the three habitats. Most uneven assemblage was observed in RF. In LS assemblage, *Canthidium ardens* (60.71%) and *O.cyclographus* (28.57%) were the most abundant species. In LBLF *Onthophagus maya* (29.33%), followed by *O.yucatanus* (19.33%), *O. cyclographus* (12%), *O. crinitus panamensis* (11.33%), and *Scatimus ovatus* (10%) were the abundant species. They accounted for 81.99% of the LBLF assemblage. *O.maya* accounted for 82.19% of the RF assemblage. *O. maya* was the most abundant species in MBWS and accounted for 54% of the overall assemblage. *Canthidium ardens*, *Coprophaneus sp1* and *Dichotomius maya* were recorded only from LS. *Ateuchus laetitiae*, *Canthidium centrale*, *Copris laeviceps*, *Eurysternus caribaeus*, *E. mexicanus* and *Scatimus ovatus* were recorded only from LBLF. *Canthon cyanellus*, *Onthophagus incensus* and *Pseudocanthon perplexus* were recorded only from RF. *Coprophaneus sp1*, *C. telamon corythus*, and *Dichotomius maya* were the singleton species in LS. *Copris laeviceps*, *C. telamon corythus*, and *Eurysternus caribaeus* were the singleton species in LBLF. *Canthon cyanellus* and *Onthophagus crinitus panamensis* were the singleton species in RF.

Tunneler guild dominated overall in MBWS (95.39%) followed by dweller (3.52%) and roller (1.08%). Guild abundance varied significantly ($H=6.995$, $df=2$, $p=.030$). Pairwise comparison using Mann Whitney U test revealed significant difference between roller and tunneler ($p=.012$) but no significant difference existed between tunneler and dweller ($p=.320$) and roller and dweller ($p=.126$). Roller and dweller guild were absent from LS, roller was absent from LBLF and dweller guild was absent from RF (Fig 7). Small beetles dominated the overall assemblage and accounted for 94.58% and large beetles 5.42%. Beetle sizes abundance varied significantly in MBWS ($p=.000$) (Fig 8). Bray Curtis similarity coefficient showed highest similarity between the dung beetle assemblages of LBLF and RF (Fig 9).

DISCUSSION

This is the first recorded study on dung beetle community structure across habitats in Belize. Dung beetle community structure varied between habitats. The forest habitats (LBLF, RF) of MBWS had higher species richness and abundance compared to LS. A direct relationship exists between forest cover and dung beetle species richness and composition (Feer, 2013; Gardener *et al.*, 2008; Halffter and Mathews 1966; Halffter *et al.*, 1992). The forest cover in LBLF and RF provides cooler, humid microclimate that is favorable for different dung beetle species. Savanna habitats are grass dominated habitats with scattered shrubs and trees with high incident light and ambient temperature when compared to forests. Such conditions reduce the time interval the dung is available to beetles as they dry up in a short period of time and increase adult and larval mortality (Klein, 1989; Galante *et al.*, 1995; Duraes *et al.*, 2005). LS is susceptible to fire in summer and waterlogging in wet season. Fire affects dung beetles indirectly by destroying vegetation and affecting microclimatic conditions (Louzada *et al.*, 2010). Flooding inundates underground brood chambers of dung beetles causing hypoxic conditions which may result in death (Lumaret, 1983; Kirk, 1983) or liquefaction of brood balls or decay by fungal attack (Hanski and Cambefort, 1991) and only tolerant species are able to survive such conditions (Tissiani *et al.*, 2015; Whipple *et al.*, 2011).

Dung resource availability is another important factor that determines dung beetle community attributes (Culot *et al.*, 2013; Nichole *et al.*, 2009). The complex vegetation structure in the forest ecosystems of MBWS provides more niche for mammals which in turn provides different and abundant food source to dung beetles (Alkemade *et al.*, 2012; Klein 1989; Nichols *et al.*, 2009). LS habitat in MBWS is poor in mammal fauna. *Onthophagus maya*, *O. yucatanus*, *O. cyclographus*, *O. crinitus panamensis* and *Scatimus ovatus* which accounted for 81% of LBLF and *O.maya* which accounted for 82% of RF assemblage seem to favor the forest habitat and can be considered as forest specialist. *Canthidium ardens* abundant in LS (60.7%) and recorded only from LS can be considered as a heliophile and open-habitat adapted species tolerant to the extreme microclimatic conditions of the savanna habitat. *D. maya* collected only from LS, was earlier collected from open habitat and disturbed forests of Pook's Hill Lodge, Cayo district and not from the pristine forests of Las Cuevas during the same collection period (Gillet, 2009). Hence, *D. maya* can be considered as a species adapted to disturbance and open habitats. *O. cyclographus* equally abundant in LS and LBLF can be considered as a generalist species with respect to habitat preference.

Tunneler guild dominated the three habitats of MBWS. Dominance of tunneler guild in Neotropical forests is a common phenomenon (Halffter *et al.*, 1992). In LS, high incident light and ambient temperature rapidly desiccates dung pads and makes dung unappealing to dung beetles, this could be a reason for the absence of roller and dweller dung beetles from LS. Buttunneler guild rapidly relocates dung beneath the soil, giving them an advantage over rollers and dwellers in LS (Halffter and Edmonds, 1982; Louzada *et al.*, 2010; Navarrete and Halffter, 2008). High grass cover on the savanna floor also hampers rolling behavior. Increased leaf litter and undergrowth in LBLF which can obstruct rolling behavior (Nichols *et al.*, 2013) could be a reason for the absence of roller dung beetles in LBLF. Dweller guild represented by *Eurysternus caribaeus* and *E. mexicanus* was found only in LBLF. Availability of moist dung pad undisturbed by tunnelers in LBLF could be the reason for their presence in this habitat and their absence in RF and LS could be attributed to frequent flooding in RF and waterlogging in LS during the eight month long wet season which washes away the dung or liquefies the dung making it unsuitable for dwellers (Hanski and Cambefort 1991).

The overall dominance of small beetles in MBWS is a clear indication of the presence of small dung pad producing mammals and low abundance and diversity of mammals in the national park (Hanski and Krikken, 1991, Nichols *et al.*, 2009). Large beetles are also negatively impacted by disturbance (Klein, 1989). Though MBWS is a protected area, the surrounding areas of the sanctuary are subjected to anthropogenic disturbance such as agriculture and human settlements which could be a hindrance for mammals to cross over to MBWS and provide dung source for dung beetles.

The present study proves that LBLF of MBWS had higher species richness and diversity of dung beetles. This result is comparable to regional studies on habitat preferences of dung beetle. The complex vegetation structure in LBLF provides favorable microclimate and also harbors more mammals which provides abundant dung source to dung beetles. LBLF is the largest habitat type in Belize and the most threatened with respect to habitat conversion. Therefore, it is important to

protect our LBLF and prevent conversion of such forests into other land use types as it has negative consequence on biodiversity of a given region.

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