Whitworth, A, Pillco Huarcaya, R, Gonzalez Mercado, H, Braunholtz, LD and MacLeod, R

Food for thought. Rainforest carrion-feeding butterflies are more sensitive indicators of disturbance history than fruit feeders

http://researchonline.ljmu.ac.uk/id/eprint/10705/

Article

Citation (please note it is advisable to refer to the publisher's version if you intend to cite from this work)

Whitworth, A, Pillco Huarcaya, R, Gonzalez Mercado, H, Braunholtz, LD and MacLeod, R (2017) Food for thought. Rainforest carrion-feeding butterflies are more sensitive indicators of disturbance history than fruit feeders. Biological Conservation, 217. pp. 383-390. ISSN 0006-3207

Full Title

Food for thought. Rainforest carrion-feeding butterflies are more sensitive indicators of disturbance history than fruit feeders

Andrew Whitworth [1, 2, 3] (Corresponding author)

andy.w.whitworth@gmail.com

Ruthmery Pillco Huarcaya [2, 3, 4]

ruth.pillcohuarcaya@gmail.com

Harryson Gonzalez Mercado [5]

jharrysongm@gmail.com

Laura Dominie Braunholtz [2]

laura_braunholtz@hotmail.co.uk

Ross MacLeod [1]

ross.macleod@glasgow.ac.uk

[1] Institute of Biodiversity, Animal Health and Comparative Medicine, College of Medical, Veterinary and Life Sciences, University of Glasgow, Glasgow, G12 8QQ, UK.

[2] The Crees Foundation, Urb. Mariscal Garmarra B-5, Zona 1, Cusco, Peru.

[3] Osa Conservation, 1012 14th Street NW, Suite 625, Washington, D.C. 20005, USA.

[4] Universidad Nacional San Antonio Abad del Cusco (UNSAAC), Perú.

[5] Museo de Historia Natural de la Universidad Nacional de San Antonio Abad del Cusco, Perú

Abstract

Tropical forests have, and in many areas continue to experience both severe and subtle forms of human disturbance; most commonly from hunting, logging and clearance for agriculture. The ability to detect a full range of impacts is essential to understanding how biodiversity responds to human disturbance. Since monitoring the entire biodiversity of a tropical forest is an impossible task, specific groups of biodiversity are often used as biological indicators. Due to their relative ease in detection and identification, their sensitivity to environmental change and their short generation time, butterflies are suggested to be one of the most effective biodiversity indicators for tropical forest monitoring. However, most biodiversity monitoring of tropical ecosystems using butterflies relies only on one sub-group, the fruit-feeding butterflies, or Nymphalidae. Here we assess for the first time if the use of carrion-feeding butterfly communities might improve our ability to detect and monitor human impacts and conservation management outcomes in tropical forests. We analysed species richness, abundance and community composition of rainforest fruit and carrion butterfly communities to see how effectively they detect known differences in forest disturbance history, between three different vertical strata of rainforest, and assess whether they provide stable results across different seasons. We found that compared to fruit-feeding butterflies, sampling carrion-feeders detected greater species richness and abundance for the same survey effort, detected more pronounced effects of known differences in historic disturbance, and showed greater temporal stability in biodiversity patterns across the year. We also identify for the first time a series of indicator butterfly species and tribes that could be used as biological indicators to study biodiversity responses to human disturbance and differences across vertical strata of the rainforest. We therefore suggest that carrion-feeding butterfly communities will be a powerful addition to the family of indicators groups that are available for monitoring the impacts of human disturbance on tropical biodiversity.

Introduction

Tropical forests worldwide have experienced, and in many cases continue to experience, both severe and subtle forms of human disturbance (Keenan et al. 2015; Kim et al. 2015); most commonly in the forms of hunting, logging and clearance for agriculture (Tyukavina et al. 2017). These disturbances have varying severity of effects on forest structure (Laurance et al. 2001), biodiversity (Putz et al. 2012; Burivalova et al. 2014; Alroy 2017), and ecosystem function (Paudel et al. 2015).

Changes in biodiversity of degraded forest (selectively logged or hunted), forest converted land (for agriculture or livestock) and secondary regenerating forest (following clearance and abandonment) are of particular interest, in particular when considering restoration and recuperation of biodiversity (Budiharta et al. 2014). However, monitoring the entire biodiversity of tropical forests is an impossible task, especially considering the tight budgets and short timeframes available for most studies (Gardner et al. 2008). This has given rise to the use of specific subset-groups of taxa that are used as biological indicators (Lawton et al. 1998). These key groups are often chosen due to their sensitivity to changes that allow them to act as indicators of general biodiversity responses to habitat disturbance or climatic changes (Lawton et al. 1998; Devries and Walla 2001; Barlow et al. 2008).

One of the most commonly used indicator taxon is butterflies (Lepidoptera), with the subset of fruit-feeding Nymphalidae often chosen to represent butterflies as a whole (DeVries, P. Murray, D. Lande 1997; Lucci Freitas et al. 2014). Butterflies have been used in assessments of tropical forest impacts such as climate change (Molina-Martínez et al. 2016), forest fires (de Andrade et al. 2017), fragmentation (Scriven et al. 2017), and post-disturbance forest recovery (Nyafwono et al. 2014). They are suggested to be effective as indicators of ecological change due to sensitivity to changes in vegetation structure and composition (Hamer et al. 2003; Bonebrake et al. 2010), and because they have a short generation time that allow for responses to change to be quickly monitored and detected (Brown 1997). Additionally, compared with other insect taxa, the taxonomy of butterflies is relatively well studied, and comprehensive field guides or local expertise are available at many localities. Despite this, Bonebrake *et al.* (2010) note that butterflies are an “imperfect indicator”. Indeed, previous studies have recognised significant seasonal variability in abundance and species richness of fruit-feeding Nymphalidae (Devries and Walla 2001; Nyafwono et al. 2014), with seasonality differentially affecting patterns across vegetation strata (Devries and Walla 2001) and butterfly body size (Ribeiro and Freitas 2011).

Given this seasonal variability, the focus on fruit-feeding butterfly communities could be one of the factors leading to a current lack of agreement about the conservation value of secondary forest and plantations based on assessments of butterfly biodiversity (Barlow *et al.*, 2007; Whitworth *et al.*, 2016). Another factor worth considering is that the use of Nymphalidae caught in fruit-based traps alone may not completely represent overall butterfly biodiversity responses to disturbance. While Horner-Devine *et al.* (2003) found that frugivorous and non-frugivorous butterfly species richness correlated across coffee farms and forest patches, the methods to trap both guilds differed greatly (fruit-baited traps vs transects with hand nets). Feeding on carrion is known in multiple species across butterfly families (Austin and Riley 1995; Hall and Willmott 2000; Molleman et al. 2005; Hamer et al. 2006; Holloway et al. 2013), and is thought to be a component of ‘puddling’ behaviour (Molleman *et al.*, 2005). The use of carrion-baited traps attracts a wider representation of the butterfly community, and often with higher capture rates (Austin and Riley, 1995; Sourakov and Emmel, 1995; J. Hall and Willmott, 2000; Hamer *et* *al.*, 2006; Whitworth *et al.*, 2016). However, to date the ecology of carrion-feeding butterflies is not well known, and their dependence on this food source versus others is not fully understood (Hall and Willmott 2000; Holloway et al. 2013). In general, information about bait attractiveness and comparisons between their effectiveness remains scarce (Freitas *et al.*, 2014). A study in Borneo that compared butterfly communities attracted to fruit vs carrion-baited traps found little similarity in species captured by the two baits (Hamer et al. 2006). Despite this, the use of carrion bait (directly compared with fruit bait) has yet to be assessed across vertical strata, replicated seasonally and in relation to tropical forest habitat disturbance. In addition to a previous study carried out at the same site as this current study (Whitworth *et al.*, 2016b), we found only one other published case that included both fruit and carrion bait to sample tropical butterflies in areas of anthropogenic disturbance; though this study did not directly compare the differences of each bait type in relation to disturbance affects (Brown and Freitas, 2000; see S1 for a summary of literature reviewed).

This paper describes, to our best knowledge, the first direct test of the relative effectiveness of fruit and carrion-baited butterfly communities as biological indicators of disturbance in tropical forest. We do so by comparing species richness, abundance and community composition of butterflies caught in fruit and carrion-baited traps to see how effectively they detect known differences in forest disturbance history, between three different vertical strata of rainforest and across different seasons. Specifically, we (1) test if both bait types detect the same directional pattern in observed species richness and abundance, (2) assess whether fruit or carrion-feeding butterfly communities show stronger responses in species richness and abundance to known differences in forest disturbance history, (3) compare the temporal and spatial patterns of both carrion and fruit feeding butterfly community composition (both at level of individual species and at the level of different tribes) across vertical rainforest strata and disturbance type; and (4) determine whether there are specialist indicator species and tribes characterising each food resource. The work was conducted in the lowland tropical rainforest of the Manu Biosphere Reserve in the Peruvian Amazon, one of the world's most biodiverse and important conservation areas.

Methods

Study location and sampling design

The data collection was carried out at the Manu Learning Centre (MLC) in the Peruvian Amazon (71°23'28"W12°47'21"S; for location map, detailed site description, and survey design, see Whitworth et al. 2016a; Whitworth et al. 2016b). In summary, a key feature of the study area was a known history of where different anthropogenic disturbance types had occurred, as previous research has indicated disturbance history to be one of the most influential factors related to biodiversity patterns (Ross et al. 2002). Disturbance types assessed within this study were: 1) selective logging (identified herein with the acronym SLR – signifying selectively logged, regenerating forest i.e. primary forest that was recovering after disturbance), 2) complete clearance due to conversion to agriculture for coffee, cacao and other subsistence crops (identified herein with the acronym CCR – signifying completely cleared and regenerating forest i.e. secondary forest), and 3) a mixed area that had previously consisted of a mosaic of small completely cleared areas used for subsistence agriculture combined with selective logging of the adjacent forest (identified herein as MXD – mixed disturbance regenerating forest). Human disturbance had started ~60 years previously and lasted for 30 years before systematic human disturbance activities were abandoned in the 1980s. Regeneration of the site occurred for at least 30 years, and from 2003 it was officially protected from all further disturbances. As such, closed canopy regenerating tropical forest covered the site at the time of the study.

Butterflies were surveyed using simple cylindrical traps (Hughes et al. 1998). Three traps were suspended at each sampling location to represent three vertical strata: understorey (1–2m), midstorey (6–10m) and canopy (>16m); for details see Whitworth et al. 2016b. In total, 18 locations were sampled across the study area based on a stratified design with six sampling locations per previously mentioned disturbance type. Total trapping effort over a 12 month period accumulated to 2160 trap days (April 2013 –March 2014) with 120 trap days at each individual sampling location. At each sampling location the traps in the three vertical strata were set to collect simultaneously, with each trap operated twice in each of four three month periods, once with fruit-banana and once with carrion-fish bait. Trapping sessions lasted for five days: accumulating to four sessions with banana (20 days) and four sessions with fish bait (20 days) for each trap over the 12 month sampling period. Traps were checked daily between 0900 and 1500, with a randomized site visiting sequence to avoid any systematic bias and bait was replaced every day to ensure similar bait freshness across all sites (Hughes et al. 1998; Devries and Walla 2001). Individuals large enough to be marked easily and safely and without transparent wings were marked with a non-toxic silver marker. Since in general, larger species are also likely to be able to travel further, this allowed a check of likely maximum recapture rates. Recapture rates were very low (1.43%) and known recaptures were excluded from the results of both methods so comparisons would not be biased and the low recapture rate meant any unidentified recaptures of smaller individuals would be insufficient to generate the patterns observed in the results. . The rotting banana bait was prepared following the methods by DeVries, Lande and Murray, (1999) and the rotten fish bait was prepared a week prior to sampling (Austin and Riley 1995; Hughes et al. 1998).

Data analysis

Abundance and Species Richness

In order to investigate biodiversity distribution patterns between the two different bait types, at different vertical levels, in forest with differences in disturbance history, and across different seasons, we assessed observed overall levels of butterfly abundance and species richness using Generalized Linear Mixed Models (GLMM's; with a negative binomial distribution, as overdispersion was detected as a result of zero-inflation; and using a log link function) in program R (R Core Team 2013). To account for repeat measures within sampling locations, sampling location identity was included as a random effect and candidate models were compared with the null model containing only this random effect. Interactions between other covariates and bait type were also included where a covariate appeared to have a significant effect. Model AICc values were compared through a stepwise modelling approach to assess the top-model; (with a ΔAICc<2; and confirmed by an analysis of variance (ANOVA) test between AICc values of top candidate models).

Community Composition

Non-metric multidimensional scaling (NMDS; using the Bray-Curtis similarity measure) was conducted to determine differences in community composition for fruit and carrion-feeding butterfly communities in each disturbance area for fruit and carrion-feeding communities separately, and to assess community composition differences between vertical strata for fruit and carrion-feeding butterflies separately. All stress values were relatively low (ranging between 0.14 and 0.25) and so results were displayed in two dimensions. To assess the statistical significance of observed differences in assemblage composition between different disturbance areas and vertical strata we performed permutational multivariate analysis of variance (PERMANOVA; using 999 permutations). This test uses pseudo-F values to compare among-group to within-group similarity (here bait, strata or disturbance area), assesses significance by permutation, and is robust in cases of balanced study designs (see Anderson and Walsh 2013). Non-metric multidimensional scaling ordinations and PERMANOVA tests were carried out in the vegan package (Oksanen et al. 2013), in program R (R Core Team 2013).

Indicators – Tribes and Species

In order to assess tribe specific preferences for bait, disturbance type, strata and season, we also carried out the same model structure described previously to assess tribe abundances (only where overall number of individuals for a tribe (n) was greater than 30 records). We also computed indicator values (IndVal, Solar *et al.* 2016, Dufrene and Legendre, 1997) for each species and each tribe in relation to their affiliation towards each bait type, vertical strata and disturbance area. The significance of IndVal indices was assessed using 1000 iterations.

Data available from the The University of Glasgow, Enlighten: Research Data repository: Datacite DOI: 10.5525/gla.researchdata.241.

Results

In total 229 species of butterfly were detected, with a total of 5219 individual records. Survey coverage was high overall (84% ±2.65% of estimated species detected in 2160 trap-days, see Whitworth et al. 2016b). It was therefore unlikely that any trends observed in the results would be driven by insufficient survey effort.

Abundance and Species Richness

Both observed sample level abundance and species richness of butterflies were higher in traps baited with carrion (compared to traps baited with fruit), higher in forest disturbed by selective logging (as opposed to secondary growth forest), higher in the understorey (compared to upper canopy strata), and higher from July to December (compared to survey sessions between January to June; see Figure 1 and Table 1). There was a significant interaction between bait and disturbance history, indicating that carrion-baited traps detected a greater difference in both abundance and species richness in relation to different types of historic rainforest disturbance than did fruit-baited traps. There was also a significant interaction between bait and season, which showed that the abundance and species richness of butterflies caught in fruit-baited traps was considerably lower from January to June, whereas carrion-baited traps only showed lower abundance and richness from April to June. This more limited seasonal difference was also to a lesser degree (see S2 for coefficient summary tables from the top models). There was no significant interaction between bait type and vertical strata, signifying similar degrees of vertical stratification in regards to species richness and abundance in both fruit and carrion-feeding butterfly communities.

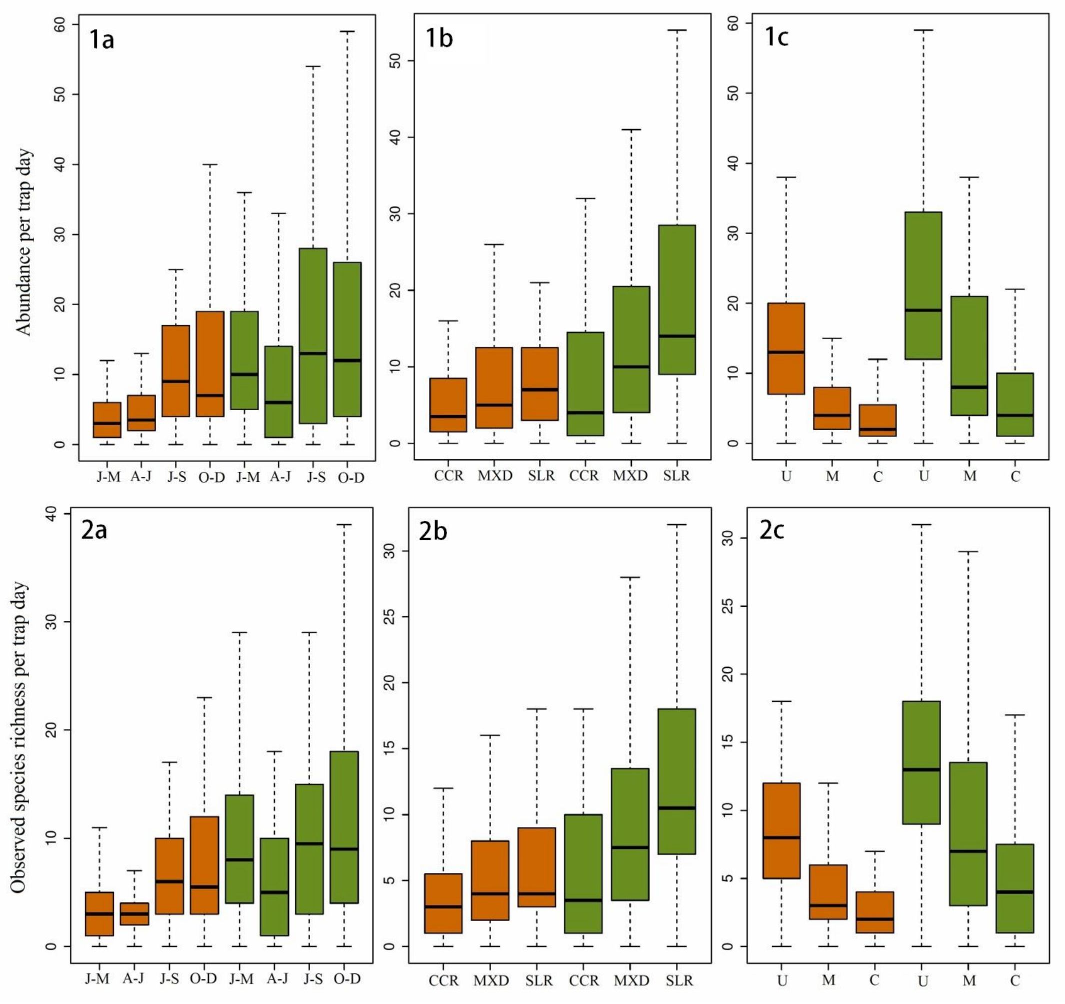

Figure 1 – The abundance (top row - 1) and observed species richness (bottom row - 2) of butterflies; 1a & 2a Between different seasons (J-M = Jan-Mar, A-J = Apr-May, J-S = Jul-Sep, O-D = Oct-Dec), 1b & 2b Between different forest types (CCR = secondary growth forest following clearance, SLR = degraded forest following selective logging and MXD = mixed disturbance regenerating forest) and 1c & 2c Between different vertical strata (U = understorey, 1-2m above ground; M = midstorey, 6-10m above ground; C = canopy, >16m above ground). Orange shaded plots (left of each plot) represent fruit-baited traps, and green shaded plots (right of each plot) represent carrion-baited traps.

Table 1 – Top models for effects on butterfly abundance (a16) and species richness (r16), along with all other weighted and null models. Top models were selected using anova analysis between the log likelihoods of the two top candidate models. For abundance there was no significant difference between top two models a16 and a17 (p=0.19) and therefore the most parsimonious model was chosen. For species richness a statistical difference (p=0.03) along with greater weighting supported r16 as the top model. See S2 for full details of candidate models.

Candidate models		Intercept	Bait type	Disturbance history	Vertical strata	Season	Bait:Strata	Bait:Season	Bait:Disturbance	df	logLik	AICc	delta	weight
Abundance	a16	1.771	+	+	+	+		+	+	16	-1375.54	2784.4	0	0.597
	a17	1.792	+	+	+	+	+	+	+	18	-1373.88	2785.4	1.03	0.357
	a14	1.573	+	+	+	+		+		14	-1380.39	2789.8	5.4	0.04
	a15	2.117	+	+	+	+			+	13	-1383.43	2793.7	9.34	0.006
	null	2.453								3	-1514.03	3034.1	249.73	0
Species richness	r16	1.45	+	+	+	+		+	+	16	-1180.31	2393.9	0	0.671
	r14	1.303	+	+	+	+		+		14	-1183.88	2396.8	2.84	0.162
	r17	1.472	+	+	+	+	+	+	+	18	-1179.58	2396.8	2.89	0.158
	r15	1.737	+	+	+	+			+	13	-1188.1	2403.1	9.14	0.007
	r1	1.593	+	+	+	+				11	-1191.88	2406.4	12.46	0.001
	null	1.981								3	-1322.74	2651.5	257.61	0

Community Composition

The composition of butterfly species between different vertical strata was distinct overall (see Figure 2 and Figure S3), was distinct for fruit and carrion-baited traps overall (see Figure 2 and Figure S4), and for both fruit and carrion-feeding communities, and across all forest types separately (see Figure 2). All these differences were statistically significant (see Figure 2: p=0.001, R values between 0.31-0.35).

Using the sampling effort and sampling in the same locations a total 211 species were detected by the carrion baited traps and 167 species by the baited banana baited traps. There was good overlap between the butterfly communities being sampled, with 65% (149 species) being trapped by both methods. The number of singletons for each bait type was very similar and slightly lower for carrion (34 carrion v 38 fruit), so the improved performance of the carrion feeder bait was not due to few 'stray' individuals from other habitats inflating the number of species. The main difference in effectiveness of methods was that the carrion bait trapped many more unique species (62 species, 27% of total butterfly species detected) compared to only 8% (18) unique species trapped using the fruit bait. There was a broader range of species visiting the carrion bait, and this resulted in the carrion bait attracting 92% of the fruit feeding species.

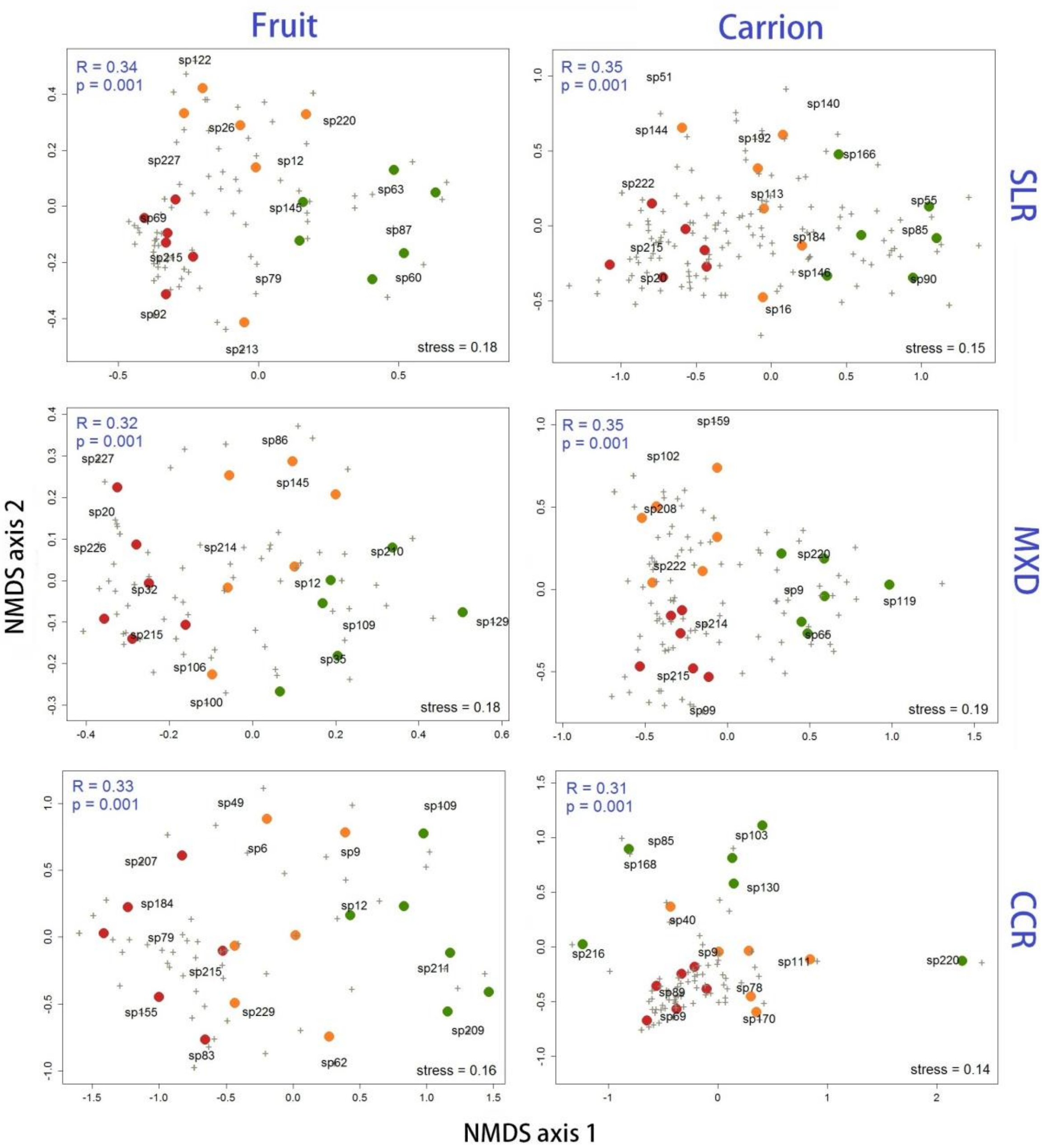

Figure 2 - NMDS plots and associated PERMANOVA test statistics showing overall that both carrion and fruit baits detect community differences between different vertical strata (red points = understorey, 1-2m above ground; orange site points = midstorey, 6-10m above ground; green site points = canopy, >16m above ground). Different plots represent different baited traps across forests with different disturbance histories (CCR = secondary growth forest following clearance, SLR = degraded forest following selective logging and MXD = mixed disturbance regenerating forest). Point labels represent species codes, with priority for those most abundant where points overlap (see S6 for ID codes related to species).

Although the composition of butterfly species between different vertical strata was distinct for both fruit and carrion-feeding communities across all forest types separately (Figure 2), community composition of butterflies was distinct between disturbance types, only within the canopy strata for the fruit-feeding community. The midstorey and understorey fruit-feeding communities displayed low R-values (both ~0.14) and were not significantly distinct between disturbance areas (p=0.2 and 0.13 respectively; see Figure 3). Carrion-baited traps however showed difference in community composition between disturbance history areas in the canopy, midstorey and the understorey (p=0.001, 0.02 and 0.01 respectively).

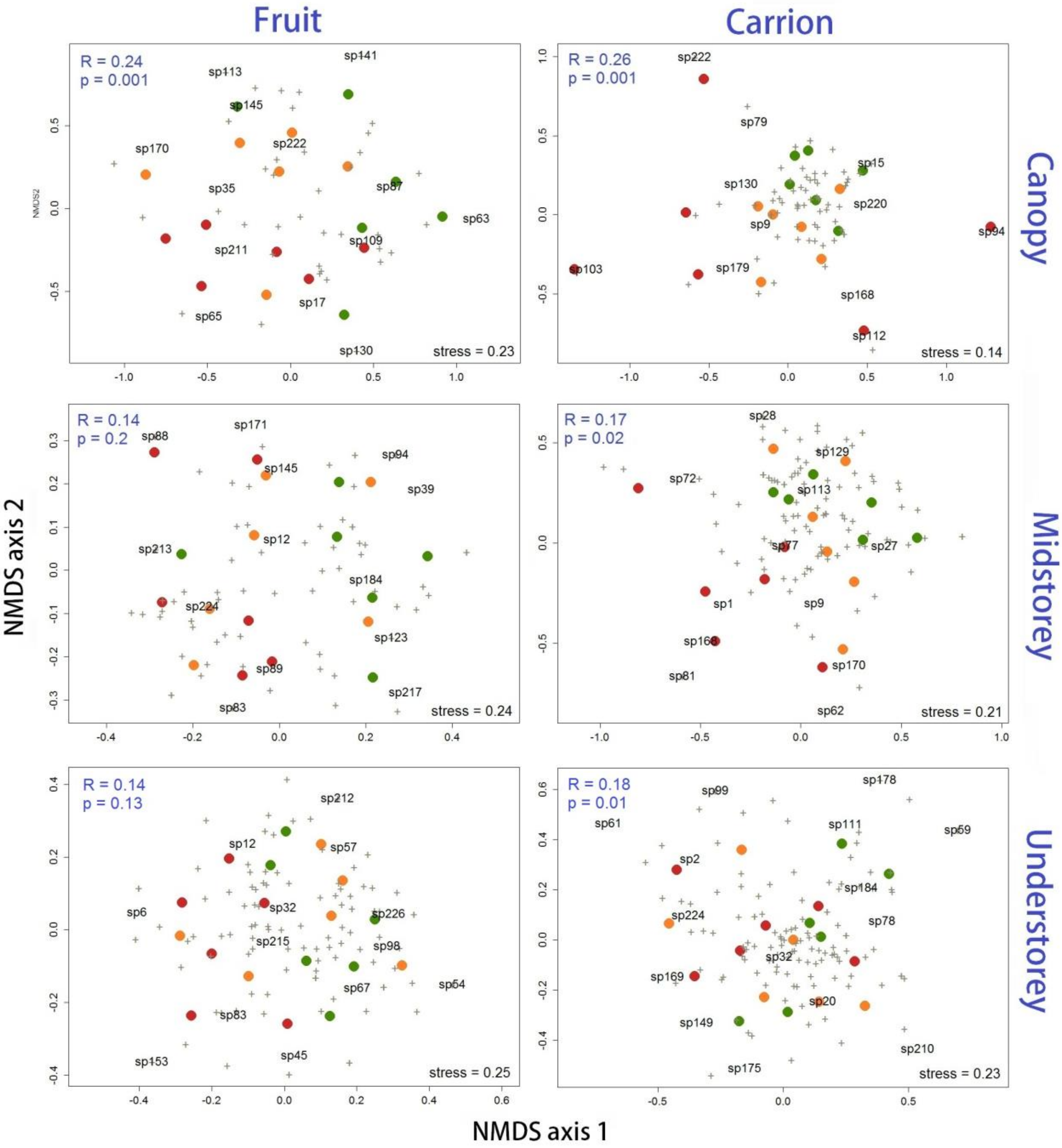

Figure 3 - NMDS plots and associated PERMANOVA test statistics showing community differences between habitats with different disturbance histories are more detectable when sampled with carrion than fruit baits (red site points = secondary growth forest following

clearance; green site points = degraded forest following selective logging; and orange site points = mixed disturbance regenerating forest). Different plots represent different baited traps across different vertical strata (understorey, 1-2m above ground; midstorey, 6-10m above ground; and canopy, >16m above ground). Point labels represent species codes, with priority for those most abundant where points overlap (see S6 for ID codes related to species).

Indicator Tribes and Species

Of the 15 tribes with >30 individuals recorded, ten were found to be indicators using carrion bait and just two of fruit bait (see Table 2). In terms of vertical strata, seven tribes were indicators of the understorey, just one tribe showed preference for the understorey-midstorey, three tribes showed a specific preference for the midstorey, two tribes preferred the midstorey-canopy levels and just a single tribe was indicative of the canopy. Seven tribes were found to be indicators of selectively logged forest, two tribes were indicators of both mixed disturbance and degraded forest, while no tribes were indicators of secondary growth forest. In terms of indicator species, over four times as many species were found to be indicators with carrion bait compared with fruit bait (40 vs 9 species respectively; see S5). Indicators species for bait types, vertical strata and forest type are listed in S6.

Table 2 – Tribes that display a preference for specific bait type, vertical strata and forest type (as suggested by an IndVal analysis). Those with an * also showed a significant preference using GLMM's. SLR = degraded logged forest, CCR = secondary growth historically cleared forest, and MXD = mixed disturbance regrowth forest.

Tribe	Number of individuals detected overall	Association with bait, strata and forest type		
		Bait	Strata	Forest type
Apaturinae	35	Carrion*	Midstorey	SLR*
Brassolini	180	Carrion	Understorey	SLR
Callicorini	34	Carrion*	Midstorey-Canopy*	
Catonephelini	608	Carrion	Understorey	MXD-SLR
Coeini	118	Fruit*	Midstorey-Canopy*	
Epiphelini	202	Carrion*	Midstorey	SLR
Euselasiini	38	Carrion*	Canopy*	SLR
Haeterini	51	Fruit	Understorey	
Heliconiini	416	Carrion		MXD-SLR
Ithomiini	53		Understorey*	SLR
Limenitidini	579	Carrion	Midstorey	SLR
Morphini	86	Fruit	Understorey*	
Nymphalini	140	Carrion	Understorey	SLR
Preponini	261	Carrion*	Understorey-Midstorey*	
Satyrini	1211		Understorey	

Discussion

We detected more pronounced effects of known differences in historic human disturbance, and greater temporal stability in biodiversity patterns across the year, in carrion-feeding butterflies than fruit-feeding butterflies. These findings suggest that performance of one of the most important groups used as indicators of biodiversity responses to anthropogenic disturbance in tropical forests could be improved by increasing the focus on carrion-feeding butterflies. As such, the use of only fruit-baited traps may be misrepresenting patterns, especially in particular seasons or in areas of different forest disturbance.

Few studies have systematically assessed the potential for different methodologies or sub-groups of indicator taxa to lead to contrasting conclusions in relation to biodiversity and conservation value of regenerating forests (Barlow et al. 2007b; Whitworth et al. 2017). Our results focus on the effect of using different bait types on detectability of patterns when using butterfly biodiversity indicators. Previous studies on other taxonomic groups also suggest that such methodological effects may be important for biodiversity assessments. For example, mist nets and point count methods used to assess the response of bird communities to tropical forest disturbance in Brazil have displayed contrasting responses of bird species richness (Barlow et al. 2007b). Likewise, an assessment of the impact of an unmarked road on bird biodiversity in the Ecuadorian Amazon found a negative response using mist nets, while point counts detected greater biodiversity near to the road (Whitworth et al. 2015).

Other studies have also suggested different biodiversity response patterns may be detectable using alternative survey methods for butterflies (Kudavidanage et al. 2012; Ribeiro and Freitas 2012). However, these studies were conducted at different survey sites and not directly compared within the same study area. An essential factor in our study in confirming that any different patterns of butterfly biodiversity could only be linked to methodological (bait-type) effects is because they were carried out within the same study site, and using traps at the same sampling locations. These results show how assessing the same taxonomic group, at the same site, using different baits can suggest different relative biodiversity value between recovering forest types. This result is consistent with a previous study that compared methodological approaches. Wood and Gillman (1998), who complemented fruit-based traps with walk-and-count transects, found that the two methods revealed different patterns of butterfly diversity in relation to tropical forest disturbance. Contrastingly, Horner-Devine *et al.* (2003) found that frugivorous (captured in baited-traps) and non-frugivorous butterflies (captured using hand nets) followed a similar trend in response to anthropogenic disturbance to tropical forest.

The results of our study indicate that fruit and carrion-feeding butterfly guilds respond in different extents to forest disturbance. This finding is comparable to results from other key taxonomic groups that compared response to forest disturbance across feeding guilds of a range of taxa including birds (Gray et al. 2007), beetles (Davies et al. 2000; Bouchard and Hébert 2016) and ants (Kwon et al. 2014). Together these results suggest that identifying which methods and taxonomic sub-groups are the best indicators of biodiversity response to disturbance is an area where further research is needed. In particular it would be interesting to investigate whether the groups, such as carrion feeding butterflies, that are good biological indicators for studying disturbance impacts differ in any systematics ways from other biodiversity to check that the use of bioindicator groups accurately reflects underlying patterns in a wider range of biodiversity. As little is currently understood about the ecology of carrion-feeding butterflies, understanding how the patterns of historical disturbance affect abundance and richness of this guild is another area that would benefit from more intensive research. Studies have suggested that carrion-feeding butterflies tend to be faster fliers with higher metabolic rates (Hall and Willmott 2000; Hamer et al. 2006), though these morphological differences have not yet been linked to ecological differences (Hall and Willmott 2000; Hamer et al. 2006).

In interpreting our results it is important to note that carrion bait predominantly attracts male butterflies (Hall and Willmott 2000; Hamer et al. 2006; Holloway et al. 2013). This is thought in some but not all species to be due to the nuptial gifting of sodium during mating with females (Molleman et al. 2005). This could make carrion an unsuitable bait where sex-bias needs to be avoided, or could make carrion a less attractive bait in certain seasons. However, the lack of strong seasonal influence on the abundance and species richness of butterflies attracted to carrion-baited traps observed in this study suggests that overall there is no complication caused by variable attractiveness of carrion-bait depending on time in mating cycle.

One limitation of the small spatial scale (~800ha) used in this study, is that transient species may enter neighbouring treatment types temporarily (Barlow et al. 2007a), which means that individuals can be detected and recorded where they might not necessarily ‘live’. However, this factor is true for all habitats and vertical strata, and given our detection of significant differences, our observed patterns can only be considered as conservative differences. Another factor to consider was highlighted by Freitas *et al.* (2014) who suggest that carrion-fish baited studies should use caution in comparative studies due to the difficulty to find the same kind of fish for bait standardization and unpredictability throughout rotting processes for each fish species. This factor however could hold equally true for fruit-baited studies. Even if researchers standardised to utilise bananas for example, bananas from different farms, or even fields with different soils, could equally risk containing varying degrees of sugar contents and pungency no matter how standardised the methodological instructions might be. Nonetheless, we suggest that future within-site assessments using baits prepared from a variety of fish species, or from different fruit mix preparations, might elucidate how strong any potential differences might be.

In conclusion, we show that sampling carrion feeding butterflies (as opposed to fruit-feeders) detects a greater species richness and abundance for the same survey effort, elucidates more pronounced effects of known differences in historic disturbance, and displays greater temporal stability in biodiversity patterns across the year. Combining survey methods is often the preferred approach where detailed species inventories are intended (Sparrow et al. 1994; Brown and Freitas 2000) and if resources allow we would suggest using both carrion and fruit baited approaches for collecting biological indicator data based on butterflies. However, methods that target sensitive community sub-sets (Beccaloni and Gaston 1995; Nyafwono et al. 2014) in order to gather the greatest amount of data per unit time, and that are less affected by seasonal patterns, are preferable when making assessments related to biodiversity value of tropical forests. As such, if multiple bait approaches are not an option, we suggest that carrion-feeding butterfly communities will be a powerful addition to the family of indicator groups available to assess the effects of habitat disturbance and forest recovery both in rainforest ecosystems and for conservation more generally. We also conclude that conducting side-by-side comparisons of survey methodologies at the same study locations are essential if we intend to effectively detangle factors related to the recovery of biodiversity in tropical forest systems.

Acknowledgements

We thank the Crees Foundation (www.crees-manu.org) and the University of Glasgow for supporting the biodiversity monitoring programme at the MLC. We gratefully acknowledge the financial support and encouragement of the TJMF Foundation and the Darwin Initiative for financial support of the Sustainable Manu project. The permit to conduct research was provided by the Ministerio de Agricultura of Peru (Authorisation Number 'Autorización No.'2904-2012-AG-DGFFS-DGEFFS). Data available from: DOI – 10.5525/gla.researchdata.242

Literature Cited

Alroy J (2017) Effects of habitat disturbance on tropical forest biodiversity. Proc Natl Acad Sci U S A 201611855. doi: 10.1073/pnas.1611855114

Anderson MJ, Walsh DCI (2013) Permanova, Anosim, Mantel Test Face Heterogeneous Dispersions : What Null Hypothesis Are You Testing? Ecol Monogr 83:557–574. doi: 10.1890/12-2010.1

Austin G, Riley T (1995) Portable bait traps for the study of butterflies. Trop Lepid 6:5–9.

Barlow J, Araujo IS, Overal WL, et al (2008) Diversity and composition of fruit-feeding butterflies in tropical Eucalyptus plantations. Biodivers Conserv 17:1089–1104. doi: 10.1007/s10531-007-9240-0

Barlow J, Gardner T a, Araujo IS, et al (2007a) Quantifying the biodiversity value of tropical primary, secondary, and plantation forests. Proc Natl Acad Sci U S A 104:18555–60. doi: 10.1073/pnas.0703333104

Barlow J, Mestre LAM, Gardner TA, Peres CA (2007b) The value of primary, secondary and plantation forests for Amazonian birds. Biol Conserv 136:212–231. doi: 10.1016/j.biocon.2006.11.021

Beccaloni GW, Gaston KJ (1995) Predicting the species richness of neotropical forest butterflies: Ithomiinae (Lepidoptera: Nymphalidae) as indicators. Biol Conserv 71:77–86. doi: 10.1016/0006-3207(94)00023-J

Bonebrake TC, Ponisio LC, Boggs CL, Ehrlich PR (2010) More than just indicators : A review of tropical butterfly ecology and conservation. Biol Conserv 143:1831–1841. doi: 10.1016/j.biocon.2010.04.044

Bouchard M, Hébert C (2016) Beetle community response to residual forest patch size in managed boreal forest landscapes: Feeding habits matter. For Ecol Manage 368:63–70. doi: 10.1016/j.foreco.2016.02.029

Brown KS (1997) Diversity, disturbance, and sustainable use of Neotropical forests: insects as indicators for conservation monitoring. J Insect Conserv 1:25–42. doi: 10.1023/A:1018422807610

Brown KS, Freitas AVL (2000) Atlantic Forest Butterflies : Indicators for Landscape Conservation '. 32:934–956.

Budiharta S, Meijaard E, Erskine PD, et al (2014) Restoring degraded tropical forests for carbon and biodiversity. Environ Res Lett 9:114020. doi: 10.1088/1748-9326/9/11/114020

Burivalova Z, Şekercioğlu ÇH, Koh LP (2014) Thresholds of Logging Intensity to Maintain Tropical Forest Biodiversity.

Davies KF, Margules CR, Lawrence JF (2000) Which traits of species predict population declines in experimental forest fragments? Ecology 81:1450–1461.

de Andrade RB, Balch JK, Carreira JYO, et al (2017) The impacts of recurrent fires on diversity of fruit-feeding butterflies in a south-eastern Amazon forest. J Trop Ecol 33:22–32. doi: 10.1017/S0266467416000559

DeVries, P. Murray, D. Lande R (1997) Species diversity in vertical, horizontal, and temporal dimensions of a fruit-feeding butterfly community in an Ecuadorian rainforest. Biol J Linn Soc 62:343–364. doi: 10.1111/j.1095-8312.1997.tb01630.x

DeVries P, Lande R, Murray D (1999) Associations of co-mimetic ithomiine butterflies on small spatial and temporal scales in a neotropical rainforest.

Devries PJ, Walla TR (2001) Species diversity and community structure in neotropical fruit-feeding butterflies. Biol J Linn Soc 74:1–15. doi: 10.1006/bijl.2001.0571

Dufrene M, Legendre P (1997) SPECIES ASSEMBLAGES AND INDICATOR SPECIES : THE NEED FOR A FLEXIBLE ASYMMETRICAL APPROACH. 67:345–366.

Gardner TA, Barlow J, Araujo IS, et al (2008) The cost-effectiveness of biodiversity surveys in tropical forests. Ecol Lett 11:139–150. doi: 10.1111/j.1461-0248.2007.01133.x

Gray MA, Baldauf SL, Mayhew PJ, Hill JK (2007) The Response of Avian Feeding Guilds to Tropical Forest Disturbance. 21:133–141. doi: 10.1111/j.1523-1739.2006.00557.x

Hall J, Willmott K (2000) Patterns of feeding behaviour in adult male riodinid butterflies and their relationship to morphology and ecology. Biol J Linn Soc 69:1–23.

Hamer KC, Hill JK, Benedick S, et al (2003) Ecology of butterflies in natural and selectively logged forests of northern Borneo: The importance of habitat heterogeneity. J Appl Ecol 40:150–162. doi: 10.1046/j.1365-2664.2003.00783.x

Hamer KC, Hill JK, Benedick S, et al (2006) Diversity and ecology of carrion- and fruit-feeding butterflies in Bornean rain forest. 22:25–33. doi: 10.1017/S0266467405002750

Holloway JD, Barlow HS, Loong HK, Khen CV (2013) Sweet or Savoury? Adult feeding preferences of Lepidoptera attracted to banana and prawn baits in the oriental tropics. Raffles Bull Zool 71–90.

Horner-Devine MC, Daily GC, Ehrlich PR, Boggs CL (2003) Countryside Biogeography of Tropical Butterflies\rBiogeografía de Paisaje de Mariposas Tropicales. Conserv Biol 17:168–177. doi: 10.1046/j.1523-1739.2003.01310.x

Hughes J, Daily G, Ehrlich P (1998) Use of fruit bait traps for monitoring of butterflies

(Lepidoptera: Nymphalidae).

Keenan RJ, Reams GA, Achard F, et al (2015) Dynamics of global forest area: Results from the FAO Global Forest Resources Assessment 2015. For Ecol Manage 352:9–20. doi: 10.1016/j.foreco.2015.06.014

Kim D-H, Sexton JO, Townshend JR (2015) Accelerated deforestation in the humid tropics from the 1990s to the 2000s. Geophys Res Lett 42:3495–3501. doi: 10.1002/2014GL062777

Kudavidanage EP, Wanger TC, Alwis C, et al (2012) Amphibian and butterfly diversity across a tropical land-use gradient in Sri Lanka; implications for conservation decision making. Anim Conserv 15:253–265. doi: 10.1111/j.1469-1795.2011.00507.x

Kwon T-S, Lee C, Sung J (2014) Diversity decrease of ant (Formicidae, Hymenoptera) after a forest disturbance: different responses among functional guilds. Zool Stud 53:37. doi: 10.1186/s40555-014-0037-z

Laurance W, Pérez-Salicrup D, Delamônica P (2001) Rain forest fragmentation and the structure of Amazonian liana communities.

Lawton JH, Bignell DE, Bolton B, et al (1998) Biodiversity inventories, indicator taxa and effects of habitat modification in tropical forest. Nat. 391:72–76.

Lucci Freitas AV, Agra Iserhard C, Pereira Santos J, et al (2014) Studies with butterfly bait traps: an overview. Rev Colomb Entomol 40:203–212.

Molina-Martínez A, León-Cortés JL, Regan HM, et al (2016) Changes in butterfly distributions and species assemblages on a Neotropical mountain range in response to global warming and anthropogenic land use. Divers Distrib 22:1085–1098. doi: 10.1111/ddi.12473

Molleman F, Grunsven RHA, Liefting M, et al (2005) Is male puddling behaviour of tropical butterflies targeted at sodium for nuptial gifts or activity? Biol J Linn Soc 86:345–361. doi: 10.1111/j.1095-8312.2005.00539.x

Nyafwono M, Valtonen A, Nyeko P, Roininen H (2014) Fruit-feeding butterfly communities as indicators of forest restoration in an Afro-tropical rainforest. Biol Conserv 174:75–83. doi: 10.1016/j.biocon.2014.03.022

Oksanen J, Blanchet FG, Kindt R, et al (2013) Package “vegan”. Community ecology package, version.

Paudel E, Dossa G, Blécourt M de (2015) Quantifying the factors affecting leaf litter decomposition across a tropical forest disturbance gradient.

Putz FE, Zuidema PA, Synnott T, et al (2012) Sustaining conservation values in selectively logged tropical forests: The attained and the attainable. Conserv Lett 5:296–303. doi: 10.1111/j.1755-263X.2012.00242.x

R Core Team (2013) R: A language and environment for statistical computing.

Ribeiro DB, Freitas AVL (2012) The effect of reduced-impact logging on fruit-feeding butterflies in Central Amazon, Brazil. J Insect Conserv 16:733–744. doi: 10.1007/s10841-012-9458-3

RIBEIRO DB, FREITAS AVL (2011) Large-sized insects show stronger seasonality than small-sized ones: a case study of fruit-feeding butterflies. Biol J Linn Soc 104:820–827. doi: 10.1111/j.1095-8312.2011.01771.x

Ross KA, Fox BJ, Fox MD (2002) Changes to plant species richness in forest fragments: Fragment age, disturbance and fire history may be as important as area. J Biogeogr 29:749–765. doi: 10.1046/j.1365-2699.2002.00722.x

Scriven SA, Beale CM, Benedick S, Hill JK (2017) Barriers to dispersal of rain forest butterflies in tropical agricultural landscapes. Biotropica 49:206–216. doi: 10.1111/btp.12397

Solar RR de C, Barlow J, Andersen AN, et al (2016) Biodiversity consequences of land-use change and forest disturbance in the Amazon: A multi-scale assessment using ant communities. Biol Conserv 197:98–107. doi: 10.1016/j.biocon.2016.03.005

Sourakov A, Emmel TC (1995) Bait trapping for butterflies in Kenya. Trop Lepid 6:1–2.

Sparrow HR, Sisk TD, Ehrlich PR, Murphy DD (1994) Techniques and guidelines for monitoring neotropical butterflies. Conserv Biol 8:800–809. doi: 10.1046/j.1523-1739.1994.08030800.x

Tyukavina A, Hansen MC, Potapov P V., et al (2017) Types and rates of forest disturbance in Brazilian Legal Amazon, 2000–2013.

Whitworth A, Beirne C, Rowe J, et al (2015) The response of faunal biodiversity to an unmarked road in the Western Amazon. Biodivers Conserv. doi: 10.1007/s10531-015-0883-y

Whitworth A, Downie R, May R Von, et al (2016a) How much potential biodiversity and conservation value can a regenerating rainforest provide ? A “ best - case scenario ” approach from the Peruvian Amazon. Trop Conserv Sci 9:224–245.

Whitworth A, Villacampa J, Brown A, Huarcaya RP (2016b) Past Human Disturbance Effects upon Biodiversity are Greatest in the Canopy ; A Case Study on Rainforest Butterflies. doi: 10.1371/journal.pone.0150520

Whitworth A, Villacampa J, Serrano Rojas SJ, et al (2017) Methods matter: Different biodiversity survey methodologies identify contrasting biodiversity patterns in a human modified rainforest — A case study with amphibians. Ecol Indic. doi: 10.1016/j.ecolind.2016.08.055

Wood B, Gillman MP (1998) The effects of disturbance on forest butter ¯ ies using two methods of sampling in Trinidad.