Estimating species diversity in a guild of Neotropical skippers (Lepidoptera: Hesperiidae) with artificial lures is a sampling problem

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Abstract. 1. Butterflies are frequently used to investigate Neotropical diversity, but the family Hesperiidae is almost never employed as a focal group. Sampling Hesperiidae with artificial lures has been used to assist in species richness estimates of males that use bird droppings as a resource, but its effectiveness for estimating total hesperiid site diversity is unknown.

2. This study characterises species richness and abundance in a diverse assemblage of Hesperiidae sampled at artificial lures. Sampled and estimated richness among taxonomic subsets were compared to the known site richness. These comparisons are used to assess artificial lures as a method for sampling total richness of Hesperiidae. We tested for potential differences in attraction in lures placed with and without the presence of army ant swarms.

3. Five years of intermittent sampling with lures recovered 65% of the known Hesperiidae species richness at the study site. We found the subfamilies were differentially attracted to lures, where 86% of Eudaminae, 56% of Hesperiinae and 55% of Pyrginae species known from the site were recovered by lures. Species accumulation and taxonomic diversity differed among lures placed alone, and those placed in the presence of army ants.

4. Our results suggest that sampling with artificial lures can be used to estimate diversity of Neotropical Eudaminae and a few tribes of Hesperiinae at some sites. Available evidence indicates that Hesperiidae feeding at bird droppings is predominately a Neotropical phenomenon. Finally, we conclude that estimating diversity in Neotropical Hesperiidae will require long-term studies that use multiple sampling techniques.

Key words. Artificial lures, bird droppings, diversity, Neotropical Hesperiidae, sampling.

The empirical measurement of diversity is central to understanding biological communities, including their dynamics and structure, and for making conservation decisions that concern them. Measuring diversity is particularly urgent in little-known tropical habitats due to their rapid destruction by both global economic and human population growth (Caughly & Gunn, 1996; Laurance & Peres, 2006; Nepstad et al., 2006). Despite interest in Neotropical habitats, few sites have been studied in any detail (Leigh et al., 1982; Gentry, 1990; McDade et al., 1994; Hubbell et al., 1999), and our understanding of focal groups in most areas is largely confined to lists of species without estimates of relative abundance (e.g., Morellato, 1992; Foster et al., 1994; Lamas, 1994; Wilson & Sandoval, 1996). Although species richness is perhaps the most universally used measure of diversity, a deeper understanding of biological diversity requires knowing something about the relative abundance among species. While there has been much discussion on what information various measures of diversity may impart about the biological communities under consideration (Gotelli & Graves, 1996; Boulinder et al., 1998; Gotelli & Colwell, 2001; Jost, 2007), the reliability of diversity measures depends on adequate sampling.

Butterflies have been used frequently to investigate various aspects of tropical insect diversity, including studies focused on temporal and spatial dynamics of communities, and the effects
of disturbance on their diversity (Brown, 1997; Shahabuddin & Terborgh, 1999; DeVries & Walla, 2001; Molleman et al., 2006). Among tropical butterfly studies, the estimation of species richness is a common theme, and while there are many short-term site estimates of tropical butterfly richness (e.g., Spitzer et al., 1997; Walpole & Sheldon, 1999; Caldas & Robbins, 2003; Lien & Yuan, 2003), long-term studies that use multiple sampling protocols provide the best estimates of tropical species diversity (DeVries, 1987; Raguso & Llorente-Bosquets, 1990; Lamas, 1994; Robbins et al., 1996).

Butterflies are composed of two monophyletic groups: Papilionoidea (true butterflies) traditionally including four or five families, and Hesperioidea (skippers) represented by the single family Hesperiidae (Ackery et al., 1999). The Hesperiidae includes seven monophyletic subfamilies accounting for about 20% of global butterfly richness, but most species are in the subfamilies Eudaminae, Hesperini, and Pyrginae (Warren, 2006; Warren et al., 2008). Despite comprising 30% of all Neotropical butterfly species, work on skippers has been primarily taxonomic or focused on the production of site lists (see Lamas et al., 1995). In contrast, few investigations have explored skipper ecology or behaviour, and no study has incorporated species abundances to describe any assemblage of Neotropical skippers.

Adult butterflies feed entirely on liquid resources that may include flower nectar, liquids from rotting fruit or carcasses, faeces and damp soil (Norris, 1936; Downes, 1973; Boggs & Jackson, 1991; Beck & Feidler, 1999). Some resources are visited only by one sex (Pliske, 1975; Ray & Andrews, 1980; Sculley & Boggs, 1996), and it has long been known that male Neotropical skippers may be found feeding on fresh faeces left by birds following swarms of raiding army ants (Zikán, 1929; Austin et al., 1993; Lamas et al., 1993; Vieira, 2004). In fact, the attraction of this feeding guild of skippers to fresh bird faeces is so strong that artificial lures resembling bird droppings have been used to facilitate surveys and ecological studies of these butterflies (Austin et al., 1993; Lamas et al., 1993; DeVries et al., 2008).

Recently, we employed standardised transects baited with artificial lures and sampled at 30-min intervals to generate a large data set from a species-rich assemblage of Neotropical skippers. Most, but not all, of that sample was used to demonstrate significant temporal diet partitions among subfamilies, genera, and species within genera, and show that differences in diet activity may contribute significantly to reproductive isolation among species in the assemblage (DeVries et al., 2008). The present investigation uses the complete sample to explore the effectiveness of artificial lures for estimating species diversity of Hesperiidae at our study site. To do so, we first provide the species abundances of the assemblage as sampled at artificial lures, and then compare the sampled and estimated richness among taxonomic subsets to the known richness of Hesperiidae at our study site. These comparisons are used to assess the effectiveness of artificial lures for sampling Neotropical Hesperiidae. We then ask if there were differences in the skipper diversity attracted to lures alone versus lures placed in the presence of army ant swarms. Finally, we discuss this guild of skippers in the context of global Hesperiidae, and suggest that the habit of feeding on bird droppings is a behaviour that occurs predominantly in the Neotropical subfamily Eudaminae.

Materials and methods

The study system

This study was conducted intermittently over 5 years (November 1992–October 1997) in lowland tropical rainforest located in the FAUTRON rainforest reserve, near Caucalândia in central Rondônia, Brazil. Varying intensities of disturbance occur at the approximately 2000-ha site, and there is a distinctly seasonal climate, with a pronounced dry season (May–September) and a wet season (October–April) marked by daily rains from late December to February. A detailed site description is provided in Emmel & Austin (1991). For over 8 years (1989–1997), the Hesperiidae were sampled extensively at this site by several workers using nets, traps and lights along forest trails, forest edges, streams, hilltops and light gaps, with particular attention to Hesperiidae that visit flowers, mud, and bird droppings. The comprehensive species list resulting from that work (G. T. Austin, unpubl.) comprises the known richness at the site, and provided a comparison to those sampled at artificial lures in the current study.

Over 800 h of sampling with lures that attract a feeding guild of male skippers (Austin et al., 1993) was conducted on 25 transects along forested trails. On each sampling day, a single 300-m transect was established by placing 100 saliva-moistened wads of toilet paper at approximately 3-m intervals on the upper surface of leaves from 0.5–1.0 m above the ground. Skippers typically landed on or near lures and attempted to feed on them (Fig. 1), whereupon they were collected with a hand net, placed into glassine envelopes labelled with sampling details, and subsequently identified to species. Transects were usually set up in the morning (between 07.00–09.00 hours) and monitored until sunset (18.00 hours) unless strong winds or heavy rain prevented observations. Transects set at different sites on successive days were walked at least twice every 30 min and samples were pooled by half-hour intervals, but all transects were pooled into a single data set and analysed here. Lures were remoistened with saliva as necessary. Occasionally lured transects coincided with active swarms of army ants in the genus Eciton (Formicidae: Ecitoninae). This observation was noted on the individual samples and used to test whether there were differences in skippers attracted to lures placed in the presence or absence of army ant swarms.

The higher classification of Hesperiidae follows Warren (2006) and Warren et al. (2008), species identifications were carried out by G.T.A., and then confirmed independently by O. H. H. Mielke, S. R. Steinhauser, and A. D. Warren. Voucher specimens are deposited at the Departamento de Zoologia, Universidade Federal do Paraná, Curitiba, Paraná, Brazil, and the McGuire Center for Lepidoptera and Biodiversity, Florida Museum of Natural History, Gainesville, Florida.

Analysis

We employed species accumulation curves and rank abundance plots to describe species richness and abundance of the total sample. To compare approximate levels of commonness and rarity among
species, we inspected rank abundance plots to divide samples into three abundance categories, and defined them as: > 200 individuals (= abundant), > 5 < 200 individuals (= common), and 5 or fewer individuals (= rare). To assess potential differential attraction to lures among subfamilies, we compared species accumulation curves of our samples taken at lures to the known total skipper richness at our study site (G. T. Austin, unpubl.).

Due to variation in behaviour of individual species and their interactions with the environment, any biodiversity sampling method must face the issue of underestimating true species richness. For this reason, estimation techniques are often used in diversity studies, particularly in samples taken intermittently over brief periods of time (Colwell & Coddington, 1994; Shen et al., 2003). Because the subfamily partitions contained multiple

Fig. 1. Various skipper species feeding on bird droppings and artificial lures in the field. (A) Astraptes anaphus (Eudaminae) feeding on a fresh bird droppping; (B) Telemiades penidas (Eudaminae), Aguna sp. (Eudaminae); (C) Muscelus assaricus (Pyrginae); (D) Phanus ecitonorum (Eudaminae); (E) Aguna asander (Eudaminae); (F) Marela tamyroides (Eudaminae) – all feeding at artificial lures.
singleton and doubleton species, we used the sampled-based species estimators jackknife 1, and its less conservative counterpart, jackknife 2 (summarised in Chao, 2005) to estimate the potential upper bounds of species richness in our lured samples. We then compared observed and the estimated lured richness to the comprehensive survey of our study site that employed various sampling methods over an 8-year period (G. T. Austin, unpubl.). This comparison allowed us to assess the efficiency of using lures for sampling Neotropical skipper diversity.

We tested whether lures placed in the presence or absence of army ant swarms attracted different sets of skippers in two ways. We first compared species accumulation curves of the subfamilies paired by the two lure placement types. Because sample sizes of Hesperiinae and particularly Pyrginae were lower than those of Eudaminae, we constructed the accumulation curves using a sample size shared among subfamilies and lure placement types (i.e., 1000 individuals). Second, we used a small subset of our data to ask if the relative proportions of collected individuals among subfamilies and species differed among lure placement types as well as among different sampling times. The subset consisted of samples taken in November 1992 and again in November 1994 because only these times had a sufficient number of both lure placement types to compare statistically. Using four separate $3 \times 2 \chi^2$ contingency tables, we tested whether the relative proportions collected among subfamilies differed (i) among years (separately for each lure placement type), and (ii) among lure placement types (separately for each year). To test whether the relative proportions of individuals collected among species differed both among lure placement types and among years, the proportions of the five most abundant species collected during both November samples were evaluated using four $5 \times 2 \chi^2$ contingency table tests. Again, we tested whether the relative proportions among the five most common species collected differed (i) among years separately for each lure placement type, and (ii) among lure placement types separately for each year.

### Table 1. Taxonomic richness sampled at lures compared with the known richness of Hesperiidae at the study site.

<table>
<thead>
<tr>
<th>Individual abundance</th>
<th>Genera</th>
<th>Percentage at lures</th>
<th>Species</th>
<th>Percentage at lures</th>
</tr>
</thead>
<tbody>
<tr>
<td>Eudaminae [8915]</td>
<td>35 (40)</td>
<td>87.5</td>
<td>158 (182)</td>
<td>86.8</td>
</tr>
<tr>
<td>Hesperiinae [4862]</td>
<td>72 (104)</td>
<td>69.2</td>
<td>152 (267)</td>
<td>56.5</td>
</tr>
<tr>
<td>Calpodini [4172]</td>
<td>21 (22)</td>
<td>95.4</td>
<td>62 (66)</td>
<td>92.4</td>
</tr>
<tr>
<td>Tribe 1 [99]</td>
<td>3 (4)</td>
<td>75.0</td>
<td>6 (7)</td>
<td>85.7</td>
</tr>
<tr>
<td>Moncini [344]</td>
<td>32 (48)</td>
<td>66.6</td>
<td>55 (130)</td>
<td>42.3</td>
</tr>
<tr>
<td>Hesperiini [141]</td>
<td>14 (26)</td>
<td>53.8</td>
<td>25 (56)</td>
<td>46.4</td>
</tr>
<tr>
<td>Erionotini [106]</td>
<td>2 (4)</td>
<td>50.0</td>
<td>4 (8)</td>
<td>50.0</td>
</tr>
<tr>
<td>Pyrginae [1494]</td>
<td>44 (67)</td>
<td>65.6</td>
<td>110 (199)</td>
<td>55.2</td>
</tr>
<tr>
<td>Pyrrhopygini [104]</td>
<td>9 (12)</td>
<td>75.0</td>
<td>27 (44)</td>
<td>61.3</td>
</tr>
<tr>
<td>Celaenorrhinini [66]</td>
<td>1 (1)</td>
<td>100</td>
<td>7 (8)</td>
<td>87.5</td>
</tr>
<tr>
<td>Carcharodini [80]</td>
<td>6 (15)</td>
<td>40.0</td>
<td>13 (47)</td>
<td>27.6</td>
</tr>
<tr>
<td>Erynnini [472]</td>
<td>10 (12)</td>
<td>83.3</td>
<td>25 (32)</td>
<td>78.1</td>
</tr>
<tr>
<td>Achlyodini [660]</td>
<td>10 (11)</td>
<td>90.9</td>
<td>25 (32)</td>
<td>78.1</td>
</tr>
<tr>
<td>Pyrgini [112]</td>
<td>8 (16)</td>
<td>50.0</td>
<td>13 (36)</td>
<td>36.1</td>
</tr>
<tr>
<td>TOTAL [15 271]</td>
<td>151 (211)</td>
<td>71.5</td>
<td>420 (648)</td>
<td>64.6</td>
</tr>
</tbody>
</table>

First column has total sample size of each taxonomic category in square brackets. Sampled generic and species richness is followed by the known richness at study site in parentheses. Note there are no tribal divisions available for the Eudaminae.

### Table 2. Categories of commonness and rarity among sampled species.

<table>
<thead>
<tr>
<th>Species abundance</th>
<th>Eudaminae</th>
<th>Hesperiinae</th>
<th>Pyrginae</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Abundant species</td>
<td>14 [5453]</td>
<td>6 [2881]</td>
<td>2 [466]</td>
<td>22 [8800]</td>
</tr>
<tr>
<td>Rare species</td>
<td>52 [134]</td>
<td>80 [175]</td>
<td>70 [164]</td>
<td>202 [473]</td>
</tr>
<tr>
<td>Doubletons</td>
<td>16 (10)</td>
<td>27 (18)</td>
<td>15 (14)</td>
<td>58 (14)</td>
</tr>
<tr>
<td>Singletons</td>
<td>14 (9)</td>
<td>31 (20)</td>
<td>27 (24)</td>
<td>72 (17)</td>
</tr>
</tbody>
</table>

The numbers in square brackets indicate the sample abundance. Categories are defined as: Rare = 5 or fewer, Common = > 5 < 200, Abundant = > 200. The categories of singletons and doubletons are included in rare species. Numbers in parentheses indicate percentage of doubleton and singleton species for the total observed species in each column category.

### Results

#### Total sample

Using artificial lures, we sampled 15 271 individual skippers (98% males) representing 420 species in three subfamilies: 158 species in 35 genera of Eudaminae, 152 species in 72 genera of Hesperiinae, and 110 species in 44 genera of Pyrginae (Table 1). There were considerable differences in sampled abundance among subfamilies with 8915 individuals in Eudaminae, 4862 in Hesperiinae, and 1494 in Pyrginae (Tables 1 & 2). Abundance distributions varied dramatically within the entire sample, among subfamily partitions, and among individual species (Figs 2, 3 and 4). There were 22 abundant species that accounted for 58% of the total abundance and only 5% of total richness, whereas the 202 rare species comprised 3% of the total abundance, but 48% of total richness (Table 2). Among subfamilies, 9% of Eudaminae, 4% of Hesperiinae, and 2% of Pyrginae richness...
was accounted for by abundant species, whereas rare species comprised 33% of Eudaminae, 53% of Hesperiinae and 64% of Pyrginae (Table 2).

**Partitions by subfamily**

The nearly asymptotic total species accumulation curve suggests we captured the majority of species that could be sampled in this assemblage (Fig. 2). Nevertheless, a very different picture emerges when subfamilies are considered independently. Here the accumulation curve for Eudaminae appears asymptotic, but curves for Hesperiinae and Pyrginae do not (Fig. 3), thus indicating that increased lure sampling effort would have recovered more species of Hesperiinae and Pyrginae (Table 1). Eudaminae had comparatively fewer singletons and doubletons than other subfamilies, further implying that this group was more thoroughly sampled (Fig. 4). However, considering these curves in light of the known richness of Hesperiidae at our site (Table 1) suggests there was strong differential attraction to lures among subfamilies, and by extension, that there are potentially different feeding habits among these groups. Thus, it seems unlikely that continued sampling with lures would have dramatically increased total species richness in Hesperiinae and Pyrginae.

**Species estimations**

To explore the effectiveness of lure sampling for capturing site diversity among groups, the upper bound jackknife estimates of our quantified observed samples were compared with the known species richness (Figs 2–4, Tables 1 and 3). As expected, both estimates inferred greater species richness than what was observed at lures, and at the family level inferred richness was
considerably higher that sampled richness (Table 3). Jackknife estimates closely approximated the known richness in Eudaminae (Table 3). Given the apparent differential attraction to lures among subfamilies (Fig. 3), however, it was not surprising that when compared to the known species richness, the jackknife methods underestimated richness at all other taxonomic partitions (Table 3). These comparisons, in concert with species accumulation (Fig. 3) and abundance distributions (Tables 1 and 2) all suggest that sampling with lures considerably underestimated total species richness of Hesperiinae and Pyrginae at the study site (Tables 1 and 3), reinforcing the idea that feeding behaviours differ among the subfamilies, and that some groups are not attracted to lures.

Comparison of lure placement

Species accumulation curves indicated differences among subfamilies with respect to samples taken at lures placed in association with army ants and samples taken at lures alone (Fig. 5). Species accumulations of Eudaminae and Hesperiinae were faster on lures alone, while species accumulation of Pyrginae was faster on lures in the presence of ants. This differential accumulation provides further support for differential attraction among subfamilies to lures (i.e., Fig. 3), including an effect of army ant swarms. Results from $\chi^2$ contingency table analyses (Table 4A) revealed significant heterogeneity among subfamily and species abundances sampled in November 1992 and November 1994 regardless of lure placement type (subfamilies: lures only: $\chi^2 = 20.8$, d.f. = 2, $P < 0.0001$; ants and lures: $\chi^2 = 36.9$, d.f. = 2, $P < 0.0001$; species: lures only: $\chi^2 = 93.9$, d.f. = 4, $P < 0.0001$; ants and lures: $\chi^2 = 68.7$, d.f. = 4, $P < 0.0001$, Table 4), indicating significant abundance variation among subfamilies and species among years. Furthermore, examining each of the November 1992 and 1994 samples separately (Table 4B), the $\chi^2$ contingency table analyses showed significant heterogeneity among subfamily and species abundances between the two sampling methods regardless of the sampling time (subfamilies: November 1992: $\chi^2 = 8.58$, d.f. = 2, $P = 0.014$; November 1994: $\chi^2 = 14.8$, d.f. = 2, $P < 0.0001$; species: November 1992: $\chi^2 = 24.9$, d.f. = 2, $P < 0.001$; November 1994: $\chi^2 = 22.7$, d.f. = 4, $P < 0.001$), providing more evidence for differential attraction among subfamilies and common species with respect to lure placement.

Table 3. Assessing lure effectiveness by comparing sampled and estimated richness with the known species richness.

<table>
<thead>
<tr>
<th></th>
<th>Known richness</th>
<th>Sampled richness</th>
<th>Jacknife 1 estimate</th>
<th>Jacknife 2 estimate</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hesperiidae</td>
<td>648</td>
<td>419</td>
<td>491</td>
<td>505</td>
</tr>
<tr>
<td>Eudaminae</td>
<td>182</td>
<td>158</td>
<td>172</td>
<td>170</td>
</tr>
<tr>
<td>Hesperiinae</td>
<td>267</td>
<td>151</td>
<td>182</td>
<td>186</td>
</tr>
<tr>
<td>Pyrginae</td>
<td>199</td>
<td>110</td>
<td>137</td>
<td>149</td>
</tr>
</tbody>
</table>

A, known, sampled, and estimated richness at the study site. B, variation in sampled and estimated richness compared to the known richness. Variation was measured as known richness – sampled or estimated richness, divided by the known richness, and this provided the percentage of under or overestimation.

Fig. 4. Rank abundance curves of individual subfamilies sampled at artificial lures. Note the variation in rare species among the subfamilies. Abbreviations: E, Eudaminae; H, Hesperiinae; P, Pyrginae.
patterns, particularly samples taken at baits or lures. It is also within a focal group are vital for uncovering real biological as well as describing biological communities. Often, however, to assess its usefulness in estimating local species diversity as important to appreciate the limitations of any sampling method unrecognised, considered insignificant, or untested. Under the effectiveness of a sample for reflecting reality is unknown, and/or artificial lures, this investigation focused on Neotropical a predominately male feeding guild that visits bird droppings and/or taxonomically better-known families Papilionidae, Pieridae, and Nymphalidae (e.g., Beccaloni & Gaston, 1995; Robbins most investigations of tropical butterfly diversity have focused 1996; Brown, 1997; Brown & Freitas, 2000). However, almost nothing is known about the comparative effectiveness among the traditional sampling of Neotropical Hesperiidae to detect otherwise rarely encountered taxa, and suggested using lures to standardise sampling of skippers associated with army ant swarms (Austin et al., 1993; Lamas et al., 1993). As a caveat, however, there is evidence accumulating that lures may not be effective everywhere in the Neotropics. For example, hesperiids were not attracted to lures in central Brazil (Vieira, 2004) nor in Guatemala (G. T. Austin, pers. obs.), and the technique was unsuccessful at one site in Amazonian Ecuador (P. J. DeVries, pers. obs.). The reasons why hesperiids in some assemblages are not attracted to artificial lures is unknown, but we do know that skippers do visit bird droppings at those same sites (G. T. Austin & P. J. DeVries, pers. obs.). Nevertheless, using lures at our study site in central Rondônia, Brazil, yielded a large sample that allowed insights into aspects of behaviour such as temporal partitioning and community structure not otherwise possible (DeVries et al., 2008, and this study).

This study revealed key strengths and weaknesses of using lures to sample Neotropical skipper diversity. For example, the accumulation curve for all lured samples (Fig. 2) gives a potentially misleading impression of sampling effectiveness at the family level inasmuch as its asymptote accounts for only about two-thirds of the known richness (Table 1). When the subfamilies were plotted separately, it was apparent that there was differential attraction to lures among them (Fig. 3). Here Eudaminae had the highest sampled richness, nearly twice the abundance of Hesperiinae, and nearly six times the abundance of Pyrginae (Fig. 4, Tables 1 and 2). The accumulation curve for Eudaminae is asymptotic at a level approaching their known richness, whereas the curves for Hesperiinae and especially Pyrginae are non-asymptotic and far below their known local richness (Tables 1 and 2). This strongly supports the observations of Austin et al. (1993) who suggested that members of the Eudaminae are particularly attracted to lures. The indication of differential attraction here is important because it emphasises that

Table 4. Heterogeneity in abundance of Hesperiidae sampled at different lure placements. A, number of individual skippers sampled within each subfamily for the periods November 1992 and November 1994. B, number of individuals sampled among the five most common species (all Eudaminae) during November 1992 and November 1994. For tables A and B, ‘lures’ represent lures placed in the absence of army ants. ‘Ants’ represent lures placed in the presence of army ants. The $\chi^2$ tests for heterogeneity performed on these data are described in the text.

<table>
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<tr>
<th></th>
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</thead>
<tbody>
<tr>
<td>Eudaminae</td>
<td>920</td>
<td>324</td>
<td>686</td>
<td>693</td>
</tr>
<tr>
<td>Hesperiinae</td>
<td>184</td>
<td>74</td>
<td>190</td>
<td>236</td>
</tr>
<tr>
<td>Pyrginae</td>
<td>114</td>
<td>99</td>
<td>81</td>
<td>156</td>
</tr>
</tbody>
</table>

Astroptes fulgerator 107 9 101 50
Aguna squamalba 42 59 9 73
Epargyreus clavicornis 51 36 27 70
Urbanus pronta 44 13 31 44
Phanus vitreus 42 24 53 27

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in studies of taxonomically rich insect families like Hesperiidae, using accumulation curves partitioned by subfamilies may provide a clearer assessment of sampling effectiveness than a single, pooled curve for an entire family.

Although lures clearly attracted more Eudaminae than either Pyrginae or Hesperiinae (Fig. 3, Table 1), members of some tribes of Hesperiinae and Pyrginae were well sampled at lures, even though they represent taxa only rarely sampled with other methods. For example, most Calpodini (sensu Warren, 2006) are regularly attracted to lures, but other tribes of Hesperiinae only rarely. Likewise, within the Pyrginae members of Erynnini and Achlyodidini are frequent at lures, while those of Carcharodini and Pyrgini are not (Table 1; G. T. Austin, pers. obs.). In general, such observations seem to indicate that the fundamental adult food resources of male Neotropical Hesperiinae and Pyrginae may differ from those of Eudaminae.

Comparing the known richness of Hesperiidae at the study site (Table 1) with lure-sampled accumulation curves partitioned by subfamilies (Fig. 3) and the upper bound estimates of jackknife procedures (Tables 1 and 3) showed that lured samples alone strongly underestimated total species richness because some groups appear not to be readily attracted to lures. Therefore, obtaining a more thorough measure of their diversity requires using other sampling methods in concert with lures. Nonetheless, we believe that by focusing on Eudaminae and select tribes of Hesperiinae (see Table 1), artificial lures have potential in future diversity studies on Hesperiidae, and for probing ecological and behavioural patterns within this skipper guild (i.e., Austin et al., 1993; DeVries et al., 2008).

This study showed significant differences in subfamily and species-level responses to artificial lures placed in the presence or absence of army ant swarms. At the level of subfamily, the Eudaminae and Hesperiinae accumulated species faster at lures alone, while the Pyrginae accumulated species faster at lures in the presence of army ants (Fig. 5). While the reason for this disparity in lure attraction remains unknown, members of Pyrginae may be responding to visual or olfactory cues that were not present in lures alone. However, once again such observations strongly imply there was differential attraction to lures among subfamilies and particular tribes (Fig. 3, Table 1), and that male skippers utilise different resources among these taxonomic groups.

Analyzing a small subset of the entire sample, we found temporal abundance differences at the subfamily and species levels with markedly different abundances between November 1992 and November 1994 (Table 4). The disparity in abundance is not unexpected given long-term studies on tropical butterflies showing year-to-year variation is the norm (e.g., DeVries & Walla, 2001, and unpublished). Hence, the significant differences in abundances at lure placement types (Table 4) may be due to inter-annual variation. We have no indication from 8 years of sampling that Hesperini and Pyrginae are proportionally less abundant than Eudaminae at this site. However, in order to better understand the temporal dynamics of this species assemblage of Hesperiidae, a more rigorous sampling regime will be required than that employed here.

The phenomenon of skippers feeding at bird droppings is well documented in the Neotropics (see Austin et al., 1993; DeVries et al., 2008 and references therein). However, the near absence of records indicating this behaviour outside of the Neotropics is remarkable. For example, in African forest some members of the genera Acleros, Gretina, Osmodes, Meza, Platylesches and Pteroetionon (Hesperini: Erionotini) may visit fresh bird droppings, while members of Abantis (Pyrginae), Kedestes (Hesperini: Erionotini) and Coeliades (Coeliadinae) do so in African savannah (S. Collins & F. Molleman, pers. comm.).

We found no reference to this behaviour for any Southeast Asian skippers, but males in the genera Bibasis, Burarana, Hasora (Coeliadinae) and Halpe, Ancistroides and Pituitaria (Hesperiinae) are reported to feed on carrion (Corbet et al., 1992; Hamet et al., 2006). Finally, in Australia members of Mesodina (Trapezitinae) are the only taxa reported to feed on bird droppings (Atkins, 1989). While the Eudaminae accounts for more than 400 Neotropical species, it is a trivial component of Old World skipper diversity (Warren, 2006), and all evidence indicates that male Eudaminae constitute the only major group of Hesperiidae that regularly use bird droppings as resources. Thus, it appears that this feeding behaviour is primarily, but not exclusively, a Neotropical phenomenon that is likely driven by birds that follow army ants (Willis, 1967; Willis & Oniki, 1978) who provide an ephemeral concentration of bird droppings that is fed upon by skippers (Austin et al., 1993). However, outside the context of army ant swarms we note that the behaviour is rarely reported, despite it being frequently observed in the field (G. T. Austin & P. J. DeVries, pers. obs.), and that no Old World avian groups can be considered analogous to Neotropical ant birds (D. W. Steadman, pers. comm.). Given that bird droppings and members of the Hesperini are ubiquitous in most terrestrial habitats, and army ant-like behaviours have evolved among various groups of ants (Hölldobler & Wilson, 1990), we hope this study serves to stimulate observations on skippers feeding at bird droppings outside of the Neotropical region. Although published observations are rare (summarised above), they suggest the tribe Erionotini (Hesperini) and the subfamily Coeliadinae may be a starting place for documenting feeding at bird droppings by Old World skippers.

Conducting and compiling complete species inventories frequently involves considerable amount of time and labour. Despite the tendency to vary widely with respect to known richness, estimation techniques are frequently employed in biodiversity surveys (see Chao, 2005; Mao & Colwell, 2005; Walther & Moor, 2005), particularly tropical ones. Regardless of how thoroughly tropical surveys are conducted, most that attempt to obtain close approximations of true species richness need to confront the fact of variation in abundance and behaviours among species at any particular sampling location. For example, long-term monthly monitoring has clearly demonstrated the dynamic nature of Neotropical fruit-feeding butterfly diversity, including the changing species composition and species abundances in space and time (e.g., DeVries & Walla, 2001; Engen et al., 2002; P. J. DeVries, unpublished). In more general surveys, some species may be genuinely common, but yet remain undetected by particular sampling methods, or they do not occur in the spatial or temporal habitat being sampled (e.g., Green & Young, 1993; Longino et al., 2002; Thompson, 2004; P. J. DeVries & G. T. Austin, pers. obs.). In this investigation, we
used multiple comparisons on a large sample of Hesperiidae to emphasise how sampling with artificial lures underestimated total species richness, particularly among certain taxonomic subsets (Tables 1, 2 and 3), and to illustrate quantitatively how sampling methods influence diversity measures in this group.

As suggested for other insect groups, and perhaps best demonstrated for ants (Longino et al., 2002), an accurate species inventory requires multiple methods, considerable effort, and time. If approximating true species richness of butterflies at a particular site is a major objective, investigations need to obtain long-term quantitative samples, use a variety of sampling methods, and to compare taxonomic partitions below the family level. After all, conducting a biological survey is a sampling problem, and to compare taxonomic partitions below the family level.

We hope this paper serves to stimulate long-term investigations into the diversity and biology of butterflies, and Neotropical Hesperiidae in particular.

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References


Vieira, R.S. (2004) *Efeito da Fragmentação Florestal Sobre Borboletas (Lepidoptera, Hesperiidae) Associadas a Formiga-de-Correição Eciton burchelli (Hymenoptera, Formicidae, Ecitoninae).* Tese, Universidade Federal do São Carlos, São Paulo, Brazil.


