Article

The Effects of Light Environment on Adult Odonate Communities in Disturbed and Intact Forest: The Importance of Small-Scale Effects

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Abstract: Deforestation in the tropics causes shifts in adult odonate community structure, from forests dominated by thermoconforming zygopteran specialists to open areas with higher representations of heliothermic anisopterans. We tested for these shifts in the Maquina and Cuecha rivers in Monteverde, Costa Rica. We compared adult odonate communities in 100 m plots (subdivided into twenty 5 m subplots) located in disturbed, partially open areas with those in 100 m plots located in intact forest and used general linear models to describe how odonate abundance, species richness, species diversity, and the Anisoptera/Zygoptera ratio varied among plots, subplots, habitat type (disturbed/forested), rivers, and as functions of percent canopy cover and light levels. Plots varied in light levels and percent canopy cover, but there were no significant differences in species richness or diversity. Community composition, however, varied across plots and subplots in NMDS and PERMANOVA analyses, largely as a consequence of the preference of *Hetaerina cruentata* and *Paltothemis lineatipes* for high light subplots and *H. majuscula* for low light subplots. NMDS axes were significantly correlated with percent canopy cover and light level in subplots, and the Anisoptera/Zygoptera ratio correlated with NMDS axes at both the plot and subplot scales, indicating that the relative abundance of anisopterans did increase with increasing light and decreasing canopy cover. Differences among plots and habitats can largely be attributed to species-specific differences in habitat selection at a small spatial scale, causing predicted shifts in the Anisoptera/Zygoptera ratio as dominance shifts from endemic forest species to wide-ranging generalists. This is one of the first studies that confirms these patterns for a cloud forest community.

Keywords: Odonata; dragonfly; community ecology; Anisoptera; Zygoptera

1. Introduction

Deforestation and the loss of riparian zones has dramatic and distinctive effects on lotic dragonfly (Insecta: Odonata) communities [1–6]. In addition to the obvious increases in sunlight, wind, and heat load affecting adults, deforestation also causes increased runoff, siltation, pollution, and a decline in the quality of aquatic systems upon which odonate larvae depend [7–9]. Indeed, because odonates are sensitive to changes in both terrestrial and aquatic components of the ecosystem, the loss of odonate diversity and changes to odonate community structure are indicators of habitat disturbance [10–13].

One of the most common metrics that reflects how odonate community structure correlates with habitat disturbance is the 'Zygoptera/Anisoptera ratio' [6,14]. Most damselflies (suborder: Zygoptera) are small, thin, shade-tolerant thermoconformers that dominate in intact forest. They have poor dispersal abilities and are dependent on the integrity of the local environment, probably contributing to the high rate of endemism among forest zygopterans in the tropics [15]. Dragonflies (suborder: Anisoptera), on the other hand, are often large and either endothermic or heliothermic [16]. Heliothermic species prefer open

habitats, including those created by forest loss. Based on these ecophysiological differences between suborders, shaded forest habitats should have a higher Zygoptera/Anisoptera ratio than open habitats [14,17–21]. In addition, these changes in the dominance of one suborder or the other may be related to differences in the dramatic structural/functional differences in their larvae, the variation in microhabitats that they exploit, and the homogenization of aquatic habitats that deforestation causes [7]. Because this ratio decreases with decreasing canopy cover, it has been promoted as a quick index for measuring habitat disturbance in lowland tropical ecosystems [6,14]. Although the ratio has been a key community descriptor in many studies in tropical savannah [22] and tropical rainforest [6,11], only a few studies have examined this pattern in high-elevation tropical or subtropical cloud forests [23–25].

Costa Rica is an excellent place to examine the effects of canopy cover on tropical odonate communities. Although the country experienced a wave of deforestation from 1970–1990, a more recent commitment to reforestation—spurred by the Mesoamerican Corridor Project—means that there are increasing amounts of secondary forests restoring canopy cover to previously impacted waterways, in addition to the large blocks of protected pristine primary forest [26–30]. Also, as a result of the efforts of several distinguished odonatologists, the odonate species are well-described [31–33].

Previous studies on Costa Rican odonate communities focused on rainforests, describing the composition of anisopteran communities [34,35] and the effects of deforestation and canopy cover on community composition [36]. As in the other studies mentioned above, Hofhansl and Schneeweis [36] found that agricultural areas were dominated by widespread species while forests were dominated by species with more restricted ranges, including forest endemics.

Given the limited range of cloud forests, their geographic isolation on montane islands, and their particular sensitivity to climate change [37], it is critical to understand the relationships between forest cover and community composition. This study had three objectives. First, we described the general composition of this cloud forest community, which is an understudied tropical odonate system. Second, we tested whether disturbed and forested areas had different communities and examined the effects of light availability and canopy cover at two spatial scales to determine which scale might be more important to adult odonate behavior. Third, we tested whether the relative abundance of zygopterans and anisopterans changes as a function of habitat disturbance and light level.

2. Methods

Adult dragonflies were surveyed in Monteverde, Costa Rica, on the Cuecha and Maquina rivers, which are approximately 1 km apart (Figure 1a). The streams were similar in their physical characteristics; they were approximately 5 m wide with sandy-bottomed pools connected by shallow riffles. Two 100 m plots were established on each river (Figure 1a). On the Maquina, the upstream plot was in a young secondary forest (Figure 1b, 10.310006 N, 84.810397 W; elevation 1401 m), and the second plot was 100 m downstream (Figure 1c, 10.309865 N, 84.811545 W; elevation 1380 m) in an open area transitioning to a disturbed successional forest upstream. On the Cuecha, the upstream plot was within a mature secondary cloud forest of the Curi Cancha Reserve (Figure 1d, 10.304416 N, 84.804914 W; elevation 1445 m); the second plot was 500 m downstream, in a successional open forest behind the Riochante Community Center, and just downstream from Monteverde Dairy and the dairy farm (Figure 1e, 10.304552 N, 84.809319 W; elevation 1400 m). As such, we sampled a mature secondary forest habitat and a more open, disturbed habitat along each river.

The four sites were each sampled seven times from May–June 2023 at the end of a protracted dry season, at approximately weekly intervals, following standard methodologies [11,38]. Each plot was subdivided into 20 5 m subplots, extending 1 m on each side of the channel. The number and species of all individuals in each subplot were recorded), with care taken not to double-count individuals from subplot to subplot. Species were identified

by eye using binoculars (Snypex© (Lynbrook, NY, USA) 10 × 32), or from photographs taken with a Sony® RX10III (Sony®, Beijing, China) with a 40–600 mm zoom-equivalent lens system. Some individuals were collected by net for in-hand identification and photographing. Species were identified by comparing photos with field guides [32,33]. Sampling was conducted between 10:00–15:00 during sunny periods.
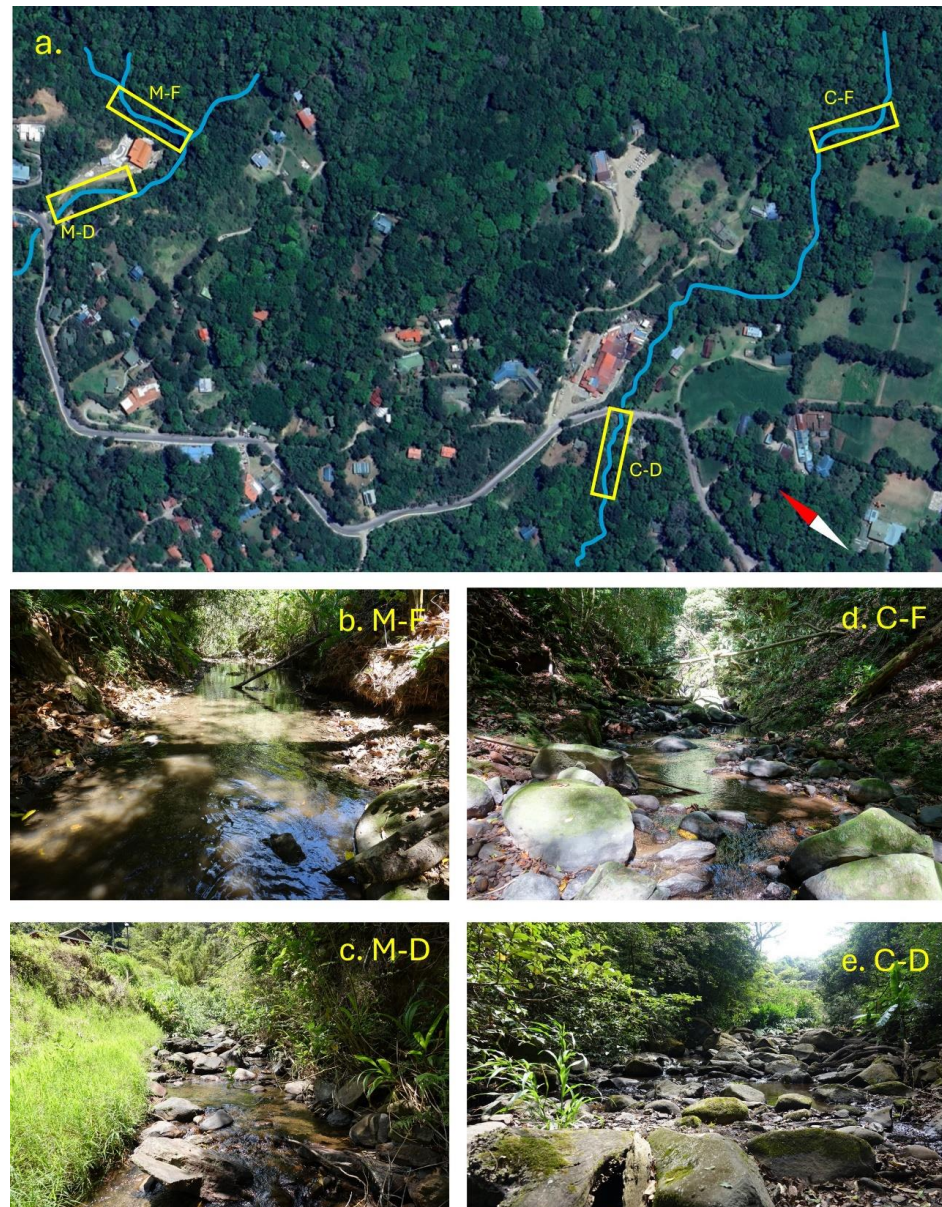

Figure 1. (**a**) Location of the plots on the Maquina and Cuecha rivers in Monteverde, Costa Rica, and photos of the sites: (**b**) Maquina forested plot (M-F), (**c**) Machina disturbed plot (M-D), (**d**) Cuecha forested plot (C-F), and (**e**) Cuecha disturbed plot (C-D). Site photo modified from Google Earth.

Light environment is critical to adult behavior, as habitat selection varies between thermoconforming, heliothermic, and endothermic species [15,18]. We measured light levels two ways. First, on a sunny day, light levels in each subplot were ranked from 1–5 (low light to high light) at each subplot. We also measured percent canopy cover in each subplot using the CanopyCapture© App for Android phones. Percent canopy cover may be a more consistent correlate of total light environment over the course of the day, and it is a component of the 'Habitat Integrity Index' commonly used in odonata surveys [3].

We evaluated the concordance between light levels and percent canopy cover in the stream plots with the Spearman rank correlation. We described the variation in percent canopy cover and light levels across streams and habitats with two-way factorial general linear models and compared estimated plot means with sequential Bonferroni *t*-tests.

We used general linear models to compare odonate communities at two spatial scales. At the plot scale, we measured total odonate abundance, zygopteran abundance, anisopteran abundance, species richness, species diversity, and Anisoptera/Zygoptera ratio in each plot each day and analyzed variation across streams and habitats. Poisson loglinear models were used for the abundance data and linear models were used for richness and diversity values. Species richness values are 'improved Chao1 estimates' [39], and species diversity values are Optimized Shannon Diversity [40]. Estimated means were compared with *t*-tests for variables exerting significant effects on dependent variables. Neither mean percent cover per plot nor mean light level per plot correlated with any dependent variable at this scale (N = 4 plots), so neither was included in the model as a covariate. Differences in the compositions of communities across plots were described with non-metric multidimensional scaling (NMDS) and analyzed with PERMANOVA to assess the direct and interactive effects of river and habitat. Bray–Curtis distances were computed on log-transformed data, with 9999 permutations in the PERMANOVA.

At the subplot scale, we examined the relationship between the abundance of each species in each subplot (summed over the seven sampling days) and the light level and percent canopy cover with Spearman rank correlations. We measured the direct and interactive effects of river, habitat, and light level on the odonate abundance, zygopteran abundance, anisopteran abundance, species richness, and species diversity in the subplots (subplot data were pooled across sampling days). Although abundances in subplots sum to plot abundances and thus are somewhat redundant, species richness and diversity patterns depend on the identities of the species, and these patterns can vary from the subplot to plot scale. For example, 20 subplots could contain the same single species, resulting in subplot and plot richness values of 1, or subplots could each have a single different species, resulting in subplot richness values of 1 with a plot richness of 20. Again, Poisson loglinear models were used for the abundance data and linear models were used for richness and diversity values. Species richness values are 'improved Chao1 estimates' [39], and species diversity values are Optimized Shannon Diversity [40], both calculated with the SpadeR online program [41]. Estimated means were compared with sequential Bonferroni tests for the factors with significant contributions to the models. Mean percent canopy cover was not significantly correlated with any of the dependent variables and therefore was not included in the general linear models. Differences in the composition of communities across subplot light levels were described with non-metric multidimensional scaling (NMDS) using Bray–Curtis distances calculated on log-transformed data. We assessed the direct and interactive effects of river, habitat, and light level in a PERMANOVA, using 9999 permutations. Because this analysis was primarily used to describe light effects, and because the order or variables in the PERMANOVA affects the explanatory strength of the variables, we included light as the third variable in the model, after river and habitat, for the most conservative assessment of this variable.

Lastly, to further describe the effects of light level and percent cover on the potential differential effects on zygopterans and anisopterans, we used Spearman rank correlations to describe the relationships between light level, percent canopy cover, NMDS scores, and the Anisoptera/Zygoptera ratio based on abundance and species richness in each suborder. Since anisopterans were only present in 16 subplots, we inverted the typical 'Zygoptera/Anisoptera' ratio (to avoid zeroes in the denominator) and also analyzed the relationships within the subset of 16 plots that contained anisopterans to eliminate the effects of the preponderance of zeros. Analyses were conducted with SPSS [42] and the vegan program in R [43].

3. Results

3.1. Overview of Odonate Communities

We made a total of 549 observations of 12 species in the four plots (Table 1, Supplementary Figure S1). The plots were dominated by zygopterans, accounting for more than 89% of individuals sampled on each stream. Only three species of anisopterans were found: two species of *Brechmorhoga* were found in all four plots, and *Paltothemis lineatipes* was found in the sunny areas of the two open disturbed plots (Table 1).

Table 1. The total number of odonate observations, by species, in plots in secondary forest and disturbed habitats on the Cuecha and Maquina rivers in Monteverde, Costa Rica.

	Cuecha		Maquina	
Species	**Forest**	**Disturb**	**Forest**	**Disturb**
Zygoptera				
Argia anceps Garrison	0	4	0	1
Argia chelata Calvert	119	116	36	5
Argia elongata Garrison & Von Ellenrieder	0	0	1	0
Argia underwoodi Calvert	0	0	1	0
Hetaerina cruentata (Rambur)	30	63	8	54
Hetaerina majuscula Selys	57	8	1	0
Heteragrion majus Selys	6	4	0	0
Lestes henshawi Calvert	0	3	0	0
Philogenia peacocki Brooks	1	0	0	0
Anisoptera				
Brechmorhoga pertinax (Hagen)	2	1	1	1
Brechmorhoga rapax Calvert	8	3	4	2
Paltothemis lineatipes Karsch	0	5	0	4
Total	223	207	52	67

3.2. Light Environments

Light levels in the subplots were significantly inversely correlated with canopy cover ($r_s = -0.634$, N = 80, $p < 0.001$). Both indices varied between habitats, and canopy cover also varied between rivers (Table 2a). However, these patterns were largely due to the differences at the plot level represented by the significant interactive effects (Table 2a). The forested Maquina plot and the disturbed Cuecha plot did not differ from one another in mean light level or canopy cover; the differences between rivers and habitats were largely driven by the significant differences between the open, well-lit disturbed Maquina plot and the closed, shady forested site on the Cuecha (Table 2b).

Table 2. (a) General linear models describing the effects of river (Cuecha vs. Maquina) and habitat (disturbed vs. forested) on light levels and percent canopy cover in subplots (Wald X^2, ns = $p > 0.05$). **(b)** Comparisons of estimated means of the dependent variables for the stream plots (sequential Bonferroni *t*-tests, means in each column followed by the same letter are not different, $p = 0.05$).

(a) Summary of GLM

		Light Level		% Canopy Cover	
Variable	**df**	**X^2**	**p**	**X^2**	**p**
River	1	0.484	ns	27.130	0.001
Habitat	1	15.022	0.001	58.671	0.001
River × Habitat	1	7.200	0.007	15.865	0.001

(b) Comparison of Estimated Means

		Light Level	% Canopy Cover
Plot	**N**	**X ± 1 se**	**X ± 1 se**
Maquina-disturbed	20	3.80 ± 0.25 a	35.75 ± 3.59 a
Cuecha-disturbed	20	2.95 ± 0.25 ab	68.75 ± 3.59 b
Maquina-forested	20	2.15 ± 0.25 b	77.55 ± 3.59 bc
Cuecha-forested	20	2.65 ± 0.25 b	81.95 ± 3.59 c

3.3. Analyses of Odonate Communities on Streams

At the plot scale, there were significant differences between rivers in mean odonate abundance/day and mean zygopterans/day (Table 3a). On average, the Cuecha had more than three times the number of total odonates and zygopterans than the plots on the Maquina (Table 3b). There were no direct or interactive effects of river or habitat on anisopteran abundance, estimated species richness, estimated diversity, or Anisoptera/Zygoptera ratio (Table 3a).

Table 3. (**a**) The results from general linear models describing the direct and interactive effects of river (Cuecha vs. Maquina) and habitat (disturbed vs. forested) on total adult odonate abundance, zygopteran abundance, anisopteran abundance, species richness, species diversity, and Anisoptera/Zygoptera ratio (An./Zyg. Ratio) per plot/day (pooled over 20 subplots/day; ns = not significant = $p > 0.05$). Species richness values are 'improved Chao1 estimates' [39], and species diversity is Optimized Shannon Diversity ([40], Hill Number = 1.0). Neither mean light level per plot nor mean percent canopy cover per plot were significantly correlated with the dependent variables and so were not included in the general linear models. (**b**) Comparisons of estimated means of the dependent variables for the Cuecha and Maquina streams (*t*-tests, means followed by the same letter are not significantly different, $p = 0.05$).

| (a) Summary of GLM		Total Odonate Abundance		Zygopteran Abundance		Anisopteran Abundance		Species Richness		Species Diversity		An./Zyg. Ratio	
Variable	df	X^2	p	X^2	p	X^2	p	X^2	p	X^2	p	X^2	p
River	1	153.59	0.001	153.44	0.001	1.61	ns	0.29	ns	0.26	ns	2.34	ns
Habitat	1	0.74	ns	0.61	ns	0.96	ns	1.13	ns	0.04	ns	1.57	ns
River × Habitat	1	2.47	ns	2.11	ns	0.35	ns	0.21	ns	2.01	ns	3.13	ns

(b) Comparisons of Estimated Means		Total Odonate Abundance	Zygopteran Abundance	Anisopteran Abundance	Species Richness	Species Diversity	An./Zyg. Ratio
River	N	X ± 1 se	X ± 1 se	X ± 1 se	X ± 1 se	X ± 1 se	X ± 1 se
Cuecha	14	30.69 ± 1.48 a	29.34 ± 1.44 a	1.36 ± 0.31 a	4.63 ± 0.73 a	3.09 ± 0.30 a	0.08 ± 0.04 a
Maquina	14	8.43 ± 0.78 b	7.59 ± 0.74 b	0.84 ± 0.25 a	4.05 ± 0.80 a	2.82 ± 0.44 a	0.16 ± 0.04 a

There were significant differences in the composition of these communities between rivers (PERMANOVA, F = 5.248, df = 1, $p = 0.001$), between habitats (F = 5.674, $p = 0.001$), and for the 'river x habitat' interaction (F = 2.468, df = 1, $p = 0.033$). The disturbed Maquina plot had a much higher relative abundance of *H. cruentata*, and lower relative abundances of *H. majuscula* and *A. chelata*, than the other three plots—even the forested plot on the same river (Figure 2a). The same pattern also occurred to a smaller degree between the Cuecha plots, where *H. cruentata* had a higher relative abundance in the disturbed plot while *H. majuscula* had a higher relative abundance in the forested plot (Figure 2a). These patterns are clear in the NMDS ordination (stress = 0.1268), where the disturbed Maquina site differs from the other three (Figure 2a), contributing to the significant direct and interactive effects in the PERMANOVA.

These differences among plots were a function of the responses of several species to the light environment at a smaller subplot scale. Across all subplots (N = 80), the abundances of *H. cruentata* and *P. lineatipes* were positively correlated with light levels ($r_s = 0.564$, $p < 0.001$ and $r_s = 0.276$, $p = 0.027$, respectively) and negatively correlated with percent canopy cover ($r_s = -0.470$, $p < 0.001$ and $r_s = -0.288$, $p = 0.010$, respectively), corresponding to their greater abundance in the disturbed Maquina plot that had the highest light levels. In contrast, the abundances of *H. majuscula* and *A. chelata* were positively correlated with percent canopy cover ($r_s = 0.313$, $p = 0.005$ and $r_s = 0.241$, $p = 0.031$, respectively), accounting for their greater proportional abundance in the shady forested sites.
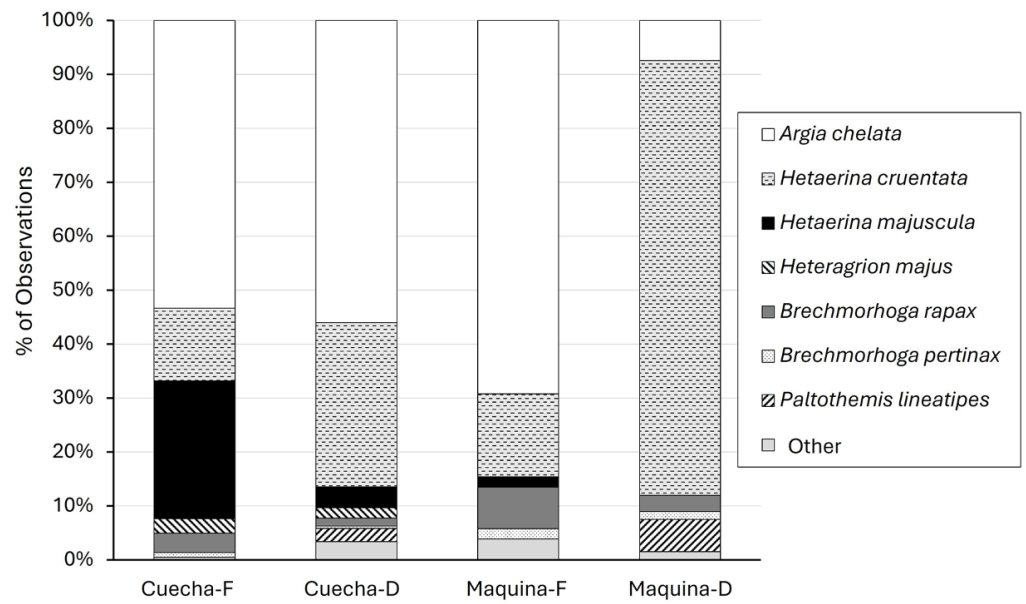
(**a**) Relative Abundances
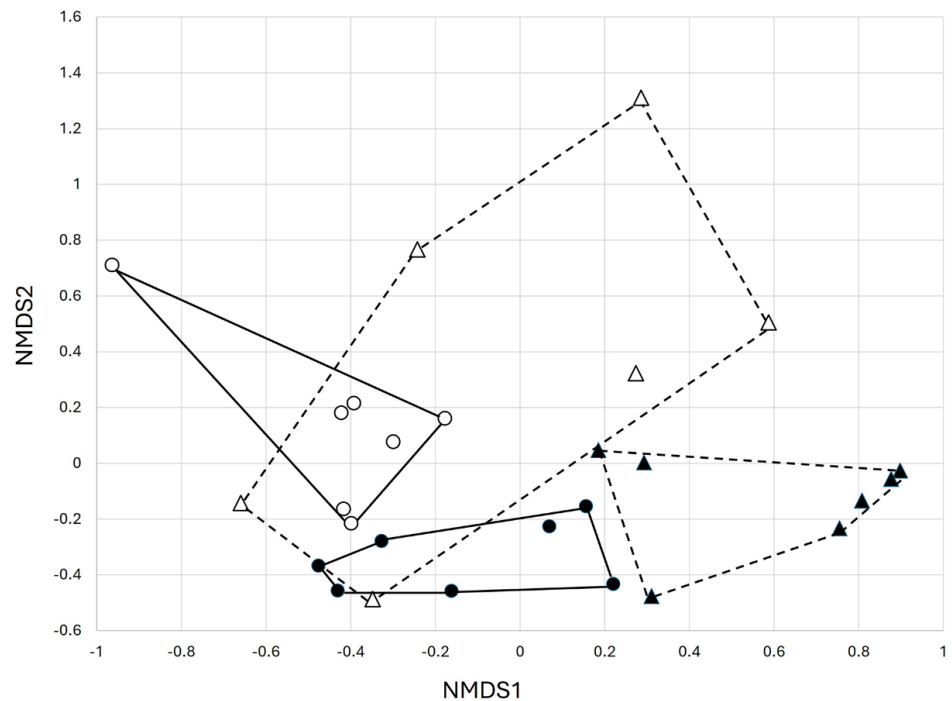
(**b**) NMDS Ordination

Figure 2. (**a**) The relative abundances of odonate species in disturbed (D) and forested (F) plots on the Cuecha and Maquina rivers in Monteverde, Costa Rica, as measured by the total number of observations pooled over seven sampling days. (**b**) NMDS ordination of communities in these plots (stress = 0.1268; circles = Cuecha River; triangles = Maquina River; open = forested plot, filled = disturbed plot).

As a consequence of these relationships with the most abundant species, light level had a significant effect on the mean abundance of all odonates, zygopterans, and anisopterans (Table 4a). Mean abundance increased for all three taxa with increasing light levels to light level 4, and then declined at light level 5 (Table 4b), with a greater proportional increase for anisopterans than zygopterans (Table 4b).

Table 4. (a) The results from general linear models describing the direct and interactive effects of light level, river, and habitat on total adult odonate abundance, zygopteran abundance, anisopteran abundance, species richness, and species diversity per subplot (pooled over 7 sampling days). Species richness values are 'improved Chao1 estimates' [39], and species diversity is Optimized Shannon Diversity ([40], Hill Number = 1.0); ns = not significant = $p > 0.05$. **(b)** Comparison of estimated means across light levels; means followed by the same letter are not significantly different (sequential Bonferroni *t*-tests, $p = 0.05$).

(a) Summary of GLM

Variable	df	Total Odonate Abundance X^2	p	Zygopteran Abundance X^2	p	Anisopteran Abundance X^2	p	Species Richness X^2	p	Species Diversity X^2	p
Light	1	36.767	0.001	23.202	0.001	12.751	0.005	6.61	ns	4.434	ns
River	4	57.936	0.001	62.518	0.001	0.400	ns	3.695	ns	0.004	ns
Habitat	1	0.163	ns	0.095	ns	2.466	ns	1.898	ns	0.037	ns
Light × River	4	4.603	ns	4.678	ns	0.470	ns	1.447	ns	3.142	ns
Light × Habitat	4	3.746	ns	3.309	ns	3.396	ns	2.886	ns	0.187	ns
River × Habitat	1	0.25	ns	0.014	ns	1.347	ns	0.447	ns	0.180	ns

(b) Comparison of Estimated Means

Light Level	N	Total Odonate Abundance $X \pm 1$ se	Zygopteran Abundance $X \pm 1$ se	Anisopteran Abundance $X \pm 1$ se	Species Richness $X \pm 1$ se	Species Diversity $X \pm 1$ se
1	11	2.94 ± 0.62 a	3.02 ± 0.64 a	0.00 ± 0.00 a	1.96 ± 0.79 a	2.23 ± 0.71 a
2	25	4.49 ± 0.53 a	4.38 ± 0.52 a	0.12 ± 0.09 a	2.06 ± 0.41 a	2.02 ± 0.61 a
3	18	5.42 ± 0.71 a	4.92 ± 0.69 a	0.32 ± 0.18 ab	2.70 ± 0.45 a	3.55 ± 0.68 a
4	14	9.50 ± 0.98 b	8.02 ± 0.92 b	1.11 ± 0.30 b	3.59 ± 0.47 a	2.59 ± 0.34 a
5	12	6.76 ± 1.82 ab	6.87 ± 1.86 ab	0.00 ± 0.00 a	2.46 ± 0.87 a	2.31 ± 0.68 a

These species-specific responses changed the composition of the communities found within subplots with different light environments (Figure 3a). The relative abundance of *H. cruentata* increased with increasing subplot light level, with concomitant declines in the relative abundance of *A. chelata* (Figure 3a). The anisopterans were absent from the lowest light level and increased in relative abundance through light level 4 (Figure 3a). These differences are represented by the lack of overlap in the NMDS analysis between communities in light level 1 and light level 4 (Figure 3b) and represent a statistically significant effect in the PERMANOVA ('Light', Table 5), even after the significant variation between rivers and habitats was accounted for in the ordered model (Table 5).

Table 5. Results from the PERMANOVA describing the direct and interactive effects of river, habitat, and light level on community composition.

Source	Df	SS	R^2	F	p
River	1	2.7161	0.1555	18.2778	0.0001
Habitat	1	2.1531	0.1233	14.4891	0.0001
Light	1	1.2071	0.0691	8.1229	0.0001
River × Habitat	1	1.6729	0.0958	11.2579	0.0001
River × Light	1	0.0759	0.0043	0.5106	0.7535
Habitat × Light	1	0.1930	0.0111	1.2986	0.2618
River × Habitat × Light	1	0.2349	0.0135	1.5808	0.1634
Residual	62	9.2133	0.5275		

(**a**) Relative Abundances
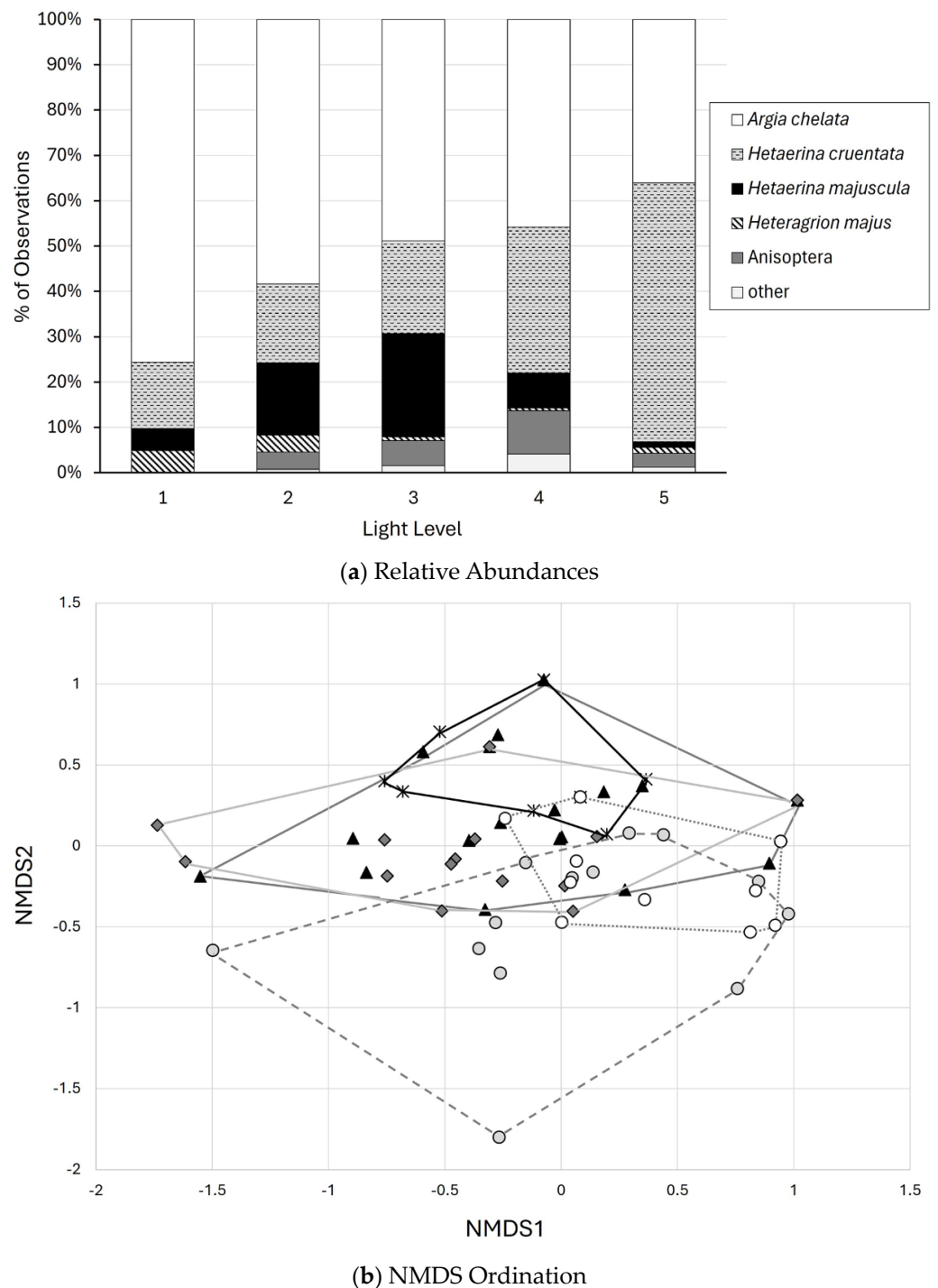

(**b**) NMDS Ordination

Figure 3. (**a**) The relative abundances of odonate species in 5 m subplots with different light levels (1 = low, 5 = high), in plots on the Cuecha and Maquina rivers in Monteverde, Costa Rica, as measured by the total number of observations pooled over seven sampling days. The three species of anisopterans (*Brechmorhoga rapax*, *B. pertinax*, and *Paltothemis lineatipes*) were pooled for visual clarity. (**b**) NMDS ordination of communities in these five light levels (stress = 0.1195; stars = light level 1, black triangles = light level 2, dark diamonds = light level 3, light grey circles = light level 4, white circles = light level 5).

Community structure, as described by the NMDS ordination axes, was strongly associated with light level and/or percent canopy cover at both the plot and subplot scales (Spearman rank correlations, Table 6). In both the complete data set and the subset containing Anisoptera, at both the plot and subplot scales, NMDS1 was positively correlated

with light level and inversely correlated with percent canopy cover (Table 6). Interestingly, even though the axes are orthogonal, NMDS2 showed opposite but significant relationships with these variables (Table 6). At the plot scale, neither Anisoptera/Zygoptera ratio was correlated with light level, percent canopy cover, or the NMDS axes in the complete data set (Table 6(a1)). In the reduced data set where Anisoptera were present, however, there was a significant inverse relationship between Anisoptera/Zygoptera richness ratio with percent canopy cover, and both ratios were positively associated with NMDS1, which correlated with light levels (Table 6(a2)). At the subplot scale, The Anisoptera/Zygoptera ratios for both abundance and richness were inversely correlated with NMDS2 in both the complete and reduced data sets (Table 6b), corresponding to the direct negative relationships with percent canopy cover (Table 6b). In the complete data set, both Anisoptera/Zygoptera ratios were also significantly correlated with light level (Table 6(b1)).

Table 6. Spearman Rank Correlations at the (**a**) plot and (**b**) subplot levels, describing the relationships between light levels, percent canopy cover, NMDS axes, and Anisoptera/Zygoptera ratios based on abundances and species richness in (**1**) complete data sets and (**2**) reduced data sets including only those sites where anisopterans were present. (one-tailed tests, + = $p < 0.10$, * = $p < 0.05$, ** = $p < 0.01$, *** = $p < 0.001$, ns = not significant = $p > 0.05$).

(**a**) Plot level (**1**) Complete data set (N = 27)	Light Level	Canopy Cover	NMDS1	NMDS2
Light Level		−0.822 ***	0.481 **	−0.423 *
Canopy Cover			−0.663 ***	0.404 *
An./Zyg. Ratio-Abundance	−0.230 ns	0.163 ns	0.171 ns	0.282 +
An./Zyg. Ratio-Richness	−0.177 ns	0.111 ns	0.171 ns	0.196 ns
(**2**) Reduced data set: Anisopterans present (N = 16)	Light Level	Canopy Cover	NMDS1	NMDS2
Light Level		−0.683 ***	0.032 ns	−0.805 ***
Canopy Cover			−0.502 *	0.459 *
An./Zyg. Ratio-Abundance	−0.160 ns	−0.318 ns	0.836 ***	0.337 ns
An./Zyg. Ratio-Richness	−0.016 ns	−0.460 *	0.856 ***	0.131 ns
(**b**) Subplot Level (**1**) Complete data Set (N = 70)	Light Level	Canopy Cover	NMDS1	NMDS2
Light Level		−0.634 ***	0.314 *	−0.603 **
Canopy Cover			−0.595 **	0.395 *
An./Zyg. Ratio-Abundance	0.239 *	−0.158 +	0.008 ns	−0.544 **
An./Zyg. Ratio-Richness	0.258 *	−0.174 +	0.010 ns	−0.560 **
(**2**) Reduced data set: Anisopterans present (N = 16)	Light Level	Canopy Cover	NMDS1	NMDS2
Light Level		−0.697 ***	0.314 *	−0.603 **
Canopy Cover			−0.595 **	0.395 *
An./Zyg. Ratio-Abundance	0.039 ns	−0.354 +	0.008 ns	−0.544 **
An./Zyg. Ratio-Richness	0.381 +	−0.462 *	0.010 ns	−0.560 **

4. Discussion

Our first goal was to describe the general composition of this cloud forest community. The relatively low species richness found in our study (12) is consistent with some previous studies of streams in tropical cloud forests [23–25], as odonate diversity tends to decrease with altitude [23]. *Argia underwoodi*, *Hetaerina majuscula*, *Lestes henshawi*, and *Philogenia peacocki* are all endemic to cloud forests of Costa Rica [33] and were primarily found in forested plots. This is not unusual, as zygopterans tend to dominate forested sites in the tropics, particularly at high elevations [15,44]. *Heteragrion majus* is a high-elevation forest species native to Costa Rica and Panama [33] that we found in the shaded parts of both plots on the Cuecha. The remaining species range at least from Mexico to Panama and use

a mix of forest and open habitats [33]. Our results are consistent with these descriptions, as all were found in the disturbed, open plots. Our results are also consistent with the general pattern found at lower elevations in the tropics, where endemic zygopterans dominate in pristine forests and are replaced by far-ranging generalist species in disturbed, open areas [45,46].

These patterns at the plot scale are a function of responses to light levels at the microhabitat scale: the abundance of *H. cruentata* and *P. lineatipes* were positively correlated with light levels in subplots, while the abundance of *H. majuscula* and *A. chelata* indicated that they preferred shadier subplots. These differences probably represent explicit choices in microhabitat selection, as the four species had access to a full range of light environments in the plots where they occurred. Again, these results are consistent with previous research. *H. cruentata* is commonly found in open areas and is apparently quite tolerant of human disturbance, dominating communities in high elevation pastures and coffee plantations in Mexico [24]. *P. lineatipes* is a mid-sized anisopteran that, like most heliotherms, perches on warm rocks in sunny areas to bask [33]. In addition, although the two *Brechmorhoga* species were more common in the forested plots and are indicators of less disturbed streams [9], they used the sunny subplots of those areas and were also found in the disturbed plots. Since these six species comprise over 95% of the observations, they are largely responsible for the significantly greater mean abundance of odonates, zygopterans, and anisopterans in higher light (level 4) subplots, and the greater proportional increase in the abundance of light dependent, heliothermic anisopterans.

Although the plots varied in light levels and percent canopy cover, these differences were not entirely responsible for the differences in odonate communities at the plot scale. Indeed, although the disturbed Maquina plot was the most open site with significantly lower percent cover and higher mean light level than the forested sites, it had significantly fewer odonates than either Cuecha plot. Indeed, the difference in odonate abundance between rivers may be responsible for the unusual decline in mean abundances at light level 5. Seven of the twelve subplots with light level 5 were in the relatively depauperate disturbed Maquina plot, potentially lowering abundances relative to other light level categories because of this unbalanced design.

Neither species richness nor species diversity varied between rivers or habitats or were affected by differences in light environments at either spatial scale. That is not particularly surprising in comparisons of stream communities [42,44]. There were, however, dramatic differences in the composition of these communities at both the plot and subplot scales, as indicated by the NMDS and PERMANOVA analyses. These differences in community structure were largely driven by differences in the light environment, indicated indirectly at the plot level by correlations of NMDS axes with light level and canopy cover, or directly at the subplot level by a significant light effect in the PERMANOVA. The preference of *H. cruentata* and *P. lineatipes* for sunny subplots explains their dominance in the sunniest plot, the lower Maquina plot. Likewise, the preference of *A. chelata* and *H. majuscula* for shady habitats explains their dominance in the forested plots. In addition, even though there were only 12 species total and only three species of Anisoptera, there was strong support for the hypothesis that the ratio of Anisoptera to Zygoptera is a good barometer of forest disturbance and canopy removal. There were significant relationships between A/Z ratios based on abundance and richness at both the plot and subplot scale: either direct relationships with light level, inverse relationships with canopy cover, or relationships with NMDS axes that correlate with these environmental indices. Strong relationships even occurred in the reduced data sets, which were limited to the 16 samples that contained Anisoptera. Again, given the small total richness, the low abundance and richness of anisopterans, and the small sample sizes in the reduced data sets, these relationships reinforce the general importance and pervasiveness of these ratios as indicators of forest canopy integrity.

Of course, there may be other environmental differences between the streams and plots that could contribute to these differences in adult odonate communities. We did not

quantify streamside vegetation, the density of perch sites, prey availability, or predation risk—which all influence adult odonate abundance and behavior and may correlate with light availability [13,34,47,48]. In addition, factors that affect larval ecology—like water quality, conductivity, dissolved oxygen, and sediment type—might contribute to differences in the abundance and composition of adult communities [17,49,50], particularly between the rivers that were ~1 km apart and may exceed the dispersal capabilities of zygopterans, in particular.

Nonetheless, this research confirms the hypotheses that odonate communities in intact tropical forests are dominated by endemic zygopterans and that more open habitats are dominated by wide-ranging generalist species. The effect of light was strongest at a small scale, which is consistent with the hypothesis that microhabitat selection by heliothermic odonates is responsible for this pattern. Even in this low diversity cloud forest community with only three species of anisopterans, the Anisoptera/Zygoptera ratio correlated with increased light and decreasing canopy cover resulting from anthropogenic habitat conversion. Given the sensitivity of cloud forests to global climate change [37], the persistence of cloud forest endemics dependent on closed canopies may be at risk.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/d16090557/s1, Figure S1: Photos of males of the 12 species observed in the four study plots in the Cuecha and Maquina Rivers, Monteverde, Costa Rica.

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