STREAM WATER QUALITY OF QUEBRADA MÁQUINA AT MONTEVERDE, COSTA RICA

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Abstract: Ecotourism has increased substantially in recent years for many Costa Rican towns, bringing potential human impacts to formerly pristine areas that attract visitors. We examined benthic macroinvertebrate communities and physical/chemical factors at 6 sites along the Quebrada Máquina stream in Monteverde to test for evidence of anthropogenic pollution. We hypothesized that stream water quality, assessed through the Hilsenhoff Biotic Index, would decrease immediately below a human development near the stream. We found no directional trend in this index, indicating that there is no direct impact of ecotourism development on stream water quality. However, both benthic macroinvertebrate species richness and Shannon-Weiner diversity decreased as the stream flowed downstream. More detailed study is needed to determine the reasons for this trend.

Key Words: Quebrada Máquina, *pollution, water quality, stream invertebrates, diversity*

INTRODUCTION

Ecotourism in Costa Rica is an important and growing part of many local economies. Monteverde alone hosts over 187,000 visitors yearly (M. Hidalgo, *pers. comm*). Rapid development in Monteverde and other ecotourism hotspots has changed in land use and may be increasing local stream pollution.

The Quebrada Máquina stream passes through areas of development as it flows from the cloud forest through the town of Monteverde. Ermentrout et al. (2003) showed that the macroinvertebrate community in a nearby stream changed substantially between upstream and downstream sites, possibly a result of changes in water quality. We hypothesized that inputs from development sites decrease Quebrada Máquina water quality as it flows through the town of Monteverde. We expected evidence of increased pollution in stream sites just below a development compared to sites just above it.

METHODS

We surveyed six sites along Quebrada Máquina in Monteverde, Costa Rica, on 20-21 January 2006: in the cloud forest (site 1); before (2) and after (3) passing a major hotel; before (4) and after (5) passing an input from a discotheque, a gas station, a residential neighborhood and road on the eastern side of town, and after an input from development on the western side of town (6). We determined the elevation and distance downstream from site 1 for each site (site 1= 1510 m above sea level; site 2= 1390 m above sea level, 900 m downstream from site 1; site 3= 1380 m, 1050 m; site 4= 1365 m, 1275 m; site 5= 1300 m, 1875 m; site 6= 1240 m, 2225 m). All sites

had approximately the same canopy composition and cover.

At each site we chose three sample spots with homogeneous rocky riffle substrates, the preferred habitat for benthic macroinvertebrates. We measured the pH, temperature, conductivity, and dissolved oxygen at each spot and kick-net sampled the benthic macroinvertebrates by disturbing the substrate in a 0.5 x 0.5 m area in front of the net for one minute. We calculated discharge at each site using stream width, depth at 11 intervals along its width, and flow (measured by timing how fast the stream carried an apple for 5 meters). We counted and identified all invertebrates collected, to family.

We estimated stream pollution levels at each site with the Hilsenhoff Biotic Index, which uses invertebrate family richness and abundance coupled with pre-assigned pollution tolerances for each family (Lehmkuhl 1979). We also calculated the Shannon Diversity Index (SDI) of invertebrate families for each site (Begon et al. 1990).

RESULTS

We collected and identified a total of 726 invertebrates representing 23 taxa. Some groups were common across all sites (two Ephemeteropteran taxa: Heptogeniidae and Siphlonuridae; and one Trichopteran taxon; Hydropsychidae). Other groups were found more commonly or exclusively upstream (two Ephemeropteran taxa: Trycorythidae and Ephemerellidae; one Plectopteran taxon: Chloropeltidae; two Coleopteran taxa: Dytiscidae and Psephenidae; and one Crustacean). No invertebrate families were found in noticeably higher numbers in downstream sites than upstream sites (Appendix A).

Hilsenhoff Biotic Index did not differ significantly between stream sites (ANOVA, $F_{5, 13} = 0.36$, $P = 0.86$; Fig.1). Abiotic conditions were also relatively constant across sites (Table 1). We found no trend in water discharge rate across sites. SDI and family richness decreased with distance downstream from site 1 (Fig.1).

Table 1: Values (mean ± SE) of selected abiotic factors across six sample sites along the Quebrada Máquina, Monteverde, Costa Rica. Measurements were taken at 3 points for each site except site 5, which had 4 measurement points.

	Temperature (°C)	DO (mg/L)	Conductivity (mV)	pH
Site 1	15.67 ± 0.09	8.83 ± 0.09	36.40 ± 1.20	6.43 ± 0.05
Site 2	16.20 ± 0.00	8.63 ± 0.03	30.90 ± 7.35	6.40 ± 0.03
Site 3	16.30 ± 0.00	8.47 ± 0.26	35.40 ± 1.00	6.42 ± 0.10
Site 4	16.00 ± 0.00	8.33 ± 0.26	42.57 ± 0.35	6.31 ± 0.03
Site 5	16.10 ± 0.00	8.88 ± 0.08	44.00 ± 0.37	6.76 ± 0.15
Site 6	15.53 ± 0.03	8.43 ± 0.07	48.40 ± 0.46	6.47 ± 0.08

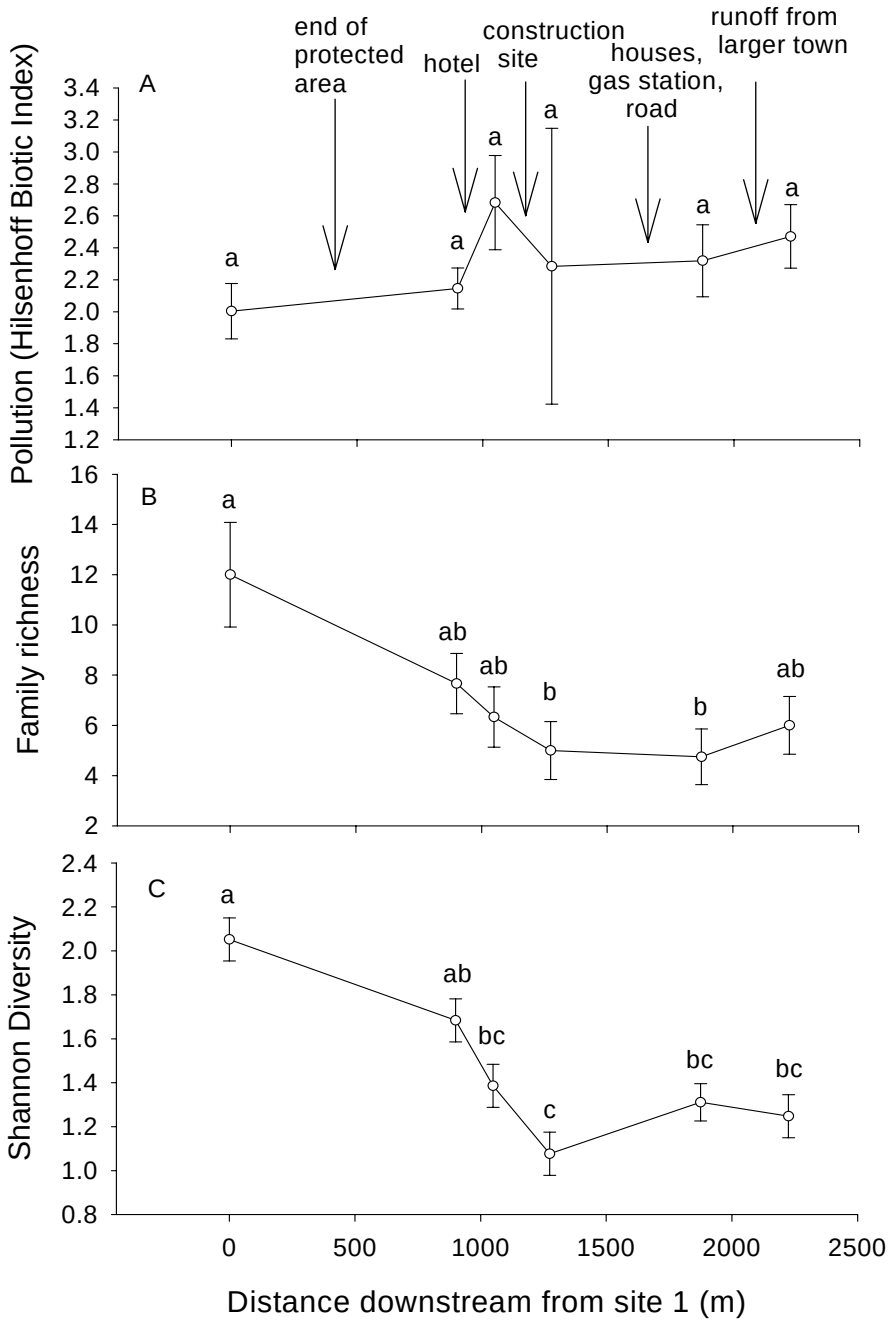

Figure 1. Comparison of trends in (A) pollution (Hilsenhoff Biotic Index), (B) family richness and (C) Shannon Diversity Index of benthic macroinvertebrates in Quebrada Maquina by sampling site, presented in distance downstream from site 1. Points and error bars represent mean ± 1 SE. Points not sharing a letter are significantly different ($p < 0.05$, with Tukey-Kramer HSD). Possible sources of pollutants into the stream are denoted with arrows

DISCUSSION

Development along the Quebrada Máquina did not obviously decrease water quality in the stream. It may be a factor in producing the decreased stream invertebrate family richness and SDI by changing habitats surrounding the stream, but there are several other possible mechanisms. Although elevation changed by 270 m across all sites, abiotic features of the stream (pH, conductivity, temperature, and dissolved oxygen) were quite stable throughout our study site (Table 1) (for review see Flowers, 1991). However, a change in elevation of that magnitude would certainly have effects on air temperature and the riparian environments surrounding the stream, which could alter allochthonous inputs. Factors such as primary production or non-benthic predators (excluding fish, which are absent) within the stream might also affect invertebrate family richness and SDI.

To identify mechanisms behind the observed changes in stream invertebrate community we recommend measuring autochthonous primary production and predation and examining riparian vegetation at each site. Another more rigorous test might involve sampling multiple streams for comparison or sampling the stream at numerous discrete intervals and comparing intervals that have a given factor that might affect water quality or invertebrate communities with intervals that do not.

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DIFFERENCES IN MOSS MORPHOLOGY AND COMMUNITY COMPOSITION BETWEEN AN OPEN AND A FOREST UNDERSTORY HABITAT

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Abstract: Mosses are dependent on high water availability for survival and reproduction, and some moss morphologies can help reduce water loss. Water availability is presumably higher in the understory than in the open. We examined how moss morphology varied within and among understory and open habitats. We also investigated whether morphological differences across habitats were due to phenotypic plasticity of individuals or fixed morphological differentiation among species. We found that moss growth form and degree of "packing" of moss tissues were significantly related to habitat: mosses in open habitat tended to grow in a cushion form and be more packed. We also found differences in community composition across habitats, which reflected morphological differences. Thus morphological differences among species may represent adaptations to habitat conditions, such as water availability.

Key Words: moss, morphology, habitat, community

INTRODUCTION

Water availability limits the growth, survival and reproduction of many plants. This is especially true for mosses (Bryophyta), which, due to their non-vascular structure, are especially reliant on access to water. Nonetheless, some mosses grow in habitats exposed to relatively high sunlight and high evaporation rates. Morphological adaptations can reduce moss water loss in these conditions. Changes in leaf form and orientation, reduction in degree of leaf branching, and increased patch density can all slow the rate at which mosses lose water (Schofield, 2001).

These morphological differences can be driven by either phenotypic plasticity of individuals, or relatively fixed morphological differentiation among species. In the first case, individual mosses might modify their growth pattern in ways that match their local environment. In the second case, species may evolve different growth patterns that make them adapted to different environments.

If water availability influenced moss morphology across habitats, we would expect to see morphological differences between mosses located in a sun-exposed, open habitat, and in an understory habitat. Mosses growing in open areas would be thicker and denser, and have less branching (lower surface area: volume ratio) than mosses growing in understory areas. These morphological differences could be due to either phenotypic plasticity or species adaptations. If it were phenotypic plasticity, we would expect to see the same moss species, with different growth patterns, in each habitat. If it were species adaptations, we would expect to see communities composed of different species in each habitat.

METHODS

We conducted our study on 20-21 January 2006 in the area surrounding Estacion Biologica de Monteverde, Costa Rica. We randomly selected 25 trees in both the cloud forest understory and open areas around the station. Sampled trees were all greater than 16 cm in diameter at breast height (1.3 m), and supported moss communities covering at least 50% of the area of a 50 x 50 cm quadrat. The quadrat was divided into 100 equal subplots (5 x 5 cm). On each tree, we randomly chose three subplots within which we measured the thickness of moss, categorized the degree of branching (ordinal scale of 1-5), and estimated the degree of packing (ordinal scale of 1-5), defined as the three-dimensional packing of moss tissue within areas occupied by moss. We also categorized moss growth form as turf (clustered moss stems), mat (spreading moss stems), or cushion (round tufts) for both habitats. We compared moss thickness (log transformed), degree of packing, and degree of branching, using ANOVA in JMP 5.0.1. We compared growth form, by nesting trees within habitat and using contingency analyses and Pearson X^2 tests.

We investigated possible differences in community composition by randomly sampling ten additional trees from each area (understory and open). We recognized taxa (probably species or genera) based on morphotype and recorded the number of 5 x 5 cm squares in which each morphotype occurred in each sample. To compare communities occupying the two habitats, we used the morphotype abundance correlation matrix (Appendix 1) to perform a multivariate Principal Component Analysis (PCA), followed by an ANOVA, comparing habitats with respect to PCA 1 and PCA 2.

RESULTS

Neither moss patch thickness ($F_{1, 48}$ = 0.81, P = 0.37) nor degree of branching differed across habitats ($F_{1, 48}$ = 0.37, P = 0.55) (Table 1). We observed turf and mat growth formations in both habitats, but we only observed cushion growth formations in open habitats (X^2 = 47.84, df = 2, P < 0.001). Degree of packing was significantly higher in open habitats ($F_{1, 48}$ = 12.02, P < 0.0011).

TABLE 1. Morphological characteristics of moss in an open and understory habitats near Estacion Biologica de Monteverde, Costa Rica. Moss thickness, degree of branching, and degree of packing are reported as mean ± standard error (branching and degree of packing on a 1-5 scale, n = 25). Growth forms indicate percentage of samples that were predominantly cushion, mat, or turf in each habitat.

Habitat	Thickness (cm)	Branching	Degree of packing	Growth Form (% cushion : mat : turf)
Open	7.15 ± 1.32	1.95 ± 0.22	2.85 ± 0.16	41 : 29 : 29
Understory	8.83 ± 0.13	1.76 ± 0.22	2.08 ± 0.16	0 : 77 : 23

Samples of the moss community from open and understory habitats separated clearly along the 1st axis of the Principal Component Analysis ($F_{1,18}$ = 59.61, P < 0.0001; Fig. 1). Examination of the loading scores for PCA 1 (Table 2) revealed that morphotypes C and A (found in low-density, mat or turf growth forms) were strong indicators of the understory habitat, and morphotypes K and M (found in

aggregated, dense cushions) were strong indicators of the open habitat. We found only one morphotype (C) in both habitats, which occurred on one sampled tree in open habitat and eight trees in the understory.

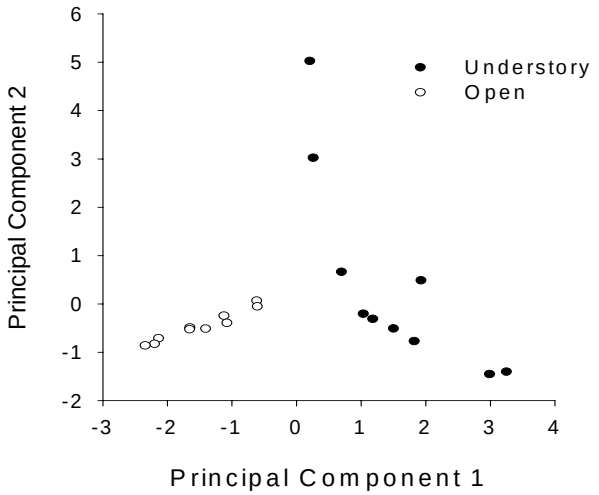

Figure 1. Moss community structure (on 20 sample trees) as represented by first two axes of Principal Component Analysis of moss morphotype abundance. Note separation of samples along PC 1 axis based on whether sample trees were in the understory or open habitat. Loading values for 16 moss morphotypes are in Table 1.

TABLE 2. Loading values from Principle Component Analysis of moss morphotypes (A-P) across 20 0.25 m^2 samples (1 sample on each of 10 trees in both open and understory habitats), near Estacion Biologica de Monteverde, Costa Rica.

PC 1		PC 2	
Moss morphotype	Loading	Moss morphotype	Loading
C	0.41	I	0.62
A	0.37	J	0.52
B	0.30	F	0.32
D	0.27	E	0.07
E	0.16	H	0.05
H	0.15	O	0.01
G	0.12	G	-0.04
F	0.04	P	-0.09
I	0.02	L	-0.10
J	0.02	N	-0.10
O	-0.05	B	-0.15
P	-0.18	K	-0.17
N	-0.22	D	-0.17
L	-0.26	M	-0.18
K	-0.39	A	-0.19
M	-0.39	C	-0.22

DISCUSSION

Some aspects of moss morphology were clearly related to habitat, and the patterns were partly interpretable with respect to differences in sunlight between habitats. In sun-exposed, open habitats, cushion growth and high percent cover can potentially reduce water loss by evaporation. However, moss patch thickness and degree of branching were not related to habitat. It is possible that microhabitats (within and among trees) influence moss patch thickness and degree of branching more than the coarse habitat (understory vs. open) conditions. For example, a patch of moss may grow better on a tree trunk with relatively high resource availability regardless of whether it is located in the open or the understory.

The clear separation between moss morphotypes across habitats suggests that differences between habitats in moss morphology mainly reflect community structure (species composition) rather than phenotypic plasticity within species. The general morphological characteristics associated with each habitat were well represented by two pairs of taxa. We were unable to test whether these morphological differences represent adaptations to different levels of sunlight and water availability, but it is plausible that open vs. understory habitats select different species from the local species pool, based partly on morphological attributes related to water retention.

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APPENDIX 1. Correlation matrix of moss morphotype (A-P) abundance across 20 0.25-m^2 samples (1 sample on each of 10 trees in both open and understory habitats).

	A	B	C	D	E	F	G	H	I	J	K	L	M	N	O	P
A	1.00	0.90	0.30	0.20	-0.06	-0.05	-0.15	-0.10	-0.14	-0.10	-0.32	-0.22	-0.29	-0.14	-0.10	-0.10
B		1.00	0.07	-0.11	-0.04	-0.11	-0.12	0.07	-0.12	-0.08	-0.26	-0.18	-0.23	-0.12	-0.08	-0.08
C			1.00	0.77	0.00	-0.18	0.34	0.05	-0.20	-0.14	-0.39	-0.30	-0.35	-0.20	-0.14	-0.14
D				1.00	-0.09	-0.09	-0.10	-0.07	-0.10	-0.07	-0.22	-0.15	-0.20	-0.10	-0.07	-0.07
E					1.00	0.03	-0.10	0.95	-0.09	-0.07	-0.21	-0.15	-0.19	-0.10	-0.07	-0.07
F						1.00	-0.10	-0.07	0.42	-0.07	-0.21	-0.15	-0.20	-0.10	-0.07	-0.07
G							1.00	-0.08	-0.11	-0.08	-0.23	-0.16	-0.21	-0.11	-0.08	-0.08
H								1.00	-0.07	-0.05	-0.16	-0.11	-0.15	-0.07	-0.05	-0.05
I									1.00	0.86	-0.23	-0.16	-0.21	-0.10	-0.07	-0.07
J										1.00	-0.16	-0.11	-0.15	-0.07	-0.05	-0.05
K											1.00	0.24	0.31	-0.05	0.10	0.50
L												1.00	0.20	0.00	-0.11	-0.11
M													1.00	0.60	-0.15	0.13
N														1.00	-0.07	-0.07
O															1.00	-0.05
P																1.00

VARIABLE SEX RATIOS IN THE MONOECIOUS PERENNIAL *BEGONIA INVOLUCRATA*

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Abstract: The sex ratio of flowers in monoecious plants can influence individual reproductive success. We tested how flower sex ratios in individual *Begonia involuncrata*, a monoecious, deceit-pollinated perennial, were related to begonia size. We expected and found a male-biased sex ratio for the species, which may help maintain pollinator visits by maintaining high levels of honest rewards (male flower pollen). However, larger begonias had a higher proportion of female flowers, perhaps because they have more resources and can therefore afford to produce more costly female flowers. Conversely, increased maleness in smaller begonias may reflect the constraints of resource limitation and/or represent optimal reproductive strategies given resource limitations. Interestingly, the different sex ratios of different sized begonias, which represent individual solutions to reproductive success, may help to maintain the male-biased sex ratio within the population, which is theoretically required to maintain the deceit-pollination system.

Key Words: Begonia involucrata, *sex ratio, deceit pollination, reproductive investment*

INTRODUCTION

Plant fitness depends on maximizing reproductive success, but lack of mobility and access to resources limit plant reproductive strategies. For monoecious plants with imperfect flowers, the ratio of female to male flowers is a variable attribute that can influence reproductive success.

We studied *Begonia involuncrata*, a monoecious, perennial plant that employs deceit pollination. In the deceit pollination system, female flowers mimic male flowers, yet do not offer nectar or pollen rewards. However, maintenance of reliable visitation rates from pollinators requires that there be sufficient rewards (honest signals) to pollinators through a male-biased sex ratio. Campbell et al. (2001) estimated that there may be an evolutionarily stable equilibrium with a sex ratio of 1:3 for females:males. For begonias, producing female flowers is more costly than male flowers because female fruits require more energy and nutrients than male pollen. Thus, the effect of sex ratio on fitness in begonias could be influenced by (1) maintenance of the deceit pollination system and (2) relative costs of male and female flowers.

We tested two competing hypotheses: (1) Begonias have a genetically fixed female:male sex ratio that is just adequate to maintain deceit pollination (possibly 1:3 female:male), or (2) begonias have plasticity in their sex ratio. Under hypothesis 2, larger begonias might produce relatively more female flowers because they have more resources to invest in reproduction.

METHODS

We conducted our study on 21 January 2006 in the Monteverde Biological Station in Costa Rica. We haphazardly sampled 26 begonias along Sendero Principal: 1 to 15 individuals from each of

12 patches (with patches defined as one or more flowering ramets within a radius of 2 m). Patches were separated from the nearest other patch by 5 - 20 m. We restricted our sample to begonias in the early stage of flower development, with female buds and male buds or open flowers containing pollen, to avoid underestimating male flower abundance due to their abscission after pollen distribution.

For each begonia, we counted the number of mature green leaves, female flowers and male flowers. We measured plant height, largest width between two consecutive leaves, and width perpendicular to the first width. We estimated competition due to crowding with an ordinal score of 1-3 for each plant (1 meaning no plants within 0.5 m and 3 meaning 4 plants within 0.5 m). We estimated canopy cover with a spherical densiometer.

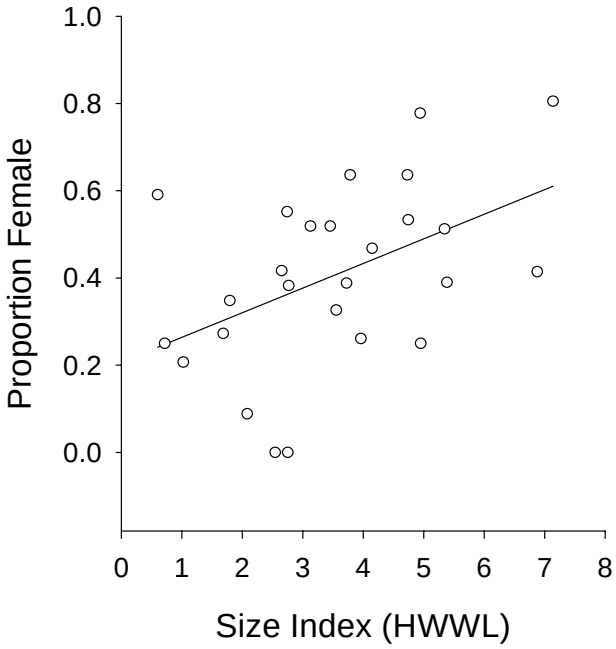

Figure 1. The proportion of female flowers in individuals of the monoecious, perennial *Begonia involucrata* increased with plant size.

We created a size index by multiplying height, both widths, and the number of leaves (HWWL index). We used linear regression to evaluate the relation between the proportion of female flowers and HWWL index, canopy cover, total number of flowers, and level of crowding (JMP v.5. 0.1).

RESULTS

The number of flowers per begonia ranged from 14 to 70 (mean ± SE = 38.3 ± 3.4). The proportion of female flowers ranged from 0 to 0.80 (mean ± SE = 0.41 ± 0.04; female:male =1:1.40). The proportion of female flowers increased significantly with begonia size (r^2=0.22, P=0.02, df=24; Fig. 1). However, proportion of female flowers was unrelated to canopy cover, total number of flowers or crowding by other plants (Fig. 2).

DISCUSSION

The ratio of female to male flowers in begonias is presumably a result of selection to maximize individual fitness but it may also have consequences for the efficacy of the deceit pollination system, which influences reproductive success of the population. For an individual, reproductive success is achieved by producing more flowers of the under-represented sex, given available resources. For the species, reproductive success requires a male-biased sex ratio that presents an honest signal to attract pollinators, but natural selection does not shape populations directly.

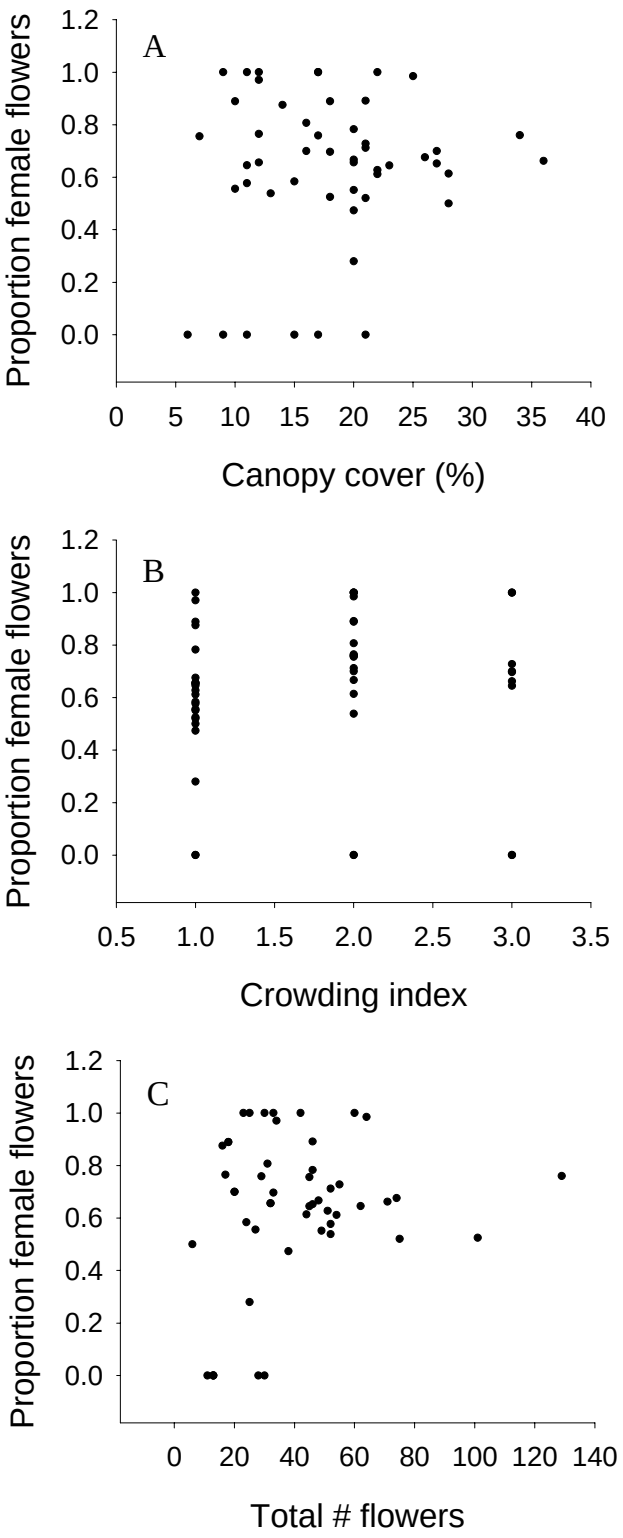

Figure 2. There was no relationship between the proportion of female flowers in begonia individuals and (a) canopy cover over the begonia ($p = 0.26$), (b) total number of flowers ($p = 0.57$), or (c) crowding by other plants ($p = 0.89$).

Bigger begonias tended to produce more female flowers, which supported the hypotheses that (1) the sex ratio of *B. involucrata* is plastic and (2) larger begonias tend to produce a larger proportion of more expensive female flowers, perhaps due to greater plant resources. We suggest that femaleness contributes more to individual fitness because having more in a male-biased system ensures that the probability of female fertilization is high. The increased maleness of smaller begonias may reflect the constraints of resource limitations and/or represent an optimal solution in the face of resource limitations. This finding raises the possibility that size constraints on sex ratios have the incidental effect of creating a male-biased sex ratio for the population as a whole, which is theoretically necessary to maintain pollinator visitation in this deceit-pollinated system.

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THE DISTRIBUTION OF *OREOPANAX XALAPENSIS* WITH RESPECT TO FOREST EDGES

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Abstract: Oreopanax xalapensis is a tropical tree species of potential medical and economic importance. Anecdotal reports characterize *O. xalapensis* as an inhabitant of cloud forest edges. We tested whether that was true in the cloud forest of Monteverde, Costa Rica. In fact, average stem density increased with increasing distance from the forest edge to 50 m within the forest, but the patterns varied among small, medium, and large size classes. Results suggest that strips of forest may be inadequate to conserve populations of *O. xalapensis*, but defensible generalizations will require sampling that is more spatially and temporally extensive than was possible here.

Key Words: Oreopanax xalapensis, *shade tolerance, edge effects, distribution*

INTRODUCTION

Oreopanax xalapensis, a tropical tree within the Araliaceae, is potentially important both medicinally and economically. Extracts from the leaves of *O. xalapensis* showed notable antifungal activity and cytotoxic activity against four different cancer cell lines, but were not toxic to normal, nonproliferating cultures of adult rat hepatocytes (Setzer 2000). Understanding the ecological distribution of this tree could aid in its conservation by helping to identify forest habitats where it occurs.

Haber et al. (1996) reported that *O. xalapensis* is more commonly found in secondary cloud forest, edge, and light gaps than in mature cloud forest but presented no data. We investigated whether this was true in the cloud forest near the biological station in Monteverde, Costa Rica, by measuring size-specific densities across replicated transects from the forest edge to interior.

METHODS

We surveyed the distribution of *O. xalapensis* on 20 - 21 January 2006 near the Monteverde Biological Station, Costa Rica. We randomly located seven points along 350 m of forest edge. These served as the origin for sample belt transects (50 m long and 10 m wide) that extended into the forest perpendicular to the edge. We identified all *O. xalapensis* individuals taller than 1 meter and calculated the stem density for ten 5 m-long segments along each belt transect. We measured diameter at breast height (DBH) for each individual, and grouped individuals into three DBH size classes (small: 0-2 cm, medium: 2-10 cm, large: 10-37 cm). We investigated how stem density changed with distance from edge, DBH size class, and transect with a general linear model using JMP 5.0.1.

RESULTS

Trees of all size classes occurred throughout the transects from forest edges to the interior (Figure 1), but average stem

density increased with distance from edge ($F_{1,204}$ = 16.6, P < 0.0001). The relationship between stem density and distance depended on size class (size x distance interaction: $F_{2,204}$ = 4.05, P = 0.02), with a positive trend in the medium class (r^2 = 0.20, df = 68, P < 0.0001) and no relationship with distance in the large or small size classes for both regressions (P > 0.1, df = 68).

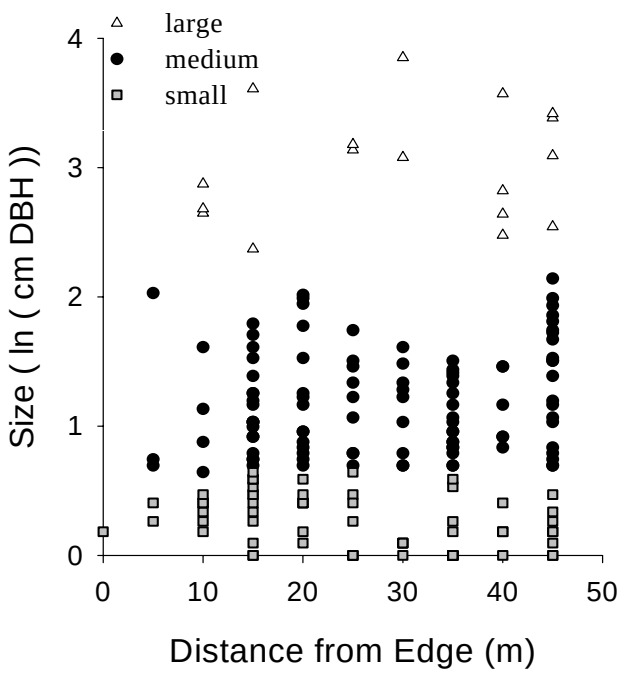

Fig. 1 Size (diameter) of *Oreopanax xalapensis* vs. distance from forest edge at Monteverde Biological Station, Costa Rica. Points are separated into 3 size classes (small: 0-2 cm; medium: 2-10 cm; large: 10-37 cm). Note increasing numbers of medium-size trees with increasing distance from edge.

DISCUSSION

We rejected the hypothesis that *O. xalapensis* density was highest at forest edges. In fact, we found that stem density increased with increasing distance from the forest edge. Because *O. xalapensis* was abundant in both small and medium size classes throughout the forest, we conclude that it must be a relatively shade tolerant species.

The interaction between stem density and distance from edge suggests some demographic tendencies in *O. xalapensis*. It may be that juveniles establish equally well from the forest edge to 50 m within the forest. In this case, higher survival rate in the interior versus the edge could generate the observed pattern of increasing stem density with distance from edge in the middle size class. The reversal to no affect of distance in the large size class could arise from higher survivorship of the medium size class closer to the edge. However, these interpretations assume a stable stage distribution. It is perhaps more likely that our data reflect historical variation in *O. xalapensis* establishment and/or survival. For example, forest re-growth into previously cleared land (i.e., edge shift) could have recently enhanced the probability that small saplings would survive to become medium-sized trees in what was the forest interior when we sampled the population. In this case, the distribution of *O. xalapensis* may differ at other edges due to different local environmental histories.

To our knowledge, this is the first quantification of the distribution and abundance of *O. xalapensis* with respect to forest edges. Given the general pattern of increased stem density with distance from forest edge, narrow strips of forest (< 100 m wide) could be inadequate to conserve sustainable populations of *O. xalapensis.*

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