EFFECTS OF DEFORESTATION AND RIPARIAN BUFFERS ON LOTIC COMMUNITIES IN SOUTHEASTERN COSTA RICA: IMPLICATIONS FOR BIODIVERSITY CONSERVATION IN TROPICAL STREAMS

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by
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AUTHORIZATION TO SUBMIT DISSERTATION

This dissertation of Christopher M. Lorion submitted for the degree of Doctor of Philosophy with a major in Natural Resources and titled “Effects of deforestation and riparian buffers on lotic communities in southeastern Costa Rica: implications for biodiversity conservation in tropical streams,” has been reviewed in final form. Permission, as indicated by the signatures and dates given below, is now granted to submit final copies to the College of Graduate Studies at the University of Idaho (UI), and to the Postgraduate School at the Centro Agronómico Tropical de Investigación y Enseñanza (CATIE) for approval.

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ABSTRACT

Freshwater biodiversity in the tropics is threatened by a variety of anthropogenic factors, and land cover change may have the most widespread and pervasive impacts. Costa Rica has experienced extensive deforestation, and there is a great need to understand how land cover change has affected stream ecosystems and investigate strategies for mitigating these impacts. Understanding the distribution of freshwater biodiversity in relatively pristine systems is also critical for effective conservation planning.

To examine the effects of deforestation on stream communities in southeastern Costa Rica and test the hypothesis that riparian forest buffers can reduce these effects, comparisons were made between forested stream reaches and stream reaches adjacent to pasture with and without a riparian forest buffer. Based on these comparisons, deforestation appeared to significantly alter the diversity and taxonomic composition of fish and benthic macroinvertebrate assemblages in the study streams, as well as stream temperatures and allochthonous inputs. Reaches with a riparian forest buffer, however, generally maintained aquatic assemblages and stream ecosystem conditions more similar to those observed in forested reaches. Overall, these results provide support for existing Costa Rican regulations mandating riparian forest protection, and suggest that appropriate riparian management could significantly reduce the impacts of deforestation on tropical stream biota.

To investigate landscape-scale patterns in fish diversity, stream fishes were sampled along an altitudinal gradient of nearly 500 m in the Sixaola River basin. Fish diversity decreased steeply with increasing elevation, and the main gradient in assemblage structure was strongly correlated with several stream habitat variables. Diadromous fishes were common throughout the altitudinal gradient and dominated the highest elevation sites, highlighting the importance of longitudinal connectivity in Mesoamerican river systems.

Systematic conservation planning that focuses on terrestrial and aquatic ecosystems, as well as biophysical linkages between these realms, could potentially identify more efficient and effective reserve networks. A critical evaluation of existing planning methods illustrates how incorporating linkages among ecosystems increases planning complexity. Several of the methods reviewed would be applicable to an example planning domain in Costa Rica and Panama, but new tools will be needed to realize the full potential of a comprehensive planning approach.

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INTRODUCTION TO THE DISSERTATION

Freshwater biodiversity is increasingly imperiled worldwide due to a variety of anthropogenic impacts that degrade aquatic habitats and fragment populations (Allan and Flecker 1993, Malmqvist and Rundle 2002). Tropical streams support extraordinarily diverse communities, and present special conservation challenges due to socioeconomic factors that constrain management options and the general lack of knowledge concerning the ecology of these systems (Pringle 2000). Therefore, there is a great need to gain a better understanding of tropical freshwater biodiversity while developing strategies to maintain the ecological integrity of tropical streams in increasingly human-dominated landscapes. The work presented here is aimed at filling current gaps in our understanding of freshwater biodiversity in an understudied area of Costa Rica, while looking for patterns that may apply more broadly to biodiversity conservation in tropical streams. The study area in southeastern Costa Rica, commonly known as Talamanca, is recognized as a globally significant center of terrestrial biodiversity (Olsen and Dinerstein 2002), but freshwater ecosystems and aquatic biodiversity also play a critical role in the sustainability of this region (Borge and Castillo 1997).

Among the factors threatening freshwater biodiversity, land cover change has particularly widespread and pervasive effects (Allan and Flecker 1993). Recent studies in diverse tropical environments have demonstrated that conversion of forests to agricultural land uses can have significant impacts on stream biodiversity (e.g. Benstead et al. 2003, Bojsen and Jacobsen 2003, Iwata et al. 2003). However, very few studies have examined the capacity of riparian forest buffers to mitigate the impacts of tropical deforestation on lotic communities. Chapters 1 and 2 of the dissertation present research examining the effects of deforestation and riparian forest buffers on fish and macroinvertebrates in lowland Costa Rican streams. In order to test the hypothesis that riparian forest buffers can mitigate the effects of deforestation, comparisons were made between forested reference stream reaches and stream reaches in pasture with and without a riparian forest buffer. Chapter 1 focuses on fish abundance and assemblage structure in the study streams, making comparisons among reach types and through time. To better understand the underlying causes of patterns observed in fish assemblages, comparisons of stream habitat and food resources available in the study reaches were also made. A similar approach is used in Chapter 2, but with a focus on benthic macroinvertebrates. In this case, comparisons among the study treatments, sampling periods and among habitat types within the sampling reaches were all of interest.

Focusing on reach-level effects of our study treatments eliminated many confounding factors when addressing research questions in the first two chapters. However, stream fishes often respond to heterogeneity in lotic systems at much larger spatial scales (Schlosser 1991, Fausch et al. 2002) and exhibit diverse responses to environmental gradients. Chapter 3 presents a study of the distribution of stream fishes along an altitudinal gradient in the Sixaola River basin, one the most intact river systems of its size on the Caribbean slope of Costa Rica. By focusing on streams with high ecological integrity within this river system, the goal of this study was to provide new insight into the factors shaping fish assemblages in the study region, develop a baseline for future comparative studies, and inform conservation planning efforts. Distributional patterns of diadromous fishes were of particular interest because the Sixaola River basin is one of a declining number of free-flowing river systems in the region.

Chapter 4 is an interdisciplinary chapter that discusses the conceptual basis and methodological framework for integrated conservation planning across terrestrial and aquatic ecosystems. Systematic conservation planning methods are widely used to identify conservation priority areas in terrestrial, freshwater, and marine ecosystems, and have traditionally considered these three realms separately. It is increasingly recognized that a more unified approach to planning is needed (e.g. Abell 2002, Stoms et al. 2004), but significant challenges remain in achieving this integration. In this chapter, the goals of systematic conservation planning across terrestrial and aquatic ecosystems are discussed, using a planning domain on the Caribbean slope of Costa Rica and Panama as an example. A critical evaluation of existing methods for integrated conservation planning based on several different criteria is presented next. The chapter ends with a discussion of how these methods could be applied in the example planning domain and the future developments needed to advance the comprehensive planning approach.

All chapters in this dissertation are formatted as manuscripts to be submitted for publication. Chapter 4 is the result of a collaborative effort by three doctoral students in the University of Idaho/CATIE IGERT Program, Beth Polidoro, Jan Schipper, and myself. Other co-authors on this chapter include Drs. J. Michael Scott and Celia Harvey.

REFERENCES

Abell, R. 2002. Conservation biology for the biodiversity crisis: a freshwater follow-up. Conservation Biology 16:1435-1437.

Allan, J. D., and A. S. Flecker. 1993. Biodiversity conservation in running waters. Bioscience 43:32-43.

Benstead, J. P., M. M. Douglas, and C. M. Pringle. 2003. Relationship of stream invertebrate communities to deforestation in eastern Madagascar. Ecological Applications 13:1473-1490.

Bojsen, B. H., and D. Jacobsen. 2003. Effects of deforestation on macroinvertebrate diversity and assemblage structure in Ecuadorian Amazon streams. Archiv für Hydrobiologie 158:317-342.

Borge, C., and R. Castillo. 1997. Cultura y conservación en la Talamanca indígena. Editorial Universidad Estatal a Distancia, San José, Costa Rica.

Fausch, K. D., C. E. Torgersen, C. V. Baxter, and H. W. Li. 2002. Landscapes to riverscapes: bridging the gap between research and conservation of stream fishes. BioScience 52:483-498.

Iwata, T., S. Nakano, and M. Inoue. 2003. Impacts of past riparian deforestation on stream communities in a tropical rainforest in Borneo. Ecological Applications 13:461-473.

Malmqvist, B., and S. Rundle. 2002. Threats to the running water ecosystems of the world. Environmental Conservation 29: 134-153.

Olsen, D. M., and E. Dinerstein. 2002. The Global 200: priority ecoregions for global conservation. Annals of the Missouri Botanical Garden 89:199-224.

Pringle, C. M. 2000. River conservation in tropical versus temperate latitudes. Pages 371-384 in P. J. Boon, B. R. Davies, and G. E. Petts, eds. Global perspectives on river conservation: science, policy, and practice. John Wiley & Sons Ltd., New York, New York, USA.

Schlosser, I. J. 1991. Stream fish ecology: a landscape perspective. BioScience 41:704-712.

Stoms, D. M., F. W. Davis, S. J. Andelman, M. H. Carr, S. D. Gaines, B. S. Halpern, R. Hoenicke, S. G. Leibowitz, A. Leydecker, E. M. P. Madin, H. Tallis, and R. R.Warner. 2005. Integrated coastal reserve planning: making the land-sea connection. Frontiers in Ecology and the Environment 3:429-436.

Chapter 1. Riparian forest buffers mitigate the effects of deforestation on fish assemblages in small tropical rainforest streams

Abstract

Riparian forest buffers may play a critical role in moderating the effects of deforestation on tropical stream ecosystems, but there is currently very little information on riparian forest buffer design or effectiveness from studies conducted in the tropics. We investigated the effects of deforestation on stream fish assemblages, and the influence of riparian forest buffers on this response, in southeastern Costa Rica in order to critically evaluate the effectiveness of forest buffers. Fish assemblages were sampled in small, lowland streams representing three different treatments: 1) forested reference stream reaches, 2) stream reaches in pasture with a riparian forest buffer averaging at least 15 m in width on each bank, and 3) stream reaches in pasture without a riparian forest buffer. Forest cover at the catchment scale was high at all sites, allowing us to isolate the reach-scale effects of our three study treatments. Fish density was consistently higher in pasture reaches than in forest and forest buffer reaches, mostly due to an increase in herbivore-detritivores, but fish biomass did not differ among reach types. Pasture reaches also had higher fish diversity than reference forest reaches, while forest buffer reaches were intermediate. Overall, fish assemblages in forest and forest buffer reaches were very similar, while those in pasture reaches were quite distinct. These patterns were persistent across three sampling periods during our 15-month long study. Stream ecosystem conditions in pasture reaches, which included higher stream temperatures and reduced fruit and seed inputs compared with forested reference reaches, appeared to favor fishes normally found in larger streams. Forest buffer reaches, in contrast, had stream temperatures and allochthonous inputs similar to forested streams in the study area. Our results provide support for existing Costa Rican legislation protecting riparian forests, and suggest that riparian areas should be given a high priority in efforts to maintain stream ecosystem integrity in the tropics.

Introduction

Tropical landscapes are increasingly dominated by agricultural land uses as a result of widespread and ongoing deforestation (Achard et al. 2002, Lambin et al. 2003). Although tropical deforestation is recognized as a critical threat to biodiversity (e.g. Pimm et al. 1995, Dirzo and Raven 2003), the effects of forest clearing on tropical stream ecosystems have received relatively little research attention and are poorly understood (Jackson and Sweeney 1995, Chapman and Chapman 2002). However, recent studies from diverse tropical environments have demonstrated that deforestation can have negative impacts on stream ecosystems and aquatic biota through degradation of stream habitats (Kamdem Toham and Teugels 1999, Iwata et al. 2003) and changes in food resources available to aquatic organisms (Bojsen and Barriga 2002, Benstead et al. 2003, Benstead and Pringle 2004).

Given the scale of tropical deforestation, the socioeconomic forces driving land use change, and the potential impacts on stream ecosystem function and aquatic biodiversity (Chapman and Chapman 2002, Benstead et al. 2003), there is a great need to develop strategies for maintaining the ecological integrity of stream ecosystems in agricultural landscapes. Protecting natural riparian vegetation along tropical streams is one of the most widely recognized strategies (e.g. Pringle and Scatena 1999, Chapman and Chapman 2002, Benstead et al. 2003, Casatti et al. 2006), and is supported by a large body of literature illustrating the importance of riparian zones to stream ecosystem function and the capacity of riparian areas to reduce the impacts of upslope land use on aquatic biota (Karr and Schlosser 1978, Gregory et al. 1991, Naiman and Décamps 1997). Specifically, forested riparian zones provide allochthonous organic matter inputs (e.g. large woody debris, leaf litter, terrestrial insects) that serve as food and habitat for stream organisms (Sweeney 1993, Pusey and Arthington 2003); filter sediments, nutrients, and pollutants from agricultural runoff (Peterjohn and Correll 1984, Osborne and Kovacic 1993, Lowrance et al. 1997); provide shade that moderates stream temperatures (Barton et al. 1985, Abell and Allan 2002); and help maintain channel features that enhance stream habitat and key ecosystem processes (Sweeney et al. 2004).

Despite the functional importance of riparian forests and their potential for maintaining stream ecosystem integrity in increasingly deforested tropical landscapes, few studies have evaluated the capacity of riparian forest buffers to mitigate the effects of deforestation on tropical stream biota. Forest buffer width recommendations from temperate studies (e.g. Castelle et al. 1994, Wenger 1999) may be equally valid in the tropics, but there is very little direct evidence to inform policy and management decisions concerning riparian forest protection. Furthermore, trophic dynamics and other stream ecosystem processes can differ substantially between temperate and tropical streams (e.g. Flecker 1992, Wootton and Oemke

1992, Wantzen and Wagner 2006), and may have significant implications for riparian buffer design and effectiveness.

In this context, we designed a study to investigate the influence of riparian forest buffers on tropical stream ecosystems, with a focus on stream fish assemblages. Fishes are an excellent indicator group for monitoring changes in stream ecosystems (Karr 1981, Fausch et al. 1990), and are strongly influenced by land use change, particularly at terrestrial-aquatic interfaces (Schlosser 1991). Many tropical fish species feed directly on allochthonous food items from surrounding forests (Goulding 1980, Lowe-McConnell 1987), and are expected to be particularly vulnerable to deforestation (Angermeier and Karr 1983). In addition, fishes can strongly influence the structure of other aquatic assemblages in tropical streams (Flecker 1992, Pringle and Hamazaki 1997, 1998). Therefore, changes in fish assemblages could have important consequences for stream ecosystem processes and biodiversity. Previous studies have demonstrated significant effects of deforestation on the trophic and taxonomic structure of neotropical stream fish assemblages (Burcham 1988, Bojsen and Barriga 2002), but the influence that riparian forest buffers might have on these effects is largely unknown.

The objectives of our study were to evaluate the capacity of riparian forest buffers to mitigate the effects of deforestation on stream fish assemblages, and to investigate specific stream habitat and food resource variables that could explain the patterns observed in fish assemblages. We pursued these objectives in a Costa Rican landscape where large patches of forest have been converted to pasture, an important land use trajectory throughout the Neotropics (Schelhas 1996, Lambin et al. 2003), but where forest cover remains relatively high overall. The patchy nature of deforestation in this area allowed us to compare pasture stream reaches with and without a riparian forest buffer to forested reference sites, while controlling to a large extent for catchment-scale land use. Stream reaches were identified as having a riparian forest buffer if they met the criteria for riparian forest protection set forth in the 1996 Costa Rican forestry law (no. 7575). This law prohibits the cutting or clearing of trees in riparian protection zones that vary in width depending on the slope of surrounding terrain and whether streams are located in rural or urban areas. The study was designed to incorporate these minimum protections as a treatment effect that could be compared with stream reaches lacking a forest buffer, and with streams in continuous forest.

Our overarching hypothesis was that riparian forest buffers could moderate the effects of deforestation on stream fish assemblages by maintaining stream ecosystem conditions similar to those found in continuous forest. Therefore, we expected that in streams affected by deforestation, the presence or absence of a riparian forest buffer would have a significant effect on fish assemblage structure, stream habitat characteristics and on the food resources available to stream fishes. Furthermore, based on previous studies of riparian buffer effectiveness in temperate environments, sites with an intact riparian forest buffer were predicted to be more similar to sites in continuous forest for all of these variables.

Methods

Study Site

The study was conducted in small, lowland streams in Limón Province, southeastern Costa Rica (9°35' N, 82°40' W). This area falls within the Bocas biogeographical province of Smith and Bermingham (2005), a region characterized by a relatively depauperate freshwater fish fauna with high endemism. Study streams drain the low hills between the Sixaola River Valley and the Caribbean coast, and are tributaries of either the lower Sixaola River or the small Gandoca stream system (Figure 1). The study area is underlain by a mix of tertiary sediments of marine origin and quaternary alluvium from several formations in the Talamanca mountain range (Bergoeing 1998). All study catchments fall within the moist tropical forest life zone of Holdridge (1967), with mean annual precipitation of approximately 2500 mm (WorldClim, Hijmans et al. 2005). Rainfall is largely aseasonal and highly variable between years, but tends to peak in November-January, while drier conditions often persist in February-April and August-September.

Twelve sampling reaches in nine first- and second-order streams were selected for the study (Figure 1). Our study design incorporated four replicates of three different stream reach types, hereafter forest, forest buffer, and pasture reaches, to investigate ecological responses to deforestation and the efficacy of riparian forest buffers. Forest reaches were selected as reference sites and had catchments dominated by primary and well-developed secondary forest with little or no past anthropogenic disturbance in near-stream areas. Riparian vegetation at these sites was characterized by a tall (>25 m), multi-strata forest canopy, a relatively open understory with many palms and some small trees and shrubs, and sparse herbaceous ground cover.

Forest buffer and pasture reaches, in contrast, had all been directly affected by deforestation and were adjacent to actively grazed pastures. Conversion of forest to pasture had occurred at least 10 years prior to our study at all of these sites. In forest buffer reaches, streams were separated from adjacent pastures by a riparian forest buffer averaging at least 15 m in width on both banks, in accordance with Costa Rican law for rural streams in flat terrain. Mean buffer width, averaging measurements on both banks, ranged from 26-45 m. Forest buffers were typically wider on one bank than the other, and buffer width on the bank with the narrower buffer averaged approximately 20 m across the four reaches. Riparian buffer width also varied considerably along the meandering stream channels and was less than 15 m at some points. Riparian vegetation in forest buffer reaches was composed of a multi-strata forest canopy with a mix of large remnant trees and younger secondary growth, a relatively dense understory, and low herbaceous ground cover. Streamside vegetation in pasture reaches was categorically different, typically having a single, often discontinuous, row of remnant and planted trees along the stream channel, and a nearly complete lack of understory trees and shrubs. Ground cover was very high, and included pasture grasses and closely cropped herbaceous plants that extended to the edge of the stream channel. There was considerable variation in tree cover among pasture reaches, representing a range of conditions common in the study area, but the canopy cover provided by the narrow strip of trees present in these sites was generally quite high (Table 1).

In addition to the riparian vegetation conditions discussed above, study reaches were selected based on having similar wetted widths and channel gradients (Table 1). The study was also designed to control, to the extent possible, for catchment-scale effects on the study streams. Therefore, we selected study reaches based on forest cover at the catchment scale, which was determined from aerial photos and extensive field reconnaissance. Forest cover was high in all of the study catchments (Table 1), but forests in the catchments of pasture and forest buffer reaches were generally more heavily disturbed by past clearing and selective logging than forests in the catchments of reference stream reaches. Deforested areas in the study catchments were used almost exclusively for pasture or small home gardens. At each study site, we established a sampling reach that was approximately 40 times the average wetted channel width, beginning at a randomly selecting starting point within a longer reach with similar habitat conditions. All study sites had pool-riffle channels (Montgomery and

Buffington 1997), and sampling reaches always captured at least three pool-riffle sequences. Stream water in the study sites had high conductivity and slightly basic pH (Table 1).

Fish Assemblages

Fish sampling was conducted with the goals of comparing stream fish assemblages among reach types and investigating how assemblages varied within and among years. Thus, we sampled all 12 study reaches once in each of following periods: February-April 2005 (sample period 1), September-October 2005 (sample period 2), and February-April 2006 (sample period 3). This sampling schedule was intended to represent dry and wet season samples in 2005 and a dry season sample in 2006. However, precipitation did not vary in a predictable seasonal pattern during the study. Furthermore, discharge in these small streams responded rapidly to rainfall events, and then quickly returned to base flow conditions when rains ceased. Given the lack of consistent seasonal patterns in precipitation and stream flow, and the fact that fish sampling was always conducted when streams were at or near base flow, the three sample periods will be referred to by their sequential order instead of by season.

Stream fishes were captured using a backpack electrofishing unit (Model B-12, Smith-Root, Inc., Vancouver, Washington, USA). Each geomorphic channel unit (pool or riffle) in the sampling reach was isolated using block nets and two thorough passes of equal effort were made through each habitat unit. Fish from the first pass were maintained in buckets, and at least 15 minutes was allowed between passes. Preliminary work indicated that a third pass yielded few fish and led to significantly higher fish mortality in these small streams. Captures from the two passes were combined for subsequent analyses. All fish were identified to species using keys in Bussing (1998), measured for length to the nearest mm, weighed to the nearest 0.1 gram, and then returned to the stream. Fishes of the genera *Eleotris* and *Sicydium* could not be identified to species reliably in the field, and so were recorded according to their respective genus. *Sicydium* was present at only one site, while *Eleotris* was present in low numbers at many of the sites. Given the similar ecology of the two species within each genus that occur in the study area (Bussing 1998, Winemiller and Ponwith 1998), we felt justified in grouping these species at the genus level in our analyses. Only fish ≥20mm were included in analyses, as smaller fish were not adequately sampled with our methods.

Fishes were also assigned to one of four trophic groups (herbivore-detritivore, omnivore, invertivore, or carnivore) based on previous studies in Costa Rica (Burcham 1988, Winemiller and Leslie 1992, Wootton and Oemke 1992, Bussing 1993, Winemiller 1993, Bussing 1998). Several species are known to consume terrestrial invertebrates, but the extent to which a particular species specialized in eating terrestrial or aquatic invertebrates varied among the studies cited above, and so all fishes that feed primarily on invertebrates were placed in a single group. Fishes were classified as carnivores if their diet regularly includes fish and freshwater shrimp, in addition to smaller invertebrates.

Stream Habitat and Food Resources

Stream habitat was evaluated in the study reaches in all three sample periods, usually 1-2 days prior to fish sampling. Based on an initial estimate of mean wetted stream width, 20 equally-spaced transects perpendicular to the stream flow were established. Stream wetted width and active channel width were measured to the nearest cm at each transect. Stream depth and substrate size were measured along transects using methods adapted from Iwata et al. (2003). Stream depth was measured at five equally-spaced points across the stream channel. At each depth sampling point, the dominant substrate in a 10 x 10 cm area surrounding the point was classified as sand or silt (particle size <2 mm), gravel (2-16 mm), pebble (17-64 mm), cobble (65-256 mm), boulder (>256 mm), or bedrock. These substrates were then coded as follows: 1, sand or silt; 2, gravel; 3, pebble; 4, cobble; 5, boulder; and 6, bedrock. The mean of the coded values for the reach was used as an index of substrate coarseness (Bain et al. 1985).

In order to estimate the relative abundance of pool and riffle habitats in the sampling reach, the total length and average width, based on three representative measurements, was recorded for each habitat unit. Channel gradient in each sampling reach was estimated using the length and slope of riffle habitats. Slope was measured using a clinometer, and pools were assumed to have a slope of zero. Stream channel canopy cover was calculated based on 20 measurements with a spherical densiometer. Measurements were made in mid-channel facing upstream, downstream and towards both banks at the top and bottom of the sampling reach, and at three equally spaced points between. At forest buffer sites, riparian buffer width on both banks was measured at the same five points. The total volume of large woody debris (LWD) in study reaches was quantified by measuring the diameter and length of all pieces ≥

10 cm in diameter and ≥ 1 m in length that were resting within the active stream channel. To compare thermal regimes between reach types, stream water temperatures in the study reaches were continuously monitored during April-May 2006 using submerged temperature loggers (HOBO Water Temp Pro, Onset Computer Corporation, Pocasset, Massachusetts, USA). Water temperatures were recorded every 30 minutes for one month at each site. Only six temperature loggers were available, and so loggers were maintained at two randomly selected sites of each reach type for one month, and then moved to the remaining sites.

Quantitative samples of potential food resources for fishes were also collected in conjunction with habitat surveys. In all three sample periods, 10 randomly placed Surber samples (sampling area 0.093 m^2) were collected to estimate benthic macroinvertebrate abundance. Surber samples were stratified by habitat type, with five samples collected from pools and five samples collected from riffles. Material captured in Surber samples was preserved in the field in 95% ethanol and invertebrates were later separated from other material in the laboratory using a dissecting microscope. Invertebrates were enumerated and identified to the lowest feasible taxonomic level. A habitat-weighted estimate of benthic macroinvertebrate density for each study reach was made based on the relative abundance of pools and riffles (Iwata et al. 2003).

Epilithic algae abundance in the study reaches was estimated in the latter two sample periods by measuring chlorophyll-*a* concentrations on natural stream substrates. At each site, one or more small stones were collected at 10 randomly-selected sampling points in the study reach. Sampling points were stratified so that five samples were collected in pools and riffles, respectively. In the laboratory, chlorophyll was extracted by placing stones in 96% ethanol for 24 hours at room temperature. Chlorophyll-*a* concentration in the extract was determined spectrophotometrically, without phaeophytin correction, using the equation of Wintermans and De Mots (1965). Stone surface area was calculated using the equation of Dall (1979), and one half of the stone area was used to estimate chlorophyll-*a* standing crop per square meter at each sampling point. Mean values in pool and riffle habitats were used to calculate a habitat-weighted estimate of chlorophyll-*a* standing crop for each study reach.

Direct aerial inputs of plant material and terrestrial invertebrates to the stream were also measured during the second two sample periods. Five clear plastic pans partially filled with soapy water, each with surface area of 0.2 m^2, were placed within or at the edge of the wetted stream channel and retrieved after 24 hours. Pan traps were used at all but one site, where repeated problems with theft and disturbance by livestock precluded data collection. In several cases, one or more samples were lost due to inclement weather and resultant increases in stream discharge. In cases where more than one sample was lost, all samples were discarded and pan traps were re-set for 24 hours. Material captured in the pan traps was preserved in 95% ethanol, and later sorted using a dissecting microscope. Terrestrial invertebrates were counted, identified to order or family, and sorted into two groups, fully terrestrial taxa and terrestrial insects with an aquatic larval stage. In some cases, particularly within the order Diptera, these distinctions required identifying insects to the family level and making assumptions based on general patterns within families. Terrestrial invertebrates, as well as fruits and seeds less than 25 mm in diameter, were dried at 60°C for 24 hours and then weighed to the nearest 0.1 mg. The few larger fruits and seeds collected in pan traps had a thick, hard pericarp and are unlikely to be consumed by fishes in the study streams.

Data Analysis

Comparisons of fish density, biomass, and species richness among reach types and sample periods were made with repeated measures ANOVAs using PROC MIXED in SAS Version 9.1 (SAS Institute Inc. 2002). Akaike Information Criterion values for small samples (AIC_c) were used to select the most appropriate covariance structure for within-subject variation in the repeated-measures models. When a significant effect of reach type was found for any variable, pairwise comparisons of the three reach types were made. We did not correct p-values for multiple pairwise comparisons in these tests, or in the tests described below, because F tests provide adequate protection against type I error for pairwise comparisons among three groups (Toothaker 1993). Species richness comparisons among sampling reaches may have been influenced by differences in sampling area and the total number of fish captured (Gotelli and Colwell 2001). Therefore, we used the program EcoSim (Gotelli and Entsminger 2005) to make individual-based rarefaction curves and estimate species richness for the study reaches based on an equal number of individuals. Diversity comparisons among reach types were made based on observed species richness and using richness estimates based on rarefaction to a common sample size.

Comparisons of fish assemblages among sampling sites were also made using two multivariate techniques, permutational multivariate analysis of variance (PERMANOVA) and non-metric multidimensional scaling (NMS). Both techniques are appropriate for analysis of community data that do not meet distributional assumptions of parametric tests (McCune and Grace 2002). Prior to analyses, fish abundance data were converted to relative abundance due to differences in sampling area among sites. Bray-Curtis dissimilarity (Bray and Curtis 1957) was used as the distance measure for PERMANOVA tests and NMS ordinations, and all species were included in the analyses.

PERMANOVA was used to test for differences in fish assemblages among reach types and sampling periods. Analyses were based on the methods of Anderson (2001) and McArdle and Anderson (2001), and were conducted using the program PERMANOVA (Anderson 2005). Data from all three sample periods were first analyzed together, with sample period and reach type as fixed factors, because we were interested in testing for significant changes in community composition through time and for possible interactions between the factors. However, because we used a repeated-measures study design, this test artificially inflated the sample size for reach type comparisons. Therefore, we also ran one-way PERMANOVA tests for the effect of reach type in each sampling period. P-values for the test statistic (pseudo F-value) in PERMANOVA tests were based on 10000 permutations of raw data. When one-way PERMANOVA tests indicated a significant effect of reach type, pairwise comparisons of reach types were made. Pairwise tests had few unique permutations, and so the Monte Carlo method of Anderson and Robinson (2003) was used to calculate p-values for the test statistic (multivariate t-statistic).

Relationships among sites based on fish relative abundance were also evaluated with NMS ordinations using the program PC-ORD (McCune and Mefford 1999). Final ordinations were based on the best configuration from 250 runs with random starting points. Appropriate dimensionality for ordinations was determined from scree plots and significance tests of stress values based on 250 Monte Carlo randomizations. Separate ordinations were run for each sample period to investigate how relationships among sites varied through time.

Habitat and food resource variables were compared among reach types and sample periods using repeated measures ANOVAs. For water temperature, a two-way ANOVA model with reach type and month as fixed effects was used. ANOVAs were conducted using SAS Version 9.1, and data transformations were made as needed to meet distributional assumptions.

Results

Fish Assemblage Structure

Over 13,000 fish representing 24 species and 11 families were captured during the study. Fish assemblages were dominated by poeciliid and characid fishes (Appendix 1), and all species encountered are native to the study area. Fish density varied significantly among reach types and sample periods (Table 2), and was higher in pasture reaches than in forest and forest buffer reaches (Figure 2a). Fish biomass also varied among sampling periods, but not among reach types (Table 2). Observed species richness varied significantly among reach types, but not among sampling periods (Table 2). Species richness was significantly higher in pasture reaches than forest reaches, while forest buffer reaches had intermediate species richness and did not differ significantly from the other two reach types (Figure 2b). Species richness estimates based on rarefaction to a common sample size yielded very similar results (Table 2), indicating that patterns in fish species richness were not an artifact of variation in sampling area or fish abundance.

Comparisons of fish density by trophic group showed no significant effect of reach type on the density of omnivores, invertivores, or carnivores (Table 2). The density of herbivore-detritivores did, however, differ significantly among reach types. Herbivore-detritivore density was highest in the pasture reaches, and differences in herbivore-detritivore density accounted for most of the variation in fish density among reach types (Figure 3). Herbivore-detritivore density was also significantly higher in forest buffer reaches than in forest reaches, but this difference was comparatively small. Among the trophic groups, only omnivores and herbivore-detritivores showed significant variation in density through time (Table 2).

PERMANOVA tests showed a highly significant effect of reach type on fish assemblages in the study reaches ($F = 14.23$, $p = 0.0001$). Sample period, in contrast, was not a significant factor ($F = 1.58$, $p = 0.14$), indicating that relative abundances were relatively stable through time. There was also no significant multivariate interaction between sample period and reach type ($F = 0.45$, $p = 0.96$). One-way PERMANOVA tests for individual sample periods also showed a significant effect of reach type (Table 3). Pairwise comparisons of reach types showed highly significant differences in fish assemblages between forest and pasture reaches and between forest buffer and pasture reaches in all three sample periods. Differences between forest and forest buffer reaches were not significant (Table 3).

NMS ordinations of sampling sites supported the conclusions of PERMANOVA tests. Ordinations produced two-dimensional configurations with relatively low stress for all three sample periods (Figure 4). The total variance explained by the two ordination axes, as indicated by coefficients of determination between distances in the ordination space and Bray-Curtis distances in the original n-dimensional space, was greater than 90% for all three ordinations. Forest reaches and forest buffer reaches consistently grouped together in multivariate space, while pasture reaches always formed a distinct group. Distances between individual reaches were also generally consistent across sampling periods (Figure 4).

Differences in fish assemblages in pasture reaches relative to other reach types were due in large part to the dramatic increase in herbivore-detritivores. However, we observed shifts in the relative abundance of species in other trophic groups, as well. For example, the small invertivore *Priapichthys annectens* was the most abundant fish in forest and forest buffer reaches, with densities three to four times those observed in pasture reaches. Decreased abundance of *P. annectens* in pasture reaches was matched by increased abundance of other invertivores, especially the poeciliid *Alfaro cultratus* and the characid *Hyphessobrycon panamensis*. Likewise, increases in the abundance of one omnivorous characid, *Astyanax aeneus*, in pasture reaches were offset to some degree by decreased abundance of another, *Bryconamericus scleroparius*, relative to forest and forest buffer reaches (Appendix 1).

Environmental Variables

We observed relatively few significant differences in comparisons of environmental variables among reach types and sample periods. Among habitat variables, only water temperature showed significant differences among reach types. Mean, minimum, and maximum daily water temperatures were all significantly higher in pasture reaches than in forest reaches during April-May 2006 (Table 4). Water temperatures in forest buffer reaches were intermediate and did not differ from the other two groups. Water temperatures also differed significantly among sampling months, but there was no significant interaction between reach type and sampling month for any of the temperature variables (Table 4). Reach type did not have a significant effect on the other stream habitat variables we measured (Table 5), but mean depth and LWD volume both varied among sample periods (Table 5).

Among food resource variables, only aerial inputs of small fruits and seeds differed significantly among reach types, with forest and forest buffer reaches having higher fruit and seed inputs than pasture reaches (Table 5, Figure 5a). In contrast, chlorophyll-*a* standing crop tended to be much higher in pasture reaches than in the other two reach types (Figure 5b). However, variation among pasture reaches was also very high and differences among reach types were not statistically significant (Table 5). There were also no significant differences or strong trends in aquatic macroinvertebrate density or aerial inputs of terrestrial invertebrates among reach types (Table 5).

Discussion

Patterns in Fish Abundance and Assemblage Structure

Comparisons of fish assemblages in our three study reach types suggest that deforestation has significant effects on stream fishes in lowland Costa Rican streams and support our hypothesis that riparian forest buffers can mitigate the effects of upslope deforestation. The differences we observed among treatments are particularly striking considering the relatively small scale of deforestation in the study catchments and the high canopy cover present in pasture reaches. Nevertheless, the geographic proximity of our sampling reaches and their similarity in size and channel gradient suggest that differences we observed among reach types were a direct consequence of land use change.

Dissimilarities in fish abundance, diversity, and assemblage structure were greatest between forest reaches and pasture reaches lacking a riparian forest buffer. In particular, pasture reaches had higher fish abundance and species richness, as well as significantly different trophic and taxonomic composition. In general, these differences were consistent with results from previous studies in neotropical streams (Burcham 1988, Bojsen and Barriga 2002), which found that deforestation significantly increased fish abundance, particularly the abundance of herbivorous species that constituted a very small component of fish assemblages in forested sites. The result was major shifts in fish assemblage trophic structure and species relative abundance patterns, with decreasing dominance by insectivorous and omnivorous fishes that feed on allochthonous material (Burcham 1988, Bojsen and Barriga 2002).

However, our results also demonstrated that these effects are strongly influenced by the presence of a riparian forest buffer. Despite similar land use patterns at the catchment and reach scale outside the riparian buffer zone in pasture and forest buffer reaches, we observed significant differences in fish abundance and assemblage structure between these reach types. Fish density was lower in forest buffer reaches than in pasture reaches for all fishes, mostly due to differences in the abundance of herbivore-detritivores. Comparisons based on the relative abundances of all fish species also revealed consistent and highly significant differences between pasture and forest buffer reaches. In all of these regards, forest buffer reaches supported fish assemblages that were much more similar to those in forested reference reaches.

Given land use patterns in the study catchments, differences between forest buffer and pasture sites appear to be driven almost completely by the ecological effects of riparian vegetation. However, comparisons of fish assemblage characteristics among the three reach types also showed that forest buffer sites were intermediate between forest and pasture reaches in many regards. This may be an indicator of catchment-scale effects that cannot be fully mitigated by riparian forest buffers, or may reflect the generally more disturbed nature of riparian vegetation in forest buffer reaches compared to forest reaches. Studies from temperate landscapes have shown mixed results regarding the relative importance of catchment versus riparian-scale land use on stream fish assemblages (Roth et al. 1996, Lammert and Allan 1999, Stauffer et al. 2000), and these relationships appear to be highly dependent on spatial scale (Lammert and Allan 1999, Allan 2004) and the extent of landscape modification (Wang et al. 2003). Nevertheless, deforestation in riparian areas can have strong impacts on fish assemblages even in watersheds with very high forest cover (Jones et al. 1999), as we observed in our study streams.

Patterns of fish abundance, diversity, and assemblage structure among the three study reach types were highly persistent across the three sample periods in our study, further reinforcing the significance of the patterns we observed. Despite low annual variation in temperature and day length, lowland tropical streams are highly dynamic environments and tropical stream fish assemblages can show marked temporal variation, often linked to seasonality of rainfall and resultant changes in stream discharge (Lowe-McConnell 1987). We did not observe any distinct seasonality in rainfall during our study, and patterns relating to surrounding land use appeared to be much stronger than temporal dynamics in fish assemblage structure. Bürnheim and Fernandes (2001) also reported very low seasonal variation in fish assemblage structure in small Amazonian rainforest streams, despite stronger seasonality in rainfall patterns. Fishes in our small study streams are subject to considerable abiotic disturbance in the form of frequent spates and extended periods of very low stream flow. Nevertheless, the relative stability of fish assemblages that we observed also suggests ample opportunities for strong biotic interactions.

Relationships between Fish Assemblages and Environmental Variables

Differences in fish assemblage structure among our three study reach types appeared to be most strongly linked to the availability of food resources. The abundance of herbivore-detritivores in pasture reaches was one the most obvious differences among reach types, and suggests that primary production was highest in pasture reaches, substantially lower in forest buffer reaches, and lowest in forest reaches. Comparisons of chlorophyll-*a* standing crop among reach types generally support this conclusion, although high variability among reaches resulted in a lack of statistically significant differences. One pasture reach, in particular, was a strong outlier, and had a very low algal standing crop throughout the study. This reach was not the most heavily shaded, but had experienced more channel degradation than other pasture reaches and had very fine, unstable sediments. Excluding this site, we did observe much higher chlorophyll-*a* standing crops in pasture reaches, consistent with many studies that have demonstrated an inverse relationship between algal abundance and stream canopy cover (e.g. Tait et al. 1994, Bojsen and Barriga 2002, Roy et al. 2006). However, we also observed large differences in chlorophyll-*a* standing crops between sites with very similar canopy cover, suggesting that other factors (e.g. nutrient availability, herbivory) were affecting algal abundance.

Allochthonous food inputs may have also influenced the differences in fish assemblages we observed among reach types. We did not observe differences in terrestrial insect inputs among reach types, contrary to other studies comparing terrestrial insect flux from forests and pastures (Edwards and Huryn 1996, Kawaguchi and Nakano 2001). However, our pasture reaches had relatively high tree cover compared to other studies. In addition, logistical constraints limited our trapping period to only 24 hours, and we observed high variability related to individual rain and wind events. A longer trapping period where all reaches were sampled simultaneously would provide a more robust comparison among reach types.

Despite these limitations, we did observe significantly higher fruit and seed inputs in forest and forest buffer reaches compared to pasture reaches. Several species in the study streams are known to feed on fruits and seeds (Bussing 1998), including two relatively large characid fishes, *B. scleroparius* and *Astyanax orthodus*. On average, these two species accounted for over 60% of fish biomass in forest and forest buffer reaches, and approximately 30% of fish biomass in pasture reaches. Stomach content analysis of specimens collected during sampling in 2006 (n=55 for *B. scleroparius*, n=60 for *A. orthodus*) revealed that allochthonous food items accounted for at least 90% of stomach contents for both species, regardless of reach type (C. M. Lorion and B. P. Kennedy, *unpublished data*). Fruits and seeds alone made up approximately 25% of the diet of *B. scleroparius* and 16% of the diet of *A. orthodus*. Pasture reaches in our study likely receive significant subsidies of allochthonous food resources from forested areas upstream, and we did not find statistically significant differences in the abundance or biomass of *A. orthodus* or *B. scleroparius* among reach types. Nevertheless, their combined biomass was, on average, approximately 40% lower in pasture reaches than in forest and forest buffer reaches. Characiform fishes are often a dominant component of neotropical fish assemblages (Lowe-McConnell 1987), and deforestation appears to have negative impacts on characids that depend on allochthonous resources (Bojsen 2005).

The influence of stream habitat on fish assemblages was less clear. Overall, we found relatively few differences in physical stream habitat among reach types, contrary to Iwata et al. (2003), who observed strong and persistent impacts of riparian deforestation on stream substrates and the distribution of channel units. The general lack of differences in habitat among reach types in our study is not entirely surprising given the small scale of deforestation in the study catchments. However, studies in Madagascar (Benstead et al. 2003) and Ecuador (Bojsen and Barriga 2002) also found very similar substrate characteristics in forested and deforested streams. Thus, the effects of deforestation on physical stream habitats appear to be highly context-specific and may depend on interactions between land use intensity in riparian areas and specific soil and climatic conditions. An understanding of these interactions may be crucial to understanding how changes in riparian vegetation will influence stream habitats and biota. As Benstead et al. (2003) note, the protection of forested headwater areas may be particularly important for minimizing sedimentation in tropical streams. In our study, substrate conditions among forest sites were relatively consistent, but forest buffer and pasture reaches showed large differences in the prevalence of fine sediments. Some forest buffer and pasture reaches were strongly affected by sedimentation, while others were not. These differences were not clearly related to land cover or buffer width, but appeared to be influenced by trampling of stream banks by livestock.

Among the habitat variables we measured, stream temperature showed the strongest differences among reach types. As expected, pasture sites had the highest temperatures, while temperatures in forested streams were significantly lower. Interestingly, forest buffer sites had intermediate temperatures that did not differ from the other two groups, suggesting that riparian vegetation can mitigate some, but not all, of the effects of deforestation on stream temperatures. The significance of the temperature differences we observed for fishes in the study streams is very difficult to determine, as we have no information on the thermal tolerance of any of the fish species present. Although the increases we observed in pasture sites were relatively small, they are substantial relative to the normal diel, and even annual, temperature variation in forested reaches, and may have important impacts on fish activity and metabolism.

Overall, we observed an increase in the importance of autochthonous production, decreased allochthonous inputs, and increased stream temperatures in pasture reaches, effects that are functionally similar to a downstream shift along the river continuum (Vannote et al. 1980). This shift was associated with an increase in herbivore-detritivore density and generally appeared to favor fish species characteristically found in larger streams (Winemiller and Leslie 1992, Bussing 1998). Although the headwater communities we observed are not dominated by endemic species, the patterns we observed are otherwise consistent with the native invasion process described by Scott and Helfman (2001) for streams in North America. Native invasions result in the homogenization of fish faunas and loss of unique headwater fish assemblages, and constitute a significant threat to freshwater biodiversity. The native invasion process also illustrates the limitations of species richness as a measure of ecosystem integrity and the importance of considering species composition when evaluating the effects of land use change on stream ecosystems (Scott and Helfman 2001).

Significantly, our results also suggest that riparian forest buffers can have a strong influence on the native invasion process by maintaining stream ecosystem conditions that favor characteristic headwater fish assemblages. Whether riparian forest buffers can serve a similar function in more extensively deforested tropical landscapes remains a key research question in general and with respect to current forestry regulations in Costa Rica. Designing studies to address this question is challenging because riparian buffer effects are nearly always confounded by catchment-scale land use patterns, longitudinal variation in buffer width, disturbance history, and natural environmental gradients (Allan 2004). By focusing on reach-scale effects, we were able to control for several of these confounding factors and demonstrate that riparian buffers have a significant influence on the response of stream biota to deforestation. Scaling these results up to more extensively deforested landscapes presents several challenges. For example, it remains unclear whether the patterns we observed among our study reaches were driven by habitat selection by adult fish or by recruitment dynamics in the study reaches. We often observed larvae and juveniles of the common poeciliid and cichlid species, but seldom encountered characids <20 mm in length. Closely-related characid species are known to move to distinct habitats for spawning and rearing (Kramer 1978), suggesting that the availability and spatial arrangement of habitats at larger spatial scales influenced the patterns we observed in our study reaches (Schlosser 1991, Fausch et al. 2002). In fact, the proximity of all study reaches to forested headwater areas may have allowed for the persistence of some species that would be absent in more extensively deforested areas. More research is clearly needed on the relationships between land use, riparian vegetation, and demographic processes of tropical stream fishes. Such information would improve our ability to predict the effects of deforestation on tropical stream communities, and would inform decisions about the trade-offs between production and conservation in agricultural landscapes.

In summary, our results demonstrate strong effects of deforestation on lowland tropical stream fish assemblages, and highlight the importance of riparian forests in mediating these impacts. Comparisons among our study reaches suggest that riparian forest buffers can reduce the effects of deforestation on lotic communities by maintaining stream ecosystem conditions similar to those found in forested catchments. Results from intensive sampling of benthic macroinvertebrates in the same study sites support this conclusion (Lorion et al. In Prep), but also illustrate that riparian forest buffers cannot fully mitigate the effects of land use change. Thus, although our results provide strong support for the principle of riparian forest protection, further research is needed in more extensively deforested catchments to determine the general effectiveness, and limitations, of riparian forest buffers. Similarly, our results support existing Costa Rican legislation protecting riparian forests, but evaluations in larger streams and in catchments with different land uses will be needed to determine whether current regulations provide sufficient protection for aquatic communities. Riparian forest buffers are not a panacea, but riparian forest protection and restoration should be given a high priority in efforts to maintain stream ecosystem integrity in tropical riverscapes.

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References

Abell, R., and J. D. Allan. 2002. Riparian shade and stream temperatures in an agricultural catchment, Michigan, USA. Verhandlungen Internationale Vereinigung fur Theoretische und Angewandte Limnologie 28:232-237.

Achard, F., H. D. Eva, H.-J. M. P. Stibig, J. Gallego, T. Richards, and J.-P. Malingreau. 2002. Determination of deforestation rates of the world's humid tropical forests. Science 297:999-1002.

Allan, J. D. 2004. Landscapes and riverscapes: the influence of land use on stream ecosystems. Annual Review of Ecology and Systematics 35:257-284.

Anderson, M. J. 2001. A new method for non-parametric multivariate analysis of variance. Austral Ecology 26:32-46.

Anderson, M. J. 2005. PERMANOVA: a FORTRAN computer program for permutational multivariate analysis of variance. Department of Statistics, University of Auckland, Auckland, New Zealand.

Anderson, M. J., and J. Robinson. 2003. Generalised discriminant analysis based on distances. Australian & New Zealand Journal of Statistics 45:301-318.

Angermeier, P. L., and J. R. Karr. 1983. Fish communities along environmental gradients in a system of tropical streams. Environmental Biology of Fishes 9:117-135.

Bain, M. B., J. T. Finn, and H. E. Booke. 1985. Quantifying stream substrate for habitat analysis studies. North American Journal of Fisheries Management 5:499-506.

Barton, D. R., W. D. Taylor, and R. M. Biette. 1985. Dimensions of riparian buffer strips required to maintain trout habitat in southern Ontario streams. North American Journal of Fisheries Management 5:364-378.

Benstead, J. P., M. M. Douglas, and C. M. Pringle. 2003. Relationship of stream invertebrate communities to deforestation in eastern Madagascar. Ecological Applications 13:1473-1490.

Benstead, J. P., and C. M. Pringle. 2004. Deforestation alters the resource base and biomass of endemic stream insects in eastern Madagascar. Freshwater Biology 49:490-501.

Bergoeing, J. P. 1998. Geomorfología de Costa Rica. Instituto Geográfico Nacional, San José, Costa Rica.

Bojsen, B. H. 2005. Diet and condition of three fish species (Characidae) of the Andean foothills in relation to deforestation. Environmental Biology of Fishes 73:61-73.

Bojsen, B. H., and R. Barriga. 2002. Effects of deforestation on fish community structure in Ecuadorian Amazon streams. Freshwater Biology 47:2246-2260.

Bray, J. R., and J. T. Curtis. 1957. An ordination of the upland forest communities of southern Wisconsin. Ecological Monographs 27:325-349.

Burcham, J. 1988. Fish communities and environmental characteristics of two lowland streams in Costa Rica. Revista de Biologia Tropical 36:273-285.

Bürnheim, C. M., and C. C. Fernandes. 2001. Low seasonal variation of fish assemblages in Amazonian rain forest streams. Ichthyological Exploration of Freshwaters 12:65-78.

Bussing, W. A. 1993. Fish communities and environmental characteristics of a tropical rain forest river in Costa Rica. Revista de Biología Tropical 41:791-809.

Bussing, W. A. 1998. Freshwater fishes of Costa Rica. Editorial de la Universidad de Costa Rica, San José, Costa Rica.

Casatti, L., F. Langeani, and C. P. Ferreira. 2006. Effects of physical habitat degradation on the stream fish assemblage structure in a pasture region. Environmental Management 38:974-982.

Castelle, A. J., A. W. Johnson, and C. Conolly. 1994. Wetland and stream buffer size requirements – a review. Journal of Environmental Quality 23:878-882.

Chapman, L. J., and Chapman, C. A. 2002. Tropical forest degradation and aquatic ecosystems: our current state of knowledge. Pages 237-249 in M. J. Collares-Pereira, M. M. Coelho, and I. G. Cowx, editors. Conservation of freshwater fishes: options for the future. Blackwell Science, Oxford, UK.

Dall, P. C. 1979. A sampling technique for littoral stone dwelling organisms. Oikos 33:106-112.

Dirzo, R., and P. H. Raven. 2003. Global state of biodiversity and loss. Annual Review of Environment and Resources 28:137-167.

Edwards, E. D., and A. D. Huryn. 1996. Effect of riparian land use on contributions of terrestrial invertebrates to streams. Hydrobiologia 337:151-159.

Fausch, K. D., J. Lyons, J. R. Karr, and P. L. Angermeier. 1990. Fish communities as indicators of environmental degradation. American Fisheries Society Symposium 8:123-144.

Fausch, K. D., C. E. Torgersen, C. V. Baxter, and H. W. Li. 2002. Landscapes to riverscapes: bridging the gap between research and conservation of stream fishes. BioScience 52:483-498.

Flecker, A. S. 1992. Fish trophic guilds and the structure of a tropical stream: weak direct vs. strong indirect effects. Ecology 73:927-940.

Gregory, S. V., F. J. Swanson, W. A. McKee, and K. W. Cummins. 1991. An ecosystem perspective of riparian zones. BioScience 41:540-551.

Gotelli, N. J., and R. K. Colwell. 2001. Quantifying biodiversity: procedures and pitfalls in the measurement and comparison of species richness. Ecology Letters 4:379-391.

Gotelli, N. J., and G. L. Entsminger. 2005. Ecosim: Null Models Software for Ecology, Version 7.72. Acquired Intelligence Inc. and Kesey-Bear, Jericho, Vermont, USA.

Goulding, M. 1980. The fishes and the forest: explorations in Amazonian natural history. University of California Press, Berkeley, California, USA.

Hijmans, R. J., S. E. Cameron, J. L. Parra, P. G. Jones, and A. Jarvis. 2005. Very high resolution interpolated climate surfaces for global land areas. International Journal of Climatology 25:1965-1978.

Holdridge, L. R. 1967. Life zone ecology. Tropical Science Center, San José, Costa Rica.

Iwata, T., S. Nakano, and M. Inoue. 2003. Impacts of past riparian deforestation on stream communities in a tropical rainforest in Borneo. Ecological Applications 13:461-473.

Jackson, J. K., and B. W. Sweeney. 1995. Present status and future directions of tropical stream research. Journal of the North American Benthological Society 14:5-11.

Jones, E. B. D., G. S. Helfman, J. O. Harper, and P. V. Bolstad. 1999. Effects of riparian forest removal on fish assemblages in southern Appalachian streams. Conservation Biology 13:1454-1465.

Karr, J. R. 1981. Assessment of biotic integrity using fish communities. Fisheries 6:21-27.

Karr, J. R., and I. J. Schlosser. 1978. Water resources and the land-water interface. Science 201:229-234.

Kamdem Toham, A., and G. G. Teugels. 1999. First data on an Index of Biotic Integrity (IBI) based on fish assemblages for the assessment of the impact of deforestation in a tropical West African river system. Hydrobiologia 397:29-38.

Kawaguchi, Y., and S. Nakano. 2001. Contribution of terrestrial invertebrates to the annual resource budget for salmonids in forest and grassland reaches of a headwater stream. Freshwater Biology 46:303-316.

Kramer, D. L. 1978. Reproductive seasonality in the fishes of a tropical stream. Ecology 59: 976-985.

Lambin, E. F., H. J. Geist, and E. Lepers. 2003. Dynamics of land-use and land-cover change in tropical regions. Annual Review of Environment and Resources 28:205-241.

Lammert, M., and J. D. Allan. 1999. Assessing biotic integrity of streams: effects of scale in measuring the influence of land use/cover and habitat structure on fish and macroinvertebrates. Environmental Management 23:257-270.

Lowe-McConnell, R. H. 1987. Ecological studies in tropical fish communities. Cambridge University Press, Cambridge, UK.

Lowrance, R., L. S. Altier, J. D. Newbold, R. R. Schnabel, P. M. Groffman, J. M. Denver, D. L. Correll, J. W. Gilliam, J. L. Robinson, R. B. Brinsfield, K. W. Staver, W. Lucas, and A. H. Todd. 1997. Water quality functions of riparian forest buffers in Chesapeake Bay watersheds. Environmental Management 21:687-712.

McArdle, B. H., and M. J. Anderson. 2001. Fitting multivariate models to community data: a comment on distance-based redundancy analysis. Ecology 82:290-297.

McCune, B., and J. B. Grace. 2002. Analysis of ecological communities. MJM Software Design, Gleneden Beach, Oregon, USA.

McCune, B., and M. J. Mefford. 1999. PC-ORD. Multivariate analysis of ecological data, Version 5.0. MjM Software, Gleneden Beach, Oregon, USA.

Montgomery, D. R., and J. M. Buffington. 1997. Channel-reach morphology in mountain drainage basins. Geological Society of America Bulletin 109:596-611.

Naiman, R. J., and H. Décamps. 1997. The ecology of interfaces: riparian zones. Annual Review of Ecology and Systematics 28:621-658.

Osborne, L. L., and D. A. Kovacic. 1993. Riparian vegetated buffer strips in water-quality restoration and stream management. Freshwater Biology 29:243-258.

Peterjohn, W. T., and D. L. Correll. 1984. Nutrient dynamics in an agricultural watershed: observations on the role of a riparian forest. Ecology 65:1466-1475.

Pimm, S. L., G. J. Russell, J. L. Gittleman, and T. M. Brooks. 1995. The future of biodiversity. Science 269:347-350.

Pringle, C. M., and T. Hamazaki. 1997. Effects of fishes on algal response to storms in a tropical stream. Ecology 78: 2432-2442.

Pringle, C. M., and T. Hamazaki. 1998. The role of omnivory in a neotropical stream: separating diurnal and nocturnal effects. Ecology 79:269-280.

Pringle, C. M., and F. N. Scatena. 1999. Aquatic ecosystem deterioration in Latin America and the Caribbean. Pages 104-113 in L. U. Hatch and M. E. Swisher, editors. Managed ecosystems: the Mesoamerican experience. Oxford University Press, New York, New York, USA.

Pusey, B. J., and A. H. Arthington. 2003. Importance of the riparian zone to the conservation and management of freshwater fish: a review. Marine and Freshwater Research 54:1-16.

Roth, N. E., J. D. Allan, and D. L. Erickson. 1996. Landscape influences on stream biotic integrity assessed at multiple spatial scales. Landscape Ecology 11:141-156.

Roy, A. H., C. L. Faust, M. C. Freeman, and J. L. Meyer. 2005. Reach-scale effects of riparian forest cover on urban stream ecosystems. Canadian Journal of Fisheries and Aquatic Science 62:2312-2329.

SAS Institute Inc. 2002. SAS Version 9.1. SAS Institute, Cary, North Carolina, USA.

Schelhas, J. 1996. Land-use choice and forest patches in Costa Rica. Pages 258-284 in J. Schelhas and R. Greenberg, editors. Forest patches in tropical landscapes. Island Press, Washington, D.C., USA.

Schlosser, I. J. 1991. Stream fish ecology: a landscape perspective. BioScience 41:704-712.

Scott, M. C. and G. S. Helfman. 2001. Native invasions, homogenization, and the mismeasure of integrity of fish assemblages. Fisheries 26:6-15.

Smith, S. A., and E. Bermingham. 2005. The biogeography of lower Mesoamerican freshwater fishes. Journal of Biogeography 32:1835-1854.

Stauffer, J. C., R. M. Goldstein, and R. M. Newman. 2000. Relationship of wooded riparian zones and runoff potential to fish community composition in agricultural streams. Canadian Journal of Fisheries and Aquatic Sciences 57:307-316.

Sweeney, B. W. 1993. Effects of streamside vegetation on macroinvertebrate communities of White Clay Creek in eastern North America. Proceedings of the Academy of Natural Sciences of Philadelphia 144:291-340.

Sweeney, B. W., T. L. Bott, J. K. Jackson, L. A. Kaplan, J. D. Newbold, L. J. Standley, W. C. Hession, and R. J. Horwitz. 2004. Riparian deforestation, stream narrowing, and loss of stream ecosystem services. Proceedings of the National Academy of Sciences of the USA 101:14132-14137.

Tait, C. K., J. L. Li, G. A. Lamberti, T. N. Pearsons, and H. W. Li. 1994. Relationships between riparian cover and the community structure of high desert streams. Journal of the North American Benthological Society 13:45-56.

Toothaker, L. E. 1993. Multiple comparison procedures. Sage Publications, Newbury Park, California, USA.

Vannote, R. L., G. W. Minshall, K. W. Cummins, J. R. Sedell, and C. E. Cushing. 1980. The river continuum concept. Canadian Journal of Fisheries and Aquatic Sciences 37:130-137.

Wang, L., J. Lyons, P. Rasmussen, P. Seelbach, T. Simon, M. Wiley, P. Kanehl, E. Baker, S. Niemela, and P. M. Stewart. 2003. Watershed, reach, and riparian influences on stream fish assemblages in the Northern Lakes and Forest Ecoregion, U.S.A. Canadian Journal of Fisheries and Aquatic Sciences 60:491-505

Wantzen, K. M., and R. Wagner. 2006. Detritus processing by invertebrate shredders: a neotropical-temperate comparison. Journal of the North American Benthological Society 25:216-232.

Wenger, S. 1999. A review of the scientific literature on riparian buffer width, extent, and vegetation. Institute of Ecology, University of Georgia, Athens, Georgia, USA.

Winemiller, K. O. 1993. Seasonality of reproduction by livebearing fishes in tropical rainforest streams. Oecologia 95:266-276.

Winemiller, K. O., and M. A. Leslie. 1992. Fish assemblages across a complex, tropical freshwater/marine ecotone. Environmental Biology of Fishes 34:29-50.

Winemiller, K. O., and B. J. Ponwith. 1998. Comparative ecology of eleotrid fishes in Central American coastal streams. Environmental Biology of Fishes 53:373-384.

Wintermans, J. F. G. M., and A. De Mots. 1965. Spectrophotometric characteristics of chlorophylls a and b and their pheophytins in ethanol. Biochimica et Biophysica Acta 109: 448-453.

Wootton, J. T. and M. P. Oemke. 1992. Latitudinal differences in fish community trophic structure, and the role of the fish herbivory in a Costa Rican stream. Environmental Biology of Fishes 35:311-319.

Table 1. Mean values (range) of physicochemical characteristics in forest, forest buffer, and pasture study reaches in southeastern Costa Rica (n = 4 for each reach type).

	Forest		Forest Buffer		Pasture	
Catchment Area (ha)	46	(26-79)	53	(15-103)	60	(15-130)
Catchment % Forest	97	(93-100)	83	(72-92)	81	(60-91)
Elevation (masl)	41	(25-55)	35	(30-38)	30	(20-37)
Channel Width (m)	2.0	(1.7-2.5)	1.7	(1.3-2.2)	1.8	(1.4-2.3)
Channel Gradient (%)	0.8	(0.6-1.1)	0.7	(0.4-0.8)	0.8	(0.5-0.9)
Canopy Cover (%)	93	(91-95)	94	(93-95)	78	(44-93)
Conductivity (μS/cm)	300	(239-403)	352	(160-442)	241	(210-286)
pH	7.9	(7.7-8.2)	8.0	(7.5-8.3)	7.6	(7.5-7.8)

Table 2. Results of repeated-measures ANOVA models of fish density, biomass, and species richness with reach type and sample period as fixed effects (n = 4 for each reach type).

	Reach Type		Sample Period		Reach Type* Sample Period	
	F	*p*	F	*p*	F	*p*
Density (individuals/m^2)	**4.87**	**0.037**	**3.96**	**0.038**	1.22	0.339
Biomass (g/m^2)	0.23	0.800	**5.36**	**0.015**	0.44	0.781
Species Richness (observed)	**6.95**	**0.015**	0.83	0.452	0.16	0.955
Species Richness (rarefaction)	**6.16**	**0.021**	1.67	0.217	0.37	0.827
Density By Trophic Group						
Herbivore-Detritivore[a]	**12.70**	**0.002**	**5.60**	**0.013**	2.16	0.115
Omnivore	1.76	0.227	**10.91**	**0.0008**	1.53	0.237
Invertivore	0.48	0.636	1.59	0.232	1.80	0.173
Carnivore[a]	0.85	0.457	2.28	0.131	0.35	0.843

Notes: Results in bold are significant at *P* < 0.05. For reach type, df = 2, 9; for sample period, df = 2, 18; for reach type*sample period interaction, df = 4, 18.

[a] $\log_{10}(x + 1)$ transformed for analysis

Table 3. Results of one-way PERMANOVA tests for differences in the species composition of fish assemblages between reach types in the three sample periods.

Sample	Reach Type		Pairwise Comparisons					
			Forest vs. Forest Buffer		Forest vs. Pasture		Forest Buffer vs. Pasture	
Period	F	*P*	t	*P*	t	*P*	t	*P*
1	**4.60**	**0.0044**	0.75	0.6359	**2.62**	**0.0072**	**2.45**	**0.0072**
2	**5.30**	**0.0015**	1.19	0.2748	**2.65**	**0.0063**	**2.45**	**0.0074**
3	**5.28**	**0.0018**	1.14	0.3013	**2.94**	**0.0038**	**2.25**	**0.0089**

Notes: Results in bold are significant at $P < 0.05$ (df = 2, 9 for F tests).

Table 4. Results of ANOVA models for the effects of reach type and sampling month on mean, minimum, and maximum daily water temperature in April-May 2006, with mean values (± SE) for each reach type.

	Reach Type		Month		Reach Type* Month		Mean ± 1 SE		
Variable	F	*P*	F	*P*	F	*P*	Forest	Forest Buffer	Pasture
Mean Temperature[1]	**8.20**	**0.019**	**20.55**	**0.004**	1.67	0.266	24.3^{a} ± 0.17	24.8^{ab} ± 0.39	25.3^{b} ± 0.29
Daily Minimum	**6.20**	**0.035**	**33.95**	**0.001**	2.29	0.182	23.9^{a} ± 0.22	24.1^{ab} ± 0.34	24.4^{b} ± 0.16
Daily Maximum[1]	**8.54**	**0.018**	**12.07**	**0.013**	1.87	0.234	24.8^{a} ± 0.12	25.5^{ab} ± 0.47	26.2^{b} ± 0.43

Notes: Results in bold are significant at $P < 0.05$. For reach type, df = 2, 6; for sampling month, df = 1, 6; for reach type*sampling month interaction, df = 2, 6. Different superscript letters indicate significant differences among reach types based on ANOVA models.
[1] $\log_{10}(x)$ transformed for analysis

Table 5. Results of repeated measures ANOVA models for habitat and food resource variables with reach type and sample period as fixed effects (n=4 for each reach type, except as noted below).

	Reach Type		Sample Period		Reach Type* Sample Period	
	F	*p*	F	*p*	F	*p*
Stream Habitat						
Mean Substrate Size	0.40	0.679	2.17	0.143	**4.59**	**0.010**
% Pool Habitat[a]	2.60	0.128	2.11	0.150	0.14	0.964
Mean Depth (cm)	3.68	0.068	**3.63**	**0.048**	0.84	0.518
LWD Volume[b]	1.96	0.196	**5.78**	**0.012**	**6.24**	**0.003**
Food Resources						
Macroinvertebrate Density (no./m²)[b]	0.37	0.700	1.32	0.292	0.07	0.990
Chlorophyll-*a* (mg/m²)[b]	1.44	0.287	**5.35**	**0.046**	3.69	0.068
Fruit and Seed Inputs (mg/m²)[b,c]	**6.08**	**0.025**	0.54	0.484	0.09	0.914
Terrestrial Insect Inputs (no./m²)[b,c]	0.34	0.720	0.11	0.745	0.88	0.451
Terrestrial Insect Biomass (mg/m²)[b,c]	2.02	0.195	**5.56**	**0.046**	1.29	0.326

Notes: Results in bold are significant at $P < 0.05$. Stream habitat variables and macroinvertebrate density were measured in all three sample periods. Other food resource variables were measured only in sample periods 2 and 3.

[a] arcsin square-root transformed for analysis

[b] $\log_{10}(x)$ transformed for analysis

[c] n=3 for pasture reaches

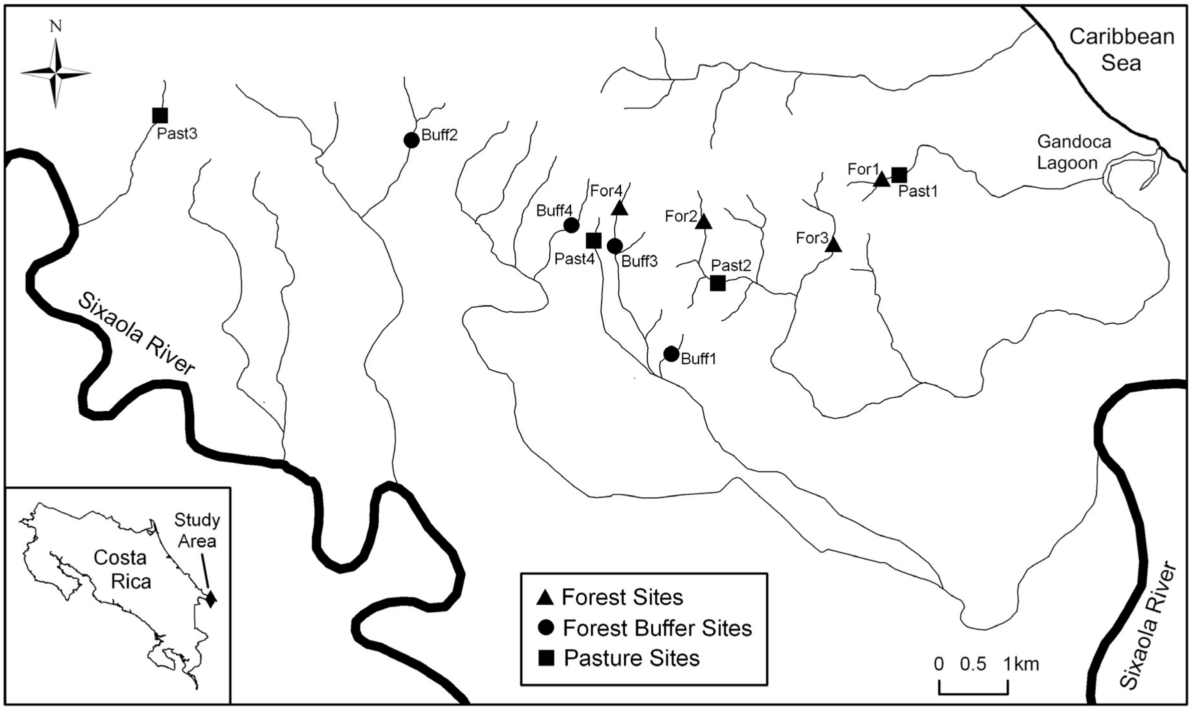


Figure 1. Map of study area in southeastern Costa Rica showing locations of the 12 study stream reaches.

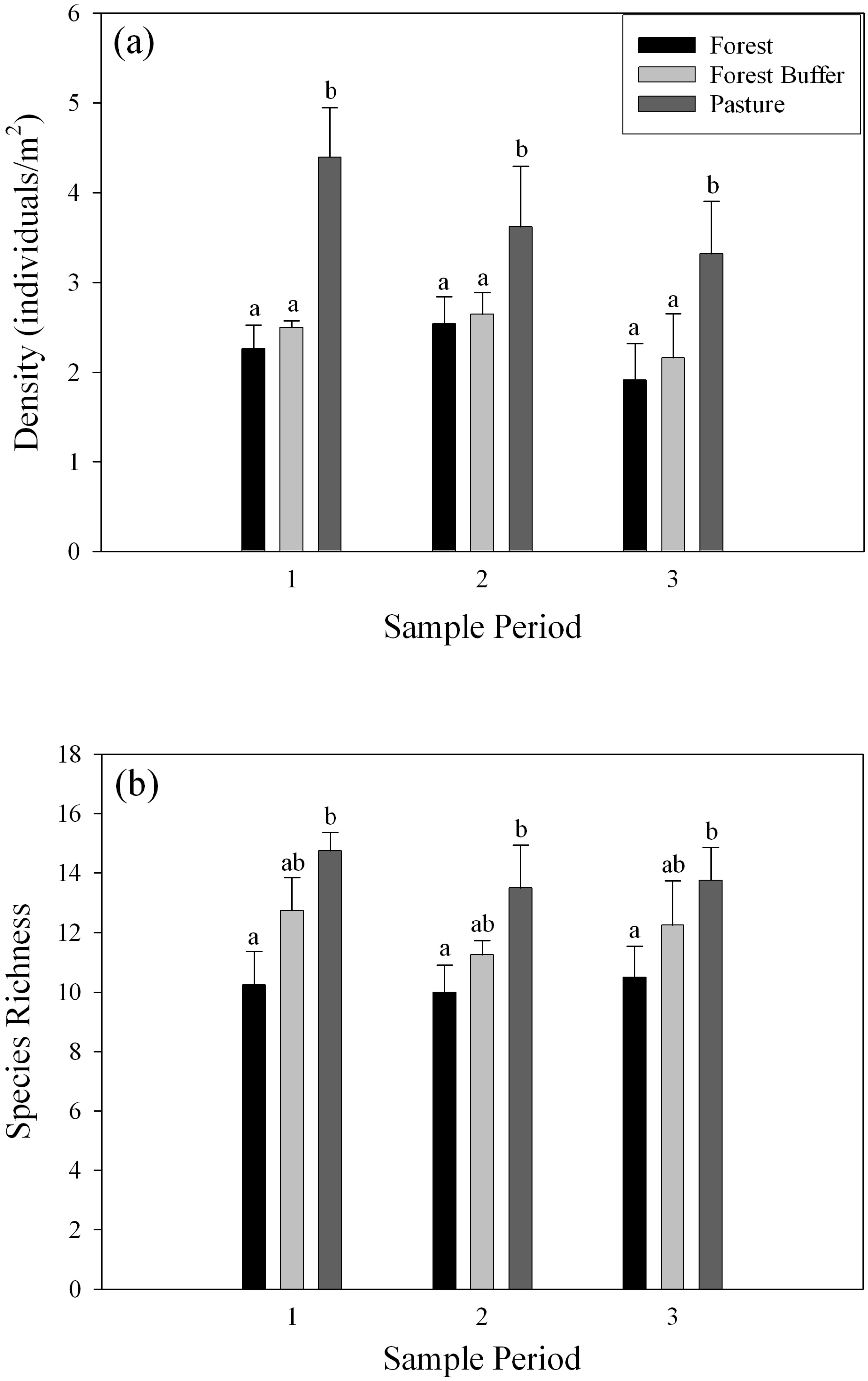


Figure 2. Plots of means (+SE) of (a) fish density (individuals/m^2), and (b) observed fish species richness for forest, forest buffer, and pasture reaches in the three sample periods. Bars with different letters indicate a significant difference between reach types ($P < 0.05$) based on repeated measures ANOVA models (see Table 2).

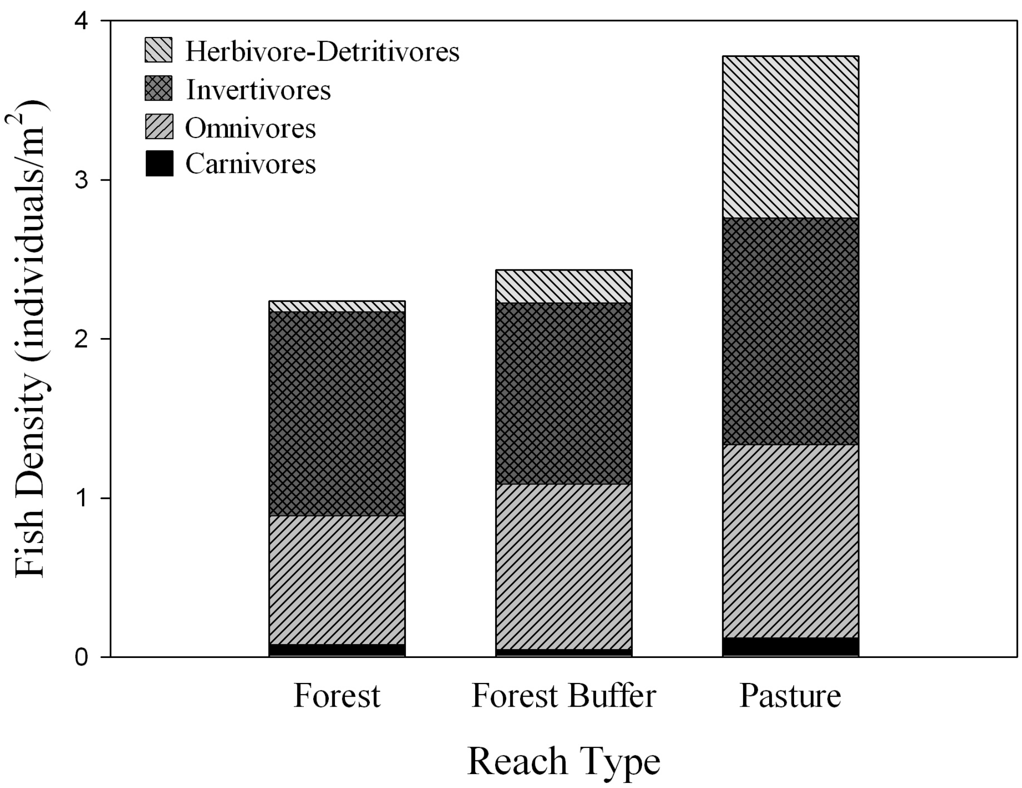
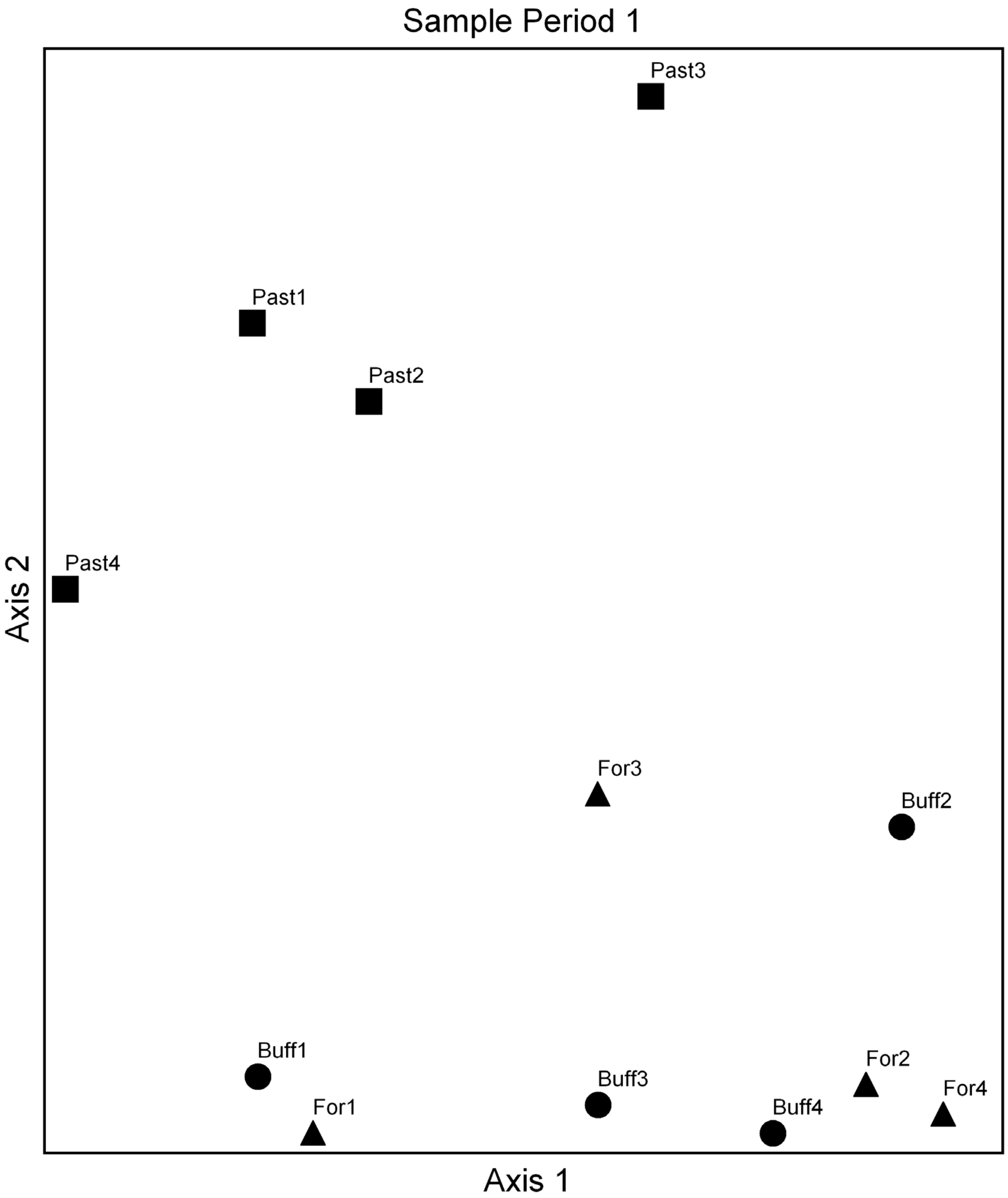
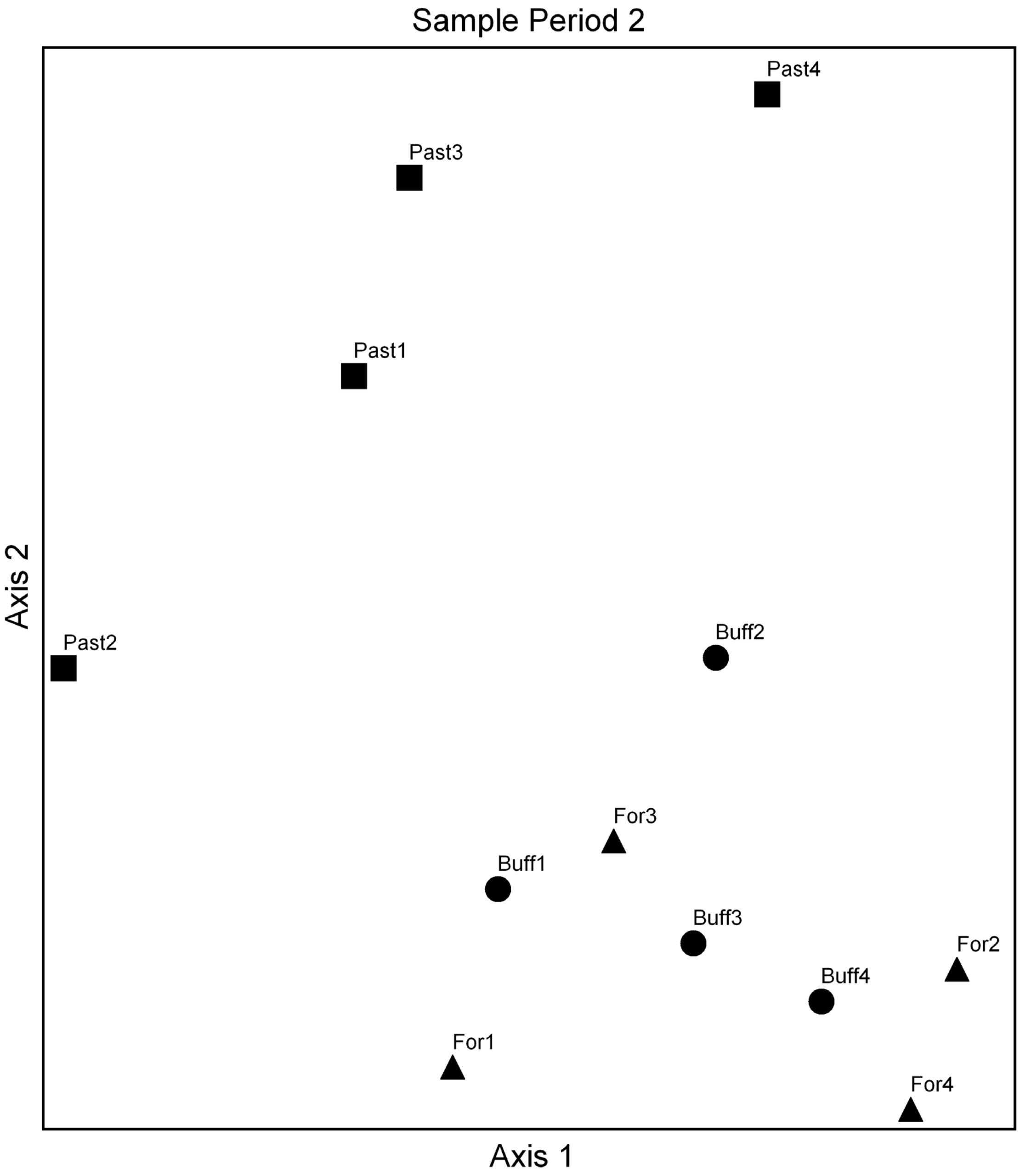


Figure 3. Mean fish density by trophic group in forest, forest buffer, and pasture reaches, all sample periods combined.

(a)


Sample Period 1
Past3
Past1
Past2
Past4
For3
Buff2
Buff1
For2
Buff3
For4
For1
Buff4
Axis 2
Axis 1

(b)


Sample Period 2
Past4
Past3
Past1
Past2
Buff2
For3
Buff1
Buff3
For2
Buff4
For1
For4
Axis 2
Axis 1

(c)

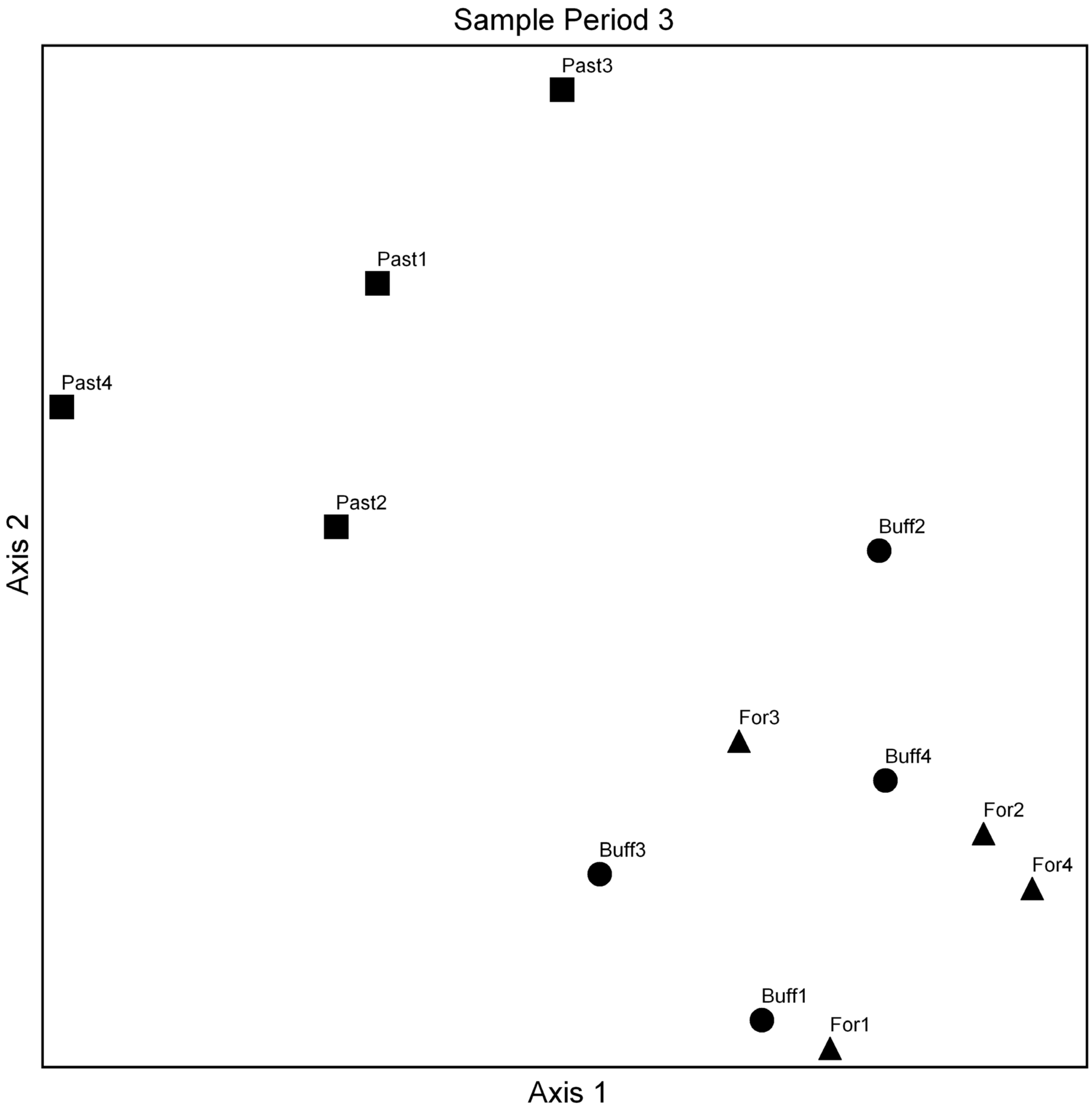


Figure 4. NMS ordinations of the 12 study reaches based on fish relative abundance for (a) sample period 1, (b) sample period 2, and (c) sample period 3. Symbols: ▲= forest; ● = forest buffer; ■ = pasture. Final stress values for the ordinations are 6.85, 8.30, and 8.19, respectively. Site codes correspond to those in Figure 1.

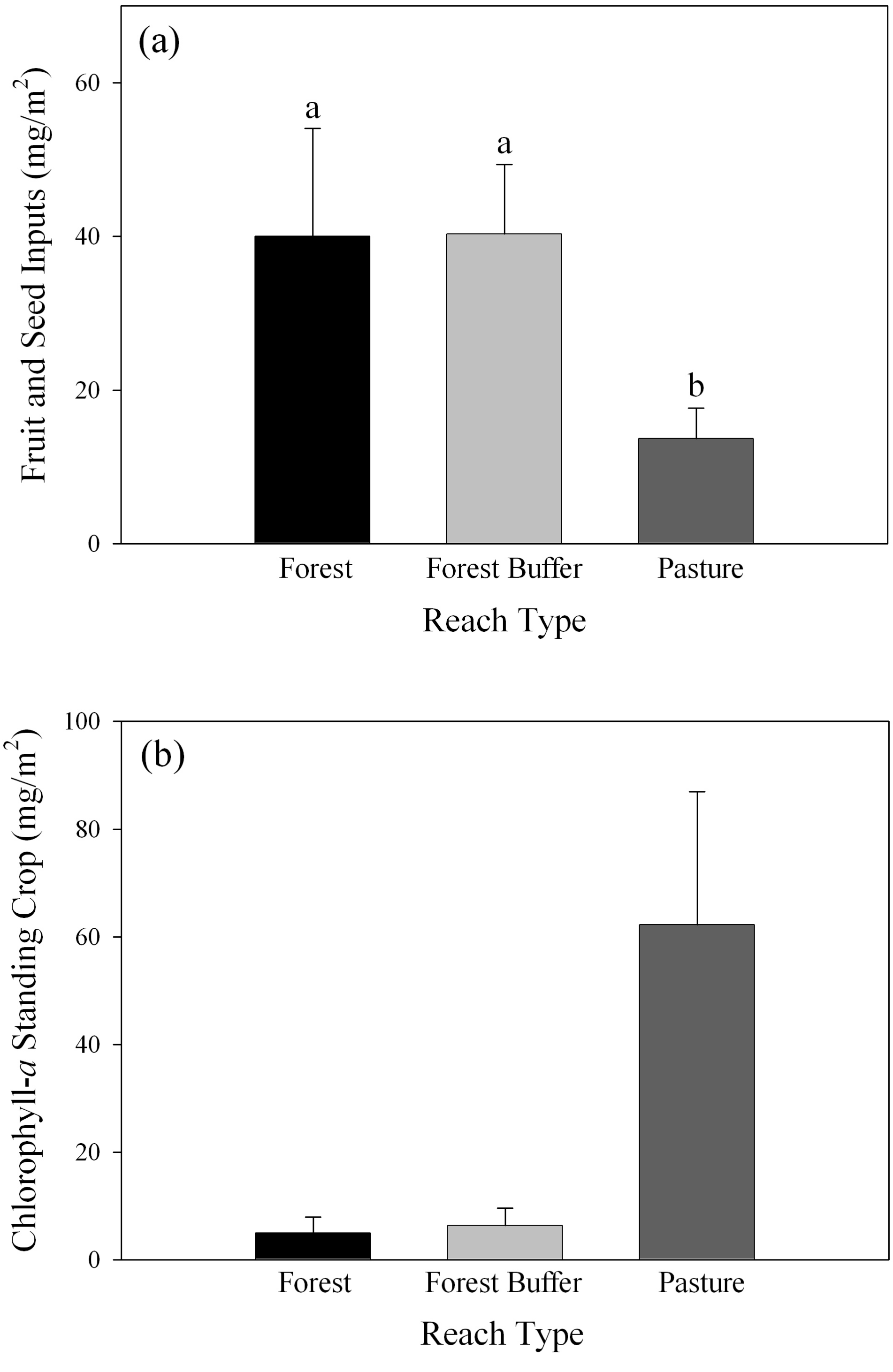


Figure 5. Mean values (+SE) for (a) aerial inputs of fruits and seeds, and (b) chlorophyll-*a* standing crop in forest, forest buffer and pasture reaches (data from sample periods 2 and 3 combined). Bars with different letters indicate significant differences among reach types ($P < 0.05$) from repeated measures ANOVA models based on log-transformed data (see Table 5).

Appendix 1. Fish species captured in the study streams, with mean density in each reach type across all sample periods and trophic classification for each species (C = carnivore, O = omnivore, I = invertivore, and H-D = herbivore-detritivore). Mean fish density and biomass for each reach type is included below.

	Fish Density (individuals/100 m^2)						Trophic Group
	Forest		Forest Buffer		Pasture		
	Mean	SD	Mean	SD	Mean	SD	
Family Anguillidae							
Anguilla rostrata	0	0	0	0	0.1	0.3	C
Family Characidae							
Astyanax aeneus	9.8	9.7	7.7	5.6	61.9	41.2	O
Astyanax orthodus	37.6	15.6	50.8	33.1	36.0	33.0	O
Bryconamericus scleroparius	30.7	28.3	37.9	29.8	15.3	18.7	O
Hyphessobrycon panamensis	0.3	0.6	0.6	1.2	15.2	17.4	I
Family Rhamphichthyidae							
Hypopomus occidentalis	0.5	0.8	0.3	1.0	0.8	1.4	I
Family Pimelodidae							
Rhamdia guatemalensis	1.3	1.7	2.3	3.6	2.5	2.9	C
Family Rivulidae							
Rivulus isthmensis	3.6	3.6	6.3	10.2	18.2	28.4	I
Family Poeciliidae							
Alfaro cultratus	13.7	15.0	20.7	14.4	79.1	26.5	I
Phallichthys amates	6.9	5.8	19.1	9.3	57.2	50.8	H-D
Phallichthys quadripunctatus	0	0	0.1	0.3	0.4	1.5	H-D
Poecilia gillii	0	0	1.4	3.1	41.3	56.4	H-D
Priapichthys annectens	109.9	51.5	86.1	34.0	28.3	17.3	I
Family Synbranchidae							
Synbranchus marmoratus	0.3	0.4	0.7	0.8	1.4	1.7	C
Family Cichlidae							
Astatheros bussingi	0.7	1.3	5.3	4.5	1.3	1.8	O
Astatheros rhytisma	0	0	0	0	0.4	0.8	O
Archocentrus myrnae	1.5	2.7	1.4	1.6	0.1	0.2	O
Archocentrus nigrofasciatus	0.4	0.7	0.6	0.7	6.1	5.8	O
Parachromis loisellei	0	0	0.1	0.3	0.7	0.7	C
Family Mugilidae							
Agonostomus monticola	0.04	0.1	0.2	0.7	0.3	0.6	O
Family Gobiidae							
Awaous banana	0.1	0.2	0	0	0.1	0.3	O
Sicydium sp.	0	0	0	0	2.8	5.5	H-D
Family Eleotridae							
Eleotris sp.	6.5	10.0	1.9	2.2	8.3	11.5	C
Gobiomorus dormitor	0.04	0.1	0	0	0	0	C
Mean Density (individuals/m^2)	2.24		2.44		3.78		
Mean Biomass (g/m^2)	5.99		6.82		7.08		

Chapter 2. Relationships between deforestation, riparian forest buffers, and benthic macroinvertebrates in lowland neotropical streams

Abstract

Riparian forest buffers could play an important role in mitigating the impacts of tropical deforestation on stream ecosystems, but few studies have documented riparian buffer effects in the tropics, and even less is known about how forest buffers might affect benthic communities. In this study, we investigated relationships between deforestation, riparian forest buffers, and benthic macroinvertebrate assemblages in small lowland streams in southeastern Costa Rica. In order to test our hypothesis that riparian forest buffers can mitigate the effects of deforestation on benthic macroinvertebrates, we made comparisons between three different stream reach types: 1) forested reference reaches, 2) stream reaches adjacent to pasture with a riparian forest buffer at least 15 m in width on both banks, and 3) stream reaches adjacent to pasture without a riparian forest buffer. Differences between forest and pasture reaches indicated that deforestation significantly altered the taxonomic composition and diversity of macroinvertebrate assemblages, and eliminated several of the most sensitive taxa. The presence of a riparian forest buffer appeared to reduce the effects of deforestation on benthic communities, as macroinvertebrate diversity and assemblage structure in forest buffer reaches were generally similar to those in forested reference reaches. One forest buffer reach was clearly an exception to this pattern, despite the presence of a wide riparian buffer. Differences among reach types were consistent in pool and riffle habitats and persisted across three sampling periods during our 15-month long study. Among the environmental variables we measured, only stream water temperature varied significantly among reach types, but trends in periphyton abundance and stream sedimentation may have also been important factors driving observed differences in macroinvertebrate assemblages. Forest cover was high upstream from all of our sites, and more research is needed to determine whether riparian forest buffers will sustain similar functions in more extensively deforested catchments. Nevertheless, our results provide support for Costa Rican regulations protecting riparian forests and suggest that proper riparian management could significantly reduce the impacts of deforestation on benthic communities in tropical streams.

Introduction

Widespread clearing of tropical forests for agriculture has transformed landscapes throughout the tropics in recent decades (Laurance 1999), and continues to affect millions of hectares annually (Achard et al. 2004). Although the effects of deforestation on tropical stream ecosystems have received relatively little research attention, there is increasing evidence that conversion of forests to agricultural land uses has pervasive impacts on benthic communities in tropical streams. In particular, deforestation can degrade stream habitats (Iwata et al. 2003, Wantzen 2006), increase stream water temperatures, and alter the abundance and diversity of basal food resources (Henry et al. 1994, Benstead et al. 2003, Bojsen and Jacobsen 2003, Benstead and Pringle 2004), leading to major shifts in benthic community structure and declines in macroinvertebrate diversity (Benstead et al. 2003, Bojsen and Jacobsen 2003, Iwata et al. 2003, Dudgeon 2006, Wantzen 2006).

Many of the effects of deforestation, including disruption of allochthonous resource subsidies, decreased stream shading, and increased sedimentation, are directly linked to processes occurring at terrestrial-aquatic interfaces (Gregory et al. 1991, Naiman and Décamps 1997), suggesting that forested riparian buffer zones could significantly reduce the impacts of deforestation on tropical streams (Pringle and Scatena 1999, Benstead et al. 2003). In fact, studies in temperate systems have demonstrated that riparian forest buffers can filter sediment and nutrients from agricultural runoff, stabilize stream banks, and provide shade that moderates stream temperatures and regulates instream primary production (Karr and Schlosser 1978, Peterjohn and Correll 1984, Osborne and Kovacic 1993, Sweeney 1993). Streamside forests also contribute leaf litter and other particulate organic matter that provides food and habitat for macroinvertebrates (Cummins et al. 1989, Sweeney 1993), often serving as the most important energy source for invertebrate production in headwater streams (Vannote et al. 1980, Wallace et al. 1997, Hall et al. 2001).

Although riparian forest buffers are expected to provide similar functions in tropical systems, studies documenting relationships between forest buffers and lotic communities in the tropics are conspicuously lacking. Recent studies have shown that riparian vegetation affects hydrological processes and sediment transport in humid tropical ecosystems (e.g. McKergow et al. 2004, Gomi et al. 2006) and is linked to physicochemical conditions in streams (Heartsill-Scalley and Aide 2003). However, information on how these factors ultimately affect aquatic communities remains remarkably scarce. Importantly, this limits our ability to collectively recommend meaningful riparian buffer strategies and predict how riparian management will influence biodiversity in tropical streams.

In this study, we investigated the extent to which riparian forest buffers moderate the effects of deforestation on benthic macroinvertebrate assemblages in small, lowland streams in southeastern Costa Rica. Costa Rica has historically had very high rates of deforestation (Sader and Joyce 1988, Sanchez-Azofeifa et al. 2001), but riparian forests are commonly maintained in agricultural areas. These forests are formally protected by the Costa Rican national forestry law (no. 7575), which prohibits the cutting or clearing of trees in riparian protection zones that vary in width depending on the slope of surrounding terrain and whether streams are located in rural or urban areas. We selected a study area where large blocks of forest have been converted to pasture, but which retains relatively high forest cover overall. These conditions allowed us to compare sites affected by deforestation with nearby forested stream reaches, while controlling to a large extent for catchment-scale land cover.

We hypothesized that riparian forest buffers would maintain habitat and trophic conditions similar to those found in forest streams, and thus reduce the impact of deforestation on macroinvertebrate assemblages. In order to test this hypothesis, we compared macroinvertebrate assemblages and a set of stream environmental variables among three stream reach types: 1) forested reference reaches, 2) stream reaches adjacent to pasture with a riparian forest buffer at least 15 m in width on both banks, and 3) stream reaches adjacent to pasture that lacked a forest buffer. The effects of land use change and other anthropogenic impacts on stream macroinvertebrates can vary considerably between different stream habitats (Kerans et al. 1992, Roy et al. 2003), so we also investigated whether relationships to deforestation and riparian forest buffers varied between pool and riffle habitats within our study reaches.

Methods

Study site

The study was conducted in Limón Province on the southern Caribbean slope of Costa Rica (9°35' N, 82°40' W). The study streams drain the hills between the Sixaola River Valley and the Caribbean coast, and are tributaries of either the Sixaola River or the Gandoca stream system (Figure 1). The study area falls within the moist tropical forest life zone of Holdridge

(1967), with mean annual temperatures around 26°C and mean annual precipitation of approximately 2500 mm (WorldClim, Hijmans et al. 2005). The area has a short dry season that generally falls between February-April. However, precipitation in the area is highly variable and does not exhibit strong seasonality, as illustrated by rainfall during our study at the nearest weather station with complete data (Figure 2).

Twelve sampling reaches in lowland streams (elevation 20-55 m) representing three different stream reach types (i.e. treatments) were selected for the study (Figure 1). Forest reaches were selected as reference sites and were located in forested catchments with little or no evidence of past anthropogenic disturbance in near-stream areas. Forest buffer and pasture reaches, in contrast, had all been directly affected by deforestation and were adjacent to actively grazed pastures. In forest buffer reaches, streams were separated from adjacent pastures by a riparian forest buffer averaging at least 15 m in width on both banks, in accordance with Costa Rican law for rural streams in flat terrain. Mean buffer width in these reaches, averaging measurements on both banks, ranged from 26-45 m. Forest buffers were typically wider on one bank than the other, however, and buffer width on the bank with the narrower buffer averaged approximately 20 m across the four reaches. Pasture reaches lacked a riparian forest buffer, but still had relatively high canopy cover (Table 1) due to the presence of a single row of remnant and planted trees along the stream channel. There was a nearly complete lack of understory trees and shrubs in pasture sites, and closely cropped grasses and herbaceous plants extended to the edge of the stream channel.

In addition to the riparian vegetation conditions discussed above, study sites were selected based on stream size and channel gradient (Table 1). Recognizing the potential importance of catchment-scale effects on the study streams, we also restricted our site selection to streams with similar levels of forest cover at the catchment scale (Table 1). In each site, we established a sampling reach that was approximately 40 times the average wetted channel width, starting at a randomly selected point in the middle of a longer reach with similar riparian conditions. All study reaches had pool-riffle channels (Montgomery and Buffington 1997), with pools generally occupying over 70% of the channel area. Riffles tended to be very shallow (average depth < 5 cm), and were also distinguished from pools by generally having higher current velocities and coarser substrates (Hawkins et al. 1993).

Environmental variables

Stream habitat conditions in the study reaches were assessed once in each of following periods: February-April 2005 (sample period 1), September-October 2005 (sample period 2), and February-April 2006 (sample period 3). This sampling schedule was intended to capture dry and wet season conditions in 2005 and dry season conditions in 2006. However, precipitation did not vary in a predictable seasonal pattern during the study (Figure 2). Furthermore, discharge in the small study streams responded rapidly to rainfall events, and then quickly returned to base flow conditions when rains ceased. Therefore, the three sampling periods will be referred to by their sequential order instead of by season.

In each study reach, we established 20 equally-spaced transects perpendicular to the stream flow to quantify channel depth and substrate characteristics following methods adapted from Iwata et al. (2003). Stream depth was measured at five equally-spaced points along each transect. At each depth sampling point, the dominant substrate in a 10 x 10 cm area surrounding the point was classified as sand or silt (particle size <2 mm), gravel (2-16 mm), pebble (17-64 mm), cobble (65-256 mm), boulder (>256 mm), or bedrock. These substrates were then coded as follows: 1, sand or silt; 2, gravel; 3, pebble; 4, cobble; 5, boulder; and 6, bedrock. The mean of the coded values for the reach was used as an index of substrate coarseness (Bain et al. 1985, Iwata et al. 2003). In sample period 3, substrate embeddedness was measured by visually estimating the percentage of the surface area of individual substrate particles embedded in fine sediments (to the nearest 25%) at each depth sampling point.

To determine the relative abundance of pool and riffle habitats in the sampling reach, we estimated the surface area of each habitat unit based on total unit length and average width from three width measurements. Channel gradient was estimated by measuring the slope of riffle habitats with a clinometer, and assuming that pools had a slope of zero. Stream channel canopy cover was calculated based on 20 measurements with a spherical densiometer. Measurements were made in mid-channel facing upstream, downstream and towards both banks at the top and bottom of the sampling reach, and at three equally spaced points between. At forest buffer sites, buffer width on each bank was measured at the same five points. Buffer width was only measured on one date because we did not observe changes in riparian vegetation at forest buffer sites during the study.

Measurements of stream water temperature, conductivity, and pH were taken at each site between 12:00 and 13:00 hours using a multi-sensor probe (YSI 556 MPS, Yellow Springs Instruments, Inc., Yellow Springs, Ohio, USA) on the day that habitat measurements were made. Two replicate water samples were also collected from all study reaches for water chemistry analysis on 1 May 2006, when streams were at base flow. Samples were placed on ice and transported to the laboratory, where they were analyzed for pH, conductivity, and concentrations of NO_3-N, NH_4-N, and PO_4-P using standard methods (APHA 1995).

Epilithic algae abundance in the study reaches was estimated in the latter two sample periods by measuring chlorophyll concentrations on natural stream substrates. At each site, one or more small stones were collected at 10 randomly-selected sampling points in the study reach. Sampling points were stratified so that five samples were collected in pools and riffles, respectively. In the laboratory, chlorophyll was extracted by placing stones in 96% ethanol for 24 hours at room temperature. Chlorophyll-*a* concentration in the extract was determined spectrophotometrically, without phaeophytin correction, using the equation of Wintermans and De Mots (1965). Stone surface area was calculated using the equation of Dall (1979), and one half of the stone area was used to estimate chlorophyll-*a* standing crop per square meter at each sampling point. Mean values from the five sampling points in each habitat type were used to estimate chlorophyll-a standing crops in pools and riffles, respectively. Estimates of leaf litter standing crops in pools and riffles were made using coarse benthic organic matter collected in Surber samples (see below). Leaf litter was sorted from woody material, dried at 60°C for 48 hours, subsampled, and then ashed at 500°C for 2 hours to determine ash-free dry mass.

Macroinvertebrate Sampling

Benthic macroinvertebrates were sampled on the same day that habitat measurements were made in all three sample periods. At each site, 10 Surber samples (0.093 m^2, 1000 μm mesh) were collected from randomly selected locations within the study reach. Sampling locations were stratified by habitat type so that five samples were collected from pools and riffles, respectively. Substrates within the sampling area were disturbed to a depth of approximately 10 cm and larger stones were scrubbed by hand to remove all attached invertebrates. Material captured in Surber samples was preserved in the field in 95% ethanol, and invertebrates were later separated from other material using a dissecting microscope.

All macroinvertebrates collected were identified to the lowest feasible taxonomic level (genus or family for aquatic insects) using Merritt and Cummins (1996), Roldán (1996), and Springer and Hanson (In Prep). Total taxa richness and Fisher's alpha diversity index were then calculated for pool and riffle habitats in each study reach. Fisher's alpha was used because it is relatively insensitive to sample size (Magurran 1988). The number of taxa in the orders Ephemeroptera, Plecoptera, and Trichoptera (EPT richness) in pools and riffles at each site was also determined because this metric is often used as an indicator of water quality (Rosenberg and Resh 1993). We measured the body length of macroinvertebrates to the nearest 1 mm, and estimated aquatic insect biomass using published length-mass equations (Benke et al. 1999) for taxa in the same genus or family, or with a similar body shape. We did not attempt to estimate biomass for other invertebrates because we lacked length-mass equations for many common non-insect taxa. Macroinvertebrates were assigned to functional feeding groups based on Merritt and Cummins (1996) and Tomanova et al. (2006) when taxonomic resolution permitted.

Data Analysis

Environmental variables and macroinvertebrate abundance and diversity measures were compared among reach types and sample periods with repeated measures ANOVAs using PROC MIXED in the program SAS (Version 9.1, SAS Institute Inc. 2002). Akaike Information Criterion values for small samples (AIC_c) were used to select the most appropriate covariance structure for within-subject variation in the repeated-measures models. For environmental variables that were only measured in one sample period, one-way ANOVAs or Kruskal-Wallis tests were used to test for significant differences among reach types, depending on whether or not distributional assumptions for a parametric test were met. For variables measured in pool and riffle habitats separately, including all macroinvertebrate variables, repeated measures ANOVA models were used to test for differences among reach types, sample periods, and habitat types (pools and riffles). Variables were log-transformed to meet distributional assumptions when necessary. Chlorophyll-a abundance in pool samples from one forest reach were below the detection limits of our measurement method, and so an estimate based on these detection limits was used as a conservative replacement for this value. When a significant effect of reach type was found for any variable, pairwise comparisons of the three reach types were made. We did not correct p-values for multiple comparisons in these tests, or in the tests described below, because F tests provide adequate protection against type I error for three or fewer pairwise comparisons (Toothaker 1993).

To investigate the effects of reach type, habitat type, and sampling period on the taxonomic composition of macroinvertebrate assemblages, we used two multivariate techniques, permutational multivariate analysis of variance (PERMANOVA) and non-metric multidimensional scaling (NMS). Both techniques are robust to departures from the distributional assumptions of parametric tests, and are therefore appropriate for the analysis of community data (McCune and Grace 2002). Prior to analysis, macroinvertebrate abundance data were $\log_{10}(x + 1)$ transformed and rare taxa, defined as taxa represented by a single individual across all sampling sites and periods, were removed from the data set. This resulted in the removal of 12 of the 105 taxa collected in the study. Bray-Curtis dissimilarity (Bray and Curtis 1957) was used as the distance measure for PERMANOVA tests and NMS ordinations.

PERMANOVA tests were based on the methods of Anderson (2001) and McArdle and Anderson (2001), and were conducted using the program PERMANOVA (Anderson 2005). Data from all three sample periods were analyzed together, with sample period, reach type (forest, buffer, or pasture), and habitat type (pool or riffle) as fixed factors. P-values for the test statistic (pseudo F-value) in PERMANOVA tests were based on 10000 permutations of raw data. When three-way PERMANOVA tests indicated a significant effect of reach type, pairwise comparisons of reach types were made, also based on 10000 permutations.

PERMANOVA tests may have overestimated differences among reach types and habitat types because we used a repeated measures study design. Therefore, NMS ordination was used to investigate how consistently samples from different habitat types and reach types were segregated in multivariate space. NMS ordination was conducted using the program PC-ORD (Version 5, McCune and Mefford 1999), and the final ordination was based on the best configuration from 250 runs with random starting points. Appropriate dimensionality for the ordination was determined from scree plots and significance tests of stress values based on 250 Monte Carlo randomizations. The contributions of individual taxa to differences between reach types and habitats were determined using the two-way crossed SIMPER routine in the program PRIMER (Version 6, Plymouth, UK).

Results

Environmental Variables

Stream habitat conditions varied considerably among the study reaches (Table 1), yet no statistically significant differences among reach types were observed for mean depth, substrate coarseness, or the proportion of riffle habitat (results not shown). Substrate embeddedness tended to be lower in forest reaches than in the other reach types (Table 1), but differences among reach types were not statistically significant ($P = 0.147$, Kruskal-Wallis test). Among the measured physicochemical variables, only stream water temperature differed significantly among reach types ($F_{2,9} = 4.29$, $P = 0.049$, data log-transformed for analysis), with pasture reaches having higher water temperatures than forest reaches. Forest buffer reaches had intermediate water temperatures and did not differ from the other two reach types. Stream temperatures also varied significantly among sample periods ($F_{2,9} = 28.13$, $P < 0.001$), and were lowest in the third sample period, when rainfall was highest. All streams had relatively high conductivity and slightly basic pH, and nutrient concentrations did not show any consistent differences among reach types (Table 1).

Leaf litter standing crops also showed no consistent differences among reach types (Table 1). In the repeated measures ANOVA, only sample period had a significant effect on leaf litter standing crop ($F_{2,36} = 6.27$, $P = 0.005$). The ANOVA for log-transformed chlorophyll-*a* standing crop showed marginally significant effects of reach type ($F_{2,18} = 3.13$, $P = 0.068$) and sample period ($F_{1,18} = 4.10$, $P = 0.058$), as well as a significant interaction between these factors ($F_{2,18} = 3.61$, $P = 0.048$). Chlorophyll-*a* standing crops tended to be greater in pasture reaches than in forest and forest buffer reaches, but there was high variability within reach types (Table 1).

Macroinvertebrate Abundance and Diversity

No significant differences in macroinvertebrate density among reach types were observed, regardless of whether data were summarized at the reach level or separated by habitat type (Table 2). There were also no significant differences in macroinvertebrate density among sample periods. However, we did observe a significant difference between habitat types (Table 2), with riffles having significantly higher macroinvertebrate densities (mean = 1396 individuals/m^2) than pools (mean = 790 individuals/m^2). Very similar patterns were observed for aquatic insect biomass, which did not differ among reach types or sample periods, but was significantly higher in riffles than in pools.

A total of 105 macroinvertebrate taxa were identified in the study streams, including 91 aquatic insect taxa representing 56 families (Appendix 1). Total taxa richness across all sampling periods ranged from 45-67 taxa for individual sampling sites, and there were no significant differences in taxa richness among reach types or sample periods. There was a significant difference in taxa richness between habitat types (Table 2), with riffles having higher taxa richness than pools. In contrast to taxa richness, alpha diversity did differ among reach types (Table 2), and was significantly higher in forest reaches than in pasture reaches (Figure 3). Alpha diversity in forest buffer reaches was intermediate and did not differ from the other two reach types (Figure 3). The difference in alpha diversity among habitat types was only marginally significant (Table 2), indicating that the difference in taxa richness between pools and riffles was mainly driven by sample size. EPT taxa richness also differed among reach types (Table 2), with forest and forest buffer reaches both having significantly higher EPT richness than pasture reaches (Figure 3). No significant differences in EPT richness were observed among sample periods or habitat types, although there was a trend toward higher EPT richness in riffles relative to pools (Figure 3).

Macroinvertebrate Assemblage Structure

Macroinvertebrate assemblages in the study streams were numerically dominated by aquatic insects (Figure 4). Non-insect taxa, especially gastropods and oligochaete worms, were much more common in pasture reaches, but still made up less than 20% of all macroinvertebrates collected in these sites. Ephemeroptera made up a much larger proportion of the insect fauna at forest and forest buffer reaches than in pasture reaches, where Diptera had a higher proportional abundance (Figure 4). Differences in taxonomic composition between pool and riffle habitats could also be seen at the ordinal level. Diptera made up a larger proportion of total macroinvertebrate abundance in pool habitats than in riffle habitats in all reach types, while Trichoptera and Coleoptera tended have higher relative abundances in riffles (Figure 4).

Multivariate comparisons of macroinvertebrate assemblages based on our finest taxonomic resolution for all taxa (see Appendix 1) also showed consistent differences among habitats and reach types. PERMANOVA tests indicated that reach type, sample period, and habitat all had significant effects on the composition of macroinvertebrate assemblages in the study reaches (Table 3). The effect of sampling period was relatively weak, while reach type and channel unit type were both highly significant (Table 3). Pairwise comparisons among reach types showed significant differences in macroinvertebrate assemblages between forest and pasture reaches (t = 2.91, *P* = 0.0001) and between forest buffer and pasture reaches (t = 2.35, *P* = 0.0001). Differences between forest and forest buffer reaches were not as pronounced, but were also significant (t = 1.51, *P* = 0.0344).

Ordination of the sampling sites based on log-transformed macroinvertebrate abundance illustrated how assemblage composition differed among reach types and between pool and riffle habitats. NMS ordination of samples from the twelve study reaches, organized by sample period and habitat type, produced a three-dimensional configuration with moderate stress (final stress = 12.06). Coefficients of determination between distances in the ordination space and Bray-Curtis distances in the original n-dimensional space indicated that the three ordination axes explained 89.7% of the total variation in assemblage structure. The plot of NMS axis 1 versus axis 3 supported the conclusion that macroinvertebrate assemblages varied significantly among reach types (Figure 4a). Forest and pasture reaches showed nearly complete segregation in multivariate space, and three of the four forest buffer reaches grouped very closely with the forest reaches (Figure 4a). One forest buffer reach, represented by six points in the ordination, consistently fell within the pasture group (Figure 4a). Differences in the composition of macroinvertebrate assemblages between pool and riffle habitats were also apparent with this combination of axes (Figure 4b). Other combinations of axes showed similar differences between reach types and habitats, but patterns were not as strong as for the combination presented above. Differences among sample periods, although significant in the PERMANOVA analysis, were not apparent in any two-dimensional combination of the ordination axes or in a three-dimensional view.

The SIMPER analysis indicated that taxa representing several different functional feeding groups contributed to differences between forest and pasture reaches. In particular, pasture reaches differed from forest reaches by generally having higher abundances of the scrapers Thiaridae and Hydrobiidae and the collector-gatherers *Caenis* and Oligochaeta and lower abundances of the collector-gatherers *Farrodes*, *Thraulodes*, *Tricorythodes*, *Neoelmis*, and *Terpides*, the shredder *Anchytarsus*, and the predators *Heteragrion*, Gomphidae, Coenagrionidae, and *Palaemnema* (Table 4). Insects of the families Chironomidae and Baetidae also tended to be more abundant in pasture reaches (Table 4). Many of the same taxa contributed to differences between forest buffer and pasture reaches (Table 4), consistent with the generally high similarity in assemblage structure between forest and forest buffer reaches. Some differences were also apparent, however, as *Tricorythodes* and Baetidae made comparatively larger contributions to differences between pasture and forest buffer sites while *Anchytarsus* was much less important (Table 4). Taxa making important contributions to dissimilarity between pools and riffles included *Thraulodes*, *Neoelmis*, *Tricorythodes*, *Smicridea*, Thiaridae, *Allenhyphes*, *Farrodes*, and Baetidae, all of which were generally more abundant in riffles, and *Caenis* and *Campsurus*, which were more abundant in pools.

Discussion

Deforestation and riparian forest buffers

Our results suggest that conversion of forests to pasture, particularly in riparian areas, leads to significant changes in benthic macroinvertebrate diversity and assemblage structure in small lowland Costa Rican streams. The strong patterns we observed among our three study reach types are particularly noteworthy considering the relatively small scale of deforestation in our study catchments and the fact that streamside trees had not been completely cleared in pasture reaches. Although we were not able to sample pasture and forest buffer reaches before deforestation occurred, the geographic proximity of our study sites and their similarities in channel size and geomorphology suggests that all sites had very similar benthic macroinvertebrate assemblages prior to forest clearing in the study area. Therefore, differences we observed among sites appear to be directly attributable to land use change.

The effects of deforestation on macroinvertebrate assemblages in our study streams appeared to depend heavily on the presence or absence of a riparian forest buffer, providing support for our hypothesis that riparian forest buffers can reduce the effects of deforestation on stream biota. Macroinvertebrates assemblages in pasture reaches, which lacked a forest buffer, had lower alpha diversity and significantly different taxonomic composition than those in our forested reference reaches. These differences were consistent with the effects of deforestation in other neotropical streams (Bojsen and Jacobsen 2003), as well as impacts associated with pastoral development in previously forested temperate catchments (Quinn 2000). Reaches with a forest buffer, in contrast, generally supported macroinvertebrate assemblages that were very similar to those in forested reference sites, both in terms of taxonomic composition and diversity. The contrast between pasture and buffer reaches was particularly evident for EPT taxa, a group commonly used as an indicator of stream degradation.

Our diversity comparisons should be interpreted with some caution because of the differences in taxonomic resolution among macroinvertebrate groups in our study. Comparisons of EPT taxa richness are more robust in this regard because most EPT taxa were identified to the genus level. The generally higher abundance of EPT taxa in forest and forest buffer reaches compared to pasture reaches likely had a strong influence on differences in EPT richness among reach types. However, we also noted that several of the EPT families considered especially sensitive to stream degradation in Costa Rica were either very rare (e.g. Perlidae, Leptoceridae) or absent (e.g. Ecnomidae) in pasture reaches. These taxa also tended to be uncommon in forest and forest buffer reaches, but were present at many different sites.

Caveats concerning taxonomic resolution do not apply to the differences in taxonomic composition that we observed among reach types, which were evident even at the ordinal level and were consistent across habitat types and sample periods. These differences were driven by a variety of taxa representing several functional feeding groups, and in some cases links to the abundance of food resources were evident. In particular, pasture reaches tended to have higher densities of scraping taxa, especially the gastropods Thiaridae and Hydrobiidae. The families Baetidae and Chironomidae, which were most abundant in pasture sites, also include many scraper and collector-gatherer taxa that can exploit in-stream primary production. Patterns of abundance among reach types for these taxa were consistent with the trend toward higher periphyton abundance in pasture reaches.

Interestingly, we found that the most common shredder in the study streams, *Anchytarsus,* was generally much more abundant in forest reaches than pasture reaches, with intermediate densities in forest buffer sites (Appendix 1). Shredder abundance is typically linked to the abundance of leaf litter (Cummins et al. 1989), and the density of ptilodactylids has been shown elsewhere to have a significant positive association with the abundance of litter detritus (Bojsen and Jacobsen 2003). However, in our study streams, the abundance of

Anchytarsus and other invertebrate shredders did not mirror patterns in leaf litter abundance, which showed no consistent differences among reach types. It remains an open question how differences in riparian vegetation among our study sites might affect the quality of litter available to shredders. Despite heavy shading by riparian vegetation and abundant leaf litter in nearly all of our streams, shredders always made up a very small component of the benthic fauna, consistent with a general pattern in tropical streams (Wantzen and Wagner 2006).

Differences in thermal regimes among reach types may have also played a significant role in producing the patterns we observed in macroinvertebrate assemblages. Point measurements in our study streams indicated that pasture reaches had significantly higher stream temperatures than forest reaches, a pattern confirmed by continuous monitoring of stream temperatures in these sites (Lorion et al. In Prep). Temperatures in forest buffer reaches tended to be intermediate, suggesting that riparian shading is an important factor. Although we know very little about the thermal ecology of benthic macroinvertebrates in our study streams, water temperature is known to have a significant influence on macroinvertebrate growth, fecundity, and survival (Sweeney 1993). The highest water temperatures observed in pasture reaches were 2-3°C outside the range of temperatures recorded during months of continuous monitoring of forest reaches. Some EPT taxa may be especially sensitive to high water temperatures, and altered thermal regimes could help explain why the EPT group was generally less abundant and diverse in pasture reaches.

Total macroinvertebrate abundance has often been found to increase in streams where canopy cover has been reduced by forest clearing (e.g. Hawkins et al. 1982, Bojsen and Jacobsen 2003), due in large part to increased primary production. Therefore, it was somewhat surprising that we did not see any consistent trends in macroinvertebrate density among our different reach types. However, a previous study in southeastern Costa Rica found that forested sites actually had higher macroinvertebrate densities than sites where forests had been cleared (Paaby et al. 1998), suggesting that the negative impacts of deforestation can counteract potential subsidy effects associated with canopy removal. Overall, we surely underestimated macroinvertebrate abundance in our study streams by using a Surber sampler with relatively coarse mesh. Nevertheless, our estimates of macroinvertebrate density were, on average, over 10 times higher than a previous study of streams in southeastern Costa Rica where macroinvertebrates were collected with a Surber sampler with the same mesh size (Paaby et al. 1998).

Spatial and temporal variation within reaches

Pool and riffle habitats in our study reaches supported macroinvertebrate assemblages with very different taxonomic composition, consistent with previous work in neotropical streams (Buss et al. 2004), including one of the few studies of stream macroinvertebrates in southeastern Costa Rica (Ramírez et al.1998). Despite these differences, the effects of deforestation, and the influence of riparian forest buffers, appeared to be consistent across pool and riffle habitats. We did not observe a significant interaction between reach type and habitat type for any of our comparisons of macroinvertebrate abundance, diversity, or taxonomic composition. Results from extensive bioassessment surveys in North America have shown that anthropogenic impacts are generally consistent across stream habitats (Gerth and Herlihy 2006). However, even when trends among habitats are similar, macroinvertebrate assemblages in different habitats may be responding to different stress gradients (Buss et al. 2004). Whether or not benthic macroinvertebrates in pools and riffles were responding to the same factors in our study streams, our results suggest that deforestation has similar impacts in both habitats. It should also be noted that the Surber sampler is likely to be more effective in riffles than in pools because it depends on invertebrates being washed downstream into the collecting net. This difference in sampling efficiency may have contributed to observed differences in macroinvertebrate abundance, taxa richness and assemblage composition between habitat types.

Temporal dynamics appeared to have less influence on macroinvertebrate assemblages than riparian conditions or stream habitat type, despite the fact that we targeted our sampling to capture seasonal variation. There were no significant changes in macroinvertebrate abundance or diversity through time, and compositional changes between sampling periods were relatively weak compared with other factors. Seasonal changes in macroinvertebrate abundance and assemblage structure in neotropical streams are strongly linked to rainfall patterns and associated disturbance by floods (Flecker and Feifarek 1994, Jacobsen and Encalada 1998, Ramírez et al. 2006). Rainfall did not show any clear seasonality during our study, and so the lack of consistent temporal trends in macroinvertebrate assemblages is not particularly surprising. The fact that high rainfall variability during our study did not obscure differences in macroinvertebrate assemblages among reach types or habitat types is more surprising and suggests that these assemblages are largely structured by deterministic processes.

Forest buffers in tropical stream management and conservation

In our study, riparian forest buffers averaging at least 15 m on both banks of headwater streams appeared to significantly reduce the effects of deforestation on benthic macroinvertebrates, generally maintaining macroinvertebrate assemblages with similar diversity and taxonomic composition as sites in continuous forest. These results suggest that proper management of riparian areas could substantially reduce the negative effects of deforestation on tropical headwater streams and provide strong support for Costa Rican regulations mandating protection of riparian forests in this zone. Forest buffers around our study streams were often considerably wider than this minimum requirement, but were also highly variable and had some areas less than 15 m in width. Despite this variability, these areas appeared to promote stream biodiversity and maintain stream ecosystem conditions similar to those found in forested reference streams.

One forest buffer reach was a clear exception to this pattern, and illustrates the limitations of riparian buffers. This reach, despite having a wide forest buffer, had been heavily disturbed by foraging pigs, was paralleled by a road that was subject to severe erosion during rain events, and had high nutrient concentrations that may have been influenced by domestic effluent from several nearby residences. Stream bank trampling by livestock and erosion from poorly maintained roads are both important sources of sediment pollution in streams (Waters 1995), and benthic habitats in this site had clearly been affected by sedimentation. It is difficult to know whether water quality was also a factor, but this reach consistently had the lowest macroinvertebrate densities of any of our sites. Riparian forest buffers can only be expected to effectively mitigate impacts on streams when factors that bypass them or overwhelm their capacity to filter sediments and nutrients are reduced or eliminated (Osborne and Kovacic 1993, Wenger 1999).

Riparian buffer functions can also be expected to vary depending on catchment-scale patterns in land use (Allan 2004). We focused on reach-scale effects in catchments with high forest cover in this study in order to eliminate many potentially confounding factors, and further research will be needed to determine whether riparian buffers will sustain similar functions in landscapes with more extensive deforestation. Although some studies in temperate streams have found that benthic communities are most strongly influenced by local riparian conditions (Sponseller et al. 2001, Lammert and Allan 1999), others have shown that catchment-scale land use is more important and can overwhelm local effects (Roth et al. 1996, Harding et al. 1998). Whichever spatial scale is most important, it appears that even in heavily forested catchments, clearing of riparian vegetation can disrupt terrestrial-aquatic linkages in headwater streams (England and Rosemond 2004).

Finally, comparisons between fish and macroinvertebrate assemblages in our study reaches suggest that riparian management has community-wide consequences in small tropical streams. In stream reaches without a forest buffer, deforestation appeared to have significant, and contrasting, effects on fish and benthic macroinvertebrate assemblages. Specifically, we found higher fish abundance and diversity in pasture reaches compared to forest reaches (Lorion et al. In Prep), while macroinvertebrates showed no change in abundance in pasture reaches and appeared to decline in diversity. These responses may not be independent, as fishes in lowland Costa Rican streams have been shown to exert top-down effects on benthic communities (Pringle and Hamazaki 1998). Stream reaches with a riparian forest buffer, in contrast, generally supported assemblages with similar diversity and taxonomic composition as forested reference sites for both fish and macroinvertebrates. This concordance between the two assemblages, which represent multiple trophic levels and diverse habitat associations, suggests that our conclusions regarding forest buffer functions are robust. Riparian forest buffers are not a replacement for continuous forest, but can promote biodiversity and stream ecosystem integrity in tropical catchments affected by deforestation.

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References

Achard, F., H. D. Eva, H.-J. M. P. Stibig, J. Gallego, T. Richards, and J.-P. Malingreau. 2002. Determination of deforestation rates of the world's humid tropical forests. Science 297:999-1002.

Allan, J. D. 2004. Landscapes and riverscapes: the influence of land use on stream ecosystems. Annual Review of Ecology and Systematics 35:257-284.

American Public Health Association (APHA). 1995. Standard methods for the examination of water and wastewater. American Public Health Association, Washington, D.C., USA.

Anderson, M. J. 2001. A new method for non-parametric multivariate analysis of variance. Austral Ecology 26:32-46.

Anderson, M. J. 2005. PERMANOVA: a FORTRAN computer program for permutational multivariate analysis of variance. Department of Statistics, University of Auckland, Auckland, New Zealand.

Bain, M. B., J. T. Finn, and H. E. Booke. 1985. Quantifying stream substrate for habitat analysis studies. North American Journal of Fisheries Management 5:499-506.

Benke, A. C., A. D. Huryn, L. A. Smock, and J. B. Wallace. 1999. Length-mass relationships for freshwater macroinvertebrates in North America with particular reference to the southeastern United States. Journal of the North American Benthological Society 18:308-343.

Benstead, J. P., M. M. Douglas, and C. M. Pringle. 2003. Relationship of stream invertebrate communities to deforestation in eastern Madagascar. Ecological Applications 13:1473-1490.

Benstead, J. P., and C. M. Pringle. 2004. Deforestation alters the resource base and biomass of endemic stream insects in eastern Madagascar. Freshwater Biology 49:490-501.

Bojsen, B. H., and D. Jacobsen. 2003. Effects of deforestation on macroinvertebrate diversity and assemblage structure in Ecuadorian Amazon streams. Archiv für Hydrobiologie 158:317-342.

Bray, J. R., and J. T. Curtis. 1957. An ordination of the upland forest communities of southern Wisconsin. Ecological Monographs 27:325-349.

Buss, D. F., D. F. Baptista, J. L. Nessimian, and M. Egler. 2004. Substrate specificity, environmental degradation, and disturbance structuring macroinvertebrate assemblages in neotropical streams. Hydrobiologia 518:179-188.

Cummins, K. W., M. A. Wilzbach, D. M. Gates, J. B. Perry, and W. B. Taliaferro. 1989. Shredders and riparian vegetation. BioScience 39:24-30.

Dall, P. C. 1979. A sampling technique for littoral stone dwelling organisms. Oikos 33:106-112.

Dudgeon, D. 2006. The impacts of human disturbance on stream benthic invertebrates and their drift in North Sulawesi, Indonesia. Freshwater Biology 51:1710-1729.

England, L. E., and A. D. Rosemond. 2004. Small reductions in forest cover weaken terrestrial-aquatic linkages in headwater streams. Freshwater Biology 49:721-734.

Flecker, A. S., and B. Feifarek. 1994. Disturbance and the temporal variability of invertebrate assemblages in two Andean streams. Freshwater Biology 31:131-142.

Gerth, W. J., and A. T. Herlihy. 2006. Effect of sampling different habitat types in regional macroinvertebrate bioassessment surveys. Journal of the North American Benthological Society 25:501-512.

Gomi, T., R. C. Sidle, S. Noguchi, J. N. Negishi, A. R. Nik, and S. Sasaki. 2006. Sediment and wood accumulations in humid tropical headwater streams: effects of logging and riparian buffers. Forest Ecology and Management 224:166-175.

Gregory, S. V., F. J. Swanson, W. A. McKee, and K. W. Cummins. 1991. An ecosystem perspective of riparian zones. BioScience 41:540-551.

Hall, R. O., G. E. Likens, and H. M. Malcom. 2001. Trophic basis of invertebrate production in 2 streams at the Hubbard Brook Experimental Forest. Journal of the North American Benthological Society 20:432-447.

Harding, J. S., E. F. Benfield, P. V. Bolstad, G. S. Helfman, and E. B. D. Jones. 1998. Stream biodiversity: the ghost of land use past. Proceedings of the National Academy of Sciences of the USA 95:14843-14847.

Hawkins, C. P., J. L. Kershner, P. A. Bisson, M. D. Bryant, L. M. Decker, S. V. Gregory, D. A. McCullough, C. K. Overton, G. H. Reeves, R. J. Steedman, and M. K. Young. 1993. A hierarchical approach to classifying stream habitat features. Fisheries 18:3-12.

Hawkins, C. P., M. L. Murphy, and N. H. Anderson. 1982. Effects of canopy cover, substrate composition, and gradient on the structure on macroinvertebrate communities in Cascade Range streams of Oregon. Ecology 63:1840-1856.

Heartsill-Scalley, T., and T. M. Aide. 2003. Riparian vegetation and stream condition in a tropical agriculture-secondary forest mosaic. Ecological Applications 13:225-234.

Henry, R., V. S. Uieda, A. A. de O. Afonso, and R. M. Kikuchi. 1994. Input of allochthonous matter and structure of fauna in a Brazilian headstream. Verhandlungen der Internationale Vereinigung für Theoretische und Angewandte Limnologie 25:1866-1870.

Hijmans, R. J., S. E. Cameron, J. L. Parra, P. G. Jones, and A. Jarvis. 2005. Very high resolution interpolated climate surfaces for global land areas. International Journal of Climatology 25:1965-1978.

Holdridge, L. R. 1967. Life zone ecology. Tropical Science Center, San José, Costa Rica.

Iwata, T., S. Nakano, and M. Inoue. 2003. Impacts of past riparian deforestation on stream communities in a tropical rainforest in Borneo. Ecological Applications 13:461-473.

Jacobsen, D., and A. Encalada. 1998. The macroinvertebrate fauna of Ecuadorian highland streams in the wet and dry season. Archiv für Hydrobiologie 142:53-70.

Karr, J. R., and I. J. Schlosser. 1978. Water resources and the land-water interface. Science 201:229-234.

Kerans, B. L., J. R. Karr, and S. A. Ahlstedt. 1992. Aquatic invertebrate assemblages: spatial and temporal differences among sampling protocols. Journal of the North American Benthological Society 11:377-390.

Lammert, M., and J. D. Allan. 1999. Assessing biotic integrity of streams: effects of scale in measuring the influence of land use/cover and habitat structure on fish and macroinvertebrates. Environmental Management 23:257-270.

Laurance, W. F. 1999. Reflections on the tropical deforestation crisis. Biological Conservation 91:109-117.

Magurran. A. E. 1988. Ecological diversity and its measurement. Princeton University Press, Princeton, New Jersey.

McArdle, B. H., and M. J. Anderson. 2001. Fitting multivariate models to community data: a comment on distance-based redundancy analysis. Ecology 82:290-297.

McCune, B., and J. B. Grace. 2002. Analysis of ecological communities. MJM Software Design, Gleneden Beach, Oregon, USA.

McCune, B., and M. J. Mefford. 1999. PC-ORD. Multivariate analysis of ecological data, Version 5.0. MjM Software, Gleneden Beach, Oregon, USA.

McKergow, L. A., I. A. Prosser, R. B. Grayson, and D. Heiner. 2004. Performance of grass and rainforest riparian buffers in the wet tropics, Far North Queensland. 1. Riparian hydrology. Australian Journal of Soil Research 42:473-484

Merritt, R. W., and K. W. Cummins. 1996. An introduction to the aquatic insects of North America, 3rd Edition. Kendall/Hunt Publishing Company, Dubuque, Iowa, USA.

Montgomery, D. R., and J. M. Buffington. 1997. Channel-reach morphology in mountain drainage basins. Geological Society of America Bulletin 109:596-611.

Naiman, R. J., and H. Décamps. 1997. The ecology of interfaces: riparian zones. Annual Review of Ecology and Systematics 28:621-658.

Osborne, L. L., and D. A. Kovacic. 1993. Riparian vegetated buffer strips in water-quality restoration and stream management. Freshwater Biology 29:243-258.

Paaby, P., A. Ramírez, and C. M. Pringle. 1998. The benthic macroinvertebrate community in Caribbean Costa Rican streams and the effect of two sampling methods. Revista de Biología Tropical 46 (Suppl. 6):185-199.

Peterjohn, W. T., and D. L. Correll. 1984. Nutrient dynamics in an agricultural watershed: observations on the role of a riparian forest. Ecology 65:1466-1475.

Pringle, C. M., and T. Hamazaki. 1998. The role of omnivory in a neotropical stream: separating diurnal and nocturnal effects. Ecology 79:269-280.

Pringle, C. M., and F. N. Scatena. 1999. Aquatic ecosystem deterioration in Latin America and the Caribbean. Pages 104-113 in L. U. Hatch and M. E. Swisher, editors. Managed ecosystems: the Mesoamerican experience. Oxford University Press, New York, USA.

Quinn, J. M. 2000. Effects of pastoral development. Pages 208-229 in K. J. Collier and M. J. Winterbourn, editors. New Zealand stream invertebrates: ecology and implications for management. New Zealand Limnological Society, Christchurch, New Zealand.

Ramírez, A., C. M. Pringle, and M. Douglas. 2006. Temporal and spatial patterns in stream physicochemistry and insect assemblages in tropical lowland streams. Journal of the North American Benthological Society 25:108-125.

Ramírez, A., P. Paaby, C. M. Pringle, and G. Agüero. 1998. Effect of habitat type on benthic macroinvertebrates in two lowland tropical streams, Costa Rica. Revista de Biología Tropical 46 (Suppl. 6):201-213.

Roldán, G. 1996. Guía para el estudio de los macroinvertebrados acuáticos del Departamento de Antioquia. Universidad de Antioquia, Colombia.

Rosenberg, D. M., and V. H. Resh. 1993. Freshwater biomonitoring and benthic macroinvertebrates. Chapman & Hall, New York, New York.

Roth, N. E., J. D. Allan, and D. L. Erickson. 1996. Landscape influences on stream biotic integrity assessed at multiple spatial scales. Landscape Ecology 11:141-156.

Roy, A. H., A. D. Rosemond, D. S. Leigh, M. J. Paul, and J. B. Wallace. 2003. Habitat-specific responses of stream insects to land cover disturbance: biological consequences and monitoring implications. Journal of the North American Benthological Society 22:292-307.

Sader, S. A., and A. T. Joyce. 1988. Deforestation rates and trends in Costa Rica, 1940 to 1983. Biotropica 20:11-19.

Sánchez-Azofeifa, G. A., R. C. Harriss, and D. L. Skole. 2001. Deforestation in Costa Rica: a quantitative analysis using remote sensing imagery. Biotropica 33:378-384.

SAS Institute Inc. 2002. SAS Version 9.1. SAS Institute, Cary, North Carolina, USA.

Sponseller, R. A., E. F. Benfield, and H. M. Valett. 2001. Relationships between land use, spatial scale, and stream macroinvertebrate communities. Freshwater Biology 46:1409-1424.

Sweeney, B. W. 1993. Effects of streamside vegetation on macroinvertebrate communities of White Clay Creek in eastern North America. Proceedings of the Academy of Natural Sciences of Philadelphia 144:291-340.

Tomanova, S., E. Goitia, and J. Helesic. 2006. Trophic levels and functional feeding groups of macroinvertebrates in neotropical streams. Hydrobiologia 556:251-264.

Toothaker, L. E. 1993. Multiple comparison procedures. Sage Publications, Newbury Park, California, USA.

Vannote, R. L., G. W. Minshall, K. W. Cummins, J. R. Sedell, and C. E. Cushing. 1980. The river continuum concept. Canadian Journal of Fisheries and Aquatic Sciences 37:130-137.

Wallace, J. B., S. L. Eggert, J. L. Meyer, and J. R. Webster. 1997. Multiple trophic levels of a forest stream linked to terrestrial litter inputs. Science 277:102-104.

Wantzen, K. M. 2006. Physical pollution: effects of gully erosion on benthic invertebrates in a tropical clear-water stream. Aquatic Conservation: Marine and Freshwater Ecosystems 16:733-749.

Wantzen, K. M., and R. Wagner. 2006. Detritus processing by invertebrate shredders: a neotropical-temperate comparison. Journal of the North American Benthological Society 25:216-232.

Waters, T. F. 1995. Sediment in streams: sources, biological effects, and control. American Fisheries Society Monograph 7. American Fisheries Society, Bethesda, Maryland, USA.

Wenger, S. 1999. A review of the scientific literature on riparian buffer width, extent, and vegetation. Institute of Ecology, University of Georgia, Athens, Georgia, USA.

Wintermans, J. F. G. M., and A. De Mots. 1965. Spectrophotometric characteristics of chlorophylls a and b and their pheophytins in ethanol. Biochimica et Biophysica Acta 109:448-453.

Table 1. Mean values (range) of environmental variables in forest, forest buffer, and pasture study reaches in southeastern Costa Rica (n = 4 for each reach type).

	Forest		Forest Buffer		Pasture	
Catchment Area (ha)	46	(26-79)	53	(15-103)	60	(15-130)
Catchment % Forest	97	(93-100)	83	(72-92)	81	(60-91)
Channel Width (m)	2.0	(1.7-2.5)	1.7	(1.3-2.2)	1.8	(1.4-2.3)
Mean Depth (cm)	11	(8-16)	12	(8-18)	17	(11-22)
Substrate Coarseness	2.8	(2.4-3.4)	2.5	(1.9-3.0)	2.6	(1.8-3.1)
Embeddedness (%)	36	(27-45)	46	(39-66)	56	(40-79)
Riffle Area (%)	28	(19-41)	26	(10-49)	17	(9-23)
Channel Gradient (%)	0.8	(0.6-1.1)	0.7	(0.4-0.8)	0.8	(0.5-0.9)
Canopy Cover (%)	93	(91-95)	94	(93-95)	78	(44-93)
Temperature (°C)	24.6	(23.0-25.9)	25.2	(24.0-27.2)	26.3	(22.9-30.5)
Conductivity (µS/cm)	300	(239-403)	352	(160-442)	241	(210-286)
pH	7.9	(7.7-8.2)	8.0	(7.5-8.3)	7.6	(7.5-7.8)
NO3-N (mg/L)	0.06	(0.04-0.09)	0.08	(0.03-0.19)	0.03	(0.0-0.07)
NH4-N (mg/L)	0.05	(0.04-0.05)	0.04	(0.03-0.06)	0.02	(0.0-0.04)
PO4-P (mg/L)	0.09	(0.05-0.12)	0.11	(0.05-0.18)	0.08	(0.05-0.17)
Leaf Litter (g AFDM/m^2)	35.1	(23.8-40.5)	31.7	(18.4-42.0)	48.2	(19.5-90.4)
Chlorophyll-a (mg/m^2)	5.0	(0.7-14.5)	6.4	(0.7-17.8)	62.2	(0.6-165.6)

Notes: Leaf litter and chlorophyll-*a* standing crop means are based on habitat-weighted estimates for the study reaches for three sample periods (leaf litter) or two sample periods (chlorophyll-*a*).

Table 2. Results of repeated-measures ANOVA models for macroinvertebrate density, biomass, and diversity with reach type (forest, forest buffer, or pasture), sample period and habitat type (pool or riffle) as fixed effects (n = 4 for each reach type).

	Reach Type		Sample Period		Habitat Type	
	$F_{2,18}$	*P*	$F_{2,36}$	*P*	$F_{1,18}$	*P*
Density (ind./m^2)[a]	0.39	0.680	2.36	0.109	**6.37**	**0.021**
Biomass (g/m^2)[a]	0.58	0.569	1.13	0.336	**7.47**	**0.014**
Taxa Richness	2.43	0.116	2.48	0.098	**10.57**	**0.004**
Alpha Diversity	**3.73**	**0.044**	0.08	0.925	3.46	0.079
EPT Taxa Richness	**4.20**	**0.032**	0.70	0.505	2.24	0.152

Notes: Results in bold are significant at $P < 0.05$. Biomass comparisons are based on aquatic insects only. Interaction terms were not significant for any of the models.

[a] $\log_{10}(x)$ transformed for analysis

Table 3. Results of three-way PERMANOVA test for the effects of reach type, sample period, and habitat type on the taxonomic composition of macroinvertebrate assemblages in study reaches in southeastern Costa Rica.

Source	df	SS	MS	F	*P*
Reach Type (R)	2	14401	7201	**6.1371**	**0.0001**
Sample Period (P)	2	4194	2097	**1.7874**	**0.0342**
Habitat Type (H)	1	14403	14403	**12.2758**	**0.0001**
R x P	4	3035	759	0.6467	0.9587
P x H	2	653	326	0.2782	0.9999
R x H	2	1962	981	0.8363	0.6481
R x P x H	4	1824	456	0.3887	0.9999
Residual	54	63358	1173		

Notes: Results in bold are significant at $P < 0.05$.

Table 4. List of taxa making the largest contributions to dissimilarity between forest and pasture reaches, and between forest buffer and pasture reaches, based on SIMPER analysis, in order of decreasing importance. Taxa in bold were generally more abundant in pasture reaches.

Rank	Forest vs. Pasture	Forest Buffer vs. Pasture
1	*Farrodes*	*Tricorythodes*
2	*Anchytarsus*	*Farrodes*
3	**Thiaridae**	**Thiaridae**
4	*Thraulodes*	*Thraulodes*
5	*Tricorythodes*	*Caenis*
6	***Caenis***	*Allenhyphes*
7	*Neoelmis*	*Neoelmis*
8	**Oligochaeta**	**Baetidae**
9	*Heteragrion*	*Campsurus*
10	Gomphidae	Chironomidae
11	*Terpides*	*Heteragrion*
12	Coenagrionidae	Coenagrionidae
13	**Hydrobiidae**	**Oligochaeta**
14	*Palaemnema*	**Hydrobiidae**
15	*Campsurus*	***Hexatoma***
16	***Hexacylloepus***	***Hexacylloepus***
17	**Chironomidae**	*Anchytarsus*
18	*Perigomphus*	*Terpides*
19	**Baetidae**	Gomphidae
20	*Hexatoma*	*Palaemnema*

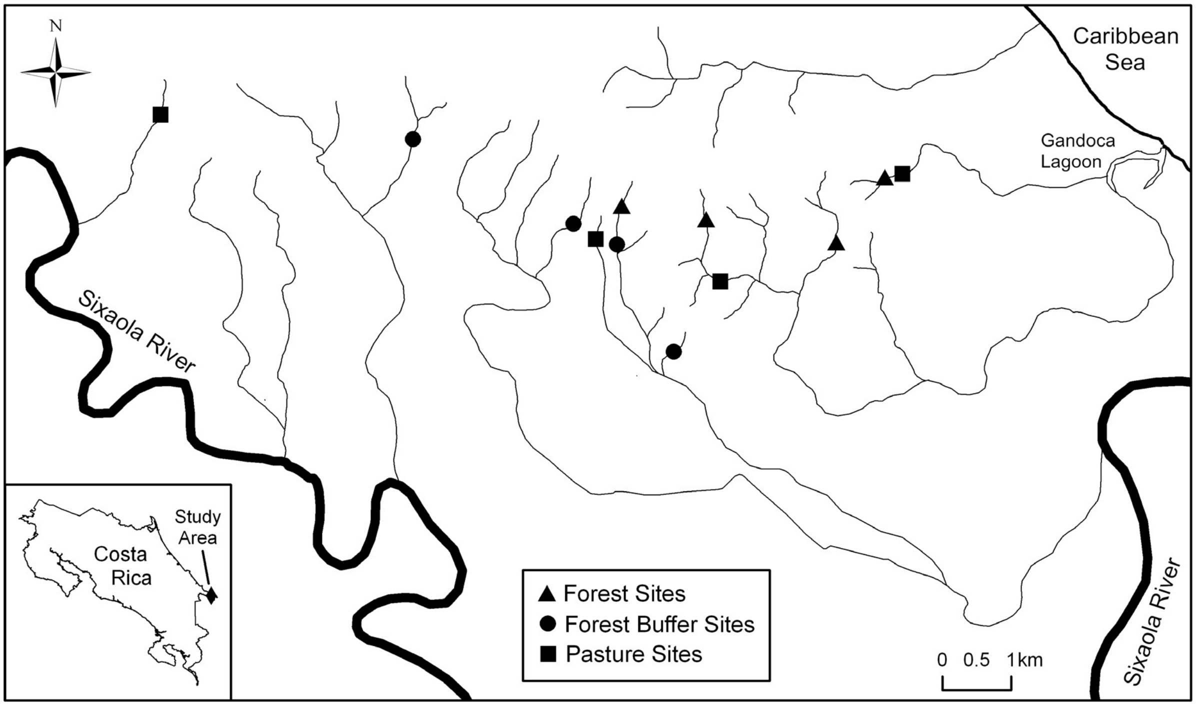


Figure 1. Map of study area in southeastern Costa Rica, with locations of the 12 sampling reaches.

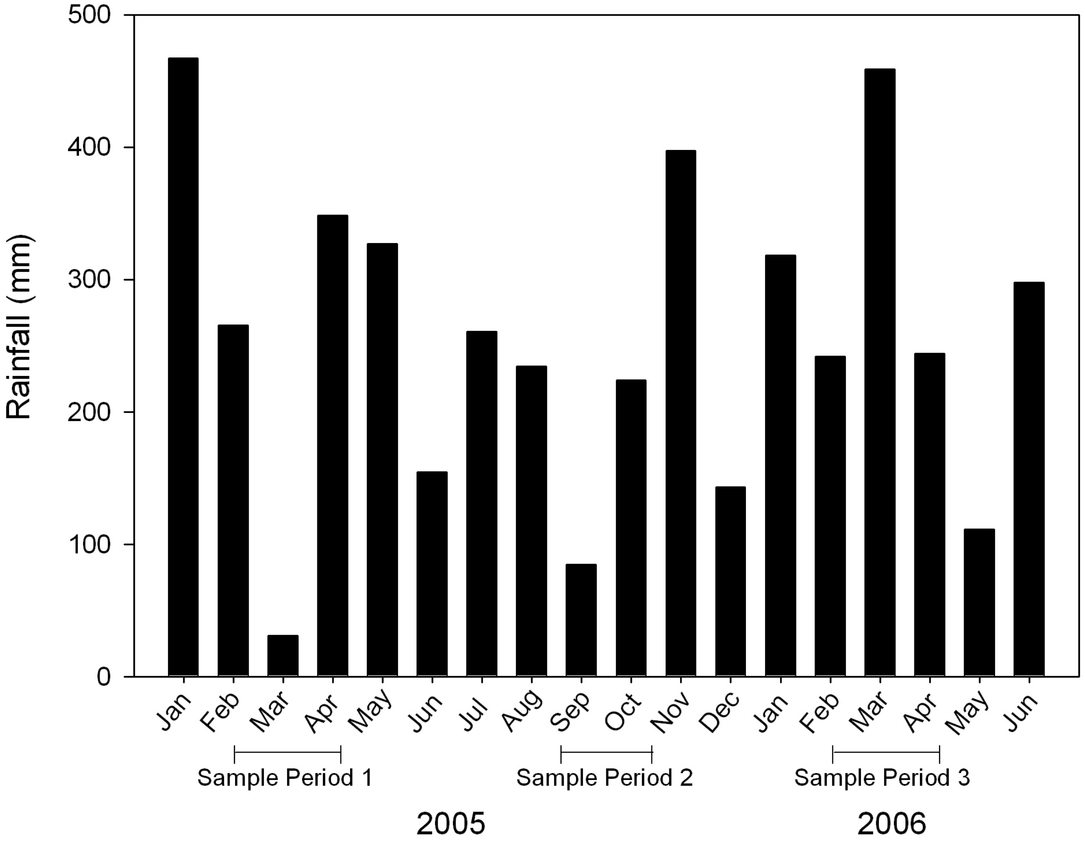


Figure 2. Monthly rainfall at Puerto Vargas, Costa Rica, January 2005-June 2006. Puerto Vargas is located approximately 25 km northwest of the study area. Data courtesy of the National Meteorological Institute (IMN), San José, Costa Rica.

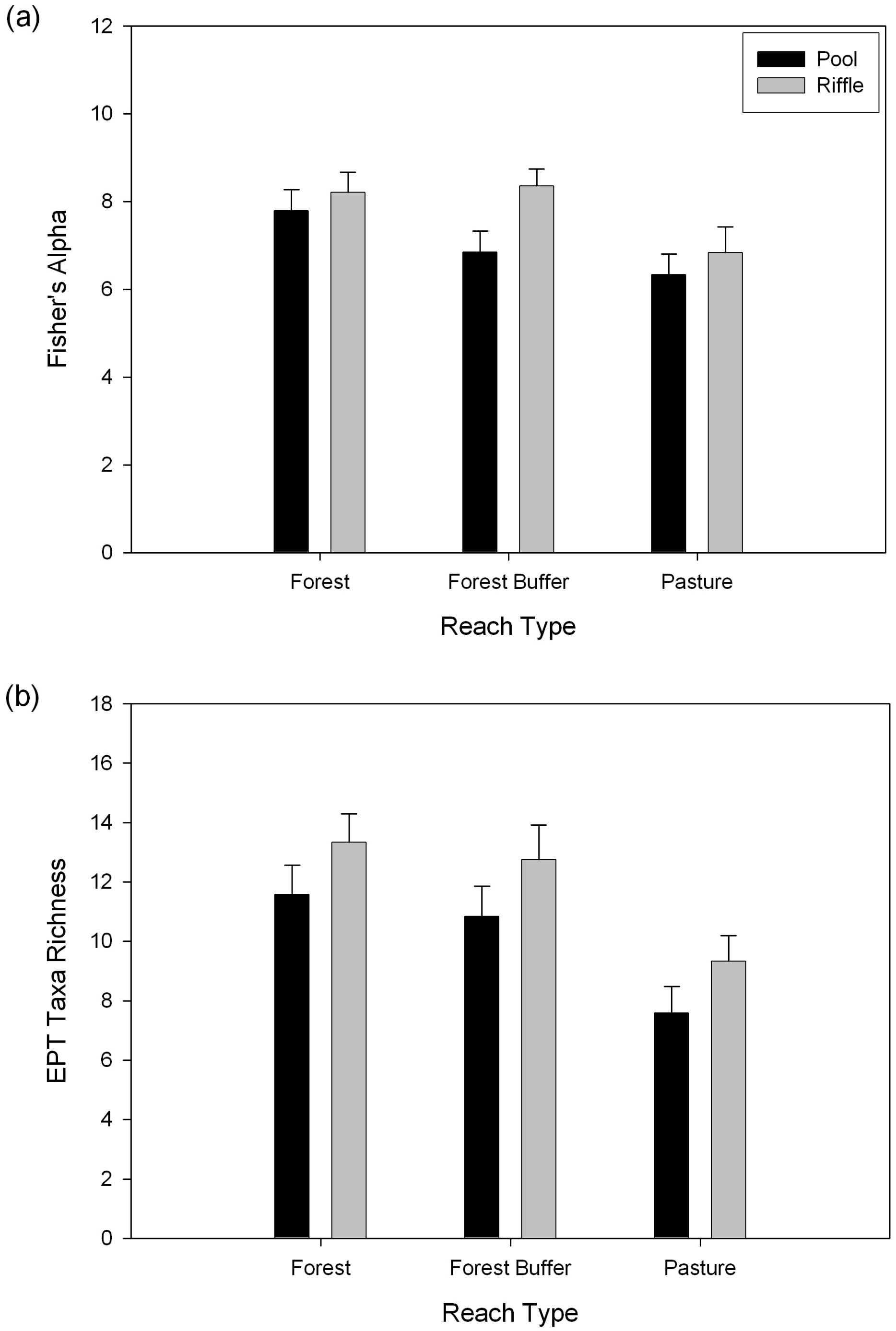


Figure 3. Plots of means (+SE) of (a) Fisher's alpha diversity index, and (b) EPT taxa richness in pool and riffle habitats in the three study reach types. Based on a repeated measures ANOVA model, Fisher's alpha values were significantly higher in forest reaches than in pasture reaches. Forest buffer reaches were intermediate and did not differ from the other two reach types. EPT taxa richness was significantly higher in forest and forest buffer reaches compared with pasture reaches. Differences between habitat types were not significant for Fisher's alpha diversity index or EPT taxa richness (see Table 2).

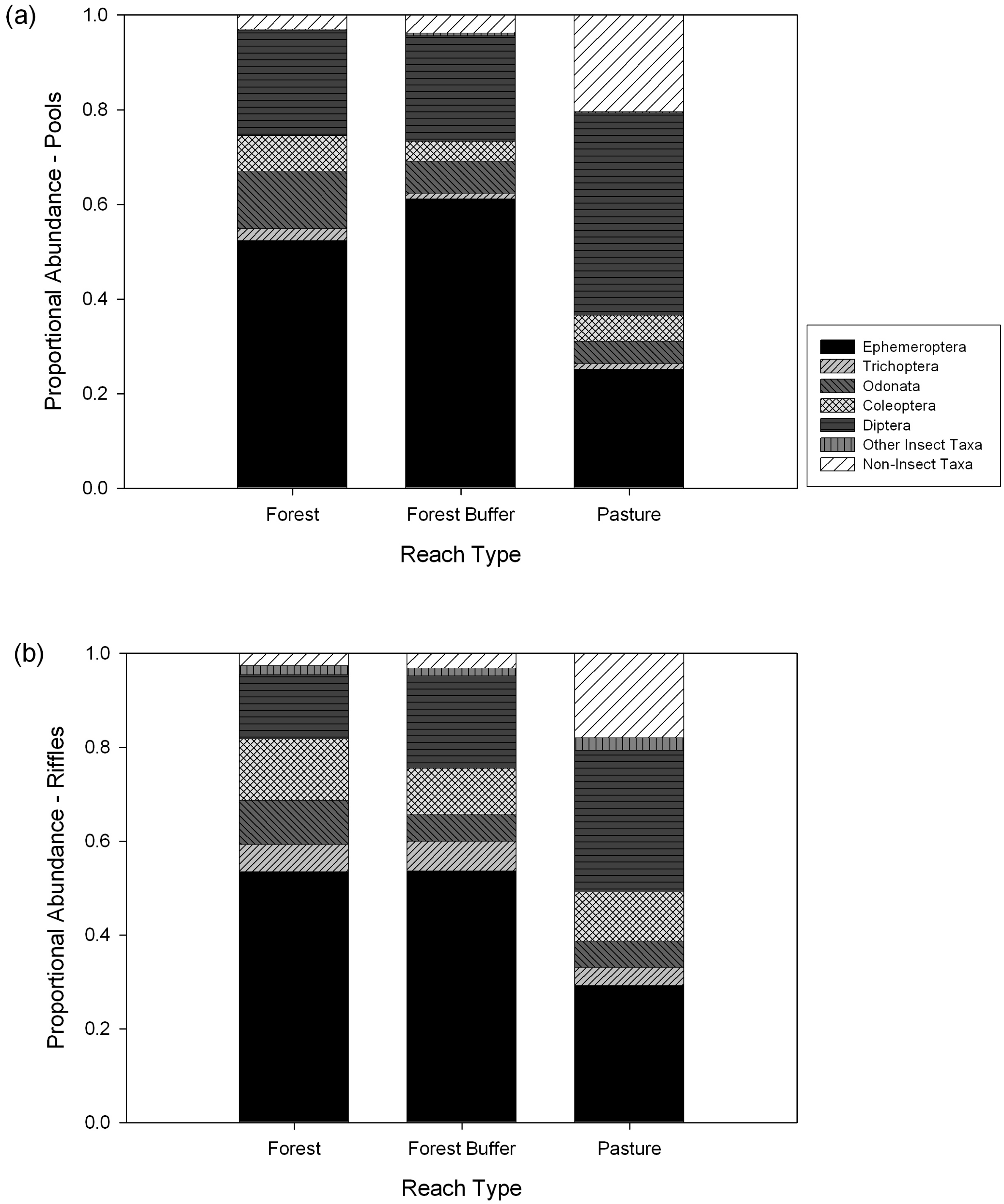
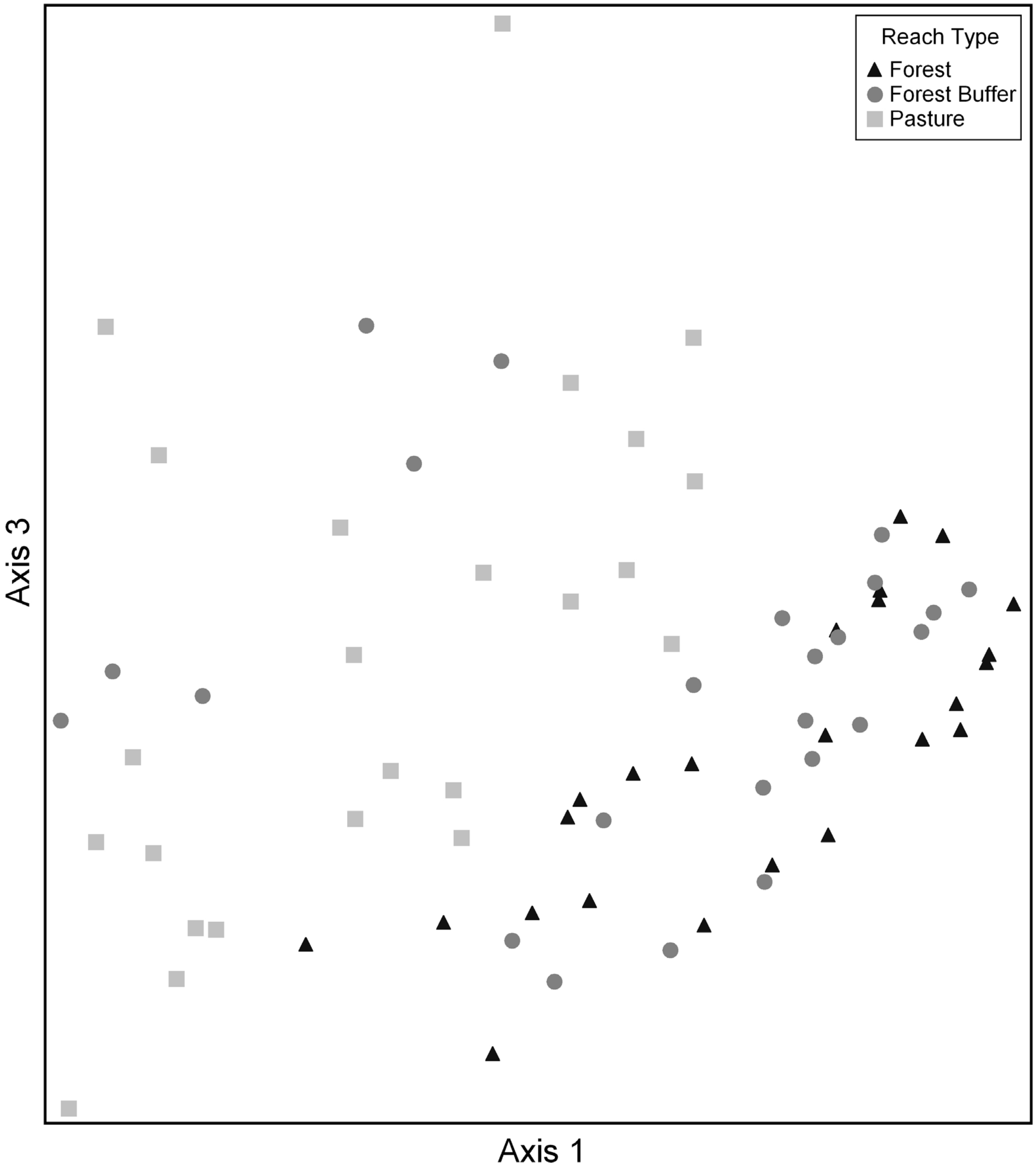


Figure 4. Taxonomic composition of macroinvertebrate assemblages in (a) pools and (b) riffles in the three study reach types in southeastern Costa Rica. Proportions are based on all macroinvertebrates captured in each reach type across all sampling periods.

(a)



(b)

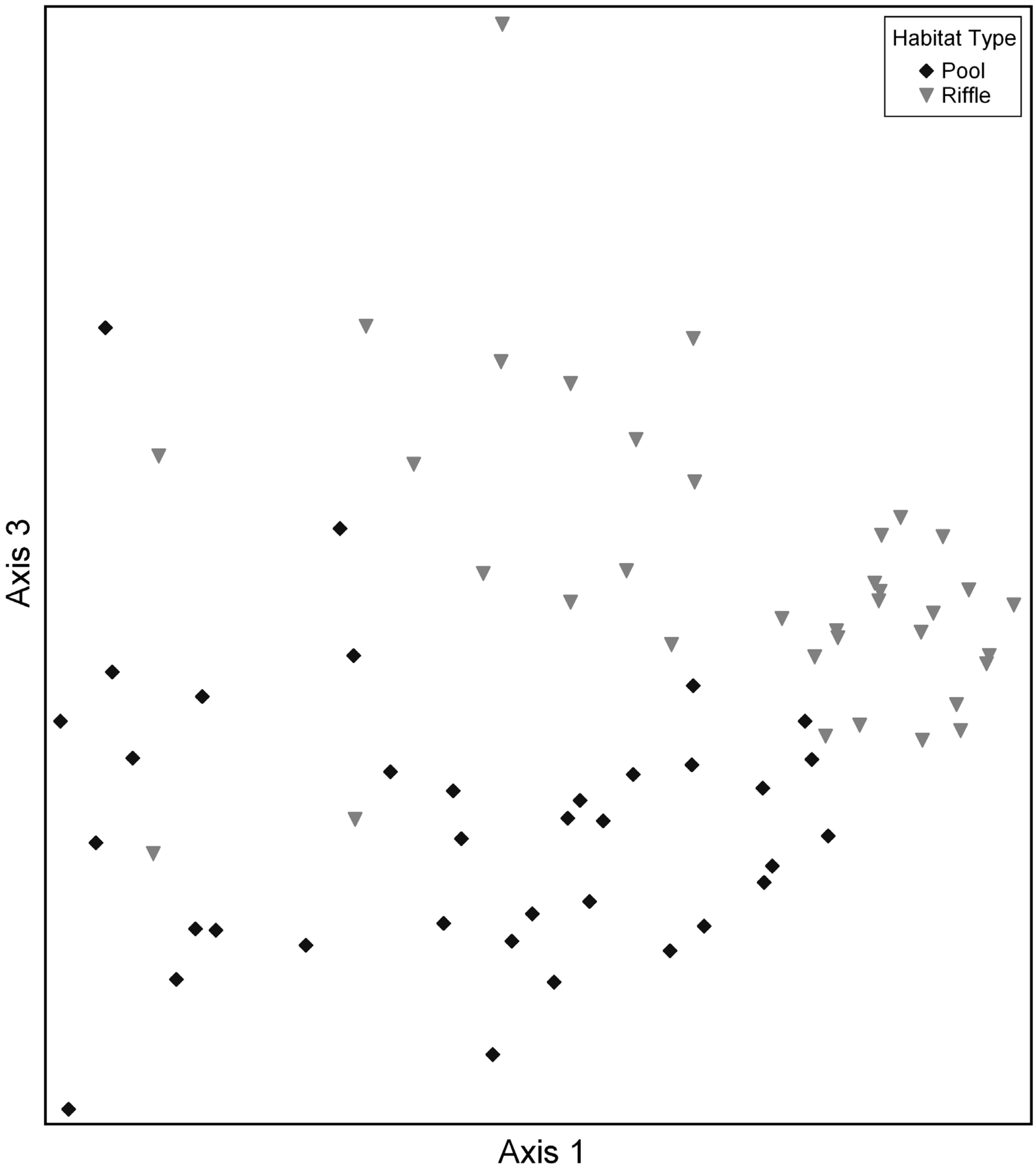


Figure 5. Plot of axis 1 versus axis 3 of the three-dimensional NMS ordination of samples from the 12 study reaches, with samples identified by (a) reach type or (b) habitat type. Each sampling reach is represented by six data points in the ordination, each one representing 5 pooled Surber samples collected in pools or riffles on one sample date.

Appendix 1. Macroinvertebrate taxa collected in the study streams and their mean density (individuals/m²) in forest (F), forest buffer (FB), and pasture (P) reaches.

Taxonomic Groups	Family	Genus	Pools			Riffles		
			F	FB	P	F	FB	P
Platyhelminthes								
Turbellaria			-	-	-	0.9	-	0.4
Annelida								
Oligochaeta			7.2	9.7	28.2	6.6	5.6	31.4
Hirudinea			-	-	0.5	0.2	0.4	0.2
Polychaeta			0.5	0.9	26.2	0.2	0.4	11.1
Mollusca								
Gastropoda	Ampullariidae		-	-	-	0.2	-	0.2
	Ancylidae		0.9	0.5	3.0	1.1	1.6	7.7
	Hydrobiidae		2.0	4.5	10.6	2.3	7.7	31.2
	Planorbidae		-	-	-	-	-	0.2
	Thiaridae		5.7	19.4	67.1	6.3	16.1	96.7
Bivalvia	Sphaeriidae		0.2	0.7	2.5	-	0.5	5.6
Arthropoda								
Arachnida								
Hydracarina			0.9	2.3	0.5	1.4	3.6	0.9
Decapoda	Atyidae		0.9	0.2	0.2	16.0	8.8	13.1
	Palaemonidae	*Macrobrachium*	0.5	1.3	1.4	3.0	2.5	5.4
	Pseudothelphusidae		-	0.5	-	1.4	0.5	0.7
Insecta								
Ephemeroptera	Baetidae		21.7	19.6	30.1	54.4	48.6	111.4
	Caenidae	*Caenis*	125.4	126.5	84.1	5.6	17.0	43.2
	Leptohyphidae	*Allenhyphes*	2.9	19.7	0.2	59.0	92.7	2.5
		Asioplax	6.6	7.2	2.3	5.2	3.8	-
		Cabecar	2.5	1.4	0.4	2.2	3.6	-
		Epiphrades	0.4	-	0.9	-	0.4	1.1
		Leptohyphes	-	-	-	-	0.5	-
		Tricorythodes	23.1	265.7	4.1	98.8	250.3	27.4
		Vacuperinus	-	2.9	-	-	0.4	-
	Leptophlebiidae	*Farrodes*	98.7	118.9	30.1	250.8	125.2	101.7
		Thraulodes	9.0	25.8	0.7	320.9	261.9	32.3
		Tikuna	0.5	-	-	-	0.2	0.5
		Terpides	18.5	11.8	0.9	10.0	10.8	6.3
		Ulmeritoides	0.2	-	2.9	-	-	-
	Polymitarcyidae	*Campsurus*	26.9	36.8	16.5	2.7	9.3	6.1
Odonata	Calopterygidae	*Hetaerina*	1.1	0.4	0.4	0.4	0.4	1.4
	Coenagrionidae		36.1	36.6	12.0	21.0	24.9	34.6
	Gomphidae		18.8	11.7	8.4	21.9	7.0	6.6
		Agriogomphus	0.5	0.9	0.5	-	-	0.2
		Perigomphus	1.4	0.4	0.2	29.6	7.2	0.9
	Lestidae		0.2	-	-	0.2	-	-
	Libellulidae		5.0	3.2	9.0	3.6	4.8	8.8
	Megapodagrionidae	*Heteragrion*	10.4	15.6	1.6	33.0	28.2	9.3
	Platystictidae	*Palaemnema*	4.3	2.7	0.4	33.0	15.1	1.8
	Polythoridae		-	-	-	0.2	-	-

Appendix 1 cont.

Taxonomic Groups	Family	Genus	Pools			Riffles		
			F	FB	P	F	FB	P
Plecoptera	Perlidae	*Anacroneuria*	1.8	3.8	-	19.7	16.9	1.1
Hemiptera	Gerridae		-	0.2	-	1.3	0.5	0.9
	Hebridae	*Hebrus*	-	-	-	-	-	0.2
	Mesoveliidae		-	-	0.4	0.5	0.5	0.4
	Naucoridae		-	0.5	-	2.7	2.7	0.5
	Veliidae	*Rhagovelia*	-	0.7	-	2.9	5.2	1.6
		Microvelia	-	-	-	0.4	0.2	0.2
Megaloptera	Corydalidae	*Chloronia*	-	-	-	0.2	-	-
		Corydalus	-	-	-	0.5	-	-
		Platyneuromus	-	-	-	0.4	-	0.2
Trichoptera	Calamoceratidae	*Phylloicus*	1.6	0.2	0.9	1.4	0.2	0.4
	Ecnomidae	*Austrotinodes*	1.8	0.5	-	0.9	1.4	-
	Helicopsychidae		-	-	0.2	0.2	0.2	0.4
	Hydropsychidae	*Macronema*	4.5	2.0	0.2	5.4	0.2	-
		Smicridea	0.7	0.7	0.2	40.0	51.8	9.3
		Leptonema	-	-	-	0.5	1.3	5.2
	Hydroptilidae	*Alisotrichia*	-	-	-	0.7	0.7	-
		Neotrichia	2.9	3.9	1.1	13.3	17.6	15.6
		Ochrotrichia	-	-	-	0.2	-	0.5
		Oxyethira	-	-	0.2	-	-	6.8
	Leptoceridae	*Nectopsyche*	2.2	1.8	-	0.5	2.2	0.4
		Oecetis	0.2	-	-	-	-	-
	Philopotamidae	*Chimarra*	-	0.4	-	16.9	20.3	2.0
	Polycentropodidae	*Polycentropus*	1.3	0.9	3.2	1.4	-	1.4
		Polyplectropus	0.7	0.2	1.1	4.1	0.2	0.4
	Xiphocentronidae		0.4	0.5	0.7	1.1	0.4	2.0
Lepidoptera	Pyralidae		-	0.2	1.8	-	-	26.2
Coleoptera	Chrysomelidae		-	0.2	-	-	-	-
	Curculionidae		-	-	-	-	0.2	-
	Dryopidae		-	-	-	-	0.5	-
	Dytiscidae		0.7	-	1.1	0.2	1.1	4.8
	Elmidae	*Austrolimnius*	-	0.4	1.3	-	0.7	6.8
		Cylloepus	1.1	-	0.5	0.7	-	-
		Heterelmis	1.6	1.3	0.7	8.1	1.8	2.9
		Hexacylloepus	7.7	12.7	16.7	2.9	11.1	10.9
		Hexanchorus	-	-	0.2	-	-	0.2
		Macrelmis	1.3	4.3	1.3	6.1	12.7	9.7
		Microcylloepus	0.2	0.7	0.7	-	2.3	7.4
		Neocylloepus	-	-	-	-	0.2	-
		Neoelmis	14.4	15.4	6.1	87.7	95.4	43.1
		Notelmis	-	-	-	0.2	-	-
		Phanocerus	-	-	-	-	-	0.2
		Stenhelmoides	-	-	-	-	0.2	-
	Gyrinidae		0.2	-	-	0.2	-	-
	Hydraenidae		-	-	0.4	-	-	1.3
	Hydrophilidae		0.4	-	0.2	0.2	-	1.1
	Hydroscaphidae		-	0.5	2.3	-	-	-

Appendix 1 cont.

Taxonomic Groups	Family	Genus	Pools			Riffles		
			F	FB	P	F	FB	P
Coleoptera	Limnichidae		0.4	-	0.2	0.2	0.2	-
	Psephenidae	Eubriinae	0.4	-	0.2	0.7	0.4	-
		Psepheninae	1.1	-	5.0	8.4	-	28.9
	Ptilodactylidae	*Anchytarsus*	18.5	7.4	0.2	82.2	23.0	0.5
	Scirtidae		0.5	0.9	0.5	0.2	1.4	0.9
	Staphylinidae		0.4	-	0.2	0.2	0.2	0.7
Diptera	Ceratopogonidae		12.6	4.5	4.3	6.1	3.6	8.3
	Chaoboridae		-	-	0.4	-	-	0.4
	Chironomidae		122.7	219.9	287.6	183.5	270.4	324.5
	Culicidae		0.9	0.2	0.4	0.7	0.4	0.7
	Dixidae		-	-	-	3.2	-	-
	Dolichopodidae		0.2	0.2	0.4	0.2	0.2	-
	Empididae		0.2	-	-	0.5	0.2	0.2
	Psychodidae		0.2	0.4	-	1.3	0.5	0.2
	Simuliidae	*Simulium*	0.2	-	0.2	1.3	18.3	6.5
	Stratiomyidae		0.2	0.2	-	0.4	0.2	-
	Tabanidae		-	-	-	0.2	-	-
	Tipulidae	*Hexatoma*	5.9	6.8	1.1	10.4	9.9	1.8
		Limonia	-	-	-	-	-	0.7
Total Density (individuals/m^2)			643	1040	688	1513	1537	1139

Chapter 3: Altitudinal gradients in diversity and diadromy in stream fish assemblages of the Sixaola River basin, Costa Rica

Abstract

Distributional patterns of stream fishes remain poorly understood in many areas of Mesoamerica, while anthropogenic impacts continue to alter freshwater ecosystems throughout the region. Diadromous fishes may be especially vulnerable to habitat degradation and hydrologic alterations, but few studies have focused on their distribution and abundance in Mesoamerican streams. In this study, we investigated how fish diversity and assemblage structure vary along an altitudinal gradient in the Sixaola River basin, one of the least-impacted river basins on the Caribbean slope of Costa Rica. Stream fishes were sampled in 20 reference-quality sites located at elevations from near sea level to 500 m. Fish diversity declined steeply with increasing elevation, with very limited species replacement. Ordination of the study sites based on fish relative abundance showed some grouping of sites, based in part on geographic proximity. The main gradient in fish assemblage structure was correlated with several stream habitat variables, especially the amount of pool habitat, substrate size, and channel slope. The location of sampling sites within the river network appeared to have a less important influence on fish diversity than altitudinal gradients in stream habitat, but may still play an important role in structuring local fish assemblages. Diadromous fishes were common at most sites, and did not show strong altitudinal gradients in species richness or abundance. As a result, their contribution to local diversity and abundance increased significantly with elevation, and diadromous fishes were dominant at the highest elevation sites. Our results illustrate how altitudinal gradients in habitat can affect stream fish assemblages and highlight the importance of riverine connectivity to the ecological integrity of streams in Mesoamerica.

Introduction

Freshwater fishes respond to a wide range of environmental gradients in lotic ecosystems, producing heterogeneity in assemblage structure at multiple spatial scales (Matthews 1998). Documenting this diversity and developing an understanding of the biotic and abiotic factors that interact to maintain it is crucial for developing effective conservation strategies (Angermeier and Winston 1998). Nevertheless, distributional patterns of stream fishes and factors influencing fish assemblage structure remain poorly understood in many areas of the tropics.

Mesoamerica has a diverse freshwater fish fauna that reflects a complex history of colonization from South America and subsequent diversification (Miller 1966, Myers 1966, Bermingham and Martin 1998). Barriers to dispersal have resulted in very limited geographic ranges for many fish species, and high levels of endemism in the region (Smith and Bermingham 2005). The freshwater ecosystems that support these fishes, however, are being heavily altered by hydroelectric development, pollution, and deforestation (Pringle and Scatena 1999, Anderson et al. 2006). Thus, there is an urgent need to document the distribution of freshwater species and assemblages in Mesoamerican river systems that are relatively intact, both as a baseline by which to compare future changes and as a goal for restoration in systems that have already been degraded.

Studies of freshwater fishes in Mesoamerica have generally emphasized variation in fish assemblages along gradients in stream size and/or distance from the ocean (Angermeier and Karr 1983, Lyons and Schneider 1990, Winemiller and Leslie 1992, Rodiles-Hernández et al. 1999, Esselman et al. 2006), and have documented positive relationships between assemblage diversity and stream size, as well as longitudinal zonation from headwater to coastal areas. Work in other regions has shown that freshwater fish assemblages can also vary greatly among streams that are similar in size, but which vary in watershed position (Osborne and Wiley 1992, Roper and Scarnecchia 2001, Pouilly et al. 2006). Comparisons among streams of similar size in different parts of a river network could provide insight into how fishes respond to environmental gradients while controlling for many confounding factors associated with longitudinal changes in channel size and stream discharge.

A variety of characteristics can be used to describe spatial variation in biological communities, including taxonomic diversity, species composition, and functional organization. The prevalence of certain life history traits can be especially informative, and may allow for more mechanistic explanations of species distributions in relation to environmental factors (Poff 1997). These traits may also provide information on how vulnerable specific species and assemblages may be to environmental change. For example, migratory species require a specific array of habitats to complete their life cycle and are particularly vulnerable to changes in hydrologic connectivity in river systems. Diadromous fishes and shrimps, which make obligatory migrations between freshwater and marine habitats during their life cycle (McDowall 1987), are widely recognized for their ecological significance and increasing imperilment in Caribbean island streams (e.g. Holmquist et al. 1998, Fiévet et al. 2001, March et al. 2003, Greathouse et al. 2006). Diadromous taxa are also common in many mainland Mesoamerican streams and face similar threats, but have generally received much less research and conservation attention (but see Cruz 1987, Anderson et al. 2006, Esselman et al. 2006).

In this study, we describe patterns in diversity and species composition of stream fish assemblages in relation to environmental conditions along an altitudinal gradient spanning nearly 500 m in the Sixaola River basin, Costa Rica. We chose to work in the Sixaola River basin because it is one of the least-impacted watersheds of its size in Costa Rica (Sanchez-Azofeifa et al. 2002), and has no hydroelectric facilities or other engineered barriers to fish passage. Furthermore, we are not aware of any published studies documenting patterns of stream fish diversity or assemblage structure in the Bocas biogeographical province of southeastern Costa Rica and western Panama (Smith and Bermingham 2005). Altitudinal gradients in fish diversity and species composition are particularly relevant to conservation in the study area because anthropogenic impacts have been concentrated in lowland areas, while many montane streams are relatively pristine. In this context, our objectives were to document the altitudinal distribution of stream fishes, particularly the distribution of diadromous species, and investigate how the structure of stream fish assemblages relates to local habitat conditions and spatial position within the river network.

Methods

Study Area

The study was conducted in the Costa Rican portion of the Sixaola River basin (9°35' N, 83°00' W), an international river basin spanning the border between Costa Rica and Panama on the Caribbean slope of the Talamanca mountains (Figure 1). The Sixaola watershed encompasses an area of approximately 2,780 km^2, and spans elevations from sea level to over 3,800 m. Over half of the Costa Rican portion of the Sixaola basin is protected by national parks or biological reserves, including the headwaters of all major tributaries. The middle portion of the watershed lies mostly within the Bribri and Cabécar Indigenous Territories, while the lower Sixaola Valley and surrounding hills are almost entirely privately owned.

In this study, we sampled streams from near sea level to an elevation of 500 m, an altitudinal gradient spanning a distance of over 50 km. Annual precipitation varies along this gradient from approximately 2500 mm in the lowlands to over 3000 mm at the highest elevation sites (WorldClim, Hijmans et al. 2005). The climate in the region is largely aseasonal, but rainfall tends to peak in November and December, while drier conditions often persist in February-April, and again in August-September (Coen 1983). Natural vegetation also varies along the altitudinal gradient, from tropical moist forest at lowland sites to premontane wet forest at the highest elevation sites (Holdridge 1967). Natural forest vegetation has been replaced with pastures and extensive banana and plantain plantations in much of the lower Sixaola River basin. Land cover in piedmont areas of the Bribri and Cabécar indigenous territories is a mix of small farms, remnant natural forest, and extensive fallows.

Twenty sites on second- and third-order streams were selected for the study and sampled on one occasion between February-June 2006. Only reference-quality sites representing the least-disturbed conditions available were selected, and streams were always at or near base flow when sampling took place. Accessibility was a major constraint, as the Sixaola basin has relatively little infrastructure and road access is limited or nonexistent in much of the study area. Potential study sites were identified from 1:50,000 scale topographic maps (Instituto Geografico Nacional, Costa Rica) and then scouted in advance of sampling to investigate current land use patterns in riparian and upland areas. We purposely avoided sites above waterfalls that would restrict fish passage. At each site, we selected a study reach twenty times the mean wetted channel width (range 65-130 m) that was representative of stream habitat conditions for several hundred meters upstream and downstream of the chosen site.

Fish Sampling

Fishes were sampled at all sites using a backpack electrofishing unit (Model B-12, Smith-Root Inc., Vancouver, Washington, USA). The goal was to capture a representative sample of the fish assemblage at each site, in terms of species presence and relative abundance, and our methods reflect time constraints associated with sampling the most remote sites. Sampling reaches were divided into 3-4 sub-reaches and one very thorough pass was made through all habitats in each sub-reach. Our preliminary work in high and low elevation streams showed that fish were concentrated in deeper pool and cascade habitats, and so a second pass was made through all pools, as well as cascade habitats >30 cm in depth. Fish from all passes at a site were combined for the analyses. Block nets were used to isolate sub-reaches when deemed necessary. All fishes >20 mm were identified to species in the field using keys in Bussing (1998), except for fishes of the genera *Eleotris* and *Sicydium*. Larger *Sicydium* individuals (>100 mm) could be identified to species in the field based on differences in tooth morphology using a hand loupe, and we found that two species, *Sicydium adelum* and *Sicydium altum*, often co-occurred at our sampling sites. However, most *Sicydium* individuals were much smaller than 100 mm, and could not be identified. Given the difficulty of reliably identifying *Eleotris* and *Sicydium* individuals, and recognizing the similar ecology of the species within each genus that occur in the study region (Bussing 1998, Winemiller and Ponwith 1998), these fish were grouped at the genus level in all analyses.

Fish species were classified as diadromous based on references from Costa Rica or other parts of their range (Gilbert and Kelso 1971, Loftus et al. 1984, Cruz 1987, 1989, Bussing 1998). For *Awaous banana*, *Eleotris*, and *Pomadasys crocro*, diadromy is assumed based on several different lines of evidence, especially their presence in *tismiches*, or mass upstream migrations of juvenile fishes and shrimps that occur in coastal rivers of the study region (Gilbert and Kelso 1971, Nordlie 1981, Winemiller and Ponwith 1998). Evidence of diadromy in *Gobiomorus dormitor* is equivocal (Gilmore 1992), but this species was rare in our collections and assuming diadromy has little effect on our results. We further classified diadromous species as catadromous or amphidromous (McDowall 1987), but migration patterns are poorly understood for all of the species collected in this study and these designations should be considered provisional pending further studies.

Environmental variables

Stream habitat measurements were made at each site in an effort to quantify key factors potentially influencing the diversity and composition of stream fish assemblages. All channel geomorphic units in the sampling reach were categorized as pool, riffles, runs, or cascades based on criteria in Hawkins et al. (1993), and the proportion of the sampling reach occupied by each channel unit type was estimated visually. Ten transects spaced two channel widths apart were established in the study reaches, and stream depth and substrate size were measured at each transect following methods adapted from Iwata et al. (2003). Stream depth was measured at five equally-spaced points across the stream channel, and at each depth sampling point, the dominant substrate in a 25 x 25 cm area surrounding the point was classified as sand or silt (particle size <2 mm), gravel (2-16 mm), pebble (17-64 mm), cobble (65-256 mm), boulder (>256 mm), or bedrock. These substrates were then coded as follows: 1, sand or silt; 2, gravel; 3, pebble; 4, cobble; 5, boulder; and 6, bedrock. The mean and standard deviation of the substrate values were used as indices of substrate coarseness and variability, respectively, for the sampling reaches (Bain et al. 1985).

Stream canopy cover was estimated visually to the nearest 10% at 5 equally spaced points in the center of the wetted channel and then averaged for the reach. Channel slope was determined using a clinometer and measuring staff. Water temperature was measured with a thermometer in a well-mixed area of the stream channel between 09:30-10:00 hours. Stream discharge data were not available for any of the sampling sites, and so we used catchment area as a proxy for stream size. Catchment boundaries were delineated using a digital elevation model of the study area with approximately 90 m resolution from the Shuttle Radar Topography Mission (SRTM; United States Geological Survey 2002). Elevation at the mid-point of each sampling reach was also determined using the SRTM elevation data. Physical characteristics of the study sites are summarized in Table 1.

Finally, in order to investigate the importance of spatial position within the watershed on fish assemblage structure, we calculated link magnitude (Shreve 1966) for all the streams in the Sixaola basin from the same 1:50,000 scale topographic maps used to select sites. Each study reach was then assigned a downstream link value based on the link magnitude at the next major tributary downstream. This is a relatively coarse method for estimating watershed position, but has been shown to adequately represent downstream processes influencing fish assemblages in tributary streams (Osborne and Wiley 1992). Streams often had a relatively small tributary immediately downstream, and so the link magnitude at the nearest confluence with the mainstem Sixaola or one of its five major tributaries (Figure 1) was used. Downstream distances to a major tributary varied from 0.2-13.4 km among the study sites and tended to be slightly lower in higher elevation sites because access routes often followed major tributaries.

Data Analysis

Relationships between elevation, environmental variables, and fish assemblage characteristics were investigated using linear regression analysis in SAS Version 9.1 (SAS Institute Inc. 2002). Log transformations were used when necessary to meet regression model assumptions. Regressions were considered significant at $\alpha = 0.05$. Fish diversity in the study reaches was expressed as species richness, based on the total number of species observed, and using Fisher's alpha diversity index. Fisher's alpha was used because it is relatively insensitive to sample size (Magurran 1988), which varied greatly among the study reaches. In order to evaluate how well our sampling methods characterized species richness in each sampling site, we used the program EcoSim (Gotelli and Entsminger 2005) to calculate individual-based rarefaction curves, and then inspected the curves to determine whether sampling produced an asymptote in species richness.

Relationships between fish assemblages and environmental gradients were investigated by extracting major gradients in fish assemblage structure using ordination and then relating these gradients to individual environmental variables. Ordination of the sampling sites was done using nonmetric multidimensional scaling (NMS) in the program PC-ORD (McCune and Mefford 1999). The distance matrix for the NMS ordination was constructed using Bray-Curtis distances among sites based on fish relative abundance, and all species were retained in the analysis. The final ordination was based on the best configuration from 250 runs with random starting points. Appropriate dimensionality for the ordination was determined from a scree plot and significance tests of stress values based on 250 Monte Carlo randomizations. Pearson correlation coefficients between environmental variables and ordination scores were calculated in PC-ORD.

Results

Altitudinal variation in fish assemblages

A total of 6689 fish representing 26 species and 11 families were captured in the study streams (Table 2). Thirteen of the 26 species belonged to the families Characidae, Poeciliidae, and Cichlidae. Characids and poeciliids were numerically dominant at all but the highest elevation sites and accounted for 38% and 30%, respectively, of all fish captured. Diadromous fishes, including two *Sicydium* species, accounted for eight of the 26 species and approximately 25% of all individuals collected in the study streams (Table 2).

Significant changes in fish diversity were observed along the altitudinal sampling gradient, with fish species richness decreasing steeply with increasing elevation (Figure 2a). Species richness rarefaction curves for the sampling sites showed that our sampling effort failed to produce an asymptote in species richness in more than half of the sites (data not shown). Most of these sites were at lower elevations, however, and so actual gradients in species richness may be even more pronounced than shown here. Our sampling methods did not allow us to make exact estimates of fish density, but fish abundance also appeared to decline markedly along the altitudinal sampling gradient (Table 3). Sample size appeared to have little influence on the altitudinal diversity gradient we observed, however, as the relationship between alpha diversity and elevation was nearly identical to the pattern observed for species richness (Figure 2b).

Changes in fish diversity with elevation were mostly driven by a pattern of species addition from the highest to lowest elevation sites, although there was considerable variation within this general pattern (Table 3). Of the 26 species captured in the study, 25 were found in at least one of the five lowest elevation sites, and these sites had 12 species that were never observed at higher elevations. Some species replacement was also evident along the altitudinal gradient, but most species that were absent from the lowest elevation sites were quickly added with increasing elevation and had broad altitudinal distributions (Table 3). In one case, we observed species replacement among congeners, with *Rhamdia rogersi* replacing *Rhamdia guatemalensis* at higher elevations.

Relationships between fish assemblage structure and environmental factors

Variation in elevation among sampling sites represented a set of highly correlated environmental gradients (Table 4), and, and as noted above, explained a large proportion of the variation in fish diversity. Elevation and downstream link magnitude were also significantly correlated, but this relationship was surprisingly weak (Table 4). Downstream link magnitude showed a significant positive correlation with alpha diversity ($r^2 = 0.34$, $P = 0.007$), but was a much poorer predictor of fish diversity than elevation. Catchment area was not significantly correlated with alpha diversity ($r^2 = 0.01$, $P = 0.73$), and also did not significantly improve model fit when added to the regression model predicting alpha diversity from elevation.

Ordination of sampling sites using NMS based on fish relative abundance produced a two-dimensional solution (Figure 3). Although the sites were selected to represent a continuous altitudinal gradient, three fairly distinct groups of sites were evident in the ordination, reflecting in part the geographic distribution of sampling sites. One group was composed of the three lowest elevation sites, all of which were located in the lower Sixaola River basin far from the other sampling sites. One of these three sites, TIG2, also showed affinities with the second group, which included 12 sites located at middle elevations. All of the sites in the second group had similar fish assemblages despite being distributed among several different sub-basins in the study area. The third group was made up of five sites located in the Coen River sub-basin (Figure 3). All of these sites were located at higher elevations and included the three highest elevation sites in the study. However, two of the sites in this group were located at similar elevations as sites in the second group, but had quite different fish assemblages.

The two NMS ordination axes together explained 94% of the variation in fish assemblages among sampling sites. The first ordination axis alone explained 72% of the total variation in fish assemblage structure, and was highly correlated with a number of habitat variables that co-varied with elevation (Table 4). In particular, Axis 1 showed strong positive correlations with channel slope, substrate coarseness, the percentage of cascade habitat, and distance from the ocean, and a weaker positive association with substrate variability. Axis 1 was negatively correlated with the percentage of pool habitat, water temperature, canopy cover, and downstream link magnitude (Table 4, Figure 3). Most environmental variables that were significantly correlated with the first NMS axis showed very similar relationships with elevation, but differences were also apparent, especially for substrate variability and water temperature. Interestingly, downstream link magnitude showed a much stronger correlation with the first NMS axis than with either elevation or fish diversity. The second ordination axis explained an additional 22% of the variation in fish assemblages, but only showed a significant correlation with one environmental variable, average depth (Table 4). Axis 2 appeared to separate the middle elevation sites, which tended to have shallower channels and more riffle habitat, from the lowest and highest elevation sites, which were dominated by pools or cascades, respectively (Figure 3). Catchment area was not significantly correlated with either of the ordination axes.

Distribution of diadromous fishes

Diadromous fishes showed very different patterns of diversity and abundance along the altitudinal gradient than fish assemblages overall. Although distance from the ocean increased with elevation, there was no significant relationship between elevation and the number of diadromous species captured in the sampling sites ($r^2 = 0.07$, $P = 0.26$). The number of diadromous individuals captured, standardized by sampling area, also showed no significant relationship with elevation ($r^2 = 0.04$, $P = 0.39$). Thus, while overall fish species richness and density declined sharply from low to high elevation sites, the diversity and abundance of diadromous species remained relatively constant. As a result, the importance of diadromous species in fish assemblages increased dramatically along the altitudinal gradient. When expressed as a percentage of total species richness at each site, diadromous species richness had a highly significant positive association with elevation (Figure 4a). Likewise, the percentage of individuals in a site with a diadromous life history also increased significantly with elevation (Figure 4b).

Although diadromous species richness did not vary significantly with elevation, we did observe changes in the species composition within the diadromous group along the altitudinal gradient (Table 3). Most of this turnover occurred at low elevations, and appeared to be related to proximity to the ocean. Distance to the ocean generally increased throughout the altitudinal gradient, but the three lowest elevation sites were much closer to the ocean than the rest of the sampling sites (Figure 1). In these sites, *Eleotris* was the dominant diadromous taxon, while *Sicydium* and *Joturus pichardi* were completely absent. Outside of the three lowest elevation sites, *Sicydium* was abundant and ubiquitous, while *J. pichardi* occurred at many sites, always in low abundance. *Agonostomus monticola* was present in all but one of the sampling sites and showed the widest altitudinal range of any species in the study. Distributional patterns of diadromous fishes were not clearly related to the different types of diadromy represented among the study species. In fact, catadromous and amphidromous taxa were both widely distributed along our sampling gradient (Table 3). In terms of abundance, catadromous species were rare at all sites, while amphidromous taxa were common in streams throughout the altitudinal gradient.

Discussion

Longitudinal gradients in fish assemblage diversity are well documented in temperate and tropical river systems (Matthews 1998, Lowe-McConnell 1987). These gradients are often driven by a pattern of species addition from headwaters to larger streams (Sheldon 1968, Horwitz 1978, Rahel and Hubert 1991, Rodiles-Hernández et al. 1999, Robinson and Rand 2005), although longitudinal zonation and species replacement can also be important (Balon and Stewart 1983, Petry and Schulz 2006). Many different factors have been proposed to explain these patterns, including increasing habitat diversity (Gorman and Karr 1978), decreasing temporal variability (Horwitz 1978), and greater habitat volume, especially the development of larger pools (Sheldon 1968), in downstream areas. These factors affect a variety of biotic and abiotic processes in streams, and interact in important ways to influence the structure and diversity of stream fish assemblages along the river continuum (Schlosser 1982, 1987).

Our results, like those of Pouilly et al. (2006), illustrate that environmental factors that co-vary with elevation can produce similar gradients in fish diversity, independent of stream size. Gradients in stream habitat from high to low elevation streams were also consistent in several ways with changes normally observed along a continuum of stream size, including increases in water temperature and the proportion of pool habitat, and decreases in channel slope and substrate coarseness. Pouilly et al. (2006) observed similar patterns in streams along an altitudinal gradient in Bolivia, and concluded that changes in stream habitat and productivity may both be important in explaining variation in fish assemblage structure.

The high covariance we observed among environmental variables complicates interpretation of their influence on fish diversity and taxonomic composition. However, geomorphic changes associated with variation in channel slope along the altitudinal gradient appeared to be especially important. In particular, we found large differences among sites in the amount of pool habitat, with the prevalence of pools increasing considerably from the highest to lowest elevation sites (Table 1). Pool development is hypothesized to be of critical importance in structuring stream fish assemblages in small temperate streams through its influence on biotic interactions, temporal variability, and habitat heterogeneity (Schlosser 1987). Although seasonal dynamics are very different in tropical streams, the availability of deeper pool habitats has similarly important effects on predation, competition and population dynamics (e.g. Power 1984).

The vast majority of fish captured in our study sites were associated with pool habitats. However, several small fish species appeared to select against deep pool habitats. *Rivulus isthmensis*, in particular, was nearly always found in riffles or in shallow pools isolated from the main channel. Biotic interactions have been shown to have a significant influence on the distribution and abundance of another *Rivulus* species in Trinidad (Gilliam et al. 1993), and may be an important factor in habitat selection for many of the fish species in our study streams.

Interestingly, although we observed a significant increase in pool habitat with decreasing elevation, we did not see any consistent trends in the mean depth of the study reaches. Averaging over the entire reach may have obscured the influence of deeper pool habitats, but maximum depths recorded in the study reaches also varied little among high and low elevation sites. Nevertheless, there were clear differences in habitat conditions in the deepest channel units along the altitudinal gradient. Low elevation pools were generally long, with very little current at base flow conditions and substrates dominated by fine organic and inorganic material. At higher elevations, the deepest habitats occurred either in relatively short pools or within cascades, both of which had much higher water velocities and coarser substrates than low elevation pools. Many fishes in the region appear to have affinities for specific water velocities (Bussing and Lopez 1976), and so flow conditions may be as important as water depth in determining habitat suitability.

The altitudinal gradient in stream canopy cover also failed to match patterns expected along a river continuum. In fact, we found a significant increase in stream canopy cover with decreasing elevation, a pattern heavily influenced by sites at the elevational extremes in our study (Table 1). Longitudinal variation in canopy cover from headwaters to larger streams is thought to play a critical role in structuring stream communities through its influence on basal food resource availability (Vannote et al. 1980). Although we did not observe analogous patterns along our altitudinal sampling gradient, there was considerable variation in stream canopy cover among sites that was influenced by stream size and channel morphology. High correlations between stream canopy cover and several other environmental factors made it difficult to isolate the effects of stream shading on fish assemblages. However, the potential for canopy cover to regulate instream primary production and fish assemblage trophic structure deserves further study.

Although local stream habitat conditions strongly affect the diversity and species composition of fish assemblages, spatial position within a watershed can also have an important influence through its effect on colonization processes (Gorman 1986, Osborne and Wiley 1992, Roper and Scarnecchia 2001). Unfortunately, spatial factors are often confounded by major habitat gradients, making it difficult to determine the relative importance of colonization dynamics versus local habitat suitability. In our study streams, sites at higher elevations were increasingly distant from potential source pools of immigrants in the largest, most temporally stable mainstem habitats, but also had very different habitat conditions than lowland streams. Not surprisingly, we found that the downstream link magnitude of our study sites was correlated with elevation. However, we also found that elevation and several co-varying habitat variables were much stronger predictors of fish diversity than downstream link magnitude. This suggests that local habitat conditions have a stronger influence on fish diversity in the study streams, but does not mean that colonization processes are unimportant. In fact, despite having a weak correlation with elevation, downstream linkage magnitude showed a relatively strong correlation with the main gradient in fish assemblage structure (Table 4).

Variation in valley morphology among the study streams supports the idea that extinction-colonization dynamics differ along the altitudinal sampling gradient. Heavy rainfall in the study region produces frequent floods with highly stochastic timing. Lowland streams typically had wide, unconstrained valleys where stream energy could dissipate during these floods. In contrast, montane sites typically had very narrow valleys where flood energy was concentrated in the stream channel. When combined with the high channel gradient in montane streams, the magnitude of flood disturbance on stream biota is likely to be much greater in these sites than in lowland streams. However, very little is known about the home range size or dispersal abilities of any the freshwater fish species in our study area, which would also influence interpretations on the importance of extinction and immigration in the study streams. By necessity, we captured a snapshot of fish assemblage structure in our study sites, but long-term studies over larger spatial scales will be needed to determine how local disturbance and larger-scale colonization dynamics influence diversity and species composition in these streams.

Understanding the consequences of fish movement is especially important for diadromous species, which make up a substantial portion of the fish fauna in the Sixaola River basin. Diadromous taxa appear to have very different patterns of abundance and diversity than non-diadromous fishes in this system, resulting in significant changes in their relative abundance along our altitudinal sampling gradient. This pattern is very different from Caribbean island streams, which generally have depauperate freshwater fish assemblages dominated by diadromous species at all elevations (Fièvet et al. 2001, Greathouse et al. 2006). In these systems, the distribution of diadromous species is influenced by their life history requirements, biotic interactions with other diadromous taxa, and physical barriers to migration (Greathouse et al. 2006), but not by interactions with a relatively rich freshwater fish fauna, as in mainland Mesoamerican streams. These biotic interactions are likely to have a strong influence on the distribution and abundance of diadromous fishes, and highlight the importance of differential abilities among diadromous taxa to penetrate higher-elevation freshwater environments (McDowall 1998). In streams of the Sixaola basin, these factors have produced a somewhat counterintuitive altitudinal gradient in the prominence of diadromous species. However, the pattern we observed is likely common on the Caribbean slope of Mesoamerica, where diadromous mugilid and gobiid fishes are able to penetrate far inland and are among the few species adapted to high gradient montane streams (Bussing and López 1977, Cruz 1987, Bussing 1998, Esselman et al. 2006).

Our findings have several important implications for conservation of stream fish diversity in the Sixaola River basin, and more broadly in Caribbean slope streams of Mesoamerica. First, small streams in the region are diverse, both physically and biologically, with major altitudinal gradients in stream fish diversity and species composition. Effective conservation of all stream fish species and assemblages along this gradient will thus require protection of a representative array of streams and their associated watersheds. Recent studies from other neotropical regions have also found high spatial heterogeneity in fish assemblages among small streams (Gerhard et al. 2004), and discussed the implications for reserve design (Mendonça et al. 2005). The current distribution of protected areas in the Sixaola River basin illustrates the difficulties of building representative reserve networks for stream fishes. While over half of the Costa Rican portion of the Sixaola watershed is currently protected by national parks or reserves, existing protected areas are concentrated in high elevation areas. There is virtually no formal protection of lowland streams and their associated watersheds, which support the most diverse fish assemblages.

Similar situations are encountered elsewhere in Costa Rica and are common worldwide, presenting further challenges when diadromous taxa are considered (Pringle 2001). Although montane streams in the Sixaola basin are protected by national park status and a lack of human infrastructure, movements by diadromous fishes connect these areas with other more heavily impacted parts of the landscape. Most of our sites were located well below the borders of La Amistad International Park, the largest protected area in the region, but we did have one site located within a small lower elevation extension of the park, and another close to the boundary of the main body of the park. In these sites, diadromous fishes made up approximately 42% and 98%, respectively, of all fishes captured. Although not specifically targeted in this study, diadromous shrimps of the families Palaemonidae and Atyidae were also common in all study streams up to the highest elevation sites (C.M. Lorion and B.P. Kennedy, unpublished data). Despite the general lack of knowledge about life histories of diadromous taxa in the region, it appears that all of these species must migrate to the ocean to complete their life cycle. As a result, downstream alterations in hydrologic connectivity, stream habitat, and water quality are likely to have a significant influence on biotic integrity upstream (Pringle 1997), including streams within protected areas (Pringle 2001).

Declines in the herbivores *Sicydium* and *J. pichardi* could have especially important consequences for the study streams, as the grazing activities of these species likely have a significant influence on nutrient cycling and other stream ecosystem processes. Our sampling methods likely underestimated the abundance of *Sicydium* gobies because of their strong benthic orientation. Nevertheless, they were among the most abundant fishes in our samples and we commonly observed feeding scars associated with their grazing activities. Although *J. pichardi* was not as common, it has traditionally been the most important species for subsistence fishing by indigenous communities in the Sixaola River basin (Borge and Castillo 1997).

In summary, we observed strong gradients in diversity and considerable heterogeneity in the species composition of stream fish assemblages along an altitudinal gradient in the

Sixaola River basin. Altitudinal variation in fish assemblage structure appeared to be strongly related to stream habitat gradients, but spatial relationships between streams and sources of colonists may also be important. Finally, diadromous species were common in all sites, highlighting the critical importance of maintaining hydrologic connectivity and natural flow regimes in Mesoamerican streams (Esselman et al. 2006), especially in the decreasing number of river systems that remain free-flowing (Anderson et al. 2006).

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References

Anderson, E. P., C. M. Pringle, and M. Rojas. 2006. Transforming tropical rivers: an environmental perspective on hydropower development in Costa Rica. Aquatic Conservation: Marine and Freshwater Ecosystems 16:679-693.

Angermeier, P. L., and J. R. Karr. 1983. Fish communities along environmental gradients in a system of tropical streams. Environmental Biology of Fishes 9:117-135.

Angermeier, P. L., and M. R. Winston. 1999. Characterizing fish community diversity across Virginia landscapes: prerequisite for conservation. Ecological Applications 9:335-349.

Bain, M. B., J. T. Finn, and H. E. Booke. 1985. Quantifying stream substrate for habitat analysis studies. North American Journal of Fisheries Management 5:499-506.

Balon, E. K., and D. J. Stewart. 1983. Fish assemblages in a river with unusual gradient (Luongo, Africa – Zaire system), reflections on river zonation, and description of another new species. Environmental Biology of Fishes 9:225-252.

Bermingham, E., and A. P. Martin. 1998. Comparative mtDNA phylogeography of neotropical freshwater fishes: testing shared history to infer the evolutionary landscape of lower Central America. Molecular Ecology 7:499-517.

Borge, C., and R. Castillo. 1997. Cultura y conservación en la Talamanca indígena. Editorial Universidad Estatal a Distancia, San José, Costa Rica.

Bussing, W. A. 1998. Freshwater fishes of Costa Rica. Editorial de la Universidad de Costa Rica, San José, Costa Rica.

Bussing, W. A., and M. I. López. 1977. Distribución y aspectos ecológicos de los peces de las cuencas hidrográficas de Arenal, Bebedero y Tempisque, Costa Rica. Revista de Biología Tropical 25:13-37.

Coen, E. 1983. Climate. Pages 35-46 in D. H. Janzen, editor. Costa Rican natural history. University of Chicago Press, Chicago, Illinois, USA.

Cruz, G. A. 1987. Reproductive biology and feeding habits of Cuyamel, *Joturus pichardi* and Tepemechín, *Agonostomus monticola* (Pisces: Mugilidae) from Rio Platano, Mosquitia, Honduras. Bulletin of Marine Science 40:63-72.

Cruz, G. A. 1989. *Joturus pichardi* (Pisces: Mugilidae) sexualmente maduros capturados en Laguna de Brus y en la desembocadura del Río Platano, Honduras. Revista de Biología Tropical 37:107-108.

Esselman, P. C., M. C. Freeman, and C. M. Pringle. 2006. Fish-assemblage variation between geologically defined regions and across a longitudinal gradient in the Monkey River Basin, Belize. Journal of the North American Benthological Society 25:142-156.

Fièvet, E., Tito de Morais, L., Tito de Morais, A., Monti, D., and H. Tachet. 2001. Impacts of an irrigation and hydroelectric scheme in a stream with a high rate of diadromy (Guadeloupe, Lesser Antilles): can downstream alterations affect upstream faunal assemblages? Archiv für Hydrobiologie 151:405-425.

Gerhard, P., R. Moraes, and S. Molander. 2004. Stream fish communities and their associations to habitat variables in a rain forest reserve in southeastern Brazil. Environmental Biology of Fishes 71:321-340.

Gilbert, C. R., and D. P. Kelso. 1971. Fishes of the Tortuguero area, Caribbean Costa Rica. Bulletin of the Florida State Museum 16:1-54.

Gilliam, J. F., D. F. Fraser, and M. Alkins-Koo. 1993. Structure of a tropical fish community: a role for biotic interactions. Ecology 74:1856-1870.

Gilmore, R. G. 1992. Bigmouth sleeper, *Gobiomorus dormitor*. Pages 105-111 in C. R. Gilbert, editor. Rare and endangered biota of Florida, Volume 2: fishes. University Press of Florida, Gainesville, Florida, USA.

Gorman, O. T. 1986. Assemblage organization of stream fishes: the effect of rivers on adventitious streams. American Naturalist 128:611-616.

Gorman, O. T., and J. R. Karr. 1978. Habitat structure and stream fish communities. Ecology 59:507-515.

Gotelli, N. J., and G. L. Entsminger. 2005. Ecosim: Null Models Software for Ecology, Version 7.72. Acquired Intelligence Inc. and Kesey-Bear, Jericho, Vermont, USA.

Greathouse, E. A., C. M. Pringle, and J. G. Holmquist. 2006. Conservation and management of migratory fauna: dams in tropical streams of Puerto Rico. Aquatic Conservation: Marine and Freshwater Ecosystems 16:695-712.

Hawkins, C. P., J. L. Kershner, P. A. Bisson, M. D. Bryant, L. M. Decker, S. V. Gregory, D. A. McCullough, C. K. Overton, G. H. Reeves, R. J. Steedman, and M. K. Young. 1993. A hierarchical approach to classifying stream habitat features. Fisheries 18:3-12.

Hijmans, R. J., S. E. Cameron, J. L. Parra, P. G. Jones, and A. Jarvis. 2005. Very high resolution interpolated climate surfaces for global land areas. International Journal of Climatology 25:1965-1978.

Holdridge, L. R. 1967. Life zone ecology. Tropical Science Center, San José, Costa Rica.

Holmquist, J. G., J. M. Schmidt-Gengenbach, and B. B. Yoshioka. 1998. High dams and marine-freshwater linkages: effects on native and introduced fauna in the Caribbean. Conservation Biology 12:621-630.

Horwitz, R. J. 1978. Temporal variability patterns and the distributional patterns of stream fishes. Ecological Monographs 48:307-321.

Iwata, T., S. Nakano, and M. Inoue. 2003. Impacts of past riparian deforestation on stream communities in a tropical rainforest in Borneo. Ecological Applications 13:461-473.

Loftus, W. F., J. A. Kushlan, and S. A. Voorhees. 1984. Status of the mountain mullet in southern Florida. Florida Scientist 47:256-263.

Lowe-McConnell, R. H. 1987. Ecological studies in tropical fish communities. Cambridge University Press, Cambridge, UK.

Lyons, J. and D. W. Schneider. 1990. Factors influencing fish distribution and community structure in a small coastal river in southwestern Costa Rica. Hydrobiologia 203:1-14.

Magurran. A. E. 1988. Ecological diversity and its measurement. Princeton University Press, Princeton, New Jersey, USA.

March, J. G., J. P. Benstead, C. M. Pringle, and F. N. Scatena. 2003. Damming tropical island streams: problems, solutions, and alternatives. BioScience 53:1069-1078.

Matthews, W. J. 1998. Patterns in freshwater fish ecology. Chapman & Hall, New York, New York, USA.

McCune, B., and M. J. Mefford. 1999. PC-ORD. Multivariate analysis of ecological data, Version 5.0. MjM Software, Gleneden Beach, Oregon, USA.

McDowall, R. M. 1987. Evolution and importance of diadromy: the occurrence and distribution of diadromy among fishes. American Fisheries Society Symposium 1:1-13.

McDowall, R. M. 1998. Fighting the flow: downstream-upstream linkages in the ecology of diadromous fish faunas in West Coast New Zealand rivers. Freshwater Biology 40:111-122.

Mendonça, F. P., W. E. Magnusson, and J. Zuanon. 2005. Relationships between habitat characteristics and fish assemblages in small streams of central Amazonia. Copeia 2005:751-764.

Miller, R. R. 1966. Geographical distribution of Central American freshwater fishes. Copeia 1966:773-802.

Myers, G. S. 1966. Derivation of the freshwater fish fauna of Central America. Copeia 1966:766-773.

Nordlie, F. G. 1981. Feeding and reproductive biology of eleotrid fishes in a tropical estuary. Journal of Fish Biology 18:97-110.

Osborne, L. L., and M. J. Wiley. 1992. Influence of tributary spatial position on the structure of warmwater fish communities. Canadian Journal of Fisheries and Aquatic Sciences 49:671-681.

Petry, A. C., and U. H. Schulz. 2006. Longitudinal changes and indicator species of the fish fauna in the subtropical Sinos River, Brazil. Journal of Fish Biology 69:272-290.

Poff, N. L. 1997. Landscape filters and species traits: towards mechanistic understanding and predictions in stream ecology. Journal of the North American Benthological Society 16:391-409.

Pouilly, M., S. Barrera, and C. Rosales. 2006. Changes in taxonomic and trophic structure of fish assemblages along an environmental gradient in the Upper Beni watershed (Bolivia). Journal of Fish Biology 68:137-156.

Power, M. E. 1984. Depth distributions of armored catfish: predator-induced resource avoidance? Ecology 65:523-528.

Pringle, C. M. 1997. Exploring how disturbance is transmitted upstream: going against the flow. Journal of the North American Benthological Society 16:425-438.

Pringle, C. M. 2001. Hydrologic connectivity and the management of biological reserves: a global perspective. Ecological Applications 11:981-998.

Pringle, C. M., and F. N. Scatena. 1999. Aquatic ecosystem deterioration in Latin America and the Caribbean. Pages 104-113 in L. U. Hatch and M. E. Swisher, editors. Managed ecosystems: the Mesoamerican experience. Oxford University Press, New York, New York, USA.

Rahel, F. J., and W. A. Hubert. 1991. Fish assemblages and habitat gradients in a Rocky Mountain-Great Plains stream: biotic zonation and additive patterns of community change. Transactions of the American Fisheries Society 120:319-332.

Robinson, J. L., and P. S. Rand. 2005. Discontinuity in fish assemblages across an elevation gradient in a southern Appalachian watershed, USA. Ecology of Freshwater Fish 14:14-23.

Rodiles-Hernández, R. E., E. Díaz-Pardo, and J. Lyons. 1999. Patterns in the species diversity and composition of the fish community of the Lacanja River, Chiapas, Mexico. Journal of Freshwater Ecology 14:455-468.

Roper, B. B., and D. L. Scarnecchia. 2001. Patterns of diversity, density, and biomass of ectothermic vertebrates in ten small streams along a North American river continuum. Northwest Science 75:168-175.

Sanchez-Azofeifa, G. A., R. C. Harriss, A. L. Storrier, and T. de Camino-Beck. 2002. Water resources and regional land cover change in Costa Rica: impacts and economics. Water Resources Development 18:409-424.

SAS Institute Inc. 2002. SAS Version 9.1. SAS Institute, Cary, North Carolina, USA.

Schlosser, I. J. 1982. Fish community structure and function along two habitat gradients in a headwater stream. Ecological Monographs 52:395-414.

Schlosser, I. J. 1987. A conceptual framework for fish communities in small warmwater streams. Pages 17-24 in W. J. Matthews and D. C. Heins, editors. Community and and evolutionary ecology of North American stream fishes. University of Oklahoma Press, Norman, Oklahoma, USA.

Sheldon, A. L. 1968. Species diversity and longitudinal succession in stream fishes. Ecology 49:193-198.

Shreve, R. L. 1966. Statistical law of stream numbers. Journal of Geology 74:17-37.

Smith, S. A., and E. Bermingham. 2005. The biogeography of lower Mesoamerican freshwater fishes. Journal of Biogeography 32:1835-1854.

United States Geological Survey. 2002. Shuttle Radar Topography Mission Dataset. Available from: http://seamless.usgs.gov.

Vannote, R. L., G. W. Minshall, K. W. Cummins, J. R. Sedell, and C. E. Cushing. 1980. The river continuum concept. Canadian Journal of Fisheries and Aquatic Sciences 37:130-137.

Winemiller, K. O., and M. A. Leslie. 1992. Fish assemblages across a complex, tropical freshwater/marine ecotone. Environmental Biology of Fishes 34:29-50.

Winemiller, K. O., and B. J. Ponwith. 1998. Comparative ecology of eleotrid fishes in Central American coastal streams. Environmental Biology of Fishes 53:373-384.

Table 1. Physical characteristics of the 20 study sites in the Sixaola River basin, Costa Rica.

Site Code	Elevation (m)	Catchment Area (km²)	Stream Width (m)	Channel Slope (%)	Mean Depth (cm)	Pool Area (%)	Temperature (°C)	Canopy Cover (%)	Distance from Ocean (km)
TG1	9	5.9	4.6	0.2	37.2	95	26	76	20
QCA	22	5.5	4.5	0.5	29.1	90	25	84	27
TG2	39	1.0	3.0	0.2	22.2	85	25	90	24
DCB	97	2.1	4.7	1.5	20.5	50	23	78	83
SH1	100	6.0	5.1	1.7	19.3	65	25	54	75
SIS	112	1.1	3.7	1.7	12.6	55	24	54	74
SH2	125	1.7	4.0	2.5	20.3	50	25	76	77
ESP	132	2.2	4.5	0.8	12.9	55	22	80	78
PLS	149	4.2	4.6	2.2	16.9	35	22	54	79
AGB	159	6.0	6.4	2.8	19.1	50	24	58	79
COC	163	3.0	4.4	5.0	14.9	40	23	68	76
DT1	168	4.2	7.2	4.0	25.2	20	23	70	87
NIA	225	2.1	4.9	9.0	24.4	25	23	78	85
LX1	261	5.5	4.7	5.2	17.9	20	23	60	91
DT2	264	2.0	4.0	4.6	15.1	45	23	72	90
SHR	266	1.7	3.8	4.0	12.5	30	21	68	83
CUR	298	6.2	5.4	6.5	28.6	10	22	52	94
LX2	350	2.7	4.0	9.0	16.3	25	23	50	92
SW1	375	1.8	3.7	6.0	19.0	40	23	60	96
SW2	500	5.3	5.4	11.0	33.7	20	21	28	98

Table 2. List of fish species captured in the 20 study sites in the Sixaola River basin, with the total number of individuals of each species captured across all sites.

Family	Species	Number of Individuals
Characidae	*Astyanax aeneus*	266
	Astyanax orthodus	294
	Bryconamericus scleroparius	1980
	Hyphessobrycon panamensis	14
Rhamphichthyidae	*Hypopomus occidentalis*	67
Pimelodidae	*Rhamdia guatemalensis*	56
	Rhamdia rogersi	79
Rivulidae	*Rivulus isthmensis*	16
Poeciliidae	*Alfaro cultratus*	368
	Brachyraphis parismina	13
	Phallichthys amates	220
	Poecilia gillii	638
	Priapichthys annectens	790
Synbranchidae	*Synbranchus marmoratus*	5
Haemulidae	*Pomadasys crocro**	1
Cichlidae	*Astatheros bussingi*	140
	Archocentrus myrnae	58
	Archocentrus nigrofasciatus	32
	Parachromis loisellei	4
Mugilidae	*Agonostomus monticola**	506
	*Joturus pichardi**	28
Gobiidae	*Awaous banana**	13
	*Sicydium adelum**	
	*Sicydium altum**	992[a]
Eleotridae	*Eleotris* sp.*	106
	*Gobiomorus dormitor**	3

* diadromous species

[a] total number of individuals of both *Sicydium* species captured

Table 3. Distribution of stream fishes along an altitudinal sampling gradient in the Sixaola River basin, with sites arranged in order of increasing elevation and fish species arranged by their general altitudinal distribution.

	Site code and elevation (m)																			
	TG1	QCA	TG2	DCB	SH1	SIS	SH2	ESP	PLS	AGB	COC	DT1	NIA	LX1	DT2	SHR	CUR	LX2	SW1	SW2
Species	9	22	39	97	100	112	125	132	149	159	163	168	225	261	264	266	298	350	375	500
Brachyraphis parismina	x																			
Pomadasys crocro [c]	x																			
Hyphessobrycon panamensis	x		x																	
Parachromis loisellei	x		x																	
***Eleotris* sp.** [a]	x	x	x																	
Astyanax aeneus	x	x	x	x																
Alfaro cultratus	x	x	x	x																
Phallichthys amates	x	x	x	x																
Archocentrus nigrofasciatus	x	x	x	x																
Synbranchus marmoratus		x	x	x																
Astyanax orthodus	x	x	x	x	x															
Gobiomorus dormitor [c]	x				x															
Awaous banana [a]	x	x			x		x	x	x	x										
Rivulus isthmensis	x											x	x		x					
Rhamdia guatemalensis	x	x	x	x	x		x				x					x				
Hypopomus occidentalis	x	x	x	x	x	x	x	x		x	x					x				
Poecilia gillii	x	x		x	x	x	x	x	x	x	x	x				x				
Astatheros bussingi	x	x	x	x	x	x	x	x	x	x	x	x			x	x				
Bryconamericus scleroparius	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x			
Agonostomus monticola [a]	x	x		x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x
Priapichthys annectens		x	x	x	x	x	x	x	x	x	x	x	x		x	x			x	
Archocentrus myrnae			x	x	x	x	x	x	x	x	x	x								
***Sicydium* sp.** [a]				x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x
Joturus pichardi [c]				x	x				x	x	x	x					x		x	x
Rhamdia rogersi								x	x	x	x	x		x	x	x	x		x	x
Total Fish Density (ind./m²)	1.7	1.7	2.2	0.9	0.7	2.0	1.2	0.6	0.5	0.7	1.4	0.5	0.4	0.2	0.7	0.9	0.3	0.2	0.6	0.2

Notes: Presence of each species in a sampling site is indicated by an 'x'. Diadromous species are in bold.
[a] amphidromous species; [c] catadromous species

Table 4. Pearson correlation coefficients of linear relationships between environmental variables and elevation, and the two NMS axes (n = 20 for all variables).

	Elevation r	NMS Axis 1 r	NMS Axis 2 r
Catchment Area	0.01	0.08	-0.44
Channel Slope	**0.90**	**0.81**	-0.43
Mean Depth	-0.02	-0.22	**-0.66**
Depth CV	-0.18	-0.16	0.41
Substrate Coarseness	**0.89***	**0.85**	0.25
Substrate SD	0.27	**0.52**	0.39
% Pool Habitat	**-0.78**	**-0.88**	0.08
% Riffle Habitat	-0.15	0.06	0.44
% Cascade Habitat	**0.82**	**0.79**	-0.34
Water Temperature	**-0.77**	**-0.61**	0.04
Canopy Cover	**-0.70**	**-0.75**	0.38
Distance from Ocean	**0.94***	**0.90**	0.05
Downstream Link Magnitude	**-0.48**	**-0.79**	0.38

Notes: Correlation coefficients in bold are significant at $P < 0.05$. Asterisks indicate linear relationships based on $\log_{10}$(elevation).

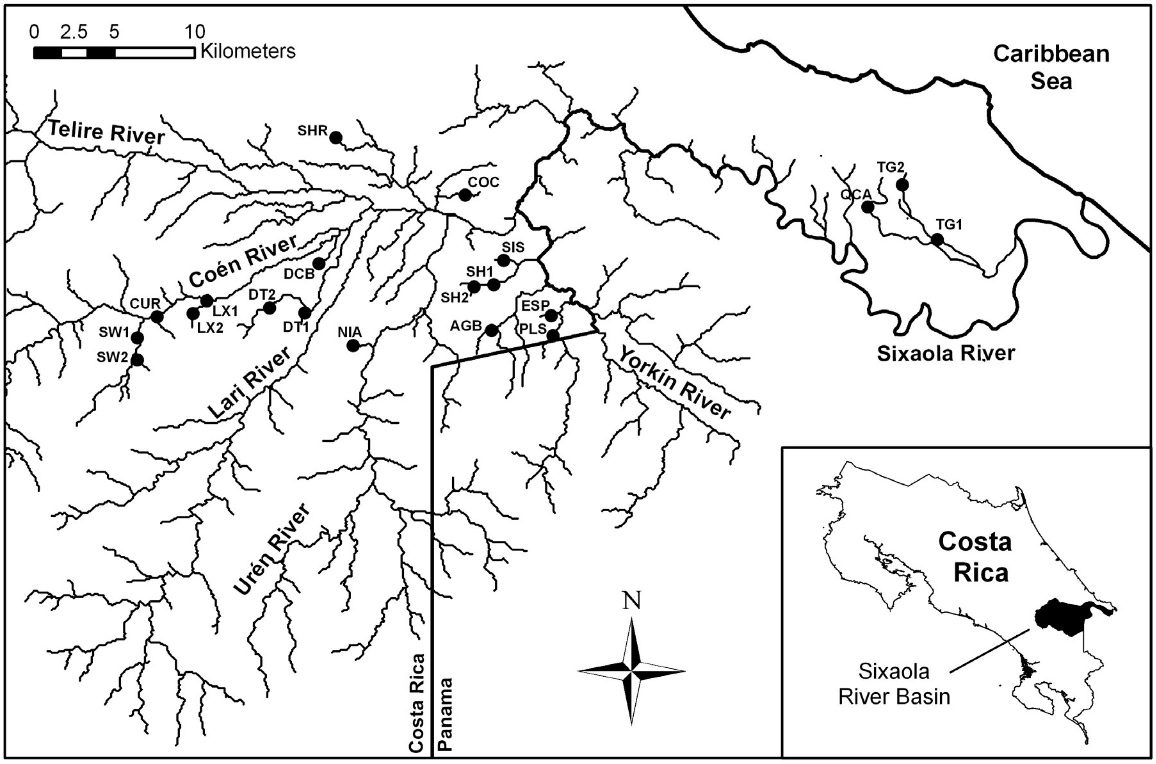

Figure 1. Map of the study area in southeastern Costa Rica, with locations of the 20 sampling sites in the Sixaola River basin.

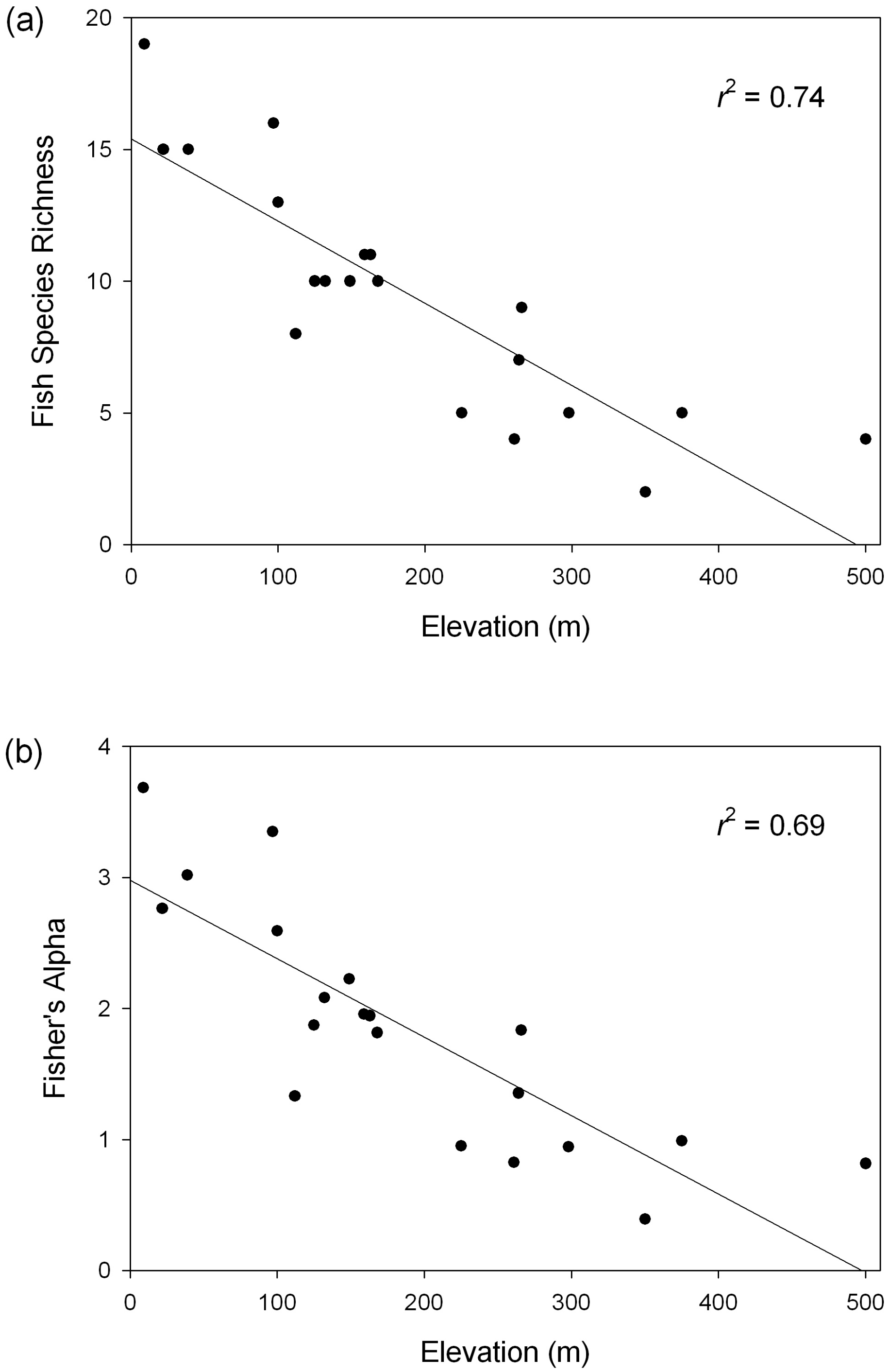

Figure 2. Plots of a) fish species richness and b) Fisher's alpha versus elevation in the 20 study sites ($P < 0.001$ for both regression relationships).

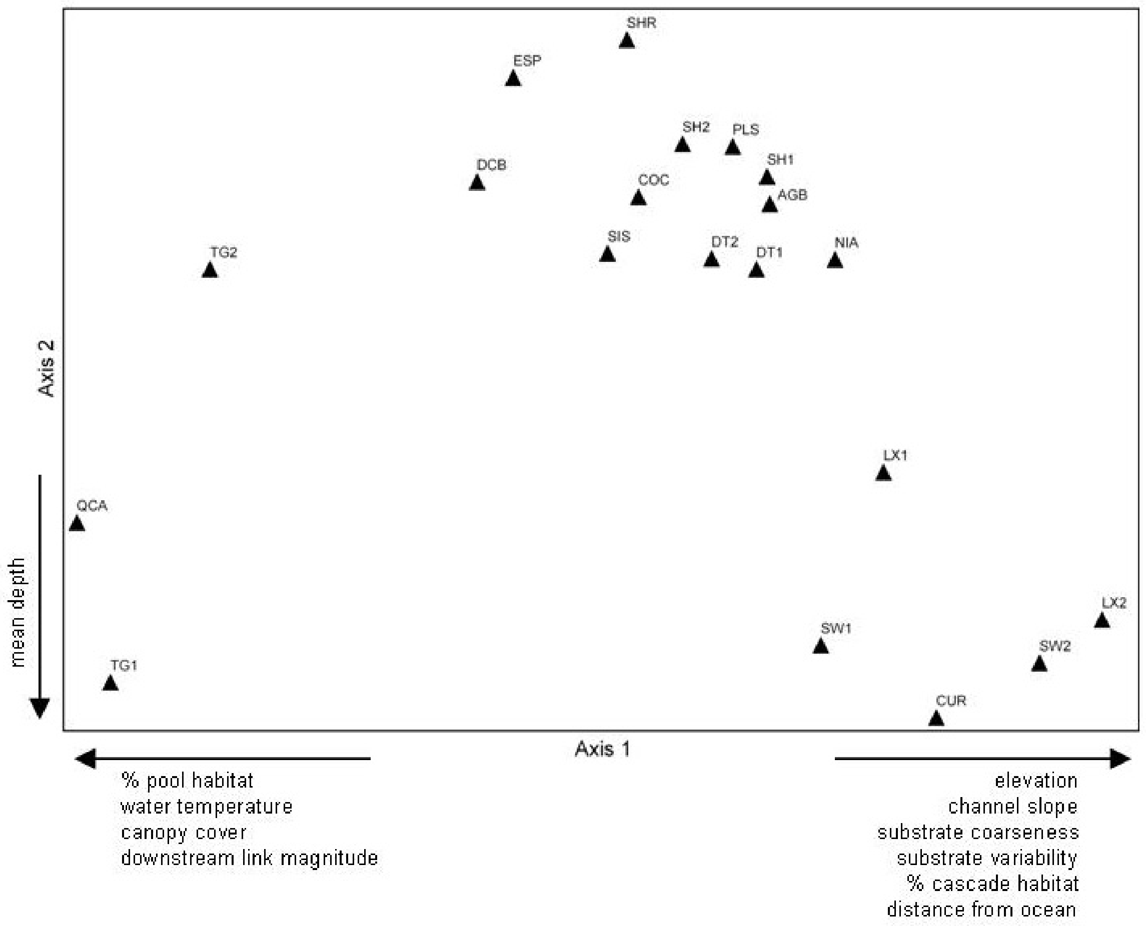

Figure 3. NMS ordination of the 20 sampling sites based on relative abundance of all fish species (final stress = 8.691). Environmental variables that were significantly correlated with NMS axes are shown, with arrows indicating the direction of increasing values for the variables listed (see Table 4 for correlation coefficients).

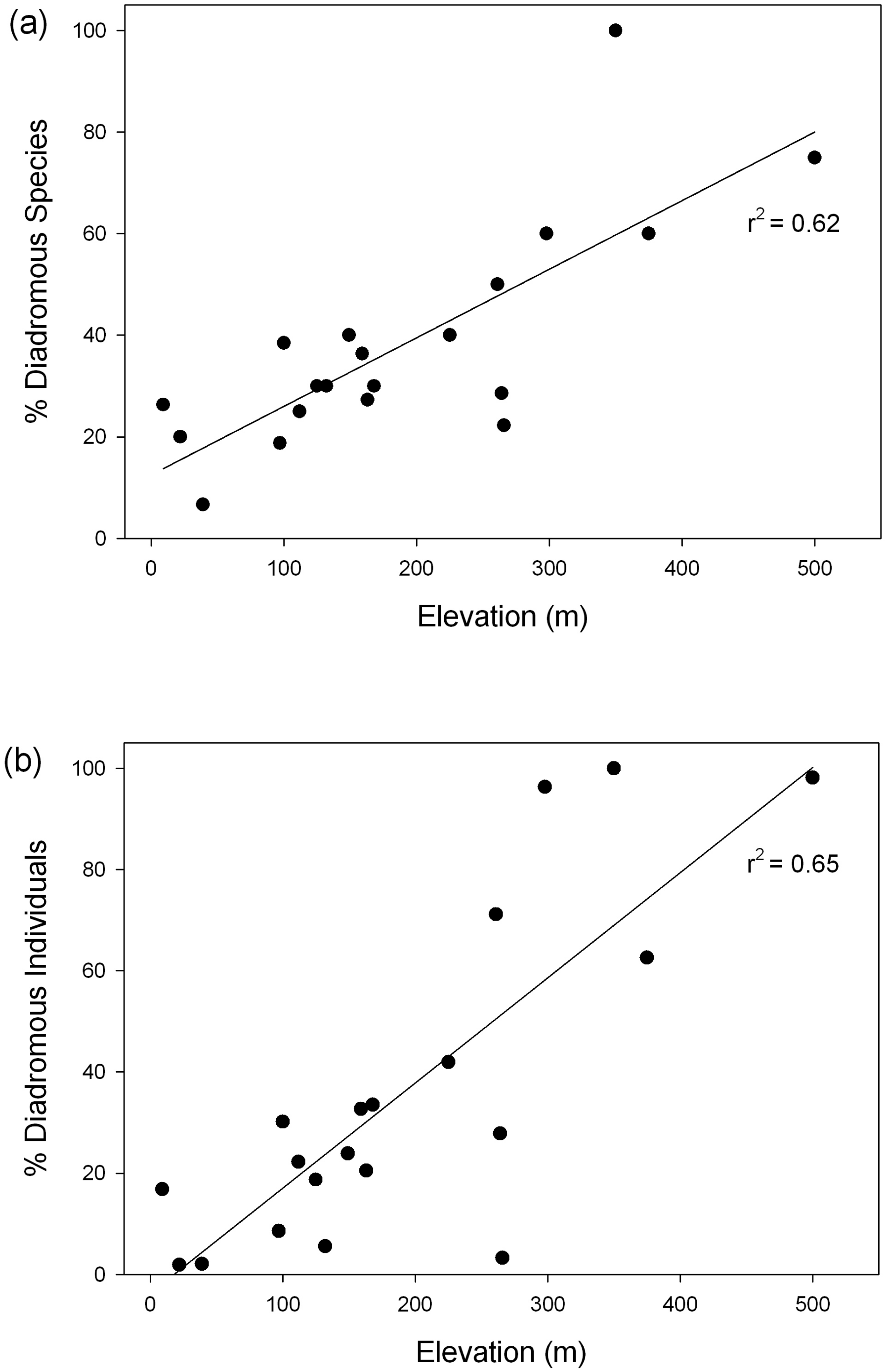

Figure 4. Linear regressions of elevation versus (a) percent diadromous species, and (b) percent diadromous individuals in the 20 study sites ($P < 0.001$ for both regressions).

Chapter 4: Comprehensive conservation planning: developing a framework for planning across terrestrial, freshwater and marine ecosystems.

Abstract

Systematic conservation planning methods are widely used to identify conservation priority areas, and for a variety of reasons have traditionally considered terrestrial, freshwater, and marine ecosystems separately. Many physical and ecological processes link these ecosystems, however, affecting biodiversity in all three realms. These interactions suggest that an integrated planning approach could more effectively conserve biodiversity. Comprehensive conservation planning for terrestrial and aquatic ecosystems poses significant challenges, and a critical evaluation of available methods is currently lacking. Here we discuss the goals of comprehensive conservation planning in general and in the specific context of a planning domain on the Caribbean slope of Costa Rica and Panama, and then review strategies currently being used to develop conservation plans for multiple realms. All of the methods we review can be used to improve integration of planning efforts for terrestrial and aquatic systems, but there is a trade-off between ease of application and the consideration of ecological linkages among realms in the planning process. Although several of the methods we review would be applicable to comprehensive planning in our study site, models needed to incorporate biophysical interactions among realms into conservation planning algorithms remain poorly developed. Given the limited capacity for expanding protected area networks in most regions, and the way that matrix lands affect existing protected areas in multiple realms, there is a great need to develop new comprehensive planning tools that can direct a broader array of conservation interventions.

Introduction

The loss of biodiversity is among the greatest challenges currently facing humanity (Pimm et al 1995, Dirzo and Raven 2003). Species and even entire ecosystems are going extinct at a rate far greater than ever before, in what has been referred to as the 6th extinction crisis (Pimm and Brooks 2000). The biodiversity crisis appears to be particularly severe in freshwater ecosystems (Abell 2002, Malmqvist and Rundle 2002), where rates of extinction and endangerment may be even higher than in terrestrial environments (Ricciardi and Rasmussen 1999, Revenga et al. 2005).

In response, there has been increased attention given to gaps in the current global protected areas network (Rodrigues et al. 2004a), as well as calls for improving conservation on private lands outside of reserves (O'Connell and Noss 1992, Knight 1999). There is continued debate about how much of the planet should be protected (Brooks 2004, Tear et al. 2004), and whether efforts should focus on species (Brooks et al. 2004), ecosystems (Higgins et al. 2004), or ecosystem function (Moss 2000). Nevertheless, the establishment of a representative network of protected areas which covers all biomes and species is a widely recognized target scientifically (Brooks 2004, Rodrigues et al. 2004a, Rodrigues et al. 2004b) and politically (Convention on Biological Diversity 1992).

Systematic conservation planning methods have been developed during the last 20 years in response to the need to move beyond ad-hoc reserve selection and to set measurable targets, strategies and goals for the design of protected areas (Margules and Pressey 2000). These methods are used to identify potential reserve networks that represent the full range of species and ecosystems in a given planning area and are spatially efficient, recognizing the scarcity of resources available for conservation and the need to balance conservation goals with other societal values (Sarkar et al. 2006). Systematic conservation planning methods are now widely used to identify conservation priority areas in both terrestrial and marine systems. Applications in freshwater environments are less common (Sarkar et al. 2006), but several recent examples illustrate how these methods can be adapted to freshwater planning (Abellán et al. 2005, Linke et al. 2007).

For a variety of institutional and logistical reasons, systematic conservation planning for terrestrial, freshwater, and marine realms has usually been conducted separately (Beger et al. In Press). Conservation planning is an inherently complex process and even in the most thoroughly studied areas in the world there is high uncertainty concerning the distribution of biodiversity (Margules et al. 2002) and threats to its persistence (Gaston et al. 2002). For aquatic ecosystems, even the most basic information needed to assess conservation priorities may be scarce (Abell 2002). Planning in different realms requires expertise in different fields of research, and entirely different governmental agencies or non-governmental organizations are often involved in management and conservation of the different ecosystems (Beger et al. In Press). Biogeographical patterns, ecological processes, and the relative importance of different threats to biodiversity also differ among terrestrial, freshwater, and marine ecosystems. As a result, conservation planning in different realms may involve very different considerations (Carr et al. 2003 Dunn 2003, Abell et al. 2007) and methods (Higgins et al. 2005). Together these factors constitute a considerable barrier to development of comprehensive planning approaches for terrestrial and aquatic ecosystems.

Despite these challenges, there is a growing recognition of the limitations of a fragmented approach to conservation planning (Beger et al. In Press). Freshwater, marine, and terrestrial ecosystems are strongly linked by fluxes of water, sediment, nutrients and organisms (e.g. Hynes 1975, Polis et al. 1997, Nakano and Murakami 2001, McDowall 1998), which have a critical influence on biodiversity in all three realms. The relevance of these interactions to conservation is particularly conspicuous for aquatic ecosystems, where many of the most important threats to biodiversity are directly linked to activities in terrestrial landscapes upslope and upstream (Allan 2004, Thrush et al. 2004, Stoms et al. 2005, Burcher et al. 2007). Resource fluxes from aquatic ecosystems can have significant effects on productivity and community composition in terrestrial environments, as well (Nakano and Murakami 2001, Polis et al. 2004, Crait et al. 2007). These interactions are particularly important in transition zones like riparian areas, which also support unique communities (Sabo et al. 2005). Given this interdependence among ecosystems, comprehensive planning approaches that consider functional interactions between terrestrial and aquatic environments are likely to produce more robust conservation assessments (Stoms et al. 2005, Beger et al., In Press).

Recognition of the potential advantages of conducting comprehensive conservation assessments has led to an increasing number of conservation plans that identify priority areas for terrestrial and aquatic biodiversity in a common planning domain. A variety of methods have been used to develop these plans, representing varying levels of integration in the planning process. However, it remains unclear how effectively these methods incorporate interactions among realms, the primary impetus for a comprehensive approach. In addition, different methods may be appropriate in different situations, depending on the availability of data and expertise, the nature of terrestrial-aquatic interactions in the planning area, and the specific goals of the conservation assessment. Unfortunately, there is currently very little information to guide decisions on planning methodology. A critical evaluation of the methods currently being used to integrate terrestrial and aquatic conservation goals could help direct efforts by conservation practitioners and suggest directions for future research.

In this paper, we begin by discussing the goals of comprehensive conservation planning, using a planning domain on the Caribbean slope of Central America as an example. Next, we summarize the strategies that have been used, or proposed, to produce conservation plans that include terrestrial and aquatic ecosystems. Each strategy is then evaluated based on several criteria, including data requirements, methodological difficulty, and the degree to which functional interactions between ecosystems are represented. Based on this evaluation, we present a set of methods that would be most useful for comprehensive planning in our example planning domain, and discuss future developments in conservation planning. Throughout our discussion, we use the term conservation planning to refer to the process of identifying potential reserve networks using systematic reserve selection algorithms and spatial decision support software, although reserve selection is just one part of the conservation planning process (Knight et al. 2006).

Goals of a comprehensive planning approach

Systematic conservation planning aims to identify an array of sites that, if managed properly, could provide for the persistence of the full complement of biodiversity in a given region (Margules and Pressey 2000). In practice, this usually entails selecting a set of priority sites based on the distribution of a few relatively well-studied groups, as well as the occurrence of habitat types that can be identified through remote sensing or unique combinations of biophysical factors like soil, elevation, and precipitation (Margules et al. 2002, Higgins et al. 2005). The extent to which plants and vertebrates, which are most often used as surrogates for biodiversity patterns, represent broader patterns of biodiversity remains an important, and unanswered, question. The ability of habitat types (e.g. land cover types) to represent patterns of biodiversity has been established in some cases, but remains poorly understood in general (Brooks et al. 2004). Despite these problems, the use of biodiversity surrogate groups and habitat types in conservation planning is a pragmatic approach to incorporating the best available information into the planning process (Pressey 2004).

Planning for the persistence of all species and ecosystems, terrestrial and aquatic, in a given area is a logical extension of the systematic planning approach. Ideally, a comprehensive plan would meet several key goals. First, it would identify a network of sites where known biodiversity features in terrestrial and aquatic ecosystems, including individual species and entire communities, are fully represented. In addition, potential reserves would be extensive enough to maintain viable populations of all species and sustain ecological processes (e.g. disturbance regimes) necessary for the long-term persistence of all biodiversity features. These goals have typically been addressed in conservation planning through the size of potential reserves, identifying corridors among these sites, and steering reserve selection away from sites with heavy human impacts (e.g. Noss et al. 1999, Cowling et al. 2003, Mattson and Angermeier 2007). In a comprehensive approach, these factors would still be relevant, but two additional considerations would also be important. First, reserve selection methods should consider spatial relationships between sites in different realms that sustain key biophysical processes (Figure 1). For example, the spatial arrangement of coral reefs and mangroves could influence the abundance and persistence of species that use both ecosystems during their life cycle (Mumby 2006). Second, a comprehensive planning approach would ideally identify a network of conservation priority areas that minimizes threats to biodiversity in all realms, recognizing that threats can move between realms and follow directional flows in aquatic ecosystems.

An example planning domain

The goals of a comprehensive assessment can be illustrated using an example planning domain that includes terrestrial, freshwater and marine ecosystems. Our example is located on the Caribbean slope of Central America in southeastern Costa Rican and northwestern Panama. The terrestrial portion of the planning domain covers approximately 3800 km^2 and encompasses the Estrella and Sixoala River basins and the smaller coastal drainages between them (Figure 2). This area is located within the Mesoamerican biodiversity hotspot (Myers et al. 2000, Mittermeier et al. 2005), and includes portions of the Talamanca montane ecoregion, a Global 200 priority ecoregion (Palminteri et al. 1999, Olson and Dinerstein 2002). The flora and fauna of this area are extremely diverse due to the wide variety of life zones, and to the fact that is it a convergence zone for fauna from North and South America (Graham 1995). The area is also rich in endemic birds, mammals, herpetofauna and plants (Palminteri et al. 1999). The inland backbone of the Talamanca Mountains is drained by a network of rivers that support diverse vertebrate and invertebrate communities, including several freshwater fish species endemic to the region (Bussing 1998). Nearshore marine environments are also included in our planning domain and are exceedingly diverse, with some of the largest coral reefs in Costa Rica (Cortes 1998). Transition zones between ecosystems in this landscape include floodplain lakes and wetlands, riparian forests, estuaries, mangroves, lagoons, and beaches. These interfaces provide critical habitat and breeding grounds for many of the region's rare and threatened species, such as estuarine nurseries for the Atlantic tarpon or beach nesting sites for sea turtles (Wilkinson 2004, Cortez 1998).

Several protected areas, including La Amistad International Park, Chirripò National Park, Hitoy-Cerere Biological Reserve, Cahuita National Park, Gandoca-Manzanillo National Wildlife Refuge, and a number of smaller private reserves have already been established in the region (Boza 1986). The current system of protected areas is extensive (Figure 2), but highly skewed toward to high-elevation montane forests and coastal swamps, leaving many of the most diverse ecosystems in the region underrepresented (Palminteri et al. 1999, Powell et al. 2000). Cahuita National Park and Gandoca-Manzanillo National Wildlife Refuge protect terrestrial and marine environments, and nearly all of the coral reefs in the area. Land tenure outside of protected areas includes private lands owned by small landholders and large corporations, as well as several indigenous reserves that together occupy nearly 30% of the planning domain and form a buffer zone around montane protected areas. Common land uses in indigenous reserves include plantain production, banana and cacao agroforestry systems, and shifting cultivation of basic grains, while intensive monocultures of banana and plantain, cattle pasture, and tourism development predominate on private lands outside reserves. Banana and plantain production is concentrated in the floodplains of the Sixaola and Estrella River systems, where plantations receive year-round pesticide applications of up to 50 kg of active ingredient/ha/yr for export-quality fruit (B. Polidoro et al., unpublished data).

Principal threats to biodiversity in this region include logging (Palminteri et al. 1999), intensive agriculture and agrochemical use (Borge and Castillo 1997, Castillo et al. 2006), illegal hunting and fishing (J. Schipper, unpublished data), and siltation in nearshore marine environments (Cortes and Risk 1985). Coastal development also poses a potential risk to biodiversity in multiple ecosystems, but the severity of this risk is largely unknown. Although most floodplain environments in the area have been severely altered, the terrestrial portion of the planning domain is one of the most intact landscapes in the region. Freshwater and marine ecosystems in the planning domain are also significant on a national level, and so there is an urgent need for strategic conservation planning in the area to ensure the long-term persistence of terrestrial and aquatic biodiversity.

A comprehensive conservation plan for our planning domain would ideally include several key components. First, the plan would identify a suite of sites that could complement the existing reserve network and allow for the persistence of species and ecosystems that are not adequately protected in existing reserves. In practice, this would mean identifying an array of sites at low to mid-elevations that support species and communities that are poorly represented in current protected areas. Ideally, these sites would be selected based on their connectivity with existing protected areas, as well as their importance for seasonal migrations of organisms, dispersal and recruitment processes, and natural disturbance regimes within terrestrial, freshwater, and marine realms. Second, the plan would target transition zones between realms, including floodplains and estuaries, which are particularly important for biodiversity conservation. These habitats could be mapped and considered as unique habitat types to be included in representation goals. Third, an effective comprehensive plan would identify a set of priority sites that minimizes threats to biodiversity throughout the reserve network. This is a particularly important aspect, as existing reserves are threatened to varying degrees by exogenous factors (e.g. sediment deposition on reefs) that threaten to undermine conservation objectives. Finally, a comprehensive plan would ideally promote the maintenance of ecosystem services (Chan et al. 2006) and maximize economic benefits to local communities (Naidoo and Ricketts 2006).

Methods for integrated conservation planning

A review of the conservation literature yielded a variety of conservation plans that use terrestrial and aquatic biodiversity features to identify a set of conservation priority areas (Table 1). These plans used diverse approaches to identify priorities, ranging from heavy dependence on expert opinion to systematic approaches driven by selection algorithms and spatial decision support systems. Methods for targeting terrestrial and aquatic biodiversity features also differed greatly among the plans we reviewed. The examples we present are not an exhaustive list, but were selected to illustrate the range of approaches that have been used. Most of these examples focus on terrestrial and freshwater ecosystems, but several recent conservation plans have also included marine conservation goals (Table 1). Based on our review of these plans and other relevant literature, we identified seven methods for developing comprehensive conservation plans for terrestrial and aquatic realms (Table 2). These approaches vary from post-hoc comparisons of conservation priorities based on independent assessments of freshwater, terrestrial, and/or marine environments in a particular geographic area to increasingly integrated approaches. Below, we evaluate each of these methods based on planning efficiency, ease of application using existing software, data requirements compared with planning for each realm separately, and the extent to which interactions between realms are considered when selecting priority areas (see Table 2).

Method 1. Develop separate conservation plans for terrestrial, freshwater, and/or marine realms and then overlay results.

This is the simplest approach methodologically, and in terms of data requirements. Once separate conservation plans for terrestrial and aquatic realms have been developed, priority areas can easily be overlaid in a geographic information system. This method is particularly well-suited to integration across freshwater and terrestrial realms because of the way that freshwater ecosystems are embedded in terrestrial landscapes. Simple overlays in this case may provide considerable insight into synergies or trade-offs between freshwater and terrestrial conservation priorities. Marine systems are more spatially distinct, but overlays may still be informative for coastal areas and islands, where protected areas could include multiple realms.

Maintaining separate planning processes for terrestrial and aquatic ecosystems may allow for more thorough and relevant assessments of biodiversity priorities in each realm, and does not require that similar data are available for both. However, the effort required to develop completely separate plans makes this a relatively inefficient approach, especially if similar data and software are being used in the planning process. In some areas, conservation assessments for one or more realms already exist, and utilizing this information in an overlay approach could significantly reduce the cost of producing a comprehensive plan. Linkages between realms are very poorly represented with this approach, although assigning higher priority to areas of spatial congruence between realms may capture sites where functional relationships are intact. Using expert opinion to modify priority areas based on overlays can also improve integration across realms.

Method 2. Target terrestrial and aquatic habitat types when selecting conservation priority areas: a coarse-filter approach.

As discussed above, targeting habitat types for protection in conservation planning is a pragmatic approach to deal with uncertainty concerning the distribution of most species. Habitat types (also referred to as land types or ecological systems) are often called coarse-filter targets because they are thought to capture large and poorly known complements of species (Groves et al. 2002). Planning using terrestrial and aquatic habitat types as coarse-filter targets is straightforward methodologically, and is an efficient way to incorporate data from both ecosystems in a single analysis. Furthermore, this approach gives equal weight to terrestrial and aquatic biodiversity through representation of all habitats in the planning domain.

Data requirements for this method may be higher than for conducting separate assessments, primarily because of the need for comparable data across realms. Maps of terrestrial habitats are often readily available or can be produced using remote sensing and other spatial data. Similar methods typically cannot be applied to aquatic systems, and so producing accurate maps of marine and freshwater habitats can be more difficult. Significant progress has recently been made in this area for freshwater systems (Higgins et al. 2005, Thieme et al. 2007), offering new opportunities for integration across realms. Other issues of scale are also relevant, especially in practice. First, the different shapes and spatial extents of ecological systems in different realms can make display very difficult, especially when spatial decision support systems are being used to explore conservation trade-offs. Boundaries between realms are particularly complicated, as they are often temporally dynamic (e.g. floodplains, tidal marshes) and difficult to delineate. Explicitly targeting transition zones for protection could alleviate many of these problems, ensuring that features in both realms, and the ecological processes they support, are adequately represented (Beger et al. In Press). Aside from capturing transition areas, this method does not effectively address linkages among realms.

Method 3. Target terrestrial and aquatic species when selecting conservation priority areas: a fine filter approach.

Although targeting specific habitat types for conservation may capture most species and assemblages, these areas may not provide adequate protection for species that depend on very specific habitats, have limited ranges, or require extraordinarily large areas to maintain viable populations. Targeting these species of special conservation concern provides a fine filter that complements the coarse-filter approach (Groves et al. 2002). The geographic distribution of focal species can be represented spatially as point occurrences or as polygons based on known areas of occupancy, habitat suitability models or expert opinion. The quality of species occurrence data varies widely, and is often heavily biased even in the best-studied areas (Margules et al. 2002, Pressey 2004). Nevertheless, once data have been standardized to a common format, fine-filter targets from different ecosystems can be treated equally in the planning process. Using fine-filter targets from terrestrial and aquatic ecosystems in this way can improve the efficiency of the planning process while using existing data and planning methods.

Like the coarse-filter approach discussed above, using fine-filter targets from terrestrial and aquatic ecosystems does little to address linkages among realms and their influence on biodiversity. Selecting focal species like amphibians, otters, or wading birds that rely on habitats in more than one realm could help, because their ranges may capture transition zones between realms. However, even for these species, there is no guarantee that key spatial relationships between habitats in different realms will be captured with this approach.

Method 4. Use conservation assessment for one realm as a "seed" for identifying priorities in other realms.

When a conservation assessment has already been made for one realm, the selected priority areas can be used to guide reserve selection in other realms. This method has been used to select potential marine reserves based on their spatial congruence with previously selected terrestrial priority areas (e.g. Floberg et al. 2004), and to modify an existing terrestrial conservation assessment so that it better represents freshwater biodiversity features (Abell et al. 2004). This approach is relatively efficient where a systematic conservation assessment for one realm has already been developed, and has minimal extra data requirements compared with planning for a single realm. Using one assessment as a starting point for others is quite similar to locking in existing protected areas when conducting a systematic conservation assessment (e.g. Cowling et al. 2003), and can be accomplished using available software tools. The extent to which ecosystem linkages are represented with this approach depends on how additional conservation priority areas are selected. For example, if spatial congruence with an existing set of sites is emphasized using a boundary length function in the selection algorithm, important functional linkages among ecosystems may be preserved incidentally. Nevertheless, because the initial set of priority areas are locked in without considering linkages with other realms, sites that are most important for maintaining these linkages and minimizing threats to biodiversity may be overlooked.

Method 5. Automatically select transition areas adjacent to aquatic conservation priority areas (e.g. floodplains).

As noted above, ecological processes that operate in transition zones where two realms meet are often vital to maintaining biodiversity in both realms. One alternative for representing these functional relationships in a comprehensive planning approach is to include parts of adjacent ecosystems when selecting priority areas. For example, riparian areas adjacent to streams targeted for protection could automatically be included as priority areas for conservation action. Shorelines adjacent to marine protected areas could also be targeted. This strategy is relatively straightforward to apply and, similar to the explicit mapping of transition zones, may protect unique biological communities and ecological processes that occur in transition zones.

Deciding exactly how much area to protect in the adjacent ecosystem is not as straightforward, and additional spatial data on the occurrence of certain vegetation types or other indicator species may be needed to determine appropriate dimensions for transition areas. Digital elevation models can be used to map geomorphic surfaces and provide information on the spatial extent of floodplains and other transition zones, even in data-poor regions (Thieme et al. 2007). Despite the advantages of this method, aquatic ecosystems are affected by land use and other impacts in their catchments at multiple spatial scales (Allan 2004), and so protecting narrow transition zones alone may not be sufficient to maintain important functional linkages among ecosystems.

Method 6. Use river basin boundaries to define the planning domain and small catchments as planning units

Freshwater ecosystems are strongly linked to the surrounding landscape, and so freshwater protected areas function best when reserves protect entire catchments (Crivelli 2002, Saunders et al. 2002, Abell et al. 2007). Using catchments as planning units, therefore, is an efficient approach to identifying priority areas that are relevant to terrestrial and aquatic conservation goals. Applying this approach is relatively simple, as watersheds can be mapped at multiple spatial scales using digital elevation models to match the target size for planning units. In addition, catchments are naturally nested in a way that facilitates spatial aggregation of planning units into larger reserves. Upstream-downstream connectivity rules can also be used to ensure that priority sub-catchments are not selected downstream from unprotected areas (Linke et al. 2007). Nearshore marine environments are similarly affected by landscapes "upstream" (Stoms et al. 2005), and could benefit from conservation at the catchment scale.

The major drawback to using catchments as the basis for comprehensive conservation planning is the mismatch between terrestrial and freshwater planning domains. Terrestrial ecoregions, the most common planning domain for terrestrial assessments, are rarely congruent with the catchment boundaries that define freshwater biogeographic units (Abell et al. 2002). This creates non-trivial issues of scale for comprehensive assessments, as representation and complementarity criteria depend on how much of a river basin or terrestrial ecoregion is included in the planning domain (e.g. Floberg et al. 2004). This issue is a fundamental challenge that cuts across many of the methods reviewed here, and can only be circumvented by developing separate conservation plans for freshwater and terrestrial ecosystems.

Method 7. Explicitly consider ecological linkages between realms in reserve selection algorithms.

Conservation planning tools that incorporate biophysical interactions between terrestrial and aquatic ecosystems have the potential to produce more realistic conservation assessments and allow for more robust evaluations of the benefits of specific conservation actions (Stoms et al. 2005, Beger et al. In Press). These interactions could include natural processes that contribute to the development and maintenance of biodiversity (e.g. diadromous fish migration, allochthonous resource subsidies among realms), as well as processes that threaten biodiversity (e.g. transport of sediment and pollutants between realms). Beger et al. (In Press) provide a thorough review of these topics, as well as specific methods for targeting a variety of ecological interactions between realms in systematic conservation assessments.

The main limitation of this approach is the difficulty of accurately describing and predicting ecosystem interactions across realms (Stoms et al. 2005). Many of the most important processes can be described qualitatively, but precise quantitative descriptions are much more difficult to develop. In some cases, such detail may not be crucial and relatively simple heuristic rules can be used. This approach has often been used to steer reserve selection away from areas with high human impact, using variables like road density. Accurate, spatially-explicit models describing movement of organisms, water, sediment, nutrients, and contaminants among realms would clearly be superior to qualitative approaches. However, data needed to parameterize these models is often limited or completely absent, particularly in developing regions. Spatial and temporal variation in these processes further complicates modeling and prediction. Finally, even if there was sufficient information to accurately predict how land-use change will affect key biophysical processes operating among realms, linking these models to decision support systems to examine trade-offs among individual sites presents another level of complexity.

Application to the example planning domain

The conservation planning methods reviewed above are not mutually exclusive, and several may be appropriate when developing a comprehensive conservation plan. Here we discuss the applicability and feasibility of each method for our example planning domain. We are not aware of any systematic planning efforts in the study region that have been conducted at a scale relevant to our planning domain, and so building on an existing plan (Method 4) is not an option in this area. The lack of previous planning efforts and high connectivity among ecosystems suggests that conducting separate conservation assessments for terrestrial, freshwater and marine environments (Method 1) would also be an inefficient and ineffective method.

Among the remaining methods, targeting specific habitats and species (Methods 2 and 3) would both be feasible in our study area. Maps of terrestrial habitat types (Kappelle et al. 2002) and nearshore marine ecosystems are available for the planning domain, and freshwater habitats could be mapped using available digital elevation data. In addition, the most important transitional ecosystems could be mapped using existing digital elevation data, aerial photos, and satellite imagery. Occurrence data within the planning domain exist for many terrestrial and aquatic species of conservation concern, including birds and amphibians (www.natureserve.org), mammals (Harvey et al. 2006, J. Schipper, unpublished data), freshwater fishes (Bussing 1998, C. Lorion, unpublished data), and reef-building corals

(Cortes 1998). Available occurrence data may not necessarily represent viable populations because most data are from short-term surveys rather than demographic studies. Nevertheless, targeting known biodiversity features in all realms would be an essential component of a comprehensive planning approach, and priority areas could be updated as new data became available. Boundary length constraints could also be used during reserve selection to improve connectivity within the reserve network, which could increase the long-term viability of small, fragmented populations and species with large area requirements.

Targeting specific species and ecosystems would not adequately address linkages between ecosystems and their influence on biodiversity, and so other methods would also be needed to produce an effective comprehensive plan. Including transition areas adjacent to freshwater and marine protected areas (Method 5) could help maintain functional relationships among realms, but may be redundant if transition zones are targeted directly. Furthermore, transition areas may already be protected by law (e.g. Costa Rican law prohibits clearing streamside forests). The limitations of protecting transition zones are evident in Cahuita National Park, where terrestrial and freshwater systems immediately adjacent to the marine environment are protected, but reef systems continue to be affected by sediment and contaminants from outside the protected area (Cortes and Risk 1985, Cortes 1998).

Using a catchment-based approach to define conservation priority areas (Method 6) is most likely to reduce threats to aquatic ecosystems, and explains why we defined our study area by watersheds as opposed to political boundaries. Small sub-catchments 10-20 km^2 in size could be mapped as planning units for our study area using readily available digital elevation data. The major problem with this approach is the lack of spatial congruence between terrestrial ecoregions and watershed boundaries in the study region. All major river basins in the region are divided by terrestrial ecoregion boundaries, with higher elevation areas on both slopes of the Talamanca Mountains located within the Talamanca montane ecoregion, and lower elevation areas defined by different terrestrial ecoregions (Palminteri et al.1999). This arrangement makes it difficult to assess how terrestrial conservation priority areas fit into ecoregional-scale representation of species and ecosystems, but cannot be avoided if catchments are used as the basis for planning.

Finally, effective conservation in our planning domain will require the maintenance of key ecosystem processes operating between terrestrial, freshwater, and marine ecosystems, as well as mitigation of threats that move among these realms. Specifically targeting ecological processes for protection using reserve selection algorithms (Method 7) could be done in several ways. First, as mentioned above, transition areas like riparian zones and shorelines could be mapped as distinct features and linked to protected areas in adjacent realms using boundary length constraints. Longer connections among reserves, such as migratory pathways for diadromous fishes, could also be identified and targeted using available reserve selection algorithms (Beger et al. In Press). Concerning threats, simple heuristic rules could be used to prioritize potential terrestrial reserves based on their potential to affect aquatic ecosystems downstream either positively or negatively (Stoms et al. 2005).

Integrating reserve selection algorithms with process models describing the flow of water, nutrients, sediment, and contaminants could provide a much clearer picture of trade-offs between different potential reserves. However, data to parameterize these models are generally lacking for the study region. Even though sedimentation and contamination are known to threaten numerous aquatic species and ecosystems in Costa Rica and other tropical coastal landscapes (Cortes and Risk 1985, Peters et al. 1997, Wishnie and Socha 2003, Castillo et al. 2006), their rates are difficult to predict and often exhibit high natural variability (Krishnaswamy et al. 2001).

Conclusions

A comprehensive approach to conservation planning that identifies priority areas based on biodiversity patterns in terrestrial and aquatic ecosystems, as well as biophysical interactions between these realms, has great potential to produce more effective reserve networks. However, realizing this potential will require the development of new tools for assessing exactly how reserve selection will affect biodiversity in all realms. Such tools could also be used to inform decisions about a broader array of conservation interventions, including habitat restoration and application of best management practices on private lands. These interventions would be particularly important in heavily modified landscapes where conservation opportunities are limited. Even in a relatively intact landscape like our example planning domain, the capacity for setting aside more protected areas is severely limited by land tenure and land use patterns and the resources available for conservation. Furthermore, setting aside more protected areas in this landscape may do less to ameliorate threats to aquatic biodiversity than improving land management practices and simply enforcing current laws.

There is good evidence that agricultural land uses in this domain vary greatly in their capacity to support native biodiversity (Harvey et al. 2006, Harvey and Gonzalez 2007), but the effects of different land uses on biodiversity in freshwater and marine ecosystems remain very poorly understood. Incorporating trade-offs between reserve designation, land-use practices, and biophysical processes into a comprehensive planning framework may seem hopelessly complex, but it may be the only way to find conservation solutions that best serve society. Private lands will always contribute at some level to conservation, and when these values are not included in the planning process, optimal solutions to complex planning problems may be missed (Faith and Walker 2002, Polasky et al. 2005).

The need to look beyond strict reserves in comprehensive planning approaches is particularly clear when considering freshwater conservation goals. Although strict protection of entire catchments is the ideal approach for freshwater biodiversity conservation, this strategy is infeasible for most river systems. Thus, effective conservation of freshwater biodiversity will require a broader perspective on protected areas that includes areas specifically managed to meet conservation goals while providing other economic opportunities (Abell et al. 2007). Expanding terrestrial reserve networks may help to achieve this goal, but eliminating destructive land uses is just as important. Ecological risk assessment provides a ready framework for identifying sites that pose the greatest threat to biodiversity, and incorporating these methods into the planning process could significantly advance comprehensive assessments. The challenge is to link these models with conservation planning algorithms in decision support systems to explore trade-offs and search for optimal solutions. Conservation planning has a long way to go to achieve these goals, but development of comprehensive planning techniques based on a broader perspective of conservation action may significantly improve our ability to conserve biodiversity in all realms.

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References

Abell, R. 2002. Conservation biology for the biodiversity crisis: a freshwater follow-up. Conservation Biology 16:1435-1437.

Abell, R., J. D. Allan, and B. Lehner. 2007. Unlocking the potential of protected areas for freshwaters. Biological Conservation 134:48-63.

Abell, R., M. Thieme, E. Dinerstein, and D. Olson. 2002. A sourcebook for conducting biological assessments and developing biodiversity visions for ecoregion conservation. Volume II: freshwater ecoregions. World Wildlife Fund, Washington, D.C., USA.

Abell, R., M. Thieme, B. Lehner, G. Powell, S. Palminteri, J.-C. Riveros, and S. Hamilton. 2004. Southwest Amazon freshwater-terrestrial integration. World Wildlife Fund, Washington, D.C., USA.

Abellán, P., D. Sánchez-Fernández, J. Velasco, and A. Millán. 2005. Conservation of freshwater biodiversity: a comparison of different area selection methods. Biodiversity and Conservation 14:3457–3474.

Allan, J. D. 2004. Landscapes and riverscapes: the influence of land use on stream ecosystems. Annual Review of Ecology and Systematics 35:257-284.

Beger, M., H. Grantham, R. Pressey, K. Wilson, E. Peterson, D. Dorfman, P. Mumby, R. Lourival, D. Brumbaugh, and H. Possingham. In Press. Conservation planning for processes connecting marine, freshwater, and terrestrial realms. Ecology Letters.

Borge, C., and R. Castillo. 1997. Cultura y conservación en la Talamanca indígena. Editorial Universidad Estatal a Distancia, San José, Costa Rica.

Boza, M. 1986. Parques Nacionales: Costa Rica. Incafo, S.A., Madrid, Spain.

Brooks T. M. 2004. Coverage provided by the global protected-area system: Is it enough? BioScience. 54:1081–1091

Brooks T. M., G. A. B. Fonseca, and A. S. L. Rodrigues. 2004. Species, data, and conservation planning. Conservation Biology 18:1682-1688.

Burcher, C. L., H. M. Valett, and E. F. Benfield. 2007. The land-cover cascade: relationships coupling land and water. Ecology 88:228-242.

Bussing, W. A. 1998. Freshwater fishes of Costa Rica. Editorial de la Universidad de Costa Rica, San José, Costa Rica.

Carr, M. H., J. E. Neigel, J. A. Estes, S. Andelman, R. R. Warner, and J. L. Largier. 2003. Comparing marine and terrestrial ecosystems: implications for the design of coastal marine reserves. Ecological Applications 13:S90-S107.

Castillo, L. E., E. Martinez, C. Ruepert, C. Savage, M. Gilek, M. Pinnock, and E. Solis. 2006. Water quality and macroinvertebrate community response following pesticide applications in a banana plantation, Limon, Costa Rica. Science of the Total Environment. 367:418-432.

Chan K. M. A., M. R. Shaw, D. R. Cameron, E. C. Underwood, and G. C. Daily. 2006. Conservation planning for ecosystem services. PLOS Biology 4:2138-2152.

Cortes, J., and M. Risk. 1985. A reef under siltation stress: Cahuita, Costa Rica. Bulletin of Marine Science 36:339-356.

Cortes, J. 1998. Cahuita and Laguna Gondoca, Costa Rica. In B. Kjerfve, B., ed. CARICOMP Carribbean Coral Reef, Seagrass and Mangrove Sites. UNESCO, France.

Cowling, R. M., R. L. Pressey, M. Rouget, and A. T. Lombard. 2003. A conservation plan for a global biodiversity hotspot – the Cape Floristic Region, South Africa. Biological Conservation 112:191-216.

Crait, J. R., and M. Ben-David. 2007. Effects of river otter activity on terrestrial plants in trophically altered Yellowstone Lake. Ecology 88:1040-1052.

Crivelli, A. J. 2002. The role of protected areas in freshwater fish conservation. Pages 373-388 in M. J. Collares-Pereira, M. M. Coelho, and I. G. Cowx, eds. Conservation of freshwater fishes: options for the future. Blackwell Science Ltd., Oxford, UK.

Dirzo, R., and P. H. Raven. 2003. Global state of biodiversity and loss. Annual Review of Environment and Resources 28:137-167.

Dunn, H. 2003. Can conservation assessment criteria developed for terrestrial systems by applied to riverine systems? Aquatic Ecosystem Health & Management 6:81-95.

Faith, D. P., and P. A. Walker. 2002. The role of trade-offs in biodiversity conservation planning: linking local management, regional planning, and global conservation efforts. Journal of Biosciences 27:393-407.

Floberg, J., M. Goering, G. Wilhere, C. MacDonald, C. Chappell, C. Rumsey, Z. Ferdana, A. Holt, P. Skidmore, T. Horsman, E. Alverson, C. Tanner, M. Bryer, P. Iachetti, A. Harcombe, B. McDonald, T. Cook, M. Summers, D. Rolph. 2004. Willamette Valley-Puget Trough-Georgia Basin Ecoregional Assessment, Volume One: Report. Prepared by The Nature Conservancy with support from the Nature Conservancy of Canada, Washington Department of Fish and Wildlife, Washington Department of Natural Resources (Natural Heritage and Nearshore Habitat programs), Oregon State Natural Heritage Information Center and the British Columbia Conservation Data Centre.

Gaston, K. J., R. L. Pressey, and C. R. Margules. 2002. Persistence and vulnerability: retaining biodiversity in the landscape and in protected areas. Journal of Biosciences 27:361–384.

Graham, A. 1995. Development of affinities between Mexico/Central America and northern South American lowlands and lower montane vegetation during the Tertiary. Pages 11-22. in S. P. Churchill, H. Balslev, E. Forero, and J. L. Luteyn, eds. Biodiversity and Conservation of Neotropical Montane Forests. The New York Botanical Gardens, New York, New York, USA.

Groves, C. R., D. B. Jensen, L. L. Valutis, K. H. Redford, M. L. Shaffer, J. M. Scout, J. V. Baumgartner, J. V. Higgins, M. W. Beck, and M. G. Anderson. 2002. Planning for biodiversity conservation: putting conservation science into practice. BioScience 52:499-512.

Harvey, C. A., and J. Gonzalez. 2007. Agroforestry systems conserve species-rich but modified assemblages of tropical birds and bats. Biodiversity and Conservation 16:2257-2292.

Harvey, C. A., J. Gonzalez, and E. Somarriba. 2006. Dung beetle and terrestrial mammal diversity in forests, indigenous agroforestry systems and plantain monocultures in Talamanca, Costa Rica. Biodiversity and Conservation 15:555-585.

Higgins, J. V., M. T. Bryer, M. L. Khoury, and T. W. Fitzhugh. 2005. A freshwater classification approach for biodiversity conservation planning. Conservation Biology 19:432-445.

Higgins J. V., T. H. Ricketts, J. D. Parrish, E. Dinerstein, G. V. N. Powell. 2004. Beyond Noah: saving species is not enough. Conservation Biology 18:1672-1673.

Huggins, A. E., S. Keel, P. Kramer, F. Núñez, S. Schill, R. Jeo, A. Chatwin, K. Thurlow, M. McPearson, M. Libby, R. Tingey, M. Palmer and R. Seybert. 2007. Biodiversity Conservation Assessment of the Insular Caribbean Using the Caribbean Decision Support System, Technical Report. The Nature Conservancy, USA.

Hynes, H. B. N. 1975. The stream and its valley. Verhandlungen Internationale Vereinigung fur Theoretische und Angewandte Limnologie 19:1-15.

Kappelle, M., M. Castro, H. Acevedo, P. Cordero, L. González, E. Méndez & H. Monge. 2002. A rapid method in ecosystem mapping and monitoring as a tool for managing Costa Rican ecosystem health. In D.J. Rapport, W.L. Lasley, D.E. Rolston, N.O. Nielsen, C.O. Qualset, and A.B. Damania, eds. Managing for Healthy Ecosystems. Lewis Publishers, Boca Raton, Florida, USA.

Knight R. L. 1999. Private lands: neglected geography. Conservation Biology 13:223-224.

Knight, R. L., R. M. Cowling, and B. M. Campbell. 2006. An operational model for implementing conservation action. Conservation Biology 20:408-419.

Krishnaswamy, J., P. N. Halpin, and D. D. Richter. 2001. Dynamics of sediment discharge in relation to land-use and hydroclimatology in a humid tropical watershed in Costa Rica. Journal of Hydrology 253:91-109.

Linke, S., R. L. Pressey, R. C. Bailey, and R. H. Norris. 2007. Management options for river conservation planning: condition and conservation re-visited. Freshwater Biology 52:918-938.

Malmqvist, B., and S. Rundle. 2002. Threats to the running water ecosystems of the world. Environmental Conservation 29: 134-153.

Margules, C. R., and R. L. Pressey. 2000. Systematic conservation planning. Nature 405:243-253.

Margules, C. R., R. L. Pressey, and P. H. Williams. 2002. Representing biodiversity: data and procedures for identifying priority areas for conservation. Journal of Biosciences 27:309–326.

Marshall, R. M., D. Turner, A. Gondor, D. Gori, C. Enquist, G. Luna, R. Paredes Aguilar, S. Anderson, S. Schwartz, C. Watts, E. Lopez, and P. Comer. 2004. An ecological analysis of conservation priorities in the Apache Highlands Ecoregion. Prepared by the Nature

Conservancy of Arizona and the Instituto del Medio Ambiente y el Desarollo Sustentable del Estado de Sonora.

Mattson, K. M., and P. L. Angermeier. 2007. Integrating human impacts and ecological integrity into a risk-based protocol for conservation planning. Environmental Management 39:125-138.

McDowall, R. M. 1988. Diadromy in fishes: migrations between freshwater and marine environments. Croom Helm, London, UK.

Mittermeier, R. A., P. R. Gil, M. Hoffman, J. Pilgrim, T. Brooks, C. G. Mittermeier, J. Lamoreux, and G. A. B. da Fonseca. 2005. Hotspots revisited: earth's biologically richest and most endangered terrestrial ecoregions. University of Chicago Press, Chicago, Illinois, USA

Moss, B. 2000. Biodiversity in fresh waters – an issue of species preservation or system functioning? Environmental Conservation 27: 1-4.

Mumby, P. J. 2006. Connectivity of reef fishes between mangroves and coral reefs: algorithms for the design of marine reserves at seascape scales. Biological Conservation 128:215-222.

Myers, N., R. A. Mittermeier, C. G. Mittermeier, G. A. B. da Fonseca, and J. Kent. 2000. Biodiversity hotspots for conservation priorities. Nature 403:853-858.

Naidoo, R., and T. H. Ricketts. 2006. Mapping the economic costs and benefits of conservation. PLOS Biology 4:2153-2164.

Nakano, S., and M. Murakami. 2001. Reciprocal subsidies: dynamic interdependence between terrestrial and aquatic food webs. Proceedings of the National Academy of Science of the USA 98: 166-170

Noss, R. F., J. R. Strittholt, K. Vance-Borland, C. Carroll, and P. Frost. 1999. A conservation plan for the Klamath-Siskiyou Ecoregion. Natural Areas Journal 19:392-411.

Noss, R.F. C. Carroll, K. Vance-Borland, and G. Wuerthner. 2002. A multicriteria assessment of the irreplaceability and vulnerability of sites in the Greater Yellowstone Ecosystem. Conservation Biology 16:895-908.

O'Connell M. A. O., and R. F. Noss. 1992. Private land management for biological diversity. Environmental Management 16:435-450

Olsen, D. M., and E. Dinerstein. 2002. The Global 200: priority ecoregions for global conservation. Annals of the Missouri Botanical Garden 89:199-224.

Palminteri, S., G. Powell, A. Fernandéz, and D. Tovar. 1999. Talamanca Montane-Isthmian Pacific Ecoregion-Based conservation plan: Preliminary reconnaissance phase. Tropical Science Center, San José, Costa Rica.

Peters, E. C., N. J. Gassman, J. C. Firman, R. H. Richmond, and E. A. Power. 1997. Ecotoxicology of tropical marine ecosystems. Environmental Toxicology and Chemistry 16:12-40.

Pimm, S. L. and T. M. Brooks. 2000 The Sixth Extinction: How large, how soon, and where? Pages 46-62 in P. Raven, ed. Nature and Human Society: the quest for a sustainable world. National Academy Press, Washington, D.C., USA.

Pimm, S. L., G. J. Russell, J. L. Gittleman, and T. M. Brooks. 1995. The future of biodiversity. Science 269:347-350.

Polasky, S., E. Nelson, E. Lonsdorf, P. Fackler, and A. Starfield. 2005. Conserving species in a working landscape: land use with biological and economic objectives. Ecological Applications 15:1387-1401.

Polis, G. A., W. B. Anderson, and R. D. Holt. 1997. Toward an integration of landscape and food web ecology: the dynamics of spatially subsidized food webs. Annual Review of Ecology and Systematics 28:289-316.

Polis, G. A., F. Sánchez-Piñero, P. T. Stapp, W. B. Anderson, and M. D. Rose. 2004. Trophic flows from water to land: marine input affects food webs of islands and coastal ecosystems worldwide. Pages 200-216 in G. A. Polis, M. E. Power, and G. R. Huxel, eds. Food webs at the landscape level. University of Chicago Press, Chicago, Illinois, USA.

Powell, G. V. N., J. Barborak, and M. Rodriguez S. 2000. Assessing representativeness of protected natural areas in Costa Rica for conserving biodiversity: a preliminary gap analysis. Biological Conservation 93:35-41.

Pressey, R. L. 2004. Conservation planning and biodiversity: assembling the best data for the job. Conservation Biology 18:1677-1681.

Revenga, C., I. Campbell, R. Abell, P. de Villiers, and M. Bryer. 2005. Prospects for monitoring freshwater ecosystems towards the 2010 targets. Philosophical Transactions of the Royal Society B 360:397-413.

Ricciardi, A., and J. B. Rasmussen. 1999. Extinction rates of North American freshwater fauna. Conservation Biology 13:1220-1222.

Rodrigues, A. S. L., S. J. Andelman, M. I. Bakarr, L. Boitani, T. M. Brooks, R. M. Cowling, L. D. C. Fishpool, G. A. B. Fonseca, K. J. Gaston, M. Hoffmann, J. S. Long, P. A. Marquet, J. D. Pilgrim, R. L. Pressey, J. Schipper, W. Sechrest, S. N. Stuart, L.G. Underhill, R. W. Waller, M. E. J. Watts & X. Yan. 2004a. Effectiveness of the global protected area network in representing species diversity. Nature 428:640-643.

Rodrigues, A. S. L., H. R. Akçakaya, S. J. Andelman, M. I. Bakarr, L. Boitani, T. M. Brooks, J. S. Chanson, L. D. C. Fishpool, G. A. B. Fonseca, K. J. Gaston, M. Hoffmann, P. A. Marquet, J. D. Pilgrim, R. L. Pressey, J. Schipper, W. Sechrest, S. N. Stuart, L. G. Underhill, R. W. Waller, M. E. J. Watts & X. Yan. 2004b. Global gap analysis: Priority regions for expanding the global protected-area network. BioScience 54:1092-1100.

Sabo, J. L., R. Sponseller, M. Dixon, K. Gade, T. Harms, J. Heffernan, A. Jani, G. Katz, C. Soykan, J. Watts, and J. Welter. 2005. Riparian zones increase regional species richness by harboring different, not more, species. Ecology 86:56-62.

Sarkar, S., R. L. Pressey, D. P. Faith, C. R. Margules, T. Fuller, D. M. Stoms, A. Moffett, K. A. Wilson, K. J. Williams, P. H. Williams, and S. Andelman. 2006. Biodiversity conservation planning tools: present status and challenges for the future. Annual Review of Environment and Resources 31:123-159.

Saunders, D. L., J. J. Meeuwig, and A. C. Vincent. 2002. Freshwater protected areas: strategies for conservation. Conservation Biology 16:30-41.

Schoen, J., and D. Albert. 2007. Southeastern Alaska conservation strategy: a conceptual approach. In J. Schoen and E. Dovichin, eds. The coastal forests and mountains ecoregions of Southeastern Alaska and the Tongass National Forest: a conservation assessment and resource synthesis. Audubon Alaska and The Nature Conservancy, Anchorage, Alaska, USA.

Stoms, D. M., F. W. Davis, S. J. Andelman, M. H. Carr, S. D. Gaines, B. S. Halpern, R. Hoenicke, S. G. Leibowitz, A. Leydecker, E. M. P. Madin, H. Tallis, and R. R.Warner. 2005. Integrated coastal reserve planning: making the land-sea connection. Frontiers in Ecology and the Environment 3:429-436

Tear T. H., P. Kareiva, P. L. Angermeier, P. Comer, B. Czech, R. Kautz, L. Landon, D. Mehlman, K. Murphy, M. Ruckelshaus, J. M. Scott, and G. Wilhere. 2005. How much is enough? The recurrent problem of setting measurable objectives in conservation. BioScience 55:835-849.

The Nature Conservancy. 2000. Middle Rockies-Blue Mountains Ecoregional Conservation Plan. Prepared by the Middle Rockies-Blue Mountains Planning Team.

The Nature Conservancy. 2001. California North Coast Ecoregional Plan. Prepared by the The Nature Conservancy of California.

The Nature Conservancy. 2002. Chesapeake Bay Lowlands Ecoregional Plan. Prepared by the Chesapeake Bay Lowlands Ecoregional Planning Team.

The Nature Conservancy. 2003a. Cook Inlet Basin Ecoregional Assessment. Prepared by The Nature Conservancy of Alaska.

The Nature Conservancy. 2003b. The West Gulf Coastal Plain Ecoregional Conservation Plan. West Gulf Coastal Plain Ecoregional Planning Team, The Nature Conservancy, San Antonio, Texas, USA.

The Nature Conservancy. 2004. A blueprint for conserving the biodiversity of the Federated States of Micronesia. The Nature Conservancy, USA.

Thieme, M., B. Lehner, R. Abell, S. K. Hamilton, J. Kellndorfer, G. Powell, and J. C. Riveros. 2007. Freshwater conservation planning in data-poor areas: an example from a remote Amazonian basin (Madre de Dios River, Peru and Bolivia). Biological Conservation 135:484-501.

Thrush, S. F., J. E. Hewitt, V. J. Cummings, J. I. Ellis, C. Hatton, A. Lohrer, and A. Norkko. 2004. Muddy waters: elevating sediment input to coastal and estuarine habitats. Frontiers in Ecology and the Environment 2:299-306.

Weitzell, R. E, M. L. Khoury, P. Gagnon, B. Schreurs, D. Grossman, and J. Higgins. 2003. Conservation Priorities for Freshwater Biodiversity in the Upper Mississippi River Basin. Nature Serve and The Nature Conservancy.

Wilkinson, C. 2004. Status of Coral Reefs of the World: 2004 Vol. 2. Australian Institute of Marine Science, Townsville, Australia.

Wishnie, M. H., and G. Socha. 2003. Watershed management in the Pacific Slope Buffer Zone of the La Amistad Biosphere Reserve, Costa Rica. Journal of Sustainable Forestry 16:65-102.

WWF. 2006. A Vision for Biodiversity Conservation in Central Africa: Biological Priorities for Conservation in the Guinean-Congolian Forest and Freshwater Region.WWF-US/Central African Regional Program Office, Washington, D.C., USA.

Table 1. Examples of conservation assessments that include targets and/or identify priority areas for terrestrial and aquatic ecosystems, arranged in chronological order.

	Author and date	**Geographic Area**	**Terrestrial**	**Freshwater**	**Marine**	**Reserve Selection Software**
1	Noss et al. 1999	Klamath-Siskiyou Ecoregion, USA	x	x		None[a]
2	TNC 2000	Middle Rockies-Blue Mountains Ecoregion, USA	x	x		SITES
3	TNC 2001	North Coast California Ecoregion, USA	x	x		None[a]
4	Noss et al. 2002	Greater Yellowstone Ecosystem, USA	x	x		SITES
5	TNC 2002	Chesapeake Bay Lowlands Ecoregion, USA	x	x	x	None[b]
6	Cowling et al. 2003	Cape Floristic Province, South Africa	x	x		C-PLAN
8	TNC 2003a	Cook Inlet Basin Ecoregion, USA	x	x	x	SITES
7	TNC 2003a	West Gulf Coastal Plain Ecoregion, USA	x	x		None[b]
9	Weitzell et al. 2003	Upper Mississippi River Basin, USA	x	x		None[b]
10	Floberg et al. 2004	Willamette Valley-Puget Trough-Georgia Basin Ecoregion, North America	x	x	x	SITES
11	Marshall et al. 2004	Apache Highlands Ecoregion, USA	x	x		SITES
12	TNC 2004	Federated States of Micronesia	x	x	x	None[b]
13	WWF 2006	Guinean-Congolian Region, Africa	x	x		None[b]
14	Huggins et al. 2007	Insular Caribbean	x	x	x	MARXAN
15	Schoen and Albert 2007	Southeast Alaska, USA	x	x		MARXAN

[a] priority areas selected based on number of targets and suitability criteria

[b] conservation portfolio designed based on expert opinion

Table 2. Methods for developing comprehensive conservation plans across terrestrial and aquatic ecosystems, with references that discuss or apply each approach.

Planning Method	References	Planning Efficiency	Technical Difficulty	Data Requirements	Ecosystem Linkages
1. Develop separate conservation plans for terrestrial and aquatic ecosystems and then overlay results	TNC 2002, Weitzell et al. 2003 TNC 2003b	Low	Low	Low	Low
2. Target terrestrial and aquatic habitat types when selecting conservation priority areas	TNC 2000, TNC 2001, Noss et al. 2002 TNC 2003a, Huggins et al. 2007	Moderate	Low	Moderate	Low
3. Target terrestrial and aquatic species when selecting conservation priority areas	TNC 2000, TNC 2001, Noss et al. 2002 Cowling et al. 2003, TNC 2003a	Moderate	Low	Low	Low
4. Use conservation assessment for one realm as a "seed" for identifying priorities in other realms	Abell et al. 2004, Floberg et al. 2004	Moderate	Low	Low	Moderate
5. Automatically select transition areas adjacent to aquatic conservation priority areas (e.g. floodplains)	TNC 2003b	Moderate	Low	Low	Moderate
6. Use river basin boundaries to define planning domain and small catchments as planning units	TNC 2000, TNC 2001 Noss et al. 2002	High	Low	Low	Moderate
7. Explicitly consider ecological linkages between ecosystems in reserve selection algorithms	Stoms et al. 2005, Beger et al. In Press	High	High	High	High

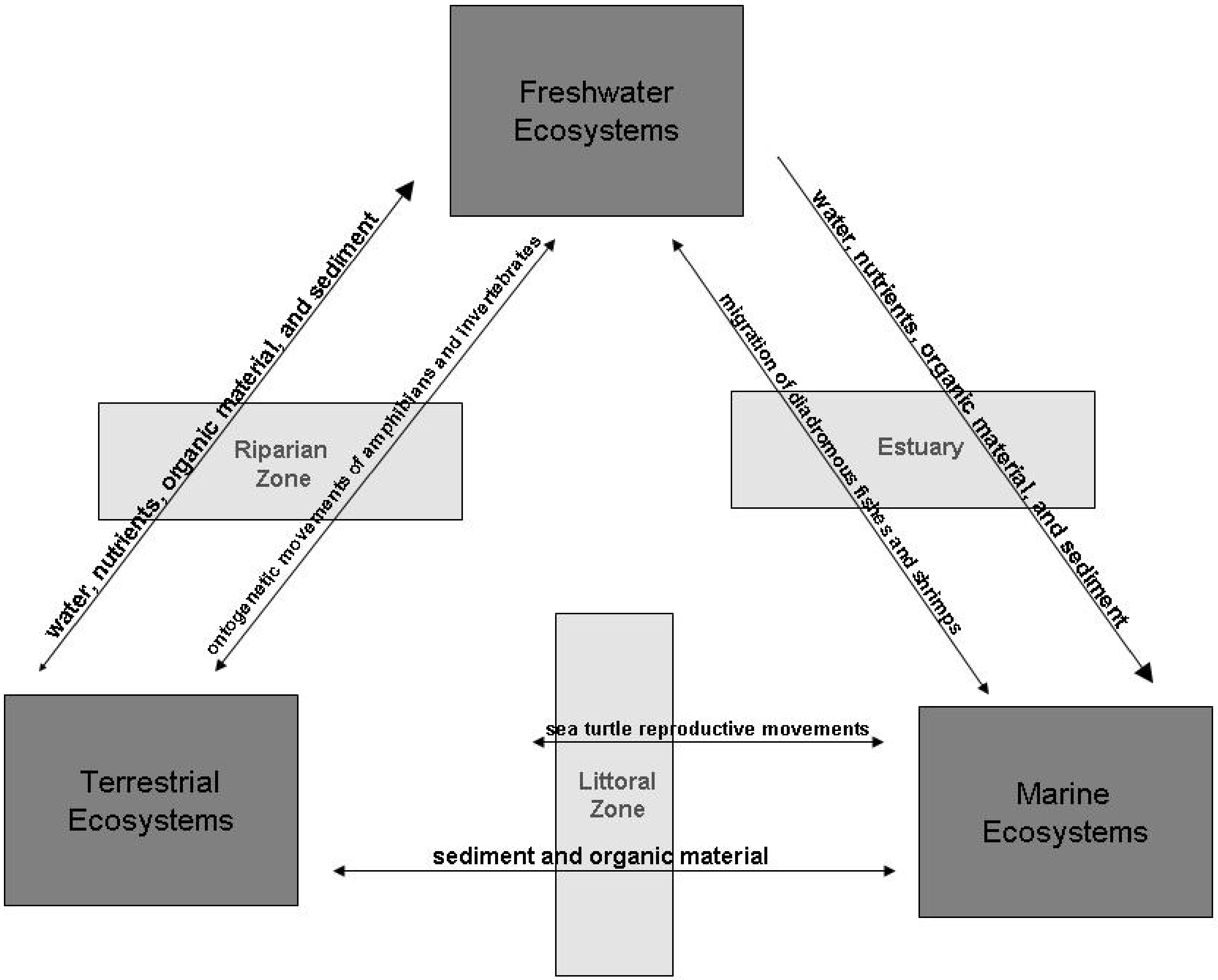

Figure 1. Examples of biotic and abiotic linkages among terrestrial, freshwater, and marine ecosystems.

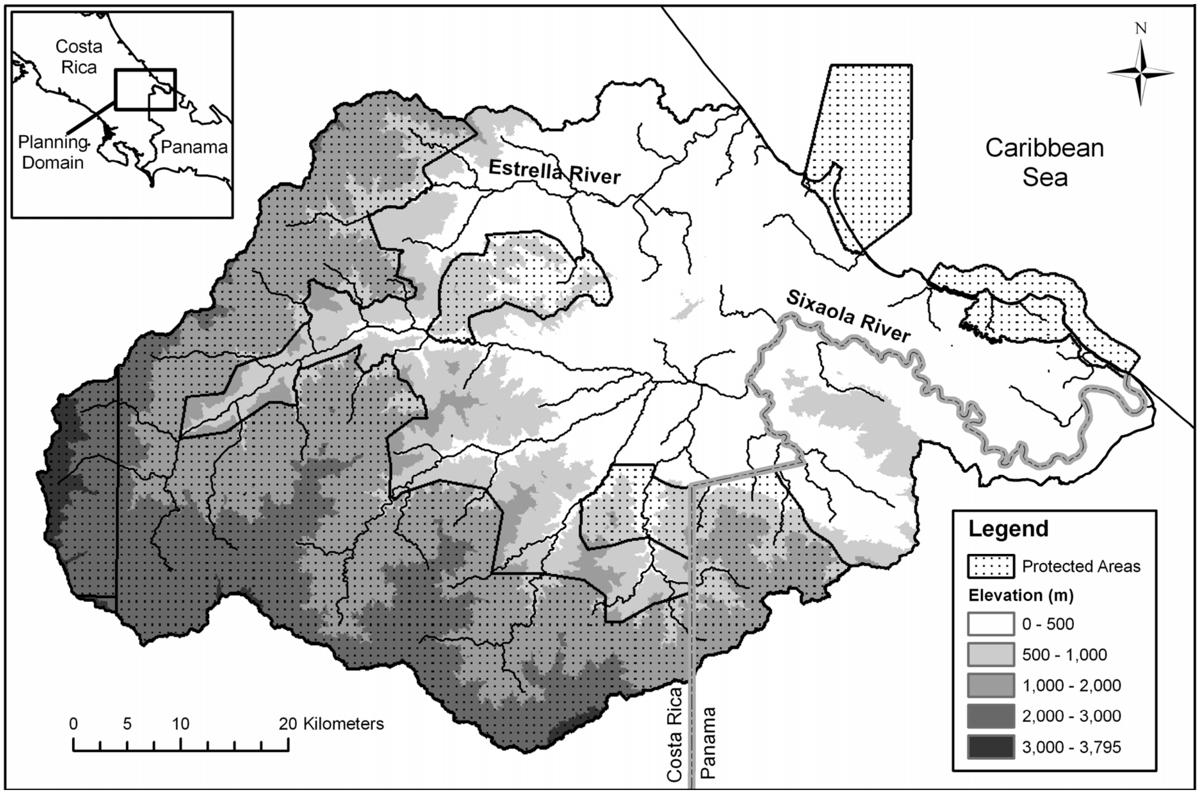

Figure 2. Map of an example planning domain on the Caribbean slope of Costa Rica and Panama.