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Centro Interdisciplinario de Investigación para el Desarrollo Integral Regional Unidad Oaxaca.

Maestría en Ciencias en Conservación y Aprovechamiento de Recursos Naturales

(Patrones y Procesos de la Biodiversidad del Neotrópico)

IMPACTO DE LA PERTURBACIÓN HUMANA EN ARROYOS DE BOSQUE MESÓFILO DE MONTAÑA SOBRE ANUROS NATIVOS EN SANTA CRUZ TEPETOTUTLA OAXACA

Tesis que para obtener el grado de:

Maestro en Ciencias

Presenta

Biól. Pablo Rogelio Simón Salvador

Directora de Tesis:

Edna Leticia González Bernal

Santa Cruz Xoxocotlán. Oaxaca. Enero 2020

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INSTITUTO POLITÉCNICO NACIONAL
SECRETARÍA DE INVESTIGACIÓN Y POSGRADO

ACTA DE REVISIÓN DE TESIS

En la Ciudad de Oaxaca siendo las 02:30 horas del día 27 del mes de noviembre del 2019 se reunieron los miembros de la Comisión Revisora de la Tesis, designada por el Colegio de Profesores de Estudios de Posgrado e Investigación de CIIDIR OAXACA para examinar la tesis titulada:

"Impacto de la perturbación humana en arroyos de bosque mesófilo de montaña sobre anuros nativos en Santa Cruz Tepetotutla Oaxaca"

Presentada por el alumno:

Simón	Salvador
Apellido paterno	Apellido materno

Nombre(s): Pablo Rogelio

Con registro: | B | 1 | 7 | 0 | 2 | 6 | 8 |

Maestría en Ciencias en Conservación y Aprovechamiento de Recursos Naturales

Después de intercambiar opiniones, los miembros de la Comisión manifestaron ***APROBAR LA TESIS***, en virtud de que satisface los requisitos señalados por las disposiciones reglamentarias vigentes.

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En la Ciudad de Oaxaca el día 27 del mes de noviembre el año 2019, el (la) que suscribe Pablo Rogelio Simón Salvador alumno(a) del Programa de Maestría en Ciencias en Conservación y Aprovechamiento de Recursos Naturales con número de registro B170268, adscrito a Centro Interdisciplinario de Investigación para el Desarrollo Integral Regional Unidad Oaxaca, manifiesta que es autor (a) intelectual del presente trabajo de Tesis bajo la dirección de la Dra. Edna Leticia González Bernal y cede los derechos del trabajo titulado: "Impacto de la perturbación humana en arroyos de bosque mesófilo de montaña sobre anuros nativos en Santa Cruz Tepetotutla Oaxaca" al Instituto Politécnico Nacional para su difusión, con fines académicos y de investigación.

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Pablo Rogelio Simón Salvador

Nombre y firma


"Nuestra lealtad es para las especies y el planeta. Nuestra obligación de sobrevivir no es sólo para nosotros mismos sino también para ese cosmos, antiguo y vasto, del cual derivamos"
Carl Sagan

Dedicatorias.

“Mis padres no sabían casi nada de ciencia. Pero al iniciarme simultáneamente al escepticismo y hacerme preguntas, me enseñaron los dos modos de pensamiento fundamentales para el método científico” C. S.

A mis padres, por enseñarme el amor hacia la naturaleza, por su, esfuerzo y apoyo incondicional, a mi hermana por sus constantes regaños y momentos de diversión.

A las comunidades indígenas de Oaxaca, que día a día trabajan por la conservación de los recursos naturales y por su lucha constante por la defensa de su territorio.

Resumen

El declive en poblaciones de anfibios en todo el mundo se ha relacionado con múltiples factores, sin embargo, el factor principal es la alteración del hábitat, generada por actividades humanas. La mayoría de los estudios relacionados registran disminución en la riqueza de especies en sitios antropizados, sin embargo; pocos han examinado efectos a nivel individual. Aquí exploramos, cómo distintos grados de perturbación antropogénica en un bosque mesófilo de montaña afectan la abundancia, condición corporal, temperatura corporal y uso de hábitat de individuos de tres especies de hílidos de arroyo. Se seleccionaron nueve arroyos en bosque mesófilo de montaña a través de un gradiente de perturbación antropogénica, desde bosque sin perturbación antropogénica hasta moderadamente perturbado. Durante 2018 se realizaron muestreos estandarizados siguiendo la técnica de encuentros visuales en transectos de 50 metros. Además, se realizó caracterización de la estructura de vegetación en los arroyos y el bosque aledaño. Encontramos una respuesta diferencial a la perturbación, por ejemplo, la mayor tasa de observación de *Charadrahyla nephila* fue mayor en corrientes no perturbadas, para *Duellmanohyla ignicolor* en los arroyos recuperados y *Ptychohyla. zophodes* en las corrientes perturbadas, así mismo este mismo patrón se reflejó en términos de condición corporal, temperatura corporal y uso de hábitat para estas tres especies, Según nuestros resultados, sugerimos que *Charadrahyla nephila* es una especie sensible a la perturbación, presentando incremento de temperatura y disminución de condición corporal conforme aumento la perturbación, mientras que otras especies (por ejemplo. *Duellmanohyla ignicolor* y *Ptychohyla. zophodes*) pueden beneficiarse de los remanentes de vegetación arbustiva que les ofrecen los arroyos en sitios recuperados y perturbados respectivamente. La identificación de los efectos de la perturbación del hábitat sobre las especies a nivel fisiológico y de uso del hábitat es fundamental para la evaluación del impacto y las actividades de mitigación, incluido el diseño de estrategias y propuestas para el manejo de áreas sujetas a perturbaciones, que permitan no solo la prevalencia de poblaciones, sino también poblaciones saludables de ranas en bosques mesófilos con manejo agrícola y forestal.

Palabras clave: Perturbación antropogénica, Hílidos, Arroyos, Temperatura corporal, Condición corporal, Uso de Hábitat

Abstract

Worldwide amphibian declines have been linked to multiple factors. However, the main factor is habitat disturbance generated by human activities. related studies have recorded a decrease in amphibian species richness in anthropized sites, however; few have examined effects at the individual level. Here we explore how different degrees of anthropogenic disturbance in a tropical montane cloud forest (TMCF) affects frog abundance, body condition, body temperature and habitat use of individuals of three species of stream dwelling hylids. Nine streams on TMFC were selected through an anthropogenic disturbance gradient from pristine forest to moderately disturbed. Along 50 m transects on these streams we looked for frogs, recorded individual traits and habitat use. In addition, characterization of the vegetation structure in the streams and the surrounding forest was carried out. We found a differential response to the habitat disturbance by species. For *Charadrahyla nephila* we recorded a higher observation rate at undisturbed streams, for *Duellmanohyla ignicolor* in the recovered streams and *Ptychohyla. zophodes* in the disturbed streams. Likewise, this same pattern was reflected in terms of body condition, body temperature and habitat use for these three species. According to our results, we suggest that *Charadrahyla nephila* is a species sensitive to disturbance, presenting body temperature increase and body condition decrease as the disturbance increases, while the other species (*Duellmanohyla ignicolor* and *Ptychohyla. zophodes*) may benefit from some characteristics offered by streams in recovered and disturbed sites respectively. The identification of habitat disturbance effects over species at physiological and habitat use level is critical for assessment impact and mitigation activities, including strategies design and proposals for the management of areas subject to disturbance that allow not only population prevalence, but also healthy populations of frogs on TMCF managed.

Keywords: Anthropogenic disturbance, Hylids, Streams, Body temperature, Body condition, Habitat use

Introducción general

Actualmente más del 80% de los ecosistemas terrestres han sido modificados principalmente por actividades humanas, reduciendo en gran medida la cobertura vegetal original (MacDougall, 2013; Saunders et al. 1991). Estos remanentes de vegetación experimentan cambios ambientales, que afectan la composición de especies y las interacciones bióticas (Murcia 1995).

Uno de los ecosistemas más amenazados por las actividades humanas es el bosque nuboso tropical, conocido en México como bosque mesófilo de montaña (BMM) (Rzedowski, 1978; Ponce-Reyes et al., 2012). Este bioma ocupa el 1% del territorio nacional, sin embargo, alberga la mayor diversidad de flora y fauna del país, y constituye el segundo ecosistema con mayor riqueza de vertebrados terrestres endémicos en el país (Flores Villela, 1994).

En el sur de México, estos bosques albergan una gran diversidad de anfibios, que además es el grupo de vertebrados mayormente amenazado a nivel mundial (Ponce-Reyes et al., 2012;). Particularmente los BMM de la Sierra de Juárez de Oaxaca, son la zona con mayor riqueza y endemismos de anfibios (42 especies) en Mesoamérica (Mata-Silva et al.,2015; Ochoa-Ochoa et al., 2014). En esta zona, la Alianza para la Extinción Cero identificó 22 especies prioritarias únicas a esta región geográfica, lo que la convierte en una zona importante para la conservación de este grupo ante un panorama actual no tan alentador (Lamoreux, Mcknight, & Hernandez, 2015; Mata-Silva et al.,2015; www.zeroextinction.org).

Los anfibios en esta región se encuentran amenazados principalmente por perturbación antropogénica del hábitat derivada de prácticas agrícolas de subsistencia como la roza-tumba y quema y en menor medida por la apertura de brechas y carreteras en áreas de bosque conservado (Campbell *et al*., 1989, Pazos-Almada y Ray 2018).

Sin embargo, adyacente a las zonas de manejo agrícola y carreteras aún existen parches de vegetación de bosque mesófilo conservado, resultando en un área con diversos regímenes de perturbación, que no llegan a ser tan drásticos *i.e.* tala completa del bosque. Bajo este escenario, los regímenes de perturbación del BMM en esta región pueden ofrecer condiciones ambientales que permitan la subsistencia de especies de anfibios nativos. No obstante, la perturbación antropogénica puede inducir cambios morfológicos o de comportamiento en los individuos, los cuales pueden ser adaptativos o inducidos ambientalmente (Delgado-Acevedo y Restrepo, 2008). Por ejemplo, se tiene registro de que la temperatura corporal, el tamaño, peso, condición corporal, así como la forma general de los anfibios puede variar de manera predecible a lo largo de gradientes ambientales (Dunson *et al.*, 1992; Pough *et al*., 1998)

En este sentido, la presente tesis se enfocó en el efecto de la perturbación antropogénica sobre tres especies de hílidos evaluando cuatro aspectos principales (1) abundancia de especies (2) índice de condición corporal (3) temperatura corporal (4) uso del hábitat, y la relación de la abundancia de cada una de las especies con la estructura de la vegetación y distancia a la perturbación. Esto se realizó en tres regímenes de perturbación antropogénica: arroyos inmersos en bosque mesófilo conservados, arroyos en áreas de bosque mesófilo recuperado y arroyos inmersos en bosque mesófilo perturbado.

La relevancia de esta investigación radica en comprender como distintas especies de anuros endémicos responden a la perturbación humana presentando ajustes tanto en características individuales como conductuales que les permiten mantenerse ante los cambios de hábitat. Dado que son pocos los estudios que abordan el efecto de las perturbaciones desde esta perspectiva, la información generada es esta tesis puede servir para el diseño y planificación de estrategias para la conservación de estas y otras especies vulnerables, restringidas al bosque mesófilo.

Impact of human disturbance in a tropical montane cloud forest streams on native anuran from Santa Cruz Tepetotutla Oaxaca

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Resumen

El declive en poblaciones de anfibios en todo el mundo se ha relacionado con múltiples factores, sin embargo, el factor principal es la alteración del hábitat, generada por actividades humanas. La mayoría de los estudios relacionados registran disminución en la riqueza de especies en sitios antropizados, sin embargo; pocos han examinado efectos a nivel individual. Aquí exploramos, cómo distintos grados de perturbación antropogénica en un bosque mesófilo de montaña afectan la abundancia, condición corporal, temperatura corporal y uso de hábitat de individuos de tres especies de hylidos de arroyo. Se seleccionaron nueve arroyos en bosque mesófilo de montaña a través de un gradiente de perturbación antropogénica, desde bosque sin perturbación antropogénica hasta moderadamente perturbado. Durante 2018 se realizaron muestreos estandarizados siguiendo la técnica de encuentros visuales en transectos de 50 metros. Además, se realizó caracterización de la estructura de vegetación en los arroyos y el bosque aledaño. Encontramos una respuesta diferencial a la perturbación, por ejemplo, la mayor tasa de observación de *Charadrahyla nephila* fue mayor en corrientes no perturbadas, para *Duellmanohyla ignicolor* en los arroyos recuperados y *Ptychohyla zophodes* en las corrientes perturbados, así mismo este mismo patrón se reflejó en términos de condición corporal, temperatura corporal y uso de hábitat para estas tres especies, Según nuestros resultados, sugerimos que *C. nephila* es una especie sensible a la perturbación, presentando incremento de temperatura y disminución de condición corporal conforme aumento la perturbación, mientras que otras especies (por ejemplo. *D. ignicolor* y *P. zophodes*) pueden beneficiarse de los remanentes de vegetación arbustiva que les ofrecen los arroyos en sitios recuperados y perturbados respectivamente. La identificación de los efectos de la perturbación del hábitat sobre las especies a nivel fisiológico y de uso del hábitat es fundamental para la evaluación del impacto y las actividades de mitigación, incluido el diseño de estrategias y propuestas para el manejo de áreas sujetas a perturbaciones, que permitan no solo la prevalencia de poblaciones, sino también poblaciones saludables de ranas en bosques mesófilos con manejo agrícola y forestal.

Palabras clave: Perturbación antropogénica, Hílidos, Arroyos, Temperatura corporal, Condición corporal, Uso de Hábitat

Introduction

Currently, the main factor that directly affects biodiversity is the high rate at which habitat is lost (Hanski, 2011). Habitat modification by human activities until the 21st century has affected 75% of the ice-free land to some extent, associated with agriculture, logging and urbanization (Fletcher et al., 2018; Pardini, Nichols and Püttker, 2018) This phenomena can alter resources availability, affecting individual performance, populations, communities and ecosystems' structure. due to changes in resources availability mainly associated to local changes in temperature and humidity due to canopy cover decrease and deterioration (Albuquerque et al., 2017; Grimm et al., 2017; Newman, 2019).

Disturbance ecology has been tremendously successful over the past decades at elucidating the interactions between disturbances, biodiversity, and ecosystems, however, most of the research focused on anthropogenic disturbance has been centered at evaluating effects on species richness and/or abundance reporting a mixture of positive, negative or insignificant effects on communities (Gardner, Barlow, & Peres, 2007; Supp & Ernest, 2014). For example, studies with vertebrates including birds, amphibians, reptiles and bats have reported variation in species composition, but not richness, along anthropogenic disturbance gradients; indicating loss of sensitive species and the propagation of tolerant species (Asefa, et al., 2017; Díaz-García, Pineda et al.,2017; Gillespie et al., 2015). But, even for cases in which species richness and abundance in altered habitats remains high, this not necessarily means that under those conditions, communities will remain functional (Thompson, Nowakowski, & Donnelly, 2015). Species respond differentially to perturbation due to variation in their sensitivity to habitat changes and specific ecological traits (Díaz-García et al., 2017).

From this perspective, there has been an increasing interest in understanding disturbance effects at an individual level, for example with metrics to quantify the physiological state of individuals i.e. body condition (BCI) (Bourbonnais et al., 2014; Jakob, Marshall, & Uetz, 1996; Labocha, Schutz, & Hayes, 2014). Several studies have shown that anthropogenic negatively affects individual body condition. For example, individuals of *Anolis antonii* at agricultural habitat presented poorer body condition than individuals from a population at a secondary forest (Gallego-carmona, Castro-arango, & Bernal-bautista, 2016). This same pattern has been shown for different vertebrates such as: *Ursus arctocs* (Brown bear), *Charadrius melodus* (Piping plover), and *Dendropsophus ebraccatus* (Hourglass treefrog) (Bourbonnais et al., 2014; Kight & Swaddle, 2007; Matias & Escalante, 2015) .

Another, even less explored approach, is the way in which species that persist in anthropized sites adjust habitat use. Usually, individual abundances would correlate with preferred resources availability at space (Murray, 2002; Venier & Fahrig, 1996). For example, abundance and presence of lizards of the genus *Anolis* is related to characteristics of the plant physiognomy related to the maturity of the forest (Zuluaga & Cárdenas, 2007). For some species of frogs it has been reported that they use particular plant species (Beard, McCullough, & Eschtruth, 2003; Watson, Mcallister, & Pierce, 2003), or structural components in a vegetation community such as perches (Parris, 2001). In addition, it has been shown that characteristics such as height, diameter and microclimate of the perch are characteristics that limit its abundance and distribution (Zuluaga & Cárdenas, 2007). Information on the habitat use of a species provides clues and their responses to disturbance processes is particularly important when aiming to identify proper habitat for the persistence and conservation of a species (Shuker & Hero, 2012).

It is known that the responses to disturbance vary depending on the ecological characteristics of each taxon (Cardoso et al., 2013; Vandewalle et al., 2010). In this sense, amphibians, given their eco-physiological characteristics in addition to a low dispersal capacity, are more sensitive to anthropogenic disturbances than other vertebrates (Hopkins, 2007). being the most threatened group of vertebrates that records the highest number of species classified as endangered according to the International Union for Conservation of Nature (IUCN) (Stuart et al., 2008). This problem is concentrated in tropical regions where diversity is high and there are many threatened species with restricted distributions (Brooks et al., 2002; Roberts, Boivin, & Kaplan, 2018).

Tropical montane cloud forest is a restricted ecosystem characterized by harboring high species richness at a small area and by being one of the most threatened ecosystems on earth (Ponce-Reyes et al., 2012). In México, the cloud forest from the Sierra Juárez of Oaxaca have the largest amount of endemic amphibians in the country at a relatively small area (Lamoreux, Mcknight, & Hernandez, 2015; Mata-Silva et al., 2015). However, historically this region has been subjected to various anthropogenic pressures, related to subsistence farming practices such as slash-and-burn, logging and to a lesser extent due to the opening of gaps and roads in relatively pristine areas (Campbell, Lamar, & Hillis, David, 1989; Flores & Manzanero, 1999; Mathews, 2003). Although when disturbance processes in this region are less severe than large-scale logging, it is important to know the response of species to these anthropogenic pressures (Burivalova, Şekercioğlu, & Lian, 2014; Gillespie et al., 2015).

Therefore, it is necessary to conduct research with a focus on small-scale habitat disturbance in order to contribute to the understanding of differential species responses and to be able to propose conservation actions for the species that inhabit tropical areas with similar regimes

In this sense, we investigated the response to differential anthropogenic disturbance of three sympatric hylid frog's species: Oaxacan cloud forest tree frog (*Charadrahyla nephila*), Sierra Juarez brook frog (*Duellmanohyla ignicolor*), and gloomy mountain stream frog (*Ptychohyla zophodes*) at streams from a tropical montane cloud forest (TMFC). We explored three main aspects (1) species abundance (2) individual variables such as body condition and body temperature, (3) habitat use. We related these aspects to structural changes at three disturbance regimes: streams from undisturbed TMFC, streams at recovered areas and streams at disturbed forest. This allowed us to understand the way in which these organisms respond to different perturbation magnitudes and define other possible ways in which habitat perturbation might have a long-term effect.

2 Materials and Methods

2.1 Study area

This study was conducted in Santa Cruz Tepetotutla (N 17°45'28.5 W 96° 33' 17.5") a community located on the northern slopes of the Sierra Juárez, Oaxaca. In this region the tropical montane cloud forest (TMCF) is distributed between 700 and 2500 m.a.s.l. (Flores & Manzanero, 1999). The maximum rainfall ranges between 3,000 and 4,500 mm/year. The area is characterized by presenting two seasons: a dry season that goes from November to April and a wet season from May to October. The mean annual temperature ranges from 12 to 18°C at 2,300 m.a.s.l. elevations and from 18 to 22 ° C at 1,200 m.a.s.l. (INEGI, 2008).

Since 2004, this community preserves 9670 ha of montane cloud forest, making this forest one of the largest preserved patches of this ecosystem in the country (Ponce-Reyes et al., 2012). The conservation model is known as Voluntary Conservation Areas (VCAs). Under this mechanism, the territory decreed by indigenous and community conserved areas (ICCAs) are officially recognized and certified by the National Protected Area Commission in Mexico (CONANP) by its acronym in Spanish). People manage their territory by having specific sites for subsistence activities such as agriculture (corn and coffee plantations) and cattle paddocks. This situation provides with a range of habitats along the perturbation-conservation axis which allows a monitoring design to compare different perturbation degrees without any of them being so drastic (i.e. total deforestation), to make it impossible for the focal species to survive.

2.2 Study species

Stream dwellers

Based on their distribution (presence in montane cloud forest) and their vulnerability due to habitat transformation according to International Union for Conservation of Nature (IUCN) evaluations, we selected three species of stream-dwelling amphibians to test ways in which they respond to human perturbation. These species are:

Charadrahyla nephila (Mendelson & Campbell, 1999).Medium size hylid (snout vent length SVL) range from 52.9 – 61.9 mm in males and 59.6 - 80.7 mm.in females**.** It inhabits tropical montane cloud forest through three mountainous systems in the Sierra Madre de Oaxaca (Sierra Juárez, Sierra Mazateca and Sierra Mixe), from elevations of 680 to 2256 m.a.s.l. It is endemic to Mexico and is categorized as Critically Endangered according to IUCN.

Duellmanohyla ignicolor (Duellman, 1961). Small size hylid, snout-vent length (SVL) of 26.3-30.0 mm. Occurs in mountain streams of cloud forests. The species is endemic to Oaxaca state, Mexico, in three localities at the lower northern slopes of Sierra de Juarez, range elevation at 600 - 1850 m.a.s.l Categorized as threatened according to IUCN.

Ptychohyla zophodes (Campbell & Duellman, 2000). Small size hylid SVL of 37.4 mm in males and 43.6 mm from females. Is restricted to the mesic forest of the Atlantic slopes of northern Oaxaca and southern Veracruz, México. Inhabits cloud forest and lower montane rainforest from 400-1500 masl, where it is generally found in vegetation around streams. Categorized as data deficient according to IUCN.

2.3 Field work

Stream selection

As the selected species are mostly stream dwellers, we monitored them in streams under three different conditions following a conservation-perturbation gradient. These conditions are: 1) Disturbed sites: streams adjacent to agriculture or cattle raising areas, 2) Recovered areas: streams located in cloud forest with around a 25 years period of recovery after a logging event and close to roads and paths 3) Undisturbed areas: streams located in mature forest and located more than one kilometer away from the nearest anthropogenic disturbance. A total of nine streams were monitored, three under each condition.

Field surveys

We conducted surveys during the dry season (March and May 2018 and March 2019) and the wet season (August, October and November 2018). Frogs were searched in potential sites along a 50 m long transect from ground level up to 3 m at streams with different perturbation regimes (see previous section) by two persons following visual encounter survey methodology (Crump & Scott, 1994). On each field trip all transects were sampled twice in two different nights. Sampling occurred between 19:00 to 02:00 hrs. (CST) standardizing the sampling length to one hour per stream. The survey order and schedules were randomized. Each night we sampled as many streams as possible. To avoid recording the same individual twice, all searches were made walking along the transect in one single direction.

Every time a frog was encountered, its species, weight, snout-vent length (SVL), body temperature (with an infrared thermometer), sex, life stage and activity at the time of encounter were recorded as quickly as possible. Afterwards, all organisms were released in the site where they were found. In addition, we recorded air temperature, relative humidity, substrate temperature, perch height and diameter and distance to the water.

2.4 Vegetation structure

To compare the structure of woody vegetation at each stream, we followed the point-centered quarter method (Cottam & Curtis, 1956). We established three 50 m long transects perpendicular to the stream. At each perpendicular transect we collected vegetation variables on three points at 15, 30, and 45 m from the stream to the interior of forest.

Each sampling point was divided into four 90° quadrants. Within each quadrat, the closest smallest tree (Diameter at breast height (DBH) between 10 - 31.5 cm) and biggest tree (DBH greater than 31.5 cm) to the sampling point were identified. For each tree, in addition to DBH, we recorded its height and its distance to the central point of the quadrant. We also measured canopy cover, leaf litter depth, slope, and percentage of available substrates.

2.5 Stream characteristics

At each stream, every 10 m along the 50 m transect, we recorded water velocity, stream width and depth, canopy cover using a spherical convex densitometer, and we made visual estimates of stream substrate composition (percentage of bedrock, gravel, sediment, sand or coarse woody debris).

2.6 Habitat use

To determine habitat utilization patterns , we recorded five parameters of the "structural habitat" (Rand, 1964): 1) type of perch categorized as herbaceous plant, shrub or tree, 2) structure of perch categorized as leaves, branch or stalk 3) perch diameter in the case of branches and stems 4) perch height above ground and 5) perch distance to water.

In order to determine perch quality and availability among the perturbation regimes we measured perch diameter and density (adapted from Dix, 1961) At each stream transect, we marked vertical quadrants every 10 meters locating an origin point at a height of 1.30 meters.

Each sampling point was divided into four 90° quadrants, each considered as a sampling unit. Within each sampling unit, the distances from the origin point to the two closest live shoots were recorded, and their diameter measured: a shoot is defined as a stem plus its appendages (Foster 1949). Perch densities are expressed in terms of aerial shoots per unit area.

2.7 Statistical analysis

Abundances

To compare the observed abundance between sites with different disturbance degree, we calculated the encounter rate (ER), weighting the number of frogs found with the applied sampling effort (person-hours), as follows:

$$Encounter\ rate\ (ER) = \frac{Total\ number\ of\ frogs\ found\ per\ site}{Sampling\ effort\ \ (person\ hours)}$$

To determine whether observed abundance of each species was affected by the disturbance degree we used a Chi squared test (Zar, 1999). The expected abundance value for each disturbance regime was equal to one third of the total abundance given that the sampling effort was the same in each disturbance degree.

Body condition index and body temperature

The body condition index (BCI) was calculated from the residuals of a linear regression between snout–vent length (SVL) and weight (Jakob et al., 1996; Rodríguez-Prieto, Martín, & Fernández-Juricic, 2010). Data were not transformed because they met all the assumptions of linear regression (Green, 2001): We used ANOVA to assess whether the anthropogenic disturbance affected the body condition index by species. Only adult male data by species were included in the analysis due to two reasons: females were less abundant and to avoid biases in the BCI calculation because females are considerably heavier than males. Subsequently, to evaluate the perturbation effect on the body condition of individuals by season, t tests were used.

To compare the effect of anthropogenic disturbance on body temperature (T_b) we used Kruskal–Wallis ANOVA, followed by pairwise post hoc Mann–Whitney U-tests. We also evaluated the relationship between T_b, substrate temperature (T_s) and air temperature (T_a) at each disturbance regime, by simple correlation (Navas et al., 2013). All analyses were performed with the statistical software GraphPad Prism 5.0.

Habitat use

We calculated the proportion of perch type and perch structure used by frogs in each degree of disturbance by species, the frequency data of frog sightings between the different types of perches were compared with $\chi 2$ test. Also, we used Kruskal-Wallis ANOVA to assess whether the anthropogenic alteration influenced the perch selection (water distance and height perch by each of the species) for each of the species.

Perch density (N° shoots/m^2) was estimated following the formulas proposed in (Dix, 1961).

$$\textit{Perch density} = {}^{10{,}000}\!/_{(Mean\ distance^2)}$$

To compare perch availability, the diameter of available perches *vs.* preferred perch diameter were compared between disturbance degrees by a Kruskal–Wallis test and a post hoc Mann– Whitney U test. We used simple lineal regression with JMP software to analyze the relationship between species (that used branches and stems) abundances and perch density in the studied streams.

Vegetation structure and stream characteristics

For each stream we calculated tree density (trees/ha) and basal area (m^2 /Ha) following the formulas proposed in (Cottam & Curtis, 1956).

$$Tree\ density = {}^{10,000}/_{(dM^2)}$$

Where dM= Average distance (m) of tree records closest to the origin of each quadrant with a DBH> 31.4.

$$Basal\ area = \ Mean\ basal\ area\ per\ tree\ (m^2) * \ Tree\ density\ (\frac{tree}{ha})$$

$$Mean\ basal\ area\ per\ tree = \pi(average\ DBH^2 /4000) * Tree\ density$$

We used Kruskal–Wallis test and post hoc Mann– Whitney U test, to compare woody vegetation structure parameters (tree density, basal area, canopy cover, DBH, tree height), between disturbed, recovered and undisturbed sites, using the software GraphPad Prism 5.0.

Stream parameters

Stream characteristics: flow, canopy cover, coverage type (percentage of bedrock, sediment, sand or coarse woody debris), were compared with a Kruskal–Wallis test and a post hoc Mann– Whitney U test, using software GraphPad Prism 5.0.

Relationships between species abundance vegetation structure and stream characteristics

We ran canonical correspondence analysis (CCA) using abundance of anuran species as the response variables. The explaining variables in CCA were tree density, height, DBH, canopy cover, litter coverage, herbaceous coverage, stream flow, distance to disturbance and slope. Analyses were made in R using the Vegan package (Oksanen et al., 2016).

3 Results

3.1 Abundance

We recorded a total of 126 individuals of *Duellmanohyla ignicolor*, 94 *Charadrahyla nephila* and 61 *Ptychohyla zophodes* with a total sampling effort of 144 person hours. Frog abundance varied with the disturbance degree: the highest number of individuals (151) was recorded at streams located in recovered cloud forest followed by streams in undisturbed cloud forest (81 individuals) and streams in disturbed sites (49 individuals; $X^2 = 72.60$, df = 4, $p < 0.0001$, Figure 1a). Frog abundance was also influenced by the season; 69.75% (196 individuals) of the frogs were recorded during the rainy season, while 30.25 % (85 individuals) were recorded during dry season, ($X^2 = 21.463$, df=2, $p = <0.0001$) (Figure 1b) being the disturbed streams where we found less individuals (4 individuals).

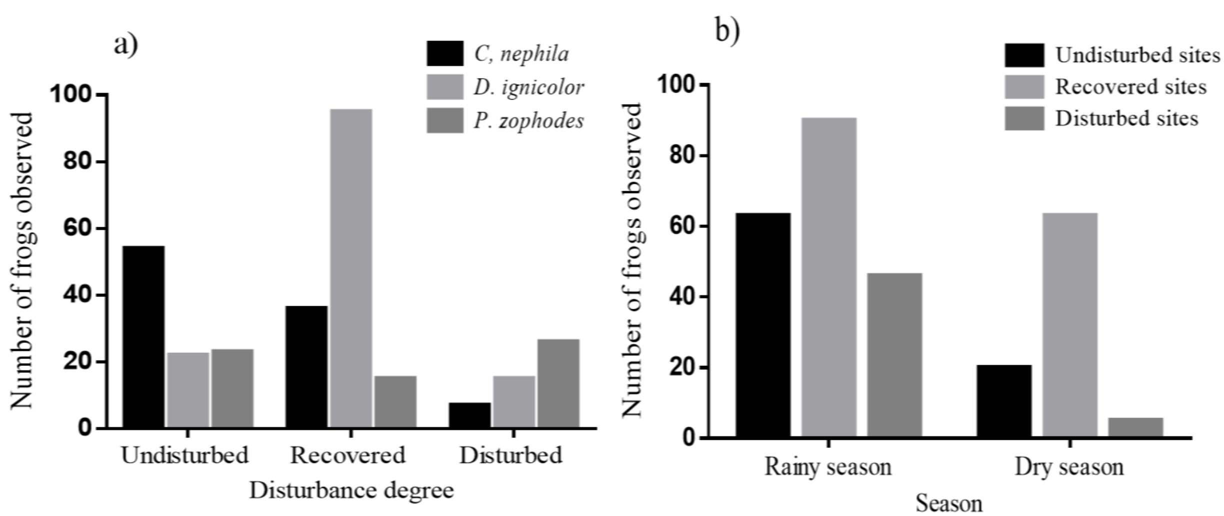

Figure 1. Hylid frogs abundance at streams with different disturbance degrees. (a) individuals observed by species (b) Abundance shifts by season

Despite the decrease in frog abundance as disturbance increased, the results showed different patterns when evaluating responses by species. *C. nephila* showed an increase in abundance as the disturbance decreased ($X^2 = 5.53$, df = 2, p= 0.4547) undisturbed sites (53 individuals), recovered sites (35 individuals) and disturbed sites (6 individuals), while during the dry season there were no records of this species in the disturbed sites (Figure 2a). *D. ignicolor* showed a greater record of individuals in recovered sites than in disturbed and undisturbed sites ($X^2 = 10.71$, df = 2, p = 0.0047; Figure 2b), on the other hand *P. zophodes* showed an increase in abundance as the anthropogenic disturbance increased ($X^2 = 9{,}864$, df = 2, p = 0.0072), however this pattern was evident only during the rainy season since during the dry season only one organism of this species was recorded (Figure 2c).

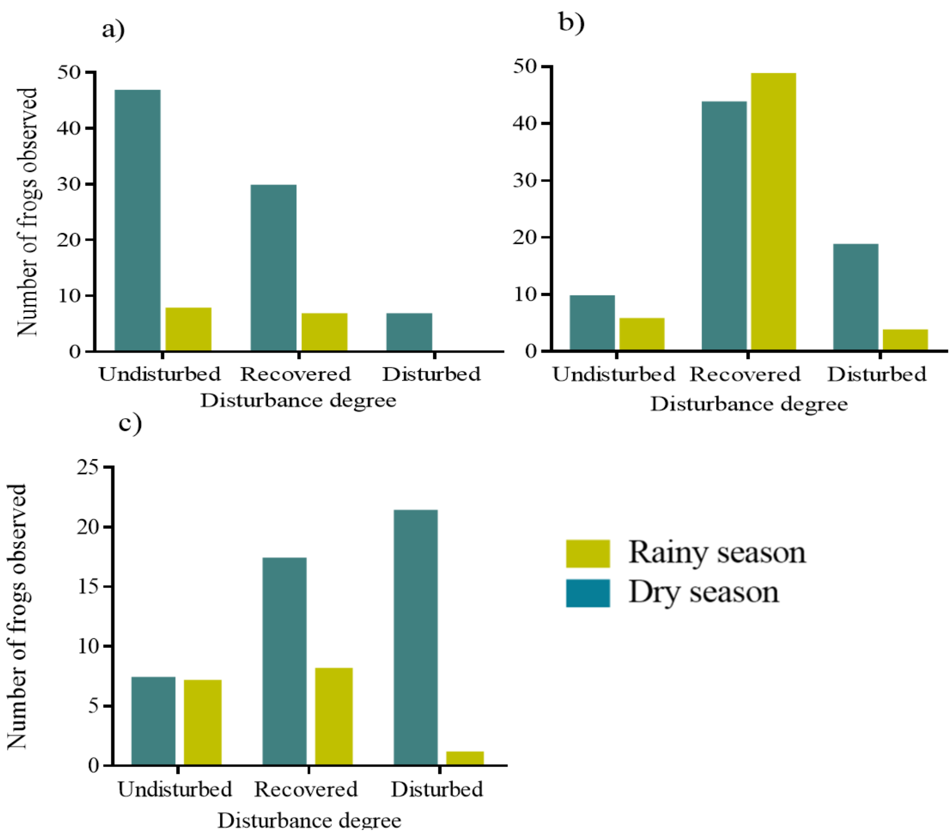

Figure 2. Number of individuals found at streams located in forest with different disturbance degree by season (a) *Charadrahyla nephila;* (b) *Duellmanohyla ignicolor* and (c) *Ptychohyla zophodes*

3.2 Body condition index

When analyzed per year, organisms didn't present differences in body condition neither by species nor by disturbance degree *Charadrahyla nephila* ($F_{2,86}$= 0.147, *P*= 0.862); *Duellmanohyla ignicolor* ($F_{2,109}$= 0.603, *P*= 0.054) *Ptychohyla zophodes* ($F_{2,49}$=0.248, *P*= 0.781). Although, when evaluating the seasonality effect, *Charadrahyla nephila* and *Ptychohyla zophodes* showed differences between perturbation regimes during the dry season. Individuals of *Charadrahyla nephila* presented a better condition in undisturbed sites *vs.* recovered sites ($t_{(10)}$=2.998, *p*= 0.0134; Figure 3a) while individuals of *Ptychohyla zophodes* showed an inverse pattern being those in recovered sites in better condition than in undisturbed sites ($t_{(11)}$=3.948, *p*= 0.0025; see Figure 3b).

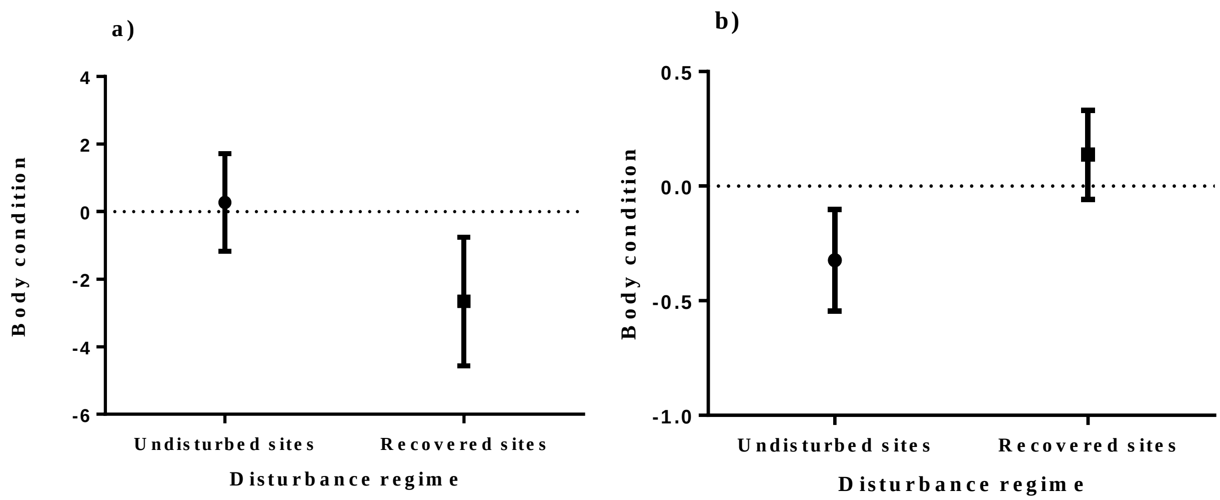

Figure 3. Body condition of individuals located at recovered and undisturbed sites during the dry season. (a) *C. nephila* and (b) *P. zophodes*. The graphs show mean values and associated standard errors.

3.3 The body temperature

Elevation was not a determining factor of air temperature (T_a) at the sampling sites ($F_{1,254}$=4.961 P= 0.353), contrary to the disturbance degree, that showed an influence on it. (Kruskal–Wallis ANOVA: $\chi2_{(2)}$ = 6.041, p=0.048). The lowest mean temperature was recorded in disturbed areas ($\bar{\bar{X}}$= 15.91 °C Min = 11 Max = 20) compared to recovered sites ($\bar{\bar{X}}$= 16.85 °C Min = 12.6 Max = 21.3) and undisturbed cloud forest ($\bar{\bar{X}}$= 16.25 Min = 11 Max = 21.6). Within the studied species, body temperature was positively correlated with substrate temperature and air temperature, however the substrate temperature showed a higher correlation coefficient (r^2); always greater than 0.96 (p <0.005) for the three species.

The species had differential responses in T_b due to anthropogenic disturbance. Species presented different T_b along the disturbance gradient. *C. nephila* individuals showed higher T_b as the degree of disturbance increased (Kruskal–Wallis ANOVA: $\chi2_{(2)}$ = 28.90, p <0.0001; Fig 4a), with a mean Tb of 16.99 °C at undisturbed streams (SE 0.10), 17.43 °C at recovered streams (SE 0.24) and 18.70 °C at disturbed sites SE 0.22). On the other hand, *P. zophodes* showed lower T_b in disturbed forest streams = 15.60 °C SE 0.47 (Kruskal-Wallis ANOVA:$\chi2_{(2)}$=10.79, p=0.0045; Fig 4c) in comparison to recovered areas (17.69°C SE 0.33) and streams at undisturbed cloud forests (16.79°C SE 0.42). *D. ignicolor* showed no differences in T_b among the different regimes (undisturbed cloud forest streams=16.40 °C SE 0.36: recovered forest streams=16.71 SE 0.19; disturbed forest streams=16.24 SE 0.46; Kruskal-Wallis ANOVA:$\chi2_{(2)}$=0.514, p=0.773; Fig 4b).

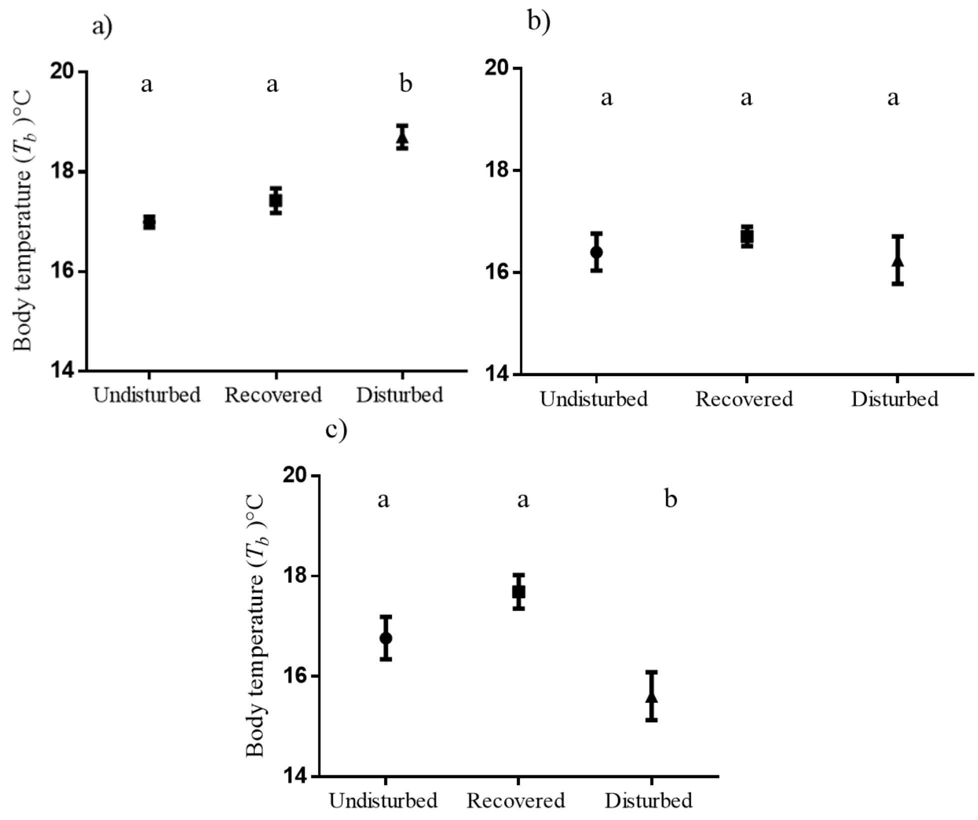

Figure 4. Body temperature (T_b) comparisons of individuals at undisturbed, recovered and disturbed sites throughout the year (2018-2019) a) *C. nephila* b) *D. ignicolor and C) P. zophodes.* The graphs show mean values and associated standard errors; different letters indicate statistical significance,

In addition, when comparing T_b, T_s and T_a by disturbance degree, *C. nephila* showed high T_b (undisturbed=16.99 °C SE 0.10; disturbed=18.70 SE 0.22) respect to T_a at undisturbed sites (16.58 °C SE 0.17) and disturbed sites (17.37 °C SE 0.35). T_b was close to T_s (undisturbed=16.98 °C SE 0.10; disturbed=18.80 SE 0.18; Fig. 5a). While *D. ignicolor* showed homogeneous T_b (undisturbed=16.40°C SE 0.36; recovered=16.71 SE 0.19 and disturbed=16.24 SE 0.46) and was near to T_a (undisturbed=16.14°C SE 0.66; recovered=16.47 SE 0.24 and disturbed=16.54 SE 0.60) and T_s (undisturbed=16.68°C SE 0.40; recovered=16.75 SE 0.18 and disturbed=16.33 SE 0.45) in each of the degrees of disturbance (Fig 5b). On the other hand *P. zophodes* recorded higher T_b =16.91°C SE 0.48 and closer to T_s =16.84°C SE 0.50 respect to T_a =14.83°C SE 0.70 only in undisturbed sites (Fig 5c).

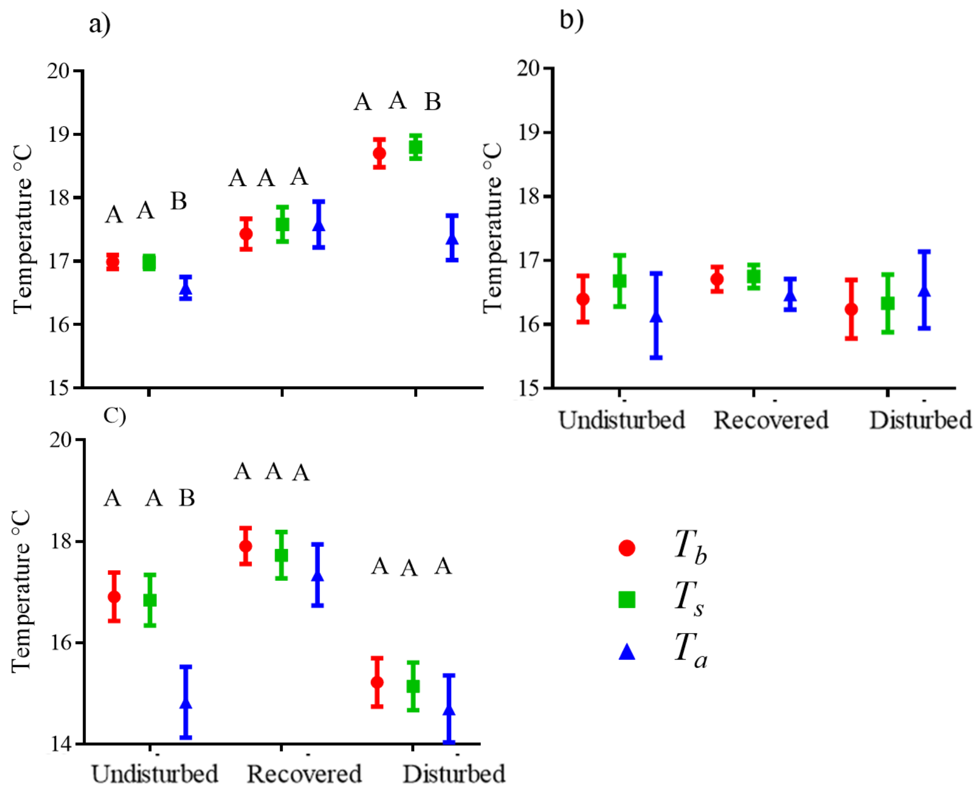

Figure 5. Comparisons of mean body temperature, substrate temperature and air temperature in a) *C. nephila* b) *D. ignicolor,* and c) *P. zophodes* at each disturbance degree. The graphs show mean values and associated standard errors. Different letters over means within same disturbance degree indicate statistical significance.

3.4 Habitat Use

Overall, a differential preference of perch type was observed by species (χ^2 = 107.49, df = 4, *P* = <0.0001). For *D. ignicolor* and *P. zophodes* 77% and 57% of individuals respectively were found laying on top of leaves, unlike *C. nephila* where 68.75% of the individuals were found grabbed of branches (Figure 6a). The substrate used also changed by species (χ2 = 34.73, df = 4, P = <0.0001; figure 6b). *D. ignicolor* and *P. zophodes* individuals were found mostly on herbaceous plants, 88.14% and 74.52% respectively contrasting with *C. nephila* where the individuals were divided almost equally between trees and shrubs (47%) and herbaceous (53%).

Likewise, the distance at which individuals were from the water also varied depending on the species, *Charadrahyla nephila* (10. 22 cm, SE 4.95), *D. Ignicolor* (53.48 cm SE 4.44) and *Ptychohyla zophodes* (37.84 cm, SE 6.58; $F_{(2,250)}$ = 21.22 P = <0.0001).

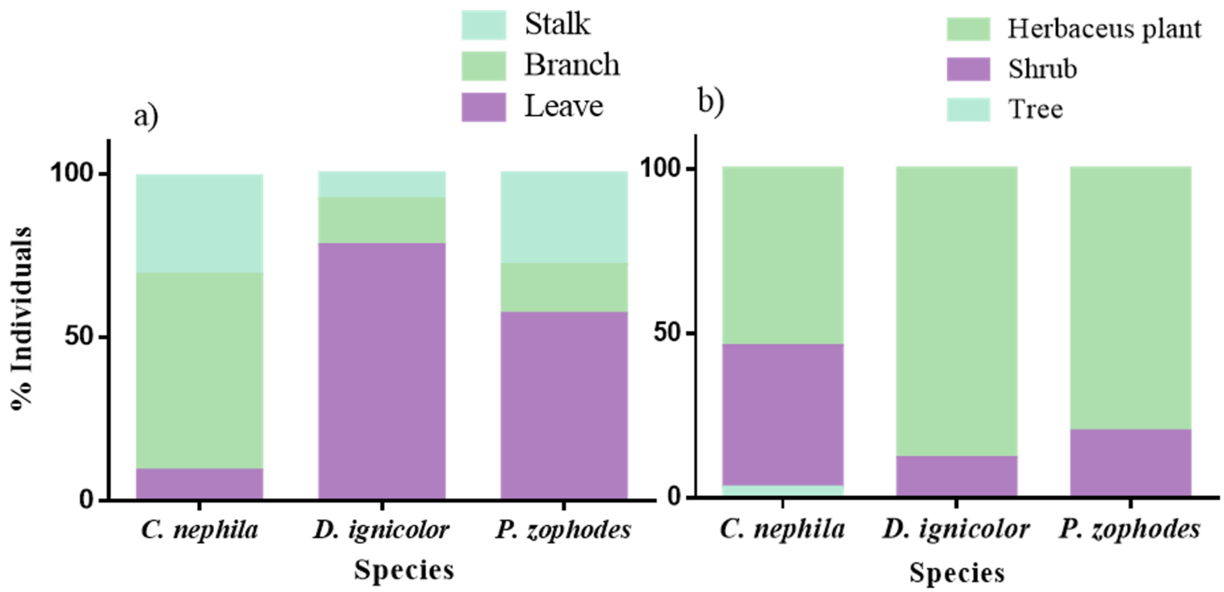

Figure 6. Percentage of individuals by species and preferred perch, a) stratum, b) perch structure.

Perch used among perturbance degrees only changed for *C. nephila* ($\chi 2$ = 13.95, df = 4, p = <0.0074) for which, in conserved sites 70.91% of the individuals was found on branches of herbaceous and shrubs, whereas at regenerated sites 47% of the individuals were found on herbaceous stalks and 75% of individuals at perturbed sites on herbaceous stalks. At undisturbed they used both herbaceous and shrubby stratum while at recovered and disturbed sites they were mostly found on herbaceous stratum ($\chi 2$ = 73.42, df = 3, p = <0.0001;Fig 7a).

For the species *D. ignicolor and P. zophodes,* neither the perch site (leaves) nor the stratum (herbaceous plants) used varied among the disturbance regimes (P > 0.05).

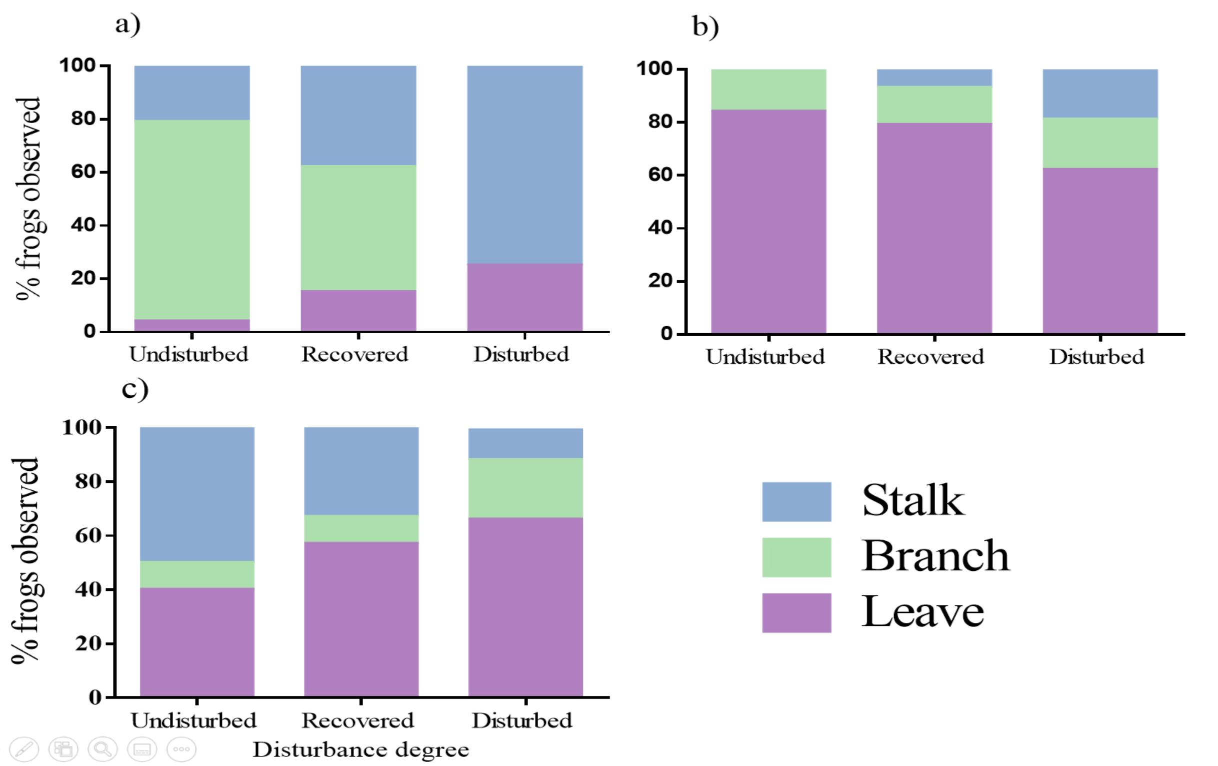

Figure 7. Percentage of frogs by species and preferred perch structure, a) *C. nephila*, b) *D. ignicolor* c) *P. zophodes*

Perch density of herbaceous plants varied depending on the degree of disturbance (Kruskal–Wallis ANOVA: $\chi2_{(2)}$ =6.974, *p*= 0.0221 Fig. 8a) being higher in undisturbed streams ($\bar{\bar{X}}$= 3.36/m^2 Min= 1.80 Max=5.67) and streams from recovered sites ($\bar{\bar{X}}$= 3.24/m^2 Min= 1.38 Max=4.85) than disturbed streams ($\bar{\bar{X}}$= 1.04m^2 Min= 0.19 Max=2.80). On the other hand, perch density of lignified was not different between streams with different degree of disturbance Kruskal–Wallis ANOVA: $\chi2_{(2)}$ =6.974, *p*= 0.91 Fig. 8b).

The diameter of the available perches of herb plants was smaller in the undisturbed cloud forest (Kruskal–Wallis ANOVA:$\chi2_{(2)}$ = 87.12, *p* = <0.0001: $\bar{\bar{X}}$= 0.35 cm Min= 0.10 Max=1.25) and recovered sites ($\bar{\bar{X}}$= 0.47 cm Min= 0.11 Max=3.80) than disturbed sites ($\bar{\bar{X}}$= 0.98 cm Min= 0.11 Max= 4.57; Fig 8c).

On the other hand, available lignified perches were thicker in disturbed sites (Kruskal–Wallis ANOVA: $\chi2_{(2)} = 38.70$, $p = <0.0001$: $\bar{\bar{X}}= 2.96$ cm Min= 0.47 Max=8.30) than recovered ($\bar{\bar{X}}= 2.15$ cm Min= 0.11 Max=8.43) and undisturbed sites ($\bar{\bar{X}}= 2.30$ cm Min= 0.11 Max= 10.75; Fig 8c). We observed a differential diameter of used perch by *C. nephila* depending on the degree of disturbance (Kruskal - Wallis ANOVA: $\chi2_{(2)} = 20.92$, $p = <0.0001$; 8c). At streams of undisturbed sites, *C. nephila* tended to use stems of small diameters ($\bar{\bar{X}}= 0.56$ cm Min = 0.22 Max = 1.42) than at recovered sites ($\bar{\bar{X}}= 1.24$ cm Min = 0.32 Max = 1.37) and disturbed sites ($\bar{\bar{X}} = 1.43$ cm Min = 0.75 Max = 2.30), this probably related to the decrease of available perches (shoots/m^2) in the recovered and disturbed streams which was 70% lower than undisturbed sites (fig 8a). Following with this interpretation, we observed an increase in individual's abundance as perch availability increased ($r^2 = 0.828$; p <0.005) (figure 8d).

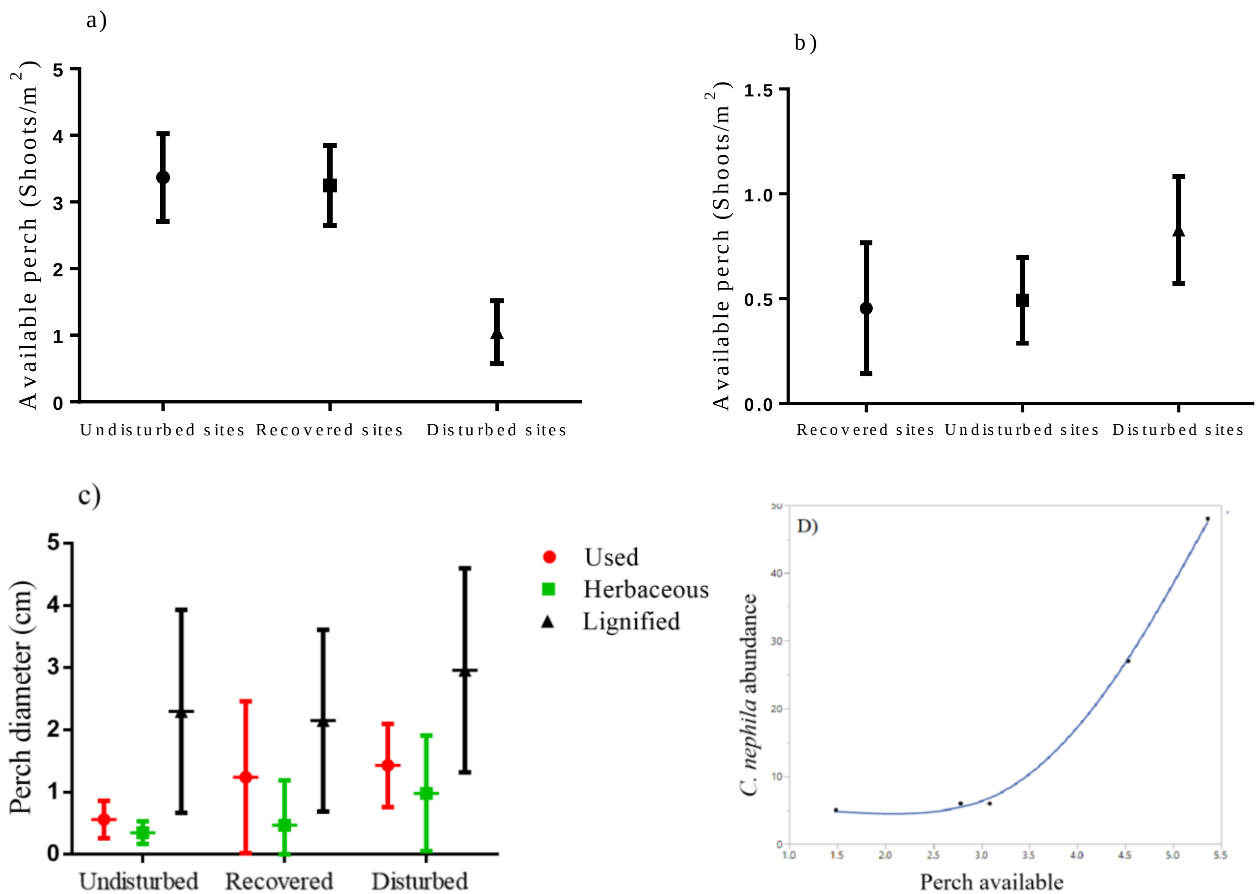

Figure 8. Different types of available perches at streams under three disturbance regimes in Santa Cruz Tepetotutla, Mexico. (a) Herbaceous plants, (b), lignified plants (c), comparisons between preferred perch diameter and diameter of available perches in herbaceous and lignified plants, (d). correlation between *C nephila* abundances and available perch. The graphs show mean values and associated standard errors.

3.5 Vegetation structure and local environmental temperature

Habitat structure characterization revealed differences on tree height, DBH and canopy cover between the three disturbance regimes (Kruskal–Wallis ANOVA: $p < 0.001$) being lower in the recovered sites compared to those of cloud forest (Fig. 9 b, c, d). The recovered and undisturbed sites generally showed greater percentage of leaf litter in comparison to disturbed sites (fig. 9 f). In contrast, herbaceous coverage in disturbed sites was greater than at the other sites (Fig. 9 g).

Atmospheric temperature varied among disturbance degrees (Kruskal–Wallis ANOVA: $\chi2_{(2)}$ = 6.041, P 0.048 Fig. 9h) The lowest mean temperature was recorded in disturbed areas ($\bar{\bar{X}}$= 15.91 °C Min = 11 Max = 20) compared to the recovered sites ($\bar{\bar{X}}$= 16.85 °C Min = 12.6 Max = 21.3) and undisturbed cloud forest ($\bar{\bar{X}}$ = 16.25 Min = 11 Max = 21.6). Relative humidity was higher in the undisturbed cloud forest (Kruskal–Wallis ANOVA:$\chi2_{(2)}$ = 12.48, P = 0.0019: $\bar{\bar{X}}$= 91.87% Min = 64 Max = 99) than recovered sites ($\bar{\bar{X}}$= 87 % Min = 59 Max = 99) and disturbed sites ($\bar{\bar{X}}$ = 86.59 % Min = 59 Max = 99; Fig. 9i).

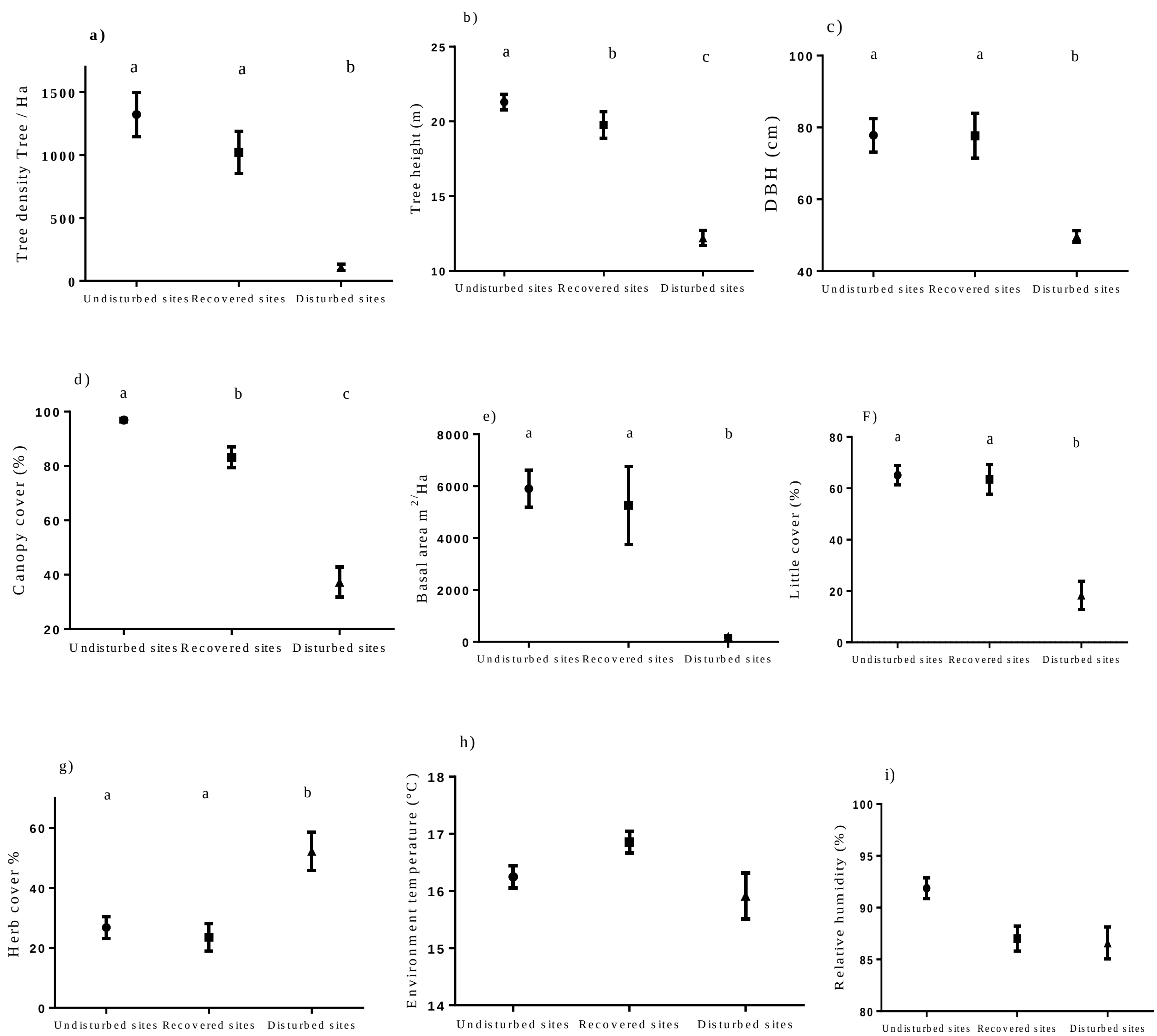

Figure 9. Variables for the woody vegetation structure (a–e), substrate percentage (f and g) and microclimate (h and i) for three disturbance regimes in Santa Cruz Tepetotutla, Mexico. The graphs show mean values and associated standard errors. Letters above the boxplots indicate a statistically significant difference (see Materials and Methods)

3.7 Relationships between species abundance vegetation structure and stream characteristics

Canonical correspondence analysis (CCA) showed that 86% of species abundance can be explained by the structure and diversity of the woody vegetation. The analysis was significant when subjected to the permutation test ($F = 0.361$, $p = 0.001$). *C. nephila* was related to undisturbed cloud forest streams, characterized by being away from anthropogenic disturbance, immersed in sites with high density of trees and streams with high flow rates. *D. ignicolor* is associated to recovered sites with streams with high percentages of leaf litter, roots of trees and shrubs at the stream. *P. zophodes*, is associated to disturbed streams, characterized by having a large amount of sand, bare ground, and woody debris in the stream. (Fig. 10).

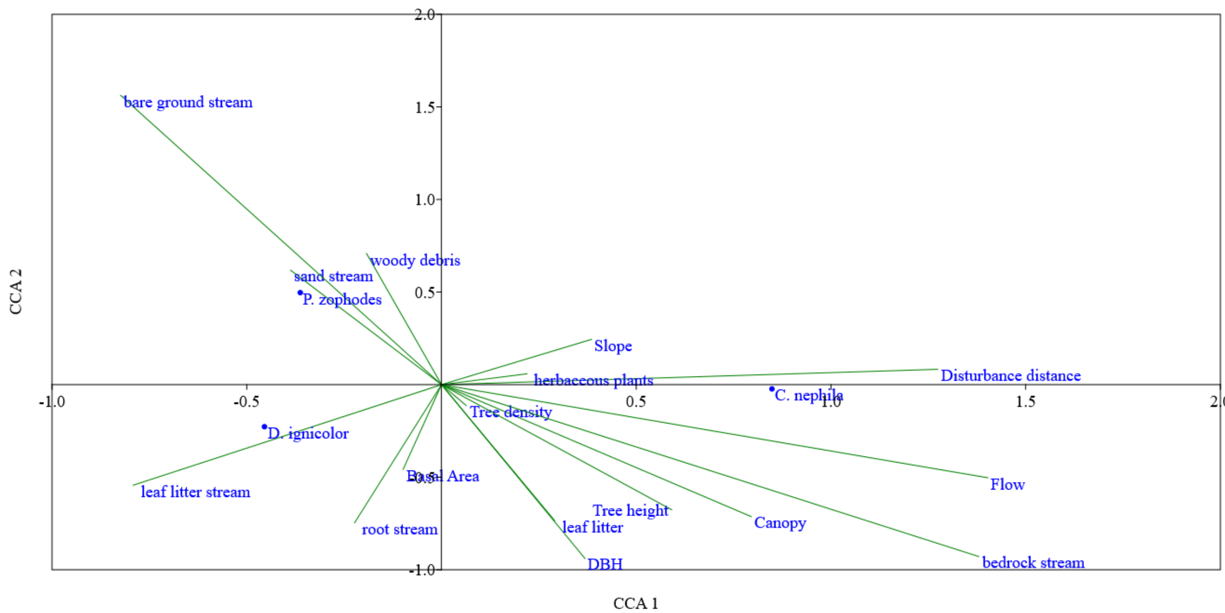

Figure 10 CCA of anuran species abundance, woody vegetation structure and stream characteristics

4 Discussion

Montane cloud forest is one of the most threatened ecosystems in the world mainly due to human activities (Ponce-Reyes et al., 2012). As TMCF management continues as happens with other tropical forests, it is necessary to understand how species respond to habitat pressures, and if they do it differentially in order to provide species-specific strategies for maintenance of resident populations in the long term.(Ledo, Montes, & Condes, 2009; Williams-Linera, 2002). This study demonstrates that anthropogenic disturbance at two levels differentially affected three species of tree frogs. Effects were observed on abundance, body condition and body temperature in addition to habitat preference Our results support the idea that amphibian responses to environmental changes are unique for each species and that the disturbance degrees differ considerably in their condition to serve as a habitat for anurans (Burton et al., 2009; Cushman, 2006).

We found a differential response regarding the encounter rates between degrees of disturbance. For example, the observation rate of *C. nephila* was higher in undisturbed streams, showing a gradual decrease as disturbance increased. This pattern is most likely related to habitat reduction that leads to a decrease of possible niches for specialist's species compared with generalist species (Steinicke, Pe´er, & Henle, 2018). It is known that increasing stressful factors on an ecosystem reduces microhabitat quality within forests which particularly affects forest specialist species(Laurance, 2000). For example, *Anolis uniformis* and *Lepidophyma flavimaculatum* reptiles linked to humid closed-canopy forest areas, declined in abundance from the old growth forest sites to the youngest successional category, where it was absent. (Hernandez-Ordóñez, Urbina-Cardona, & Martinez-ramos, 2015)

Therefore, highly specialized species often disappear, or their abundance decreases due to anthropogenic disturbance. (Laurance, 2000; Steinicke et al., 2018). As our studied species are all arboreal specialists, we anticipated that all of them would exhibit negative responses to anthropogenic disturbance. Contrary to forecasts, the species *P. zophodes* showed the highest observation rate in disturbed streams, in addition to findings of tadpoles, metamorphic and juvenile individuals of this species at those sites, which is indicative of their use as breeding and refuge places.

We hypothesize that *P. zophodes* is a resilient species to anthropogenic disturbance effects due to the management level to which these streams are subjected. Usually shrubs and herbaceous substrates, associated to streams adjacent to paddocks and farmland, are preserved, allowing the permanence of perch sites useful to perform different activities (reproduction, thermoregulation and foraging). We propose that arboreal specialist species remain at disturbed sites due to the mantainance of shrubby and herbaceous structures on the stream's edge, as observed on arboreal species *Pristimantis shrevei* (Rodriguez et al., 2017) and *Anolis carolinensis* (Schaefer et al., 2009)Schaefer et al., 2009) where their abundance is determined by shrubs density, since they depend on these structures as foraging, breeding and resting sites.

D. ignicolor showed a considerably higher observation rate at streams in recovered cloud forest which reflects the recovery of the microclimate conditions and habitat structure that this species needs for its subsistence (Audino, Louzada, & Comita, 2014; Hernandez-Ordóñez et al., 2015)**.**

Even when undisturbed cloud forest streams presented similar vegetation structure and microhabitats availability. however, recovered sites, presented higher environmental temperature which could allow a greater food availability for this species and therefore a greater abundance.

For example, *Hyla cinerea* a tree frog from North America, showed greater abundance at places with greater food availability at areas where small-scale disturbances, such as canopy gaps also provide low-growing vegetative structure, favorable for insects and tree frogs. (Horn et al., 2005). This is consistent with our results lower percentages of canopy cover in recovered sites (83.1%), compared to undisturbed streams (96.8 %), allow a higher solar incidence and probably greater food abundance and perches. Overall, we propose that variation in abundance patterns of these arboreal specialist species reflects their ability to tolerate small-scale disturbance events differentially and the variations in spatial and feeding resources requirements of these species.

Body condition.

The body condition index (BCI) is an effective indicator of the physiological and nutritional condition of individuals at a population (Hayes & Shonkwiler, 2001; Jakob et al., 1996), and is commonly used also to assess the individual health of frogs (Hedge & Krishnamurthy, 2014). Since the BCI is defined as the accumulated reserve energy in the body, it is taken as an indicator of animal health (Peig & Green, 2009) and it is expected to deteriorate as basic resources deplete when disturbance increases (Ellis & Mcwhorter, 2012). This pattern is clearly reflected in *C. nephila*, for which, organisms from undisturbed were in better condition than those in recovered sites particularly during the dry season, which is, the period, of greatest stress for this group (Pounds & Crump, 1994).

Considering that perturbation can abate basic resources and that during the dry season food and water availability reduces (Allen et al., 2017) this could be the determinant moment for a species at perturbed sites to persist. In the studied site, we consider that for *C. nephila* this could be the effect of an increase in solar radiation and temperature, as well as a lower availability of specific preferred perching sites as perturbation increases For example, individuals of *D. ebraccatus* also presented a decrease in body condition as pond perturbation increased, related to increased solar radiation and decreased humidity in disturbed habitats leads to pond desiccation directly affecting breeding sites and food availability. (Matias-Ferrer & Escalante, 2015).

We also propose that the difference in body conditions between recovered and undisturbed sites could be influenced by the decrease of available perches with preferred diameters in recovered sites. Given that for arboreal organism such as lizards of the genus *Anolis* and raptors, the number of available perches is directly related to the feeding latency and prey capture success (Andersson, Wallander, & Isaksson, 2009; Drakeley, Lapiedra, & Kolbe, 2015). In other words, the lack of enough perches or detrimental perch's quality reduces foraging success and number of prey catch.

Predation may also influence the reduction in body condition of *C nephila*, as at disturbed habitats, due to lower plant cover, organisms might require higher energy expenditure for predator avoidance (Bradley, Werner, & Skelly, 2000; Eakin, Calhoun, & Hunter, 2019). The lack of plant cover might increase detection by predators of larger individuals thus diminishing body condition due to stress and reduced time for foraging (Sinervo et al., 2010).

C nephila at undisturbed TMCF streams, with a higher BCI, would have a greater ability to compete for available resources and survive than those of undisturbed streams. Consequently, this work suggests that anthropogenic disturbance, which extreme conditions might be potentiated during the dry season, puts these individuals under greater stress representing an imminent threat for its conservation. The three species in question showed a differential effect on the index of body condition, and we hypothesized it could be related to frog abundance at each disturbance regime.

On the other hand, greater body condition of *P. zophodes* at recovered streams than in undisturbed sites, during the dry season, might suggests that perch height differences at the sites had an impact on corporal condition of the individuals, taking account that for arboreal animals, both perch sites and the height at which they are located are important for foraging (Menin, Rossa-Feres, & Giaretta, 2005; Shuker & Hero, 2012). We observed that by *P. zophodes* on undisturbed sites perch at 75 cm of height and recovered sites used perches at higher 90 cm , this effect has already been observed in *Anolis bimaculatus,* in places where it is sympatric with *A. wattsi,* it competes for breeding sites being relegated to lower height hangers and directly affects the food volumes that this species may access, as a direct result of the perch heights being below positions that they need to feed normally (Rummel & Roughgarden, 1985).

We highlight the importance of understanding the effects of environmental disturbance on species' body condition to maintain not only abundance, but also healthy populations of frogs at managed forests. Body condition is of the utmost importance for the prevalence of viable populations, since is related to high physical status in several amphibian species, given that energy obtained from food is mainly spent on functions including foraging, reproduction, metabolism, endocrine control, and immune response (Pough, 2007; Taigen & Pough, 1983). Therefore, it can mediate several life history trade-offs related with survival and reproduction (Ferrie et al., 2014).

On the other hand, it is related to the ability to survive long periods of fasting and potential predatory pressures (Hertz, Huey, & Stevenson, 1993; Huey, 1991). Future research in our study sites should evaluate whether prey availability and diversity vary across disturbance degrees.

Body temperature

Given the strong physiological links between ectotherms and their environments, understanding the influence of environmental gradients on anurans is essential to interpret their activity and habitat use. (Hertz et al., 1993; Porter et al., 2002).

This study shows anthropogenic disturbance effects on body temperature of tree frogs. It is known that temperature rise in naturally disturbed environments (fires) opens up opportunities within the preferred temperature range, positively affecting thermoregulation of terrestrial species (*Bufo boreas*) in terms of prey abundance and efficiency of digestion (Hossack et al., 2009). However, air temperature variation that exists on anthropized environments could affect arboreal species from environments with greater thermal stability to physiological level, on locomotor performance, food digestion and absorption, given exhibit a limited variation on body temperature, and with a close relationship between body temperature and substrate temperature (Navas, Gomes, & Carvalho, 2008; Navas et al., 2013)

Our results show that changes in structural characteristics in a cloud forest result in changes in the body temperature of *C. nephila*, which presented a higher body temperature as disturbance increased. These changes are probably due to habitat transformation i.e. logging that causes a fluctuation in local temperature (Fig 5 a).

For example, at transformed sites, local temperature tends to increase during the day as a consequence of a reduction of canopy cover which allows a higher incidence of solar radiation along the brushwood to the ground level but tends to be lower at night due to wind exposure (Bakken, 1989; Chenn et al., 1999). This effect is determinant on amphibians since they have no control internal over body temperature (Huey, 1991; Raske et al., 2012). We also found that substrate temperature was closely related to body temperature, however, on disturbed sites the average body temperature was higher than average air temperature.

There are two possible scenarios, either this species is selecting microhabitats with higher temperature or due to disturbance only sites with high temperature are left. The first scenario suggests that *C. nephila* can regulate T_b by selection of available of microenvironments, choosing in this case sites with higher temperature. The second scenario suggests a reduction in availability of sites with preferred temperatures being only available structures with higher temperature as a result of disturbance (Bakken, 1989). The capacity of this species to resist both conditions might be caused by the resistance to water loss capacity that some tree frogs have , which allows them to select microhabitats with a higher temperature even at sites with lower relative humidity (Angilletta, Niewiarowski, & Navas, 2002; Wygoda, 1988; Wygoda & Williams, 1991)

Habitat disturbance did not influence the body temperature of *D. ignicolor*, which was also highly correlated to substrate temperature. This pattern is common among other species of the *Hylidae* family with arboreal and nocturnal habits that lack a thermoregulatory behavior thus presenting a body temperature influenced mainly by the environmental temperature (Snyder & Weathers, 1975; Wygoda & Williams, 1991).

Taking this into account, thermal ecology of *D. ignicolor* obeys to the general pattern presented by other tropical anurans of similar habits. Nevertheless, this study shows that the body temperature of *D. ignicolor* is related circumstantially to the substrate temperature, regardless of the air temperature.

In addition, this species did not show differences in body temperature among the perturbation gradient. Taking into account that the evaluated habitats are contrasting in terms of temperature and structure, it is possible that *D. ignicolor* is having thermoregulatory behaviours, such as changes in posture or basking, that directly affect their body temperature; these thermoregulatory behaviours are expected especially in thermally challenging environments as habitats subjected to disturbance (Row & BlouIn-Demers, 2006).

In *P zophodes,* the substrate temperature was closely related to body temperature, and similarly related to air temperature in recovered and disturbed sites. However, in undisturbed sites the body temperature differed significantly from the air temperature. Given the greater amount of microhabitats in these sites, it is also possible that this species has thermoregulatory behaviours that allowed individuals to reach higher temperatures, probably related to temperatures that some amphibians need to carry out digestion (Fontaine., Novarro, & Kevin D, 2018). However, we cannot make greater inferences by not having data on preferred (selected) temperatures (T_{set}).

We propose to explore the thermal ecology of these species in a disturbance gradient, taking data from preferred (selected) temperatures (T_{set}), operative environmental temperatures (T_e) and critical minimum and maximum temperatures (CT_{min} and CT_{max}) in order to have a broader comprehension of the environment thermal quality and the thermoregulatory accuracy of the species.

Habitat use and perch availability

The results of this study suggest that there is a differential use of perches among the three study species that coexist in the same streams. Overall for sympatric anurans to coexist in the same ponds or streams, species come to have differences in the use spatial resources (Caldwell, 1974; Donnelly & Guyer, 1994). *C. nephila*, prefers stalks and branches of shrubs and herbaceous stems basically due to its perching behaviour (where frogs hold to the branches) probably related to their larger sizes and heavier weights. In contrast, the smaller and lighter species *D. ignicolor* and P. *zophodes* used leaves and leaves or stalks respectively. This coincides with the assumption that stream dwellers tree frogs, spread space resources (perches) according to their individual sizes (Caldwell, 1974).

In addition, our results show a habitat use change of *Charadrahyla nephila*, according to the degree of disturbance. At undisturbed sites it used mostly shrubby branches as their preferred perches contrasting to disturbed sites this species used herbaceous stalks. according to what we observed at conserved sites, are reduced at disturbed sites as a direct effect of farmland management, where the shrubby substrate decreases. This pattern has already been observed in species that depend on plant structures, such as *Anolis carolinensis* for which abundance varies in response to the shrub-level vegetation density (Schaefer et al.,2015).

Likewise, we observe that this species in conserved streams preferred perches of smaller diameter (0.56 cm) and used perches of greater diameter in the recovered sites (1.24 cm) and disturbed streams (1.43 cm). The decrease of hangers with the preferred diameter is directly related to the loss of vegetal structure, derived from the farmland management in the disturbed sites, and that have a direct effect on the density of hangers that this species prefers. Which, accordingly, also directly affects the density of individuals found in the disturbed sites.

D. ignicolor and *P. zophodes* were not affected in terms of habitat use among disturbance regimes, we assume that these species were not directly affected since their small size allows them to use the leaves and branches of the shrub-herbaceous substrate, that prevails in the streams studied even at the disturbed sites where high herbaceous structures are preserved due to the management type that the community makes on streams within farmland and paddocks areas.

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Conclusiones

Este estudio demuestra que la perturbación antropogénica en dos niveles afecta diferencialmente a tres especies de ranas arborícolas del bosque mesófilo de montaña. En términos de abundancia, condición y temperatura corporal, además de uso de hábitat, las especies de estudio mostraron respuestas únicas para cada grado de perturbación antrópica. demostrando que los arroyos sometidos a diferentes grados de perturbación difieren considerablemente en su condición para servir como hábitat para estas especies.

C. nephila es una especie susceptible a la perturbación antropogénica, mostrando una disminución gradual en abundancia a medida que aumentó la perturbación. Debido a que en sitios perturbados disminuyo la cantidad recursos espaciales disponibles para la especie (perchas preferidas), además de mostrar una disminución en la condición corporal en arroyos inmersos en bosque recuperado *vs* bosque conservado, durante la estación seca, que es el período de mayor estrés para anfibios. Además, los cambios en la temperatura local incidieron en la temperatura corporal de *C. nephila* mostrando una temperatura corporal más alta a medida que aumentó la perturbación, esto como resultado de la diferencia en las características estructurales de vegetación leñosa entre regímenes de perturbación.

A pesar de que las mayores abundancias de *D. ignicolor* se observaron en arroyos del bosque recuperado, ésta no mostró ser afectada en términos de condición corporal, temperatura corporal y uso de hábitat entre los regímenes de perturbación evaluados. Aparentemente esta es una especie tolerante a perturbaciones antropogénicas ligeras en BMM con alta conectividad.

P. zophodes muestra ser una especie resiliente a la perturbación antropogénica al presentar mayores abundancias conforme aumentó la perturbación. Por otro lado, se detectó, presencia de renacuajos, individuos metamórficos y juveniles de esta especie en arroyos sometidos a perturbación lo cual muestra esta especie utiliza dichos arroyos para reproducción y refugio. Además, mostró tener mejores condiciones corporales en los arroyos adyacente a bosques recuperados que en arroyos inmersos en bosque conservado, indicativo de una mayor tasa de captura de presas en sitios recuperados. Sin embargo, la temperatura corporal de esta especie disminuyó en los sitios perturbados como resultado de las fluctuaciones en temperatura del aire en los mismos.

En general, proponemos que la variación en los patrones de abundancia de estas ranas especialistas arborícolas refleja su capacidad para tolerar eventos de perturbación a pequeña escala de manera diferencial y las variaciones en los requisitos de recursos espaciales, térmicos y de alimentación de estas especies.

Implicaciones para la conservación de anuros

Nuestros resultados indican que incluso los arroyos en áreas recuperadas y perturbadas fungen como refugios para algunos anuros nativos, debido al nivel de manejo al que está sometida la vegetación aledaña a estas corrientes. Por ejemplo, en arroyos inmersos en potreros y tierras de cultivo, los arbustos y plantas herbáceas adyacentes a las corrientes se conservan, lo que permite la permanencia de sitios de percha para ranas arborícolas útiles para realizar diferentes actividades (reproducción, termorregulación y forrajeo). Sin embargo, cabe mencionar que estas perchas no son óptimas para hylidos de tallas más grandes i.e *C. nephila*

Además, proponemos que debido al manejo de uso de la tierra que lleva a cabo la comunidad chinanteca Santa Cruz Tepetotutla, donde se realiza rotación de cultivos y se mantiene el estrato arbustivo y herbáceo adyacente a arroyos en zonas de manejo agrícola, se amortigua la pérdida de especies que incluso se consideraban extintas como *Duellmanohyla ignicolor.* Esto además permite la prevalencia de especies endémicas residentes del bosque mesófilo de montaña (*Ptychohyla zophodes*). Sin embargo, la perturbación a pequeña escala, es decir la tala del bosque mesófilo primario adyacente a arroyos, puede comprometer la prevalencia de especies más sensibles a las perturbaciones antropogénicas, como: *Charadrahyla nephila.*